

AGENDA

BOARD OF SUPERVISORS, COUNTY OF MONO STATE OF CALIFORNIA

Regular Meetings: The First, Second, and Third Tuesday of each month. Location of meeting is specified just below.

MEETING LOCATION Suite Z, 2nd Floor Minaret Mall, 437 Old Mammoth Rd., Suite Z, Mammoth Lakes, CA 93546

Regular Meeting February 21, 2017

TELECONFERENCE LOCATIONS: 1) First and Second Meetings of Each Month: Mammoth Lakes CAO Conference Room, 3rd Floor Sierra Center Mall, 452 Old Mammoth Road, Mammoth Lakes, California, 93546; 2) Third Meeting of Each Month: Mono County Courthouse, 278 Main, 2nd Floor Board Chambers, Bridgeport, CA 93517. Board Members may participate from a teleconference location. Note: Members of the public may attend the open-session portion of the meeting from a teleconference location, and may address the board during any one of the opportunities provided on the agenda under Opportunity for the Public to Address the Board. **NOTE:** In compliance with the Americans with Disabilities Act if you need special assistance to participate in this meeting, please contact the Clerk of the Board at (760) 932-5534. Notification 48 hours prior to the meeting will enable the County to make reasonable arrangements to ensure accessibility to this meeting (See 42 USCS 12132, 28CFR 35.130).

Full agenda packets are available for the public to review in the Office of the Clerk of the Board (Annex I - 74 North School Street, Bridgeport, CA 93517). Any writing distributed less than 72 hours prior to the meeting will be available for public inspection in the Office of the Clerk of the Board (Annex I - 74 North School Street, Bridgeport, CA 93517). **ON THE WEB**: You can view the upcoming agenda at http://monocounty.ca.gov. If you would like to receive an automatic copy of this agenda by email, please subscribe to the Board of Supervisors Agendas on our website at http://monocounty.ca.gov/bos.

UNLESS OTHERWISE SPECIFIED BY TIME, ITEMS SCHEDULED FOR EITHER THE MORNING OR AFTERNOON SESSIONS WILL BE HEARD ACCORDING TO AVAILABLE TIME AND PRESENCE OF INTERESTED PERSONS. PUBLIC MAY COMMENT ON AGENDA ITEMS AT THE TIME THE ITEM IS HEARD.

9:00 AM Call meeting to Order

Pledge of Allegiance

1. OPPORTUNITY FOR THE PUBLIC TO ADDRESS THE BOARD

on items of public interest that are within the subject matter jurisdiction of the Board. (Speakers may be limited in speaking time dependent upon the press of business and number of persons wishing to address the Board.)

2. APPROVAL OF MINUTES - NONE

3. RECOGNITIONS - NONE

4. BOARD MEMBER REPORTS

The Board may, if time permits, take Board Reports at any time during the meeting and not at a specific time.

5. COUNTY ADMINISTRATIVE OFFICE

CAO Report regarding Board Assignments Receive brief oral report by County Administrative Officer (CAO) regarding work activities.

6. DEPARTMENT/COMMISSION REPORTS

7. CONSENT AGENDA

(All matters on the consent agenda are to be approved on one motion unless a board member requests separate action on a specific item.)

A. Out of State Travel Request - Penny Galvin

Departments: Finance

This conference will provide Penny Galvin with enhanced knowledge on the EMS billing software ImageTrend and allow her to share her knowledge of EMS billing in order to assist in the expansion and fine tuning of the ImageTrend software.

Recommended Action: Approve out of state travel request for Penny Galvin, in order to attend ImageTrend Connect 2017 Conference in St. Paul, Minnesota, leaving July 18, 2017 and returning on July 21, 2017.

Fiscal Impact: The fiscal impact for this travel and training is approximately \$1790, which includes registration, hotel, airfare, and per diem. Finance Department has sufficient budget to cover the cost of this training.

B. Out of State Travel Request - Janet Dutcher

Departments: Finance

This annual conference features many opportunities for finance officers to hone their leadership and management skills with a chance to learn about fiscal strategies, policies and practices for managing governmental financial resources and to implement these best practices here in Mono County.

Recommended Action: Approve out of state travel request for Janet Dutcher, to attend the Government Finance Officers' Association (GFOA) annual conference being held in Denver, Colorado this year, leaving May 21 and returning May 24, 2017.

Fiscal Impact: The fiscal impact for this travel and training is approximately

\$1,520, which includes registration, hotel, airfare and per diem. The cost of this training event is included in the department's budget.

C. Out of State Travel NADCP Training Conference

Departments: Probation

(Karin Humiston) - Seeking approval for out of state travel from July 9, 2017 through July 12, 2017 for the National Association of Drug Court Professionals (NADCP) Annual Training Conference in Washington D.C. Attendees are Jon Himelhoch, Stacie Casabian and Rich Bonneau.

Recommended Action: Approve out of state travel for Probation employees Jon Himelhoch and Stacie Casabian and for Behavioral Health employee Rich Bonneau to attend the NADCP Annual Training Conference in Washington DC July 9-12, 2017 with a travel day July 8, 2017.

Fiscal Impact: No fiscal impact to the General Fund. Registration \$1,800.00; Hotel \$2,640; Airfare (currently) \$1,530; Per Diem meals \$810; Airport Parking \$70; Mileage to/from Reno Airport \$185; Taxi to/from Washington DC Airport \$50. Total \$7,085. Probation Department has budgeted this travel expense and it will be paid for from Drug Court Grant funding and SB678 Evidence Based Practices funding.

D. Planning Commission Appointments

Departments: Community Development and Board of Supervisors

Appointment of three planning commissioners to new four-year terms.

Recommended Action: 1. Reappoint Roberta Lagomarsini, with term expiring March 1, 2021, to the Mono County Planning Commission as recommended by Supervisor Stump; 2. Reappoint Daniel Roberts, with term expiring March 1, 2021, to the Mono County Planning Commission as recommended by Supervisor Gardner; 3. Reappoint Scott Bush, with term expiring March 1, 2021, to the Mono County Planning Commission as recommended by Supervisor Planning Commission as recommended by Supervis

Fiscal Impact: No impact.

E. Recruitment of EMS Chief

Departments: EMS Department

The current EMS Chief, Robert Rooks, will exceed his 960 hours allocated in his employment contract by the end of April, 2017. Therefore, recruitment for his replacement should begin as soon as possible.

Recommended Action: That the Board of Supervisors authorize the Mono County Human Resources Department to begin recruitment for a full-time EMS Chief.

Fiscal Impact: The allocated salary for this position is between \$96,000 to \$108,000 depending on qualifications and experience. The full cost of this position

with benefits will be between \$173,879 and \$192,142 for a full year.

8. CORRESPONDENCE RECEIVED - NONE

All items listed are located in the Office of the Clerk of the Board, and are available for review. Direction may be given to staff regarding, and/or the Board may discuss, any item of correspondence listed on the agenda.

9. **REGULAR AGENDA - MORNING**

A. Presentation on Sierra Nevada Bighorn Sheep

Departments: Public Works

1.5 hours (30 minute presentation; 1 hour discussion)

(CA DFW staff and USFWS staff) - Presentation by CA DFW and USFWS regarding Sierra Nevada Bighorn Sheep Recovery efforts.

Recommended Action: None (informational only). Provide any desired direction to staff.

Fiscal Impact: None.

B. Direction to Staff re Conway Ranch Request for Grazing Proposals Departments: Public Works

2.5 hours (15 minute presentation; 2.25 hour discussion)

(Tony Dublino) - Presentation by Tony Dublino regarding potential issuance by County of a Request for Proposals for Grazing at Conway Ranch.

Recommended Action: Receive presentation and provide direction to staff regarding the issuance of an RFP for grazing on Conway Ranch, including, but not limited to, one of the following options: 1. Direct staff to prepare RFP for sheep grazing at Conway and Mattly Ranch. Any such proposal will require indemnification as well as applicant funding of any necessary CEQA. Once prepared, present to Board for approval, posting and publishing. 2. Direct staff to prepare RFP for cattle grazing at Conway and Mattly Ranch. Any such proposal will require indemnification as well as applicant funding of any necessary CEQA. Once prepared, present to Board for approval, posting and publishing. 2. Direct staff to prepare RFP for cattle grazing at Conway and Mattly Ranch. Any such proposal will require indemnification as well as applicant funding of any necessary CEQA. Once prepared, present to Board for approval, posting and publishing. 3. Do not direct staff to prepare an RFP – allow current grazing lease to expire without subsequent lease in place.

Fiscal Impact: None at this time.

10. OPPORTUNITY FOR THE PUBLIC TO ADDRESS THE BOARD

on items of public interest that are within the subject matter jurisdiction of the Board. (Speakers may be limited in speaking time dependent upon the press of business and number of persons wishing to address the Board.)

11. CLOSED SESSION

A. Closed Session--Human Resources

CONFERENCE WITH LABOR NEGOTIATORS. Government Code Section 54957.6. Agency designated representative(s): Stacey Simon, Leslie Chapman, and Dave Butters. Employee Organization(s): Mono County Sheriff's Officers Association (aka Deputy Sheriff's Association), Local 39--majority representative of Mono County Public Employees (MCPE) and Deputy Probation Officers Unit (DPOU), Mono County Paramedic Rescue Association (PARA), Mono County Public Safety Officers Association (PSO), and Mono County Sheriff Department's Management Association (SO Mgmt). Unrepresented employees: All.

12. REGULAR AGENDA - AFTERNOON

A. Review of Need for Continuation of Local Emergency

Departments: CAO, Sheriff 10 minutes (5 minute presentation; 5 minute discussion)

(Leslie Chapman, Ingrid Braun) - On January 31, 2017 the Mono County Sheriff declared a state of local emergency as a result of extreme winter weather. The Board of Supervisors ratified this declaration on February 7, 2017, and further declared a continuing state of emergency. Mono County Code Section 2.60.080 requires that the Board of Supervisors review the need for continuing the local emergency every 14 days until it is terminated. This item is provided for that purpose.

Recommended Action: Review need for continuing the local emergency. If Board determines that need no longer exists, direct staff to prepare a declaration terminating local emergency.

Fiscal Impact: None

B. Mid-Year Budget Review

Departments: CAO, Finance

1 hour 20 minutes (20 minutes presentation, 60 minutes discussion)

(Leslie Chapman, Janet Dutcher) -

Receive analysis of the County's General Fund fiscal performance for the year ended June 30, 2016. Present mid-year budget review and discuss budget updates.

To view documents related to this item which are too large to attach to the agenda, please click on the link below:

http://www.monocounty.ca.gov/auditor/page/2016-17-mono-county-mid-yearbudget-review

Recommended Action: Receive analytical analysis of General Fund fiscal performance for the year ended June 30, 2016. Hear budget updates and approve the mid-year budget adjustments (4/5ths vote required). Provide any desired direction to staff.

Fiscal Impact: Increase in General Fund appropriations of \$147,347 funded with an increase in revenues of \$147,347 and the use of carryover balance of \$0. Increase in Non-General Fund appropriations of \$1,037,409 funded with an increase in revenues of \$626,948 and the use of carryover balance of \$410,461.

C. SB 844 Jail Project Proposal Package

Departments: Public Works

1 hour (15 minute presentation, 45 minute discussion)

(Garrett Higerd) -

Update on proposal for jail revenue bond funds to construct a new jail facility on the site of the old County hospital on Twin Lakes Road.

The Mono County General Plan is available

at: http://monocounty.ca.gov/planning/page/general-plan-eir

The contract documents (in template form) for the project are located at http://www.bscc.ca.gov/s_cfcformofdocuments.php. These documents would be approved as to form in the proposed resolution and then finalized and executed if the grant were awarded. County Counsel has reviewed the contracts and finds them to be legally adequate and acceptable as to form.

Recommended Action: Receive update on status of SB 844 Jail Project Proposal Package to construct a new jail facility on the site of the old County hospital on Twin Lakes Road. Approve Resolution R17-_____ authorizing application for adult detention facility construction funds under SB 844 from the Board of State and Community Corrections and adopting a General Plan EIR Addendum. Provide direction to staff.

Fiscal Impact: If Board approves submission of application, if County is awarded funds, and depending on ultimate design of project, Mono County's contribution towards this project is projected to be about \$2,053,000, which includes \$440,000 of in-kind labor for project management, construction management, transition planning and grant administration that will be required over the life of the project (the next four to five years) and a \$1,613,000 cash match of which \$203,000 has already been budgeted. We expect the remaining \$1,410,000 will come from an investment pool loan or bank line of credit having a five-year term, payable semi-annually at as low as 2.50% interest per year. Annual loan payments will be approximately \$302,000, and will cost the County \$98,750 in interest over the term of the loan. Annual loan payments could be funded through a combination of budgetary expenditure savings, expenditure deferrals and if necessary, temporary expenditure reductions.

ADJOURN



OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

💻 Print

MEETING DATE February 21, 2017

Galvin

Departments: Finance

TIME REQUIRED

SUBJECT

Out of State Travel Request - Penny

APPEARING BEFORE THE BOARD

PERSONS

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

This conference will provide Penny Galvin with enhanced knowledge on the EMS billing software ImageTrend and allow her to share her knowledge of EMS billing in order to assist in the expansion and fine tuning of the ImageTrend software.

RECOMMENDED ACTION:

Approve out of state travel request for Penny Galvin, in order to attend ImageTrend Connect 2017 Conference in St. Paul, Minnesota, leaving July 18, 2017 and returning on July 21, 2017.

FISCAL IMPACT:

The fiscal impact for this travel and training is approximately \$1790, which includes registration, hotel, airfare, and per diem. Finance Department has sufficient budget to cover the cost of this training.

CONTACT NAME: Gerald Frank

PHONE/EMAIL: 760-932-5483 / gfrank@mono.ca.gov

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR PRIOR TO 5:00 P.M. ON THE FRIDAY 32 DAYS PRECEDING THE BOARD MEETING **SEND COPIES TO:**

MINUTE ORDER REQUESTED:

T YES 🔽 NO

ATTACHMENTS:

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Staff Report

Conference Agenda

History

Time	Who	Approval
2/16/2017 12:22 PM	County Administrative Office	Yes
2/15/2017 9:46 AM	County Counsel	Yes
1/27/2017 8:19 AM	Finance	Yes



P.O. Box 495

(760) 932-5480

Fax (760) 932-5481

DEPARTMENT OF FINANCE COUNTY OF MONO

Gerald A. Frank Assistant Finance Director Treasurer-Tax Collector

Bridgeport, California 93517

Janet Dutcher, CPA, CGFM Finance Director Stephanie Butters Assistant Finance Director Auditor-Controller

P.O. Box 556 Bridgeport, California 93517 (760) 932-5490 Fax (760) 932-5491

Date:February 14, 2017To:Honorable Board of SupervisorsFrom:Finance: Janet Dutcher, Gerald Frank

Subject: Out of State Travel Request – Penny Galvin

Actions Requested:

Approve out of state travel request for Penny Galvin, in order to attend ImageTrend Connect 2017 Conference in St. Paul, Minnesota, leaving July 18, 2017 and returning on July 21, 2017.

Discussion:

The Finance Department is requesting that Penny Galvin be allowed to attend the ImageTrend Connect 2017 Conference in St. Paul, Minnesota. This is a three-day training course on the EMS Billing Bridge. This conference will provide Ms. Galvin with enhanced knowledge on the EMS billing software ImageTrend and allow her to share her knowledge of EMS billing in order to assist in the expansion and fine tuning of the ImageTrend software. This conference will also allow Ms. Galvin to connect with users from across the country to share ideas, processes, and examine key issues.

Fiscal Impact:

The fiscal impact for this travel and training is approximately \$1790, which includes registration, hotel, airfare, and per diem.

Billing Bridge Track Schedule

Billing Bridge			
Wednesday, July 19			
7-8 am	Continental Breakfast		
8 - 9 am	Welcome Address		
9-9:15 am	Vendor Break		
9:15 - 9:45	What's New in Billing Bridge		
9:45-10 am	Vendor Break		
10 am - 12 pm	Basic System Administrator Training		
12-1:30 pm	Lunch and Vendor Break		
1:30-2:30 pm	ТВА		
2:30-3 pm	Vendor Break		
3-3:30 pm	Claims in Billing Bridge		
3:30-4 pm	Vendor Break		
4-5 pm	Payment Posting in Billing Bridge		
Evening	Hooley Awards Dinner		



Thursday, July 20			
7-8 am	Continental Breakfast		
8-8:30 am	Letters and Statements		
8:30-8:45 am	Vendor Break		
8:45-9:15 am	Triggers in Billing Bridge		
9:15-9:45	Vendor Break		
9:45-10:45 am	Reporting in Billing Bridge		
10:45-11 am	Vendor Break		
11 am - 12 pm	Hooley Short		
12-1:30 pm	Lunch and Vendor Break		
1:30-2:30 pm	TBA		
2:30-3 pm	Vendor Break		
3-5 pm	Advanced System Admin Training for Billing Bridge		
Evening	Open Evening		

	Friday, July 21
7-8 am	Continental Breakfast
8-8:30 am	Closing
8:30-9 am	Break
8:45-10 am	Billing Bridge Open Discussion
10-10:15 am	Break
10:15-11:15 am	ТВА
11:15-11:30 am	Break
11:30 am-12 pm	The Future of Billing Bridge





OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

💻 Print

MEETING DATE February 21, 2017

Departments: Finance

TIME REQUIRED

SUBJECT

Out of State Travel Request - Janet Dutcher PERSONS APPEARING BEFORE THE BOARD

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

This annual conference features many opportunities for finance officers to hone their leadership and management skills with a chance to learn about fiscal strategies, policies and practices for managing governmental financial resources and to implement these best practices here in Mono County.

RECOMMENDED ACTION:

Approve out of state travel request for Janet Dutcher, to attend the Government Finance Officers' Association (GFOA) annual conference being held in Denver, Colorado this year, leaving May 21 and returning May 24, 2017.

FISCAL IMPACT:

The fiscal impact for this travel and training is approximately \$1,520, which includes registration, hotel, airfare and per diem. The cost of this training event is included in the department's budget.

CONTACT NAME: Janet Dutcher

PHONE/EMAIL: 760-932-5494 / jdutcher@mono.ca.gov

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR PRIOR TO 5:00 P.M. ON THE FRIDAY 32 DAYS PRECEDING THE BOARD MEETING SEND COPIES TO:

MINUTE ORDER REQUESTED:

🔲 YES 🔽 NO

ATTACHMENTS:

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Staff report

□ <u>2017 GFOA Conference Overview</u>

History

Time	Who	Approval
2/16/2017 6:11 AM	County Administrative Office	Yes
2/15/2017 9:43 AM	County Counsel	Yes
2/9/2017 9:17 AM	Finance	Yes



DEPARTMENT OF FINANCE AUDITOR-CONTROLLER COUNTY OF MONO

Stephanie M. Butters Assistant Finance Director Auditor-Controller Janet Dutcher, CPA, CGFM Director of Finance P.O. Box 556 Bridgeport, California 93517 (760) 932-5490 Fax (760) 932-5491

Date:	February 21, 2017
To:	Honorable Board of Supervisors
From:	Janet Dutcher, Finance Director
Subject:	Out of State Travel Request – Janet Dutcher

Action Requested:

Approve out of state travel request for Janet Dutcher, to attend the Government Finance Officers' Association (GFOA) annual conference being held in Denver, Colorado this year, leaving May 21 and returning May 24, 2017.

Discussion:

The GFOA is a professional organization representing government finance officials. The organization is dedicated to enhancing and promoting the professional management of governmental financial resources by identifying, developing and advancing fiscal strategies, policies and practices for the public benefit. The annual conference features many opportunities for finance officers to hone their leadership and management skills with a chance to share strategies, policies and practices. The conference also features sessions that focus on project management, managing teams, recruiting and identifying new employees, and how to improve communication.

The Finance Department is requesting out-of-state travel for Janet Dutcher to attend this year's GFOA annual conference scheduled for May 21-24, 2017 in Denver, Colorado. This is an opportunity to learn more about government budgeting, accounting and reporting and changing compliance requirements and to implement these best practices here in Mono County.

Fiscal Impact:

The fiscal impact for this travel and training is approximately \$1,520, which includes registration, hotel, airfare and per diem. The cost of this training event is included in the department's budget.

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Registration Information

Conference Hotels

First-time Scholarship

Mentor Program

Call for Topics

Keynote Speakers

Conference Sessions

Preconference Seminars

GFOA Social Events

Current Exhibitors

Exhibitor Information

Sponsorship Opportunities

Annual Conference FAQ

Denver Info

Tours

Future GFOA Conference Locations

Conference Brochure

- 111TH ANNUAL CONFERENCE | MAY 21-24, 2017 | DENVER, COLORADO
 Drawing on more than a century of experience, public finance professionals have widely come to view the GFOA Annual Conference as the one truly "must-attend" professional development event of the year. As in the past, the 111th GFOA Annual Conference in Denver, Colorado, will feature unparalleled opportunities for sharing ideas, sharpening skills, discovering new tools and technologies, and networking with peers from across North America and around the world.
 Why Attend?
 Soncurrent sessions. A rich array of concurrent sessions covering a broad range of topics allowing participants to tailor their conference experience to their own needs and circumstances.
 Preconference sessions. Special preconference sessions provide
- an opportunity to explore selected topics of special interest in even greater depth.
- General sessions. Experience direct contact with nationally recognized speakers of special interest to public finance professionals.



- Discussion groups. Meet peers and discuss topics relevant to specific types of organizations, such as school districts, utilities, large governments, small governments, and the next generation of finance officers.
- > Networking opportunities. Make the contacts you need to build new relationships and cement existing relationships.
- Social events. Wind down from a busy day and connect with colleagues during GFOA's Welcome Reception and Closing Event.
- > CPE credit. Earn more than 20 CPE credits, most of it directly relevant to government, with even more credits available for those who elect to participate in preconference sessions.
- Exhibit hall. GFOA's exhibit hall will put participants in contact with vendors that offer practical tools and solutions for a broad range of professional challenges. Stroll through the hall to view new products, ask questions, and see live demonstrations of services that can save your government time and money. Join us on Sunday morning to kick off this year's hall with music and a ribbon cutting ceremony! Watch for new events in the exhibit hall!

Registration

Registration is now open

First-Time Annual Conference Attendee Scholarship:

GFOA's Executive Board will be awarding fifty scholarships per state or province to first-time annual conference attendees who are GFOA active (government) members. The scholarship waives the conference registration fee to give those individuals who have never attended a GFOA conference an opportunity to do so. For information on how to apply for a first-time conference attendee scholarship, please e-mail First Annual Conference Scholarship.

Schedule of Events

- > Preconference Seminars: May 19-20, 2017
- Concurrent Sessions and Networking Groups: May 21-24, 2017
- Welcome Reception: May 21, 2017
- Exhibits: May 21-23, 2017
- Denver Rocks! Closing Event: May 23, 2017

The above events (unless otherwise noted) will be held at the Colorado Convention Center, 700 14th Street, Denver, Colorado 80202. Concurrent sessions will begin at 1:30 p.m. on Sunday and conclude at 12:10 p.m. on Wednesday.

Hotels

Reserve your room in GFOA's official hotel block and you'll have a great opportunity to network with other conference attendees outside the Colorado Convention Center.

Experient will be the official housing company for this event. Booking through GFOA's official housing provider will ensure that attendees receive the GFOA discounted rate and are working with a reputable company. Click here to read more about hotels and reserve your room.

See You in Denver!

World-class attractions. Natural wonders. A thriving arts scene. Dozens of innovative and acclaimed restaurants. Nightlife options galore. The word is getting out - Denver's got it all. While you're here, you owe it to yourself to discover the best that the Mile High City has to offer. Check out some customized itineraries, explore a few locals' favorites and transform your time in Denver into a vacation you'll never forget.

For more information on Denver, please visit www.denver.org/gfoa/



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OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST			
	E Print		
MEETING DATE	February 21, 2017		

Departments: Probation

TIME REQUIRED

SUBJECT

Out of State Travel NADCP Training Conference PERSONS APPEARING BEFORE THE BOARD Karin Humiston

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

Seeking approval for out of state travel from July 9, 2017 through July 12, 2017 for the National Association of Drug Court Professionals (NADCP) Annual Training Conference in Washington D.C. Attendees are Jon Himelhoch, Stacie Casabian and Rich Bonneau.

RECOMMENDED ACTION:

Approve out of state travel for Probation employees Jon Himelhoch and Stacie Casabian and for Behavioral Health employee Rich Bonneau to attend the NADCP Annual Training Conference in Washington DC July 9-12, 2017 with a travel day July 8, 2017.

FISCAL IMPACT:

No fiscal impact to the General Fund. Registration \$1,800.00; Hotel \$2,640; Airfare (currently) \$1,530; Per Diem meals \$810; Airport Parking \$70; Mileage to/from Reno Airport \$185; Taxi to/from Washington DC Airport \$50. Total \$7,085. Probation Department has budgeted this travel expense and it will be paid for from Drug Court Grant funding and SB678 Evidence Based Practices funding.

CONTACT NAME: Karin Humiston

PHONE/EMAIL: 760-932-5570 / khumiston@mono.ca.gov

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR **PRIOR TO 5:00 P.M. ON THE FRIDAY** 32 DAYS PRECEDING THE BOARD MEETING SEND COPIES TO:

MINUTE ORDER REQUESTED:

🔽 YES 🗖 NO

ATTACHMENTS:

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History		
Time	Who	Approval
2/16/2017 4:53 AM	County Administrative Office	Yes
2/15/2017 9:42 AM	County Counsel	Yes
2/14/2017 9:39 AM	Finance	Yes



Mailing: P.O. Box 596, Bridgeport, California 93517 Bridgeport office (760) 932-5570•Fax (760) 932-5571 MAMMOTH Office (760) 924 1730•Fax (760) 924-1731

probation@monocagov

Mark Magit Presiding Judge Superior Court

D r.K ann H um iston Chief Probation O fficer

TO: Mono County Board of Supervisors

FROM: Karin Humiston, Chief of Probation

Date: February 2, 2017

SUBJECT:

Approve out of state travel for Drug Court Team employees, Jon Himelhoch, Stacie Casabian and Rich Bonneau, to attend the National Association of Drug Court Professionals (NADCP) Annual Training Conference in Washington DC, July 9-12, 2017. July 8, 2017 is a travel day.

DISCUSSION:

Mono County implemented the collaborative court, Drug Court, in fiscal year 15/16. Drug Court team members are mandated to attend training within this Evidence Based Practice program. The Drug Court grant is a federal grant. Agreement to receive these monies includes a commitment to attend training and improve the program through Evidence Based Practices. Further, Mono County Probation Officers are under a state mandate to obtain Standards Training for Corrections (STC) training. Finally, our Drug Court was audited several months ago by subject matter experts with American University. The need for training was emphasized and required.

FISCAL IMPACT:

There is no fiscal impact to the General Fund. We will receive a \$100 per registrant discount for being members of the NADCP. Registration \$1,800; Hotel \$2,640; Airfare (at current prices) \$1,530; Per Diem meals \$810; Airport Parking \$70; Mileage to/from Reno Airport \$185; Taxi to/from Washington Airport \$50. Total \$7,085. The Probation Department has budgeted this in our travel expense account and it will be paid for from the Drug Court Grant funding and SB678 Evidence Based Practices funding.



OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

💻 Print

MEETING DATE February 21, 2017

Departments: Community Development and Board of Supervisors

TIME REQUIRED		PERSONS
SUBJECT	Planning Commission Appointments	APPEARING BEFORE THE
		DUARU

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

Appointment of three planning commissioners to new four-year terms.

RECOMMENDED ACTION:

1. Reappoint Roberta Lagomarsini, with term expiring March 1, 2021, to the Mono County Planning Commission as recommended by Supervisor Stump; 2. Reappoint Daniel Roberts, with term expiring March 1, 2021, to the Mono County Planning Commission as recommended by Supervisor Gardner; 3. Reappoint Scott Bush, with term expiring March 1, 2021, to the Mono County Planning Commission as recommended by Supervisor Planning Commission

FISCAL IMPACT:

No impact.

CONTACT NAME: Scott Burns

PHONE/EMAIL: 924.1807 / sburns@mono.ca.gov

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR **PRIOR TO 5:00 P.M. ON THE FRIDAY** 32 DAYS PRECEDING THE BOARD MEETING SEND COPIES TO:

MINUTE ORDER REQUESTED:

🗖 YES 🔽 NO

ATTACHMENTS:

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b <u>staff report</u>

History

Time	Who	Approval
2/16/2017 4:52 PM	County Administrative Office	Yes
2/16/2017 4:52 PM	County Counsel	Yes
2/16/2017 4:52 PM	Finance	Yes

Mono County Community Development Department

P.O. Box 347 Mammoth Lakes, CA 93546 (760) 924-1800, fax 924-1801 www.monocounty.ca.gov P.O. Box 8 Bridgeport, CA 93517 (760) 932-5420, fax 932-5431 www.monocounty.ca.gov

February 21, 2017

To: Honorable Mono County Board of Supervisors

- From: Scott Burns, Director
- **RE:** Planning Commission Reappointments

RECOMMENDATION:

- 1. Reappoint Roberta Lagomarsini, with term expiring March 1, 2021, to the Mono County Planning Commission as recommended by Supervisor Stump;
- 2. Reappoint Daniel Roberts, with term expiring March 1, 2021, to the Mono County Planning Commission as recommended by Supervisor Gardner; and
- 3. Reappoint Scott Bush, with term expiring March 1, 2021, to the Mono County Planning Commission as recommended by Supervisor Peters.

FISCAL IMPACT:

No impact beyond budgeted expenses.

BACKGROUND:

The Mono County Planning Commission consists of five commissioners appointed by the Board of Supervisors, with each supervisor entitled to nominate one commissioner. The term of each commissioner expires March 1st following the date of the nominating supervisor's term expiration. The Planning Commission currently has three seats with terms expiring March 1, 2017.

On the prior recommendation of District 2 Supervisor Stump, Roberta Lagomarsini was appointed by the Board of Supervisors last fall to fill the commissioner's seat previously vacated by Rodger Thompson, with the four-year term expiring March 1, 2017. Supervisor Stump requests that she be reappointed to a new four-year term.

Daniel Roberts, current Planning Commission Vice-Chair, was last appointed by Supervisor Alpers. Supervisor Gardner has nominated Commissioner Roberts for reappointment to a new four-year term.

Similarly, Commissioner Scott Bush was previously appointed by Supervisor Fesko. Supervisor Peters has nominated Scott Bush for reappointment to a new four-year term.

The attached Mono County Code chapter provides further explanation of Planning Commission purpose, composition and duties. Each Board member recommending a planning commission appointment was consulted individually and independently by staff, in compliance with public meeting laws.

Please call Scott Burns at 924.1807 if you have questions.

ATTACHMENT: Mono County Code Chapter 2.36 Excerpt

Mono County Code Excerpt

Chapter 2.36 - PLANNING COMMISSION

2.36.010 - Creation of planning commission.

The Mono County planning commission is created to advise the board of supervisors and planning department and otherwise take such actions as are authorized or required by law. (Ord. 96-01 § 1 (part), 1996.)

2.36.020 - Membership—Terms—Vacancies.

A. The planning commission consists of five members appointed by the board of supervisors, who shall be eligible voters of Mono County. Each supervisor shall be entitled to nominate one commission member.

B. The term of each member appointed after the effective date of this section shall expire on March 1st following the date of the expiration of the term of the nominating supervisor.

C. Vacancies shall be filled by appointment for the unexpired portion of the term.

D. Members of the planning commission may be removed by a majority of the board of supervisors for the following reasons:

1. Failing to meet the following attendance requirements: a commissioner shall not have three consecutive unexcused absences for regular meetings, nor may a commissioner miss five or more regular meetings in any twelve-month period;

2. Acting inappropriately, in the board's opinion, in matters regarding conflict of interest;

3. Failing to carry out commissioner duties over a period of time due to a frequent inability to vote, caused by repeated conflict of interest issues;

4. Failing to carry out the duties of commissioner by abstaining on issues when there are no apparent conflict of interest issues;

5. Other enumerated causes which, in the opinion of a majority of the board, are reflected in the commissioner's failure to carry out the duties of the commission, or bringing discredit to the county of Mono. (Ord. 07-01 § 1, 2007; Ord. 96-01 § 1 (part), 1996.)

2.36.060 - Duties.

A. The planning commission shall have such duties and take such actions as are required by this code, assigned by the board of supervisors or otherwise required by law.

B. The planning commission shall act as the principal advisory body to the board of supervisors on planning matters. (Ord. 96-01 § 1 (part), 1996.)



OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

💻 Print

MEETING DATE February 21, 2017

Departments: EMS Department

TIME REQUIRED

SUBJECT Recruitment of EMS Chief

PERSONS APPEARING BEFORE THE BOARD

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

The current EMS Chief, Robert Rooks, will exceed his 960 hours allocated in his employment contract by the end of April, 2017. Therefore, recruitment for his replacement should begin as soon as possible.

RECOMMENDED ACTION:

That the Board of Supervisors authorize the Mono County Human Resources Department to begin recruitment for a full-time EMS Chief.

FISCAL IMPACT:

The allocated salary for this position is between \$96,000 to \$108,000 depending on qualifications and experience. The full cost of this position with benefits will be between \$173,879 and \$192,142 for a full year.

CONTACT NAME: Lynda Saldico

PHONE/EMAIL: 7609241842 / Isalcido@mono.ca.gov

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR **PRIOR TO 5:00 P.M. ON THE FRIDAY** 32 DAYS PRECEDING THE BOARD MEETING SEND COPIES TO:

MINUTE ORDER REQUESTED:

🗖 YES 🔽 NO

ATTACHMENTS:

Click to download

Staff Report

History

Time	Who	Approval
2/16/2017 12:24 PM	County Administrative Office	Yes
2/16/2017 11:05 AM	County Counsel	Yes
2/16/2017 8:48 AM	Finance	Yes

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DIVISION OF EMERGENCY MEDICAL SERVICES P.O. BOX 3329 Mammoth Lakes, CA 93546 (760) 924-1832 Fax (760) 924-1831



February 21, 2017

TO: Honorable Board of Supervisors

FROM: Lynda Salcido, Public Health/EMS Director

SUBJECT: Recruitment for EMS Chief

RECOMMENDED ACTION:

That the Board of Supervisors authorize the Mono County Human Resources Department to begin recruitment for a permanent EMS Chief.

DISCUSSION:

On July 5th, 2016, the County entered into an employment agreement with Robert Rooks, a retired annuitant within the PERS system, to serve as the temporary EMS Chief. This temporary position allowed Chief Rooks, an experienced EMS administrator, to assist the County in determining what next steps should be taken to implement the changes recommended in the EMS Ad Hoc Report as reported to the Board previously. He is currently conducting and facilitating discussions with the Board to ensure that they may guide and direct the future of this vital program, representing the interests of their constituents and visitors to Mono County. Chief Rooks also assumed day to day operational oversight of the program, assured compliance with all State regulations, oversaw staff, monitored expenditures and budget and coordinated efforts to ensure services to the Tri Valley.

This agreement allowed for a maximum of 960 hours per fiscal year. Chief Rooks hours for fiscal year 2016/17 will completed by approximately the end of April, 2017. It is imperative that recruiting for a permanent EMS Chief begin as soon as possible.

FISCAL IMPACT:

The allocated salary for this position is between \$96,000 to \$108,000 depending on qualifications and experience. The full cost of this position with benefits will be between \$173,879 and \$192,142 for a full year.

For questions regarding this item, please call Lynda Salcido at 924-1842.

Thank you.



OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

🖃 Print

MEETING DATE February 21, 2017

Departments: Public Works

TIME REQUIRED	1.5 hours (30 minute presentation; 1 hour discussion)	PERSONS APPEARING
SUBJECT	Presentation on Sierra Nevada Bighorn Sheep	BEFORE THE BOARD

CA DFW staff and USFWS staff

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

Presentation by CA DFW and USFWS regarding Sierra Nevada Bighorn Sheep Recovery efforts.

RECOMMENDED ACTION:

None (informational only). Provide any desired direction to staff.

FISCAL IMPACT:

None.

CONTACT NAME: tdublino@mono.ca.gov

PHONE/EMAIL: 760.932.5453 / tdublino@mono.ca.gov

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR PRIOR TO 5:00 P.M. ON THE FRIDAY 32 DAYS PRECEDING THE BOARD MEETING **SEND COPIES TO:**

MINUTE ORDER REQUESTED:

🗌 YES 🔽 NO

ATTACHMENTS:

Click to download

CDFW letter

Report2014-2015

Besser Pneumonia

Buchalski Genetic

Wehausen et al

D <u>Theisen letter - Reno Fish and Wildlife</u>

History		
Time	Who	Approval
2/16/2017 4:02 PM	County Administrative Office	Yes
2/16/2017 2:16 PM	County Counsel	Yes
2/16/2017 2:24 PM	Finance	Yes



State of California – Natural Resources Agency DEPARTMENT OF FISH AND WILDLIFE Inland Deserts Region 787 North Main St., Suite 220 Bishop, CA 93514 Phone: 760-873-4305 Fax: 760-872-1284 Email: tom.stephenson@wildlife.ca.gov EDMUND G. BROWN JR., Governor CHARLTON H. BONHAM, Director



February 15, 2017

Board of Supervisors Mono County PO Box 715 Bridgeport, California 93517

Dear Mono County Supervisors:

The California Department of Fish and Wildlife (CDFW) has jurisdiction over the conservation, protection, and management of fish, wildlife, native plants and habitat necessary for biologically sustainable populations of such species. In that capacity, CDFW administers the California Endangered Species Act (CESA), as well as other provisions of the California Fish and Game Code that afford protection to the State's fish and wildlife resources. Furthermore, CDFW is the lead agency for implementation of recovery efforts for federally endangered Sierra Nevada bighorn sheep. It is the goal and responsibility of CDFW to protect and maintain viable populations of fish and wildlife resources throughout the State. The purpose of this letter is to clarify the risk to bighorn sheep posed by the grazing of domestic sheep.

Historically there were thousands of bighorn sheep in the Sierra Nevada but their numbers declined dramatically with the arrival of European settlers in the mid-1800s and the domestic sheep they grazed throughout the Sierra¹. Domestic sheep brought diseases to which bighorn have no immunity. By the 1980s there were less than 300 bighorn in the Sierra Nevada and by the 1990s they had declined to just over 100. Consequently, Sierra Nevada bighorn were listed as State and federally endangered in 1999. CDFW has led the implementation of recovery efforts since that time and the population has grown to more than 600 bighorn.

Recent genetic analyses have confirmed that Sierra bighorn are one of three unique subspecies of bighorn sheep in North America². Bighorn sheep are adapted to the extreme winter conditions experienced in the Sierra Nevada and are able to survive the heavy snowfall in Mono County and Yosemite National Park by wintering on alpine ridges where high winds scour away the snow. In fact, recent analyses indicate that individuals that spend their winters high in the alpine may be more successful at recruiting lambs than individuals that use lower elevation winter ranges³.

Along with CDFW, numerous cooperating agencies have worked diligently and spent millions of dollars to implement recovery actions. Sierra bighorn are approaching the numerical and geographic goals for downlisting from endangered to threatened status under the Endangered Species Act. An additional goal that must be met for downlisting is the management of domestic sheep to prevent contact with bighorn such that the risk of disease transmission is eliminated. CDFW is concerned that Mono County's continued grazing of domestic sheep on Conway ranch will prevent downlisting of Sierra bighorn. The biggest threat to Sierra bighorn is disease from domestic sheep and goats, as disease can have dramatic and prolonged detrimental population effects on bighorn sheep⁴. In this letter we will summarize the evidence

Conserving California's Wildlife Since 1870

linking domestic sheep to disease in bighorn sheep, the likely consequences if a disease outbreak were to occur, and why Conway Ranch (including both Conway and Mattly parcels) are of particular concern.

Domestic sheep transmit fatal disease to bighorn sheep

Many experimental⁵⁻⁷ and case studies⁸⁻¹⁰ have shown a direct link between domestic sheep contact with bighorn sheep and pneumonia outbreaks. The results of 11 experimental studies in which bighorn sheep were penned with domestic sheep was almost always bighorn death (98% mortality of 90 bighorn), while the domestic sheep remained healthy⁷. Research on the respiratory disease responsible for these deaths has been challenging because pneumonia outbreaks often involve multiple pathogens and some bacteria can be hard to sample⁷. There is definitive evidence, however, that the bacteria *Mannheimia haemolytica*, often associated with pneumonia in bighorn sheep¹¹, has been transferred from domestic sheep to bighorn sheep^{6,12}. This was done by genetically tagging bacteria in domestic sheep and then locating those tagged bacteria in bighorn sheep during comingling experiments⁶. In addition, disease pathogens (including *Mycoplasma ovipneumoniae*) have been transmitted between animals in separate pens spaced 25-39 feet apart¹³.

Respiratory disease outbreaks in bighorn sheep are often linked to exposure or interaction with domestic sheep. Summary of bighorn declines and die-offs include eleven cases of pneumonia that began after contact with domestic sheep and resulted in 50-100% of the population dying^{8,10}. These die-offs occurred across the west: British Columbia, Oregon, Washington, South Dakota, Nevada, Colorado, New Mexico, and California (Warner Mountains). In addition, bighorn and domestic sheep interactions were confirmed prior to two outbreaks in Montana with 68-88% mortality rates⁹. These represent only a subset of documented pneumonia outbreaks in which a connection to domestic sheep can be made.

Disease outbreaks in bighorn are often catastrophic, initially causing all age die-offs followed by long term reduced lamb recruitment¹⁴. Careful and long term study of the pneumonia outbreak in the Hell's Canyon region, which includes 14 populations of bighorn sheep has found that for decades after an outbreak most or all lambs die due to continued reoccurrence of pneumonia¹⁴. This persistent reduction in recruitment has already shifted the age structure of the population to older animals and the infected populations are projected to go locally extinct in time without intervention¹⁵.

Wildlife managers and professionals throughout the west recognize that disease can be transmitted from domestic to bighorn sheep and that domestic and bighorn sheep should be effectively separated. These concepts are clearly demonstrated in the Desert Bighorn Council's *Guidelines for Management of Domestic Sheep*¹⁰, the Western Association of Fish and Wildlife Agencies' *Recommendations for domestic sheep and goat management in wild sheep habitat*¹⁶, and The Wildlife Society and American Association of Wildlife Veterinarians Joint Issue Statement, *Domestic sheep and goats disease transmission risk to wild sheep*¹⁷. Recently, the Montana Wool Growers Association and the Montana Wild Sheep Foundation agreed that at

this time "the best method to prevent disease transmission is effective separation in time and space"¹⁸. In addition, the Bureau of Land Management (BLM) has guidelines¹⁹ and both the BLM and the US Forest Service²⁰ have issued decisions directed at effectively separating domestic and bighorn sheep.

Conway Ranch threatens Mt. Warren Herd

The Department has repeatedly expressed concern to Mono County regarding domestic sheep grazing at Conway and Mattly Ranches (Letters submitted June 5, 2014, October 14, 2014, September 29, 2015, and during numerous County meetings).

Specifically Conway Ranch is of concern because of the close proximity to the occupied Mt. Warren herd. This map includes use by 9 animals within 1 mile of Conway Ranch. The striking of ram S21 by a car on 395 indicates the area near Conway summit has been perceived by Sierra bighorn as habitat and that the highway is not a barrier to bighorn sheep movement.

Location	Date	Method	Animal Description
NW corner of Conway	11/30/03	Ground	1 ram (S21) hit by car on 395
West of Mattly	12/8/03	Ground	1 ram (S21) mortality location
Within 1 mile	2005-2013	Collar	8 collared rams (S20, S44, S69, S105,
		&	S122, S156, S158, S239), 1 uncollared
		Ground	ram

As part of a risk assessment process, directed by a subgroup of the Sierra Nevada bighorn sheep recovery team, CDFW developed a quantitative spatial model of risk that predicts the likelihood of Sierra bighorn movement onto domestic sheep allotments within 37 miles (60 km) of occupied bighorn core habitat.^{21,22} This approach uses habitat suitability and distance to estimate the likelihood that Sierra bighorn will move beyond their core habitat to areas where domestic sheep are grazed. Based on known Sierra bighorn movements, these analyses demonstrate that Conway Ranch falls within a zone of high potential for contact.^{21,22} Because of the risk of contact and disease transmission, domestic sheep grazing no longer occurs on federal land within the high risk zone for Sierra bighorn. In addition, if domestic sheep were to spread disease into bighorn occupying adjacent habitat, it would likely spread to other nearby herds in the Sierra and reduce population viability and prospects for recovery.²³



Separation is the key to minimizing disease threat

Due to the very close proximity of Conway Ranch to known bighorn use, and propensity of both Sierra bighorn and domestic sheep to wander, minimization measures such as increased use of guard and herd dogs or fencing are inadequate in creating effective separation. Both male and female Sierra bighorn are known to make forays throughout the year. In addition to a general increase in ram movements during the rut (October-December), GPS collared ewes have made long distance forays during all seasons (table includes notable GPS collared ewe movements during the last 5 years).

Ewe ID	Herd Unit	Notable Movement	Timing	Described in Annual Report
S166	Mt. Williamson	Movements throughout Mt. Baxter, Mt. Williamson, and Mt. Langley herd units	Year round	2012, 2013, 2014
S167	Mt. Baxter	Moved south and west outside of are generally used by Mt. Baxter herd	Year round	2012, 2013, 2014
S89	Mt. Warren	Migration between Lundy Canyon and Camiaca Peak	November 2013, repeatedly May- September 2014	2014
S240, S241, S242, S243, S244	Wheeler	Expansion into granite park	Summer 2013	2014

Domestic sheep are also known to stray in small groups, particularly in response to predators²⁴. We have observed small groups of domestic sheep that wandered away from their herd without knowledge of the herder. In addition, there is no scientific literature showing the effectiveness of herding or guard dogs in preventing bighorn sheep from coming in to contact with domestic sheep.²²

Pneumonia transmission from domestic to bighorn sheep does not require nose to nose contact. Disease transmission that later resulted in bighorn mortality, has been documented between animals in pens 25-39 feet apart¹³. Viable pneumonia bacteria has persisted after traveling airborne more than 60 feet²⁵. Aerosol transmission of pneumonia bacteria indicates the need for double fencing to reduce the potential for disease transmission. However, at this time there is no tested standard of fencing structure and spacing that is known to prevent disease transmission. In addition, double fencing that is robust enough to keep domestic lambs from

escaping and high enough to keep bighorn sheep from clearing, would prohibit movement of other wildlife use at Conway Ranch including mule deer and sage grouse.

Mono County has received considerable conservation recognition for their role in protecting the Bi-State sage grouse population. We encourage the County to exhibit comparable leadership in the conservation of Sierra bighorn. We hope Mono County will take advantage of the opportunity to join in the significant efforts on behalf of Sierra bighorn recovery. We appreciate the opportunity to express our concerns about Conway Ranch and look forward to answering guestions and sharing more information with you on February 21, 2017.

If you have any questions or concerns regarding wildlife issues, please contact me at (760) 937-0238 or tom.stephenson@wildlife.ca.gov.

Sincerely,

Tom Stephenson, Ph.D. Program Leader for Sierra Nevada Bighorn Sheep Recovery

cc: Alisa Ellsworth, CDFW Environmental Scientist Lacey Greene, CDFW Environmental Scientist David Elms, CDFW Environmental Program Manager Leslie MacNair, CDFW Regional Manager Erin Nordin, USFWS Carolyn Swed, USFWS

References (Many of these are available at http://www.dfg.ca.gov/snbs/Literature.html)

- 1. Buechner, H. K. The bighorn sheep in the United States: its past, present, and future. *Wildl. Monogr.* **4**, 174 (1960).
- Buchalski, M. R. et al. Phylogeographic and population genetic structure of bighorn sheep (Ovis canadensis) in North American deserts. J. Mammal. 1–16 (2016). doi:10.1093/jmammal/gyw011
- 3. Spitz, D. B. Causes and consequences of migratory behavior in Sierra Nevada bighorn sheep. (2015).
- 4. U.S. Fish and Wildlife Service. Recovery Plan for the Sierra Nevada Bighorn Sheep. 215 (2007).
- 5. Wehausen, J., Kelley, S. & Ramey II, R. Domestic sheep, bighorn sheep, and respiratory disease: a review of the experimental evidence. *Calif. Fish Game* **97**, 7–24 (2011).
- 6. Lawrence, P. K. *et al.* Transmission of Mannheimia haemolytica from domestic sheep (Ovis aries) to bighorn sheep (Ovis canadensis): unequivocal demonstration with green fluorescent protein-tagged organisms. *J. Wildl. Dis.* **46**, 706–717 (2010).
- 7. Besser, T. E. *et al.* Survival of Bighorn Sheep (Ovis canadensis) Commingled with Domestic Sheep (Ovis aries) in the Absence of Mycoplasma ovipneumoniae. *J. Wildl. Dis.* **48**, 168–172 (2012).
- 8. Cassaigne G, I., Medellín, R. a & Guasco O, J. a. Mortality during epizootics in bighorn sheep: effects of initial population size and cause. *J. Wildl. Dis.* **46**, 763–771 (2010).
- 9. Howard, T. A. Comparison of wild-domestic sheep interaction policies in bighorn disease outbreak locations in the continental U.S., 1990-2010. (2013).
- 10. Staff, T. Guidlines for management of domestic sheep in the vicinity of desert bighorn habitrat. *Desert Bighorn Counc. Trans.* (1990).
- 11. Bemani, E., Esmaeilzadeh, S., Gharibi, D. & Ghorbanpoor, M. Immunohistochemical and bacteriological investigations of Mannheimia haemolytica in sheep bronchopneumonia. *Kafkas Univ. Vet. Fak. Derg.* **23**, 1–8 (2016).
- 12. Lawrence, P. K. *et al.* CORRECTION: Transmission of Mannheimia haemolytica from domestic sheep (Ovis aries) to bighorn sheep (Ovis canadensis): unequiovocal demonstration with green fluorescent protein-tagged organisms. *J. Wildl. Dis.* **46**, 1346–1347 (2010).
- 13. Besser, T. E. *et al.* Epizootic pneumonia of bighorn sheep following experimental exposure to Mycoplasma ovipneumoniae. *PLoS One* **9**, (2014).
- 14. Besser, T. E. *et al.* Causes of pneumonia epizootics among bighorn sheep, Western United States, 2008-2010. *Emerg. Infect. Dis.* **18**, 406–414 (2012).
- 15. Manlove, K., Frances Cassirer, E., Cross, P. C., Plowright, R. K. & Hudson, P. J. Disease introduction is associated with a phase transition in bighorn sheep demographics. *Ecology* **97**, 2593–2602 (2016).
- 16. Wild Sheep Working Group. Recommendations for Domestic Sheep and Goat Management in Wild Sheep Habitat. 28 (2012).
- 17. The Wildlife Society & American Association of Wildlife Veterinarians. *Domestic sheep* and goats disease transmission risk to wild sheep. (2015).
- 18. Solan, Brian, M. & McEwen, Dave, M. Montana Wool Growers Association and Montana Wild Sheep Foundation cooperative efforts letter. (2016).
- 19. Ellis, S. A. US BLM Management of Domestic Sheep and Goats to Sustain Wild Sheep. **2**, (2016).
- 20. Rainville, Suzanne C., U. Record of decision for the: Final supplemetnal environmental impact statement and forest plan amendment identifying suitable rangeland for domestic sheep and goat grazing to maintain habitat for viable bighorn sheep populations, Payette National Forest. (2010).
- 21. Baumer, A. et al. A process for identifying and managing risk of contact between Sierra Nevada bighorn sheep and domestic sheep. (2009).
- 22. Croft, B. *et al.* A process for identifying and managing risk of contact between Sierra Nevada Bighorn Sheep and domestic sheep. (2010).
- 23. Clifford, D. L. *et al.* Assessing disease risk at the wildlife-livestock interface: A study of Sierra Nevada bighorn sheep. *Biol. Conserv.* **142**, 2559–2568 (2009).
- 24. CDFW. The efficacy of measures to minimize contact between domestic sheep and Sierra Nevada Bighorn Sheep on the Humboldt-Toiyabe National Forest, Mono County, California. (2006).
- 25. Dixon, D. M. et al. Viability of air-borne Pasteurella spp. 6–13 (2002).



2014-2015 Annual Report

of the Sierra Nevada Bighorn Sheep Recovery Program

Julia M. Runcie, Alexandra P. Few, David W. German, John D. Wehausen, and Thomas R. Stephenson

Summary of Progress

This report documents conservation and monitoring activities carried out between May 1, 2014 and April 30, 2015 by California Department of Fish and Wildlife's (CDFW) Sierra Nevada Bighorn Sheep Recovery Program (the Recovery Program). The Recovery Program works to return the population of Sierra bighorn Nevada sheep (Ovis canadensis sierrae; hereafter Sierra bighorn) to a stable level through adaptive management based on an understanding of their distribution and demographics following the guidelines established bv the Recovery Plan for Sierra Nevada Bighorn Sheep (the Recovery Plan, USFW 2007). Chief among the Recovery Program's activities are regular population counts, causespecific mortality investigations, habitat and demographic modeling, captures to deploy radio collars, and translocations increase to the distribution of bighorn throughout the range.



Figure 1. Distribution of Sierra bighorn herd units, April 30, 2015. All herd units considered essential for recovery are occupied.

As a result of the translocations completed in March and April 2015 and the recent natural colonization of the Taboose Creek herd unit, Sierra bighorn have now met the distribution requirements identified in the Recovery Plan, occupying 14 herd units (Figure 1). Survey data from this season indicate that there are now at least 288 adult and yearling ewes in the Sierra; the Recovery Plan identifies a minimum target population size of 305 females distributed among 4 recovery units. We project that the Sierra bighorn population may reach all demographic criteria required for downlisting of the species within the next 5 years.

Conservation Activities

Translocations

In March and April 2015, we captured and translocated 31 animals, reintroducing herds to the Laurel Creek area of the Kern Recovery Unit in Sequoia National Park and to the Cathedral Range of the Northern Recovery Unit in Yosemite National Park. A new deme in the Mt. Gibbs herd unit was created, and the Olancha Peak herd unit was augmented.

The Kern Recovery Unit is the most remote area currently occupied by Sierra bighorn; though limited connectivity exists with the Olancha Peak and Mt. Langley herds, we expect that this recovery unit could serve as a refuge for Sierra bighorn in the event of a disease outbreak in the more connected herds along the Sierra Crest. We reintroduced bighorn to the Big Arroyo drainage of the Kern Recovery Unit in March 2014; the recent addition of 7 ewes and 4 rams to the Laurel Creek drainage will speed the growth and increase the genetic diversity present in this recovery unit.

The Northern Recovery Unit contains two of the smallest herds in the Sierra (Mt. Gibbs and Mt. Warren). Both have exhibited slow population growth, and the Mt. Warren herd has recently experienced a high rate of mortality (Few et al. 2013, Runcie et al. 2014). In September 2012, biologists from the Recovery Program and Yosemite National Park began discussing the possibility that the Northern Recovery Unit may require an additional herd to reach its recovery goal of 50 females. The Washburn Lake area of the Merced drainage and the adjacent Cathedral Range were identified as suitable habitat (Few et al. 2015), and in March and April 2015 we introduced 10 ewes and 3 rams to this area to initiate the Cathedral Range herd. We also augmented the Mt. Gibbs herd with 5 collared ewes known to have high genetic diversity; these ewes were placed in the Alger Creek drainage below Mt. Wood, an area of high-quality habitat south of the currentlyoccupied Mt. Gibbs range.

The Olancha Peak herd unit was created in March 2013 with 10 ewes and 4 rams, and augmented with an additional 4 ewes in March 2014. Two of the rams introduced to the herd have since died. In March 2015 we captured 2 high-heterozygosity rams from the Mt. Baxter herd unit and translocated them to Olancha Peak in an effort to maintain high genetic diversity and reproductive success within this herd unit.

Disease Management

Domestic sheep and goats carry respiratory pathogens that can cause fatal pneumonia when transmitted to wild bighorn (Lawrence et al. 2010, Wehausen et al. 2011). The only effective means to prevent disease transmission is to prevent contact by maintaining separation both in time and space (Wild Sheep Working Group 2012). Domestic sheep grazing that occurs in proximity to bighorn habitat can pose a significant threat to Sierra bighorn recovery, and the Recovery Plan stipulates that measures to prevent contact must be implemented and be successful before the subspecies can be

downlisted (USFWS 2007). For decades, CDFW has worked closely with land management agencies, landowners and permit-holders to mitigate this threat by vacating high risk allotments and performing actions like double-fencing and scheduled grazing to minimize the possibility of contact between bighorn and domestic animals.

During this reporting period we applied a disease risk model, combining a resource selection function model based on ram occurrences with a cost distance analysis to quantify the proximity of domestic sheep and goat grazing to bighorn core home ranges and the risk of contact of bighorn with domestic sheep and goats. We then examined the robustness of this model to the expanding distribution of Sierra bighorn. This model will directly inform translocation efforts, allowing identification of suitable areas for future Sierra bighorn reintroductions.

Some of the highest risk grazing occurs on the Conway and Mattly Ranches, which are owned and managed by Mono County and abut the Mt. Warren herd unit. Recovery Program leaders met with Mono County to continue discussing the risk that grazing on the Conway and Mattly Ranches poses to Sierra bighorn. In 2015, CDFW will monitor domestic sheep grazing operations on the Mattly Ranch at the mouth of Lundy Canyon.

Sierra Bighorn Population Monitoring

Herd Unit Surveys

Demographic data provide a foundation for the Recovery Program's adaptive management strategy, shaping our understanding of the health and growth of the Sierra bighorn population. Each year we focus on obtaining ground surveys from multiple populations and comparing these results with data from previous years. Certain herds (specifically Mt. Baxter and Wheeler Ridge) provide better survey opportunities in the winter, when animals congregate on low-elevation range; however surveys of most herds are more feasible in the summer. When possible, we compare minimum counts with mark-resight (MR) estimates, in which the total population is estimated from the ratio of marked to unmarked animals in an unbiased sample. During this reporting period we attempted surveys of all occupied herd units except Bubbs Creek (see Table 1 for survey results).

Olancha Peak

We surveyed Olancha Peak in June and September 2014 and April 2015 and accounted for 14 adult ewes, 2 yearling ewes, 6 lambs, 2 adult rams, and 2 yearling rams. One ram (S196) died of unknown causes before these surveys, and 3 ewes (S273, S206, and S272) died between November and April. S272 was seen in very poor condition in September 2014 and was nursing a late lamb. Her poor condition likely led to her death. We classified S273's death as a probable mountain lion kill, but were unable to determine the cause of death for S206. During the March 2015 capture we augmented this herd with two collared rams (S358 and S197) from the Mt. Baxter herd. At the end of this reporting period, we estimate that this population contained 11 adult ewes, 2 yearling ewes, 2 yearling rams, 6 lambs, and 4 adult rams. All adult ewes and rams are collared.

Laurel Creek

In March 2015 we introduced 6 adult ewes, 1 yearling ewe, and 4 adult rams to the previously-vacant Laurel Creek herd unit in the Kern Recovery Unit. All of the ewes were pregnant. One ram (S364, originally from Mt. Baxter) left Laurel Creek on April 2 and traveled to Cartago Creek at the north end of the Olancha Peak herd unit. On April 28 he left Olancha Peak and started a return journey toward the Kern River; at the time of this report he was on the Boreal Plateau. Another ram (S311, originally from Sawmill Canyon) left the herd unit boundary on April 13 and traveled to the Mt. Langley herd unit, where he remained for several weeks before joining S364 on the Boreal Plateau. Ram S322 also left Laurel Creek for the Boreal Plateau in May 2015, leaving only 1 ram, S204, in the Laurel Creek herd unit.

Big Arroyo

We introduced 10 Sierra bighorn ewes and 4 rams to the Big Arroyo herd unit in March 2014. One adult ewe (S281) and one adult ram (S233) died of unknown causes during this reporting period. Summer surveys and subsequent genotyping of lamb pellets confirmed the survival of 5 lambs; therefore the population of the Big Arroyo in May 2015 was 9 adult ewes, 5 lambs, and 3 adult rams.

Mt. Langley

Surveys of the Mt. Langley herd in August 2014 accounted for 45 adult ewes, 10 yearling ewes, 18 lambs, 57 adult rams, and 8 yearling rams. One collared adult ewe (S86) was censored (due to collar failure) during this reporting period and so was not included in this count. During a capture in October 2014, 3 adult ewes and 2 yearling ewes were collared and 1 previously-collared adult ewe was recaptured and her collar replaced. In March 2015, we removed 8 uncollared adult ewes, 1 uncollared yearling ewe, and 3 previously-collared adult ewes for translocations. We also collared 1 adult ewe who was re-released at Mt. Langley. Three collared adult rams (S179, S189, and S220) and 1 collared adult ewe (S341) died during this reporting period. S179's cause of death was unknown, S189 died from rockfall, S220 was classified as a probable lion kill, and S341 was determined to be a certain lion kill. As of May 2015, we estimate that this population contained 34 adult ewes, 9 yearling ewes, 18 lambs, 54 adult rams, and 8 yearling rams. Twenty-four percent of adult ewes and nine percent of adult rams have functional telemetry collars.

Mt. Williamson

In October 2014 we conducted the first survey of the Mt. Williamson herd unit since 2010. Our observations resulted in a minimum count of 11 adult ewes, 2 yearling ewes, 4 lambs, 8 adult rams, and 2 yearling rams. This is likely a significant undercount. One adult ram (S135) was killed by rockfall in June 2014.

Bubbs Creek

We did not survey the Bubbs Creek herd during this reporting period.

Mt. Baxter

In spring 2015, ground surveys led us to a minimum count of 46 adult ewes, 6 yearling ewes, 29 lambs, 25 adult rams, and 8 yearling rams. In October 2014 we collared 7 adult ewes, 1 yearling ewe, and 7 adult rams in this herd unit. In February 2015 we collared an additional 4 adult rams and 1 yearling ram. One 5 year old collared adult ram, S318, died of malnutrition in February 2015. In March 2015 we removed 6 uncollared adult ewes and 1 uncollared yearling ewe for translocation to Laurel Creek; we also removed 5 previously-collared rams for translocation to Laurel Creek, the Cathedral Range, and Olancha Peak. We estimate that at the end of this reporting period the Mt. Baxter population contained a minimum of 40 adult ewes, 5 yearling ewes, 29 lambs, 19 adult rams, and 8 yearling rams. Based on these minimum figures, a maximum of 38% of adult ewes and 26% of adult rams carry functional collars. Rams were probably significantly undercounted in 2015; thus the percent collared is likely considerably lower than 26%.

Sawmill Canyon

A survey in August 2014 resulted in a minimum count of 77 bighorn: 38 adult ewes, 6 yearling ewes, 17 lambs, 8 adult rams (2 seen and 6 collars not seen), and 8 yearling rams. Because our survey efforts focused on ewe groups, we expect that many adult rams were missed in this count. We collared 9 adult ewes, 8 adult rams, and 1 yearling ram in October 2014. One collared ewe (S231) and 1 collared ram (S313) died during this capture. In February 2015 we collared 4 more adult rams. In March and April 2015 we removed 3 collared adult ewes for translocation to Alger Creek in the Mt. Gibbs herd unit, and 3 collared adult rams for translocation to Laurel Creek. We estimate that at the end of this reporting period the Sawmill Canyon herd contained 34 adult ewes (of which 41% wear functional collars). Without a reasonable count of adult rams we cannot estimate the percentage of rams collared, but there are currently 13 functional collars on rams in this population.

Taboose Creek

On April 24, 2014, 2 biologists saw a group of 12 bighorn in this herd unit consisting of 11 adult rams and 1 yearling ewe. This was the first occasion on which Recovery Program staff made a confirmed observation of a female in the Taboose Creek herd unit. Subsequent observations were made in July, August of 2014, and February, and April of 2015. To date, the maximum numbers of each class of animal seen at one time has been 2 adult ewes, 1 yearling ewe, 15 adult rams, and 2 yearling rams. In October and February 2015 we collared 3 rams in this herd unit; 1 on Split Mountain and 2 on Birch Mountain in the northern end of the herd unit. All 3 have since traveled between the Taboose Creek and Sawmill Canyon herd units. In addition, 3 rams collared in Sawmill Canyon have made forays into Taboose Creek. We cannot yet provide estimates of the size or composition of the population that uses the Taboose Creek herd unit, nor

can we confidently describe the relationship between this population and the Sawmill Canyon bighorn.

Wheeler Ridge

During the exceptionally dry winter of 2014-2015 bighorn did not congregate on the low-elevation winter range at Wheeler Ridge; several attempts to survey this herd during the winter months were unsuccessful. However, numerous yearlings (4 female and 9 male) were observed suggesting good recruitment. We will attempt a summer survey in 2015 to obtain better population data.

Convict Creek

June 2014 surveys counted a minimum of 13 adult ewes, 8 lambs, 2 adult rams, 1 yearling ram, and 1 unclassified yearling in the Convict Creek herd unit. One 2-year-old ram was observed in 2013 and 1 in 2012, so the adult rams seen may be the 3- and 4-year-old rams produced by this population. While it is possible there are only 2 adult rams in this herd, it seems unlikely. On December 17, 2014, a group containing 1 yearling ewe and 2 yearling rams was observed, which added 1 yearling to the summer's count. In October 2015 we collared 2 adult females, 1 adult male, and 1 male lamb in this herd unit. The collared adult male, S337, was killed by a mountain lion on April 15, 2015. Based on our counts we estimate a maximum of 38% of ewes and 0% of rams have functional telemetry collars.

Cathedral Range

In March and April 2015 we introduced 9 adult ewes, 1 yearling ewe, and 3 adult rams to this newly-designated herd unit in Yosemite National Park. The ewes were moved from the Mt. Langley herd unit and all but the yearling were pregnant. Two rams were from Mt. Baxter and 1 was from Wheeler Ridge. On April 12, 14 days after translocation, 1 ram (S359) died of unknown causes. A mortality investigation noted that he had been scavenged by a black bear. He was 12 years old which is close to the maximum age for rams. Prior to his translocation, we recognized that he might be close to the end of his life. Nevertheless, because of his high genetic diversity and that it was unlikely for him to be competitive for mates in his native herd; we opted to give him a chance to breed in a new herd.

Mt. Gibbs

Biologists surveyed the Mt. Gibbs herd unit in July and September 2014 and accounted for 10 adult ewes, 3 yearling ewes, 8 lambs, 4 adult rams, and 1 yearling ram. Preliminary analyses of fecal samples from Mt. Gibbs rams in combination with observational data indicate that there may be as many as 9 adult rams in this herd. In October 2014 we replaced 1 nonfunctional ewe collar and 2 ram collars; we also captured and collared 1 yearling ewe. In April 2015 we introduced 5 adult ewes from the Sawmill Canyon and Mt. Langley herds to the Alger Creek area of the Mt. Gibbs herd unit with the intention of creating a new deme in that habitat and continuing genetic

Herd		E	wes		Lambs		Rams		Total
	Adult	Yearling	Total	MR Est.		Adult	Yearling	Total	
Olancha	14	2	16		6	2	2	4	26
Laurel	6	1	7		0	4	0	4	11
Big Arroyo	9	0	9		5	4	0	4	18
Langley	45	10	55	68 (50-91)	18	57	8	65	138
Williamson	11	2	13		4	8	2	10	27
Baxter	46	6	52		29	25	8	33	114
Sawmill	38	6	44		17	8	8	16	77
Taboose	2	1	3		0	15	2	17	20
Convict	13	1	14		8	2	2	4	26
Cathedral	9	1	10		0	3	0	3	13
Gibbs	10	3	13*		8	4	1	5	26
Warren	11	0	11		8	7	3	10	29

rescue efforts for that population. Sixty-seven percent of adult ewes in this herd unit now wear functional collars, and 3 adult rams have working collars.

Table 1. Minimum count data and mark-resight estimates (MR Est.) from surveys conducted during the 2014-2015 reporting period. Lambs are not identified by sex. Because translocations occurred after surveys were completed, translocated animals are shown both in their original herd units and in the herd units to which they were translocated.

*These data do not include ewes translocated into this population in March 2015.

Mt. Warren

Our usual early summer survey of this herd unit in July of 2014 identified 8 adult ewes, 6 lambs, and 3 yearling rams in this herd unit. Those yearlings were consistent with 3 lambs identified during counts in 2013 and 3 male lamb genotypes identified from lamb fecal samples that year; however, the count of 8 ewes was 3 lower than expected from 2013 data. During subsequent field work in 2014, three ewes and 2 lambs were sighted unexpectedly from a long distance on the top of the Dore Cliffs south of Lundy Canyon, where no ewes have been known since a small female deme that resided in that area perished during the heavy winter of 2010-2011. Genotyping of lamb fecal pellets identified two lambs from samples collected below the Dore cliffs which were different from 6 lambs similarly sampled and identified genetically from the opposite side of Lundy Canyon. This brought the total minimum count for this herd unit to 11 ewes, 8 lambs, and 3 yearling rams. The origin of the 3 ewes seen on the Dore Cliffs in 2014 has not yet been determined.

In October 2014, a biologist observed a group of 7 adult rams including all the collared rams known to be alive (S65, S239, and S185). This observation likely accounted for all of the adult rams in the Mt. Warren herd, bringing the total population size at that time to at least 29.

At the end of October we collared 2 adult ewes, 2 adult rams, and 1 yearling ram in the Mt. Warren herd unit, and replaced 2 adult ram collars. Genetic analysis showed that the yearling ram did not match any of the 3 male lamb genotypes from the 2013 season. This indicates the existence of at least one more ewe than was counted in 2013. One adult ewe died during capture. One collared adult ram was killed by a mountain lion in January 2015. At the end of this reporting period we estimate that the Mt. Warren herd unit contained 10 adult ewes of which 3 have functional telemetry collars, 8 lambs, 3 yearling rams, and 6 adult rams, 5 of which have functional telemetry collars.

Geographic Distribution

Sierra bighorn now occupy 14 herd units in 4 recovery units spanning a nearly 150-mile stretch of the Sierra Nevada (Figure 1). The Recovery Plan designates 16 herd units historically occupied by Sierra bighorn (USFWS 2007); the recently-completed Translocation Plan demarcates 2 additional herd units identified as suitable for reintroductions (Few et al. 2015). Of these 18 areas, 12 are included in recovery goals for the subspecies. All 12 of these herd units are now inhabited. Over the next few years, continued population monitoring and augmentation of recently-introduced herds will be essential to confirm that bighorn are persisting and flourishing in these areas.

Collaring Efforts

The Recovery Program strives to maintain a high proportion (30-35%) of marked animals within each herd to facilitate accurate population surveys, monitoring of reproductive success, and cause-specific mortality investigations (Table 2). The data we collect from GPS collars are central to our ongoing studies of habitat selection, seasonal migration, home range use, and survival. We conduct annual captures to create new marks, replace nonfunctional collars, and translocate animals to new habitat in accordance with the Translocation Plan (Few et al. 2015). Captures also give us the opportunity to assess the health and reproductive status of captured animals and to collect samples for genetic analysis.

We carried out 3 captures during this reporting period. Wildlife capture specialists from Leading Edge Aviation captured Sierra bighorn from 8 herds (Mt. Langley, Mt. Baxter, Sawmill Canyon, Bubbs Creek, Taboose Creek, Convict Creek, Mt. Gibbs, and Mt. Warren) using a net-gun fired from a helicopter. During October 18-28, 2014, we captured 62 Sierra bighorn (33 ewes, 27 rams, and 2 lambs) in order to increase the percentage and distribution of collared animals in each herd to aid us in obtaining accurate counts and survival data and to obtain genetic data on rams to allow selection of members of that sex for translocations based on genetic diversity. Three mortalities occurred as a result of this capture. A previously-collared Sawmill Canyon ewe, S231, died of spinal cord trauma when she was caught in the net with another animal. A previously-uncollared Mt. Warren ewe was captured alive under ordinary circumstances but was dead on arrival at basecamp; a field necropsy revealed that the pericardium and the bottom portions of the lungs were filled with blood. S313, a newly-collared Sawmill Canyon ram, moved about half a mile after his release on October 19 and died the next

	Lan	gley	Willia	amson	Ba	xter	Sav	vmill	Tab	oose	Bu	bbs	Wh	eeler	Cor	nvict	Gil	bbs	Wa	rren
Sex	F	М	F	М	F	М	F	М	F	М	F	М	F	М	F	М	F	М	F	М
5/1/2014	7	7	5	3	11	3	12	4	0	0	4	3	12	7	3	0	4	4	1	4
Additions	+7	0	0	0	+6	+11	+6	+13	0	+3	+2	+2	0	+6	+2	+1	+6	0	+2	+3
Subtractions	-6	-2	-1	-2	-2	-9	-4	-4	0	0	-1	-1	-1	-2	0	-1	0	-1	0	-2
4/30/2015	8	5	4	1	15	5	14	13	0	3	5	4	11	11	5	0	10	3	3	5
% Collared	24	9	36	13	38	26	41	*	0	20	50	*	26	55	38	0	67	33	30	83

day; although no injuries or unusual behaviors were evident during his capture, the timing of his death indicates it was probably capture-related.

Table 2. Distribution of radio collars by herd unit; new herd units created with introduced animals are not included because 100% of adults are collared. Additions include new captures, recaptures where nonfunctional collars were replaced, and augmentations. Subtractions include removals for translocation, mortalities, censors, and nonfunctional collars. The percent of the population collared is based on functional collars and adult population size from the most recent complete minimum counts. Because the number of collars is always known, but the population data are the minima, what is presented is the maximum %.

* Indicates a population from which the minimum number of rams is not known; thus, a maximum % collared cannot be determined.

On February 19, 2015 and February 20, 2015, we captured 18 Sierra bighorn rams in an effort to gain a larger pool of individuals known to have high genetic diversity; the Translocation Plan calls for selecting rams with high heterozygosity when initiating new herds (Few et al. 2015).

Over 5 days in March and April 2015 we translocated 31 Sierra bighorn. We reintroduced bighorn to 2 previously-vacant areas of historic habitat by moving 10 ewes and 3 rams to the Cathedral Range in Yosemite National Park and 7 ewes and 4 rams to the Laurel Creek area in Sequoia National Park. We supplemented the Olancha Peak herd, which was reintroduced in 2013, with 2 rams to maximize genetic diversity. We augmented the Mt. Gibbs herd with 5 ewes to increase genetic diversity while also creating a new deme in the Alger Creek basin south of Mt. Wood.

Sierra Bighorn Population Dynamics

Population Size

When Sierra bighorn were listed as an endangered species in 1999, only about 125 animals were known to exist in the range. We now estimate the total population size at over 600 bighorn (Few et al. 2015); the largest herds contain more than 40 adult and yearling females (Figure 2).

Because we did not obtain a complete survey of the Wheeler Ridge herd this year, the estimate shown was derived from the total females found in the previous year's surveys minus 10 removals for translocation to Big Arroyo and including the 4 yearling ewes

observed during our December survey attempts. The apparent increase in the Sawmill Canyon population is likely the result of a more complete count this year.

At the end of the 2014-2015 survey season we estimate that there were at least 288 female bighorn in the Sierra (Figure 3). The Recovery Plan recommends downlisting when the female population reaches 305 animals distributed throughout the recovery units (50 in the Kern Recovery Unit, 155 in the Southern Recovery Unit, 50 in the Central Recovery Unit, and 50 in the Northern Recovery Unit; USFWS 2007). We anticipate reaching this goal within 5 years. The Southern and Central Recovery Units are 11 and 34 females, respectively, under their recovery goals.



Figure Combined 3. population trajectories for adult and yearling ewes from all occupied herds (Olancha Peak, Big Arroyo, Mt. Langley, Mt. Williamson, Bubbs Creek, Mt. Baxter, Sawmill Canyon, Taboose Creek, Wheeler Ridge, Convict Creek, Mt. Gibbs, and Mt. Warren) from 1999-2014 surveys. Population estimates in earlier years lack data for some herds. Some of the significant increases have been due to better data and cannot be construed as population gains; for example, the increase between 2012 and 2013 is the result of more complete counts in 2013.

Figure 2. Population trajectories for adult and yearling females from 1999-2014 based on a combination of minimum counts, markresight estimates, and reconstructed data for 6 herds in the Sierra Nevada with annual population data. In years when no data were available or when surveys were incomplete, survey totals from the most recent complete count were used. Data from mark-resight estimates are with plotted error bars representing 95% confidence intervals. In all figures, years are defined from May 1 to April 30 of the following year.





Figure 4. Adult and yearling females present in each recovery unit at the end of the 2014-2015 reporting period relative to the distribution of females specified in recovery goals.

Survival and Cause-Specific Mortality

Demographic rates are important tools for evaluating population health and growth. Adult female survival is the primary factor driving population growth or decline in Sierra bighorn herds (Johnson et al. 2010). Maintaining radio collars on 30-35% of females in each herd unit allows us to detect and investigate mortalities; we use this information to calculate annual Kaplan-Meier survival rates of radio-collared ewes (Kaplan and Meier 1958). Following these rates over time gives us an understanding of the year-to-year variation in adult ewe survival and the general trend of this metric in different populations.



Figure 5. Annual Kaplan-Meier survival rates of radiocollared ewes in the Northern, Central, and Southern Recovery Units for 2007-2014. The dashed line represents 90% survival.

Between 2007 and 2014, survival rates varied from 0.58 to 1.0 (Figure 5). The lowest survival rates occurred in the Northern Recovery Unit in 2012, in the Central Recovery Unit in 2010, and in the Southern Recovery Unit in 2008. In 2014 survival rates were high in all herd units; survival rates above 90% are associated with population growth (unpublished data).

The Recovery Program prioritizes prompt mortality investigations. Understanding the predominant causes of bighorn mortality can help develop conservation measures that may increase survival and population growth. During this reporting period we detected 14 natural mortalities of collared bighorn (5 female, 9 male; Figure 6). We were unable to determine the cause of 6 of these mortalities. One ram at Mt. Baxter died of malnutrition. One ram at Mt. Langley and one ram at Mt. Williamson died of physical injury (the former due to a fall from a cliff, the latter due to rockfall). We determined that 3 Sierra bighorn (1 ewe at Mt. Langley, 1 ram at Mt. Warren, and 1 ram at Convict Creek) were killed by mountain lions; 2 additional mortalities (1 ram at Mt. Langley and 1 ewe at Olancha Peak) were considered probable mountain lion kills.



Figure 6. Cause-specific natural mortalities of radio-collared bignorn from May 1 to April 30 of the following year.

Reproduction and Recruitment

Recruitment, the proportion of females that reach reproductive age, can be measured by comparing the number of adult and yearling females observed in each herd unit in one year with the total number of adult females observed there the following year. Assuming accurate minimum counts in both years and 100% survival, the two numbers would be equal. This is rarely the case; yet, in 4 herd units, Olancha Peak, Mt. Langley,

Herd		20)13		2014
	Adult Ewes	Yearling	Total Ewes	Known	Adult Ewes
		Ewes		Gains/Losses	
Olancha	14	0	14		14
Langley	38	9	47	-2	45
Baxter	40	6	46		46
Convict	12	1	13		13
Gibbs	11	1	12	-1	10
Warren	7	4	11		11

Table 3. Comparison of the number of adult ewes in 2014 to the total number of ewes in 2013 after accounting for recruitment of yearlings and known losses or gains from mortalities or translocations. Populations with poor minimum counts in either year are not included.

Mt. Baxter and Convict Creek (Table 3) the 2014 totals of adult ewes corresponded exactly to the number of adult and yearling ewes observed in 2013 after known losses were subtracted. 2014 totals in the two remaining herd units, Mt. Gibbs and Mt. Warren (Table 3), are only one ewe short of projected totals based on 2013 data. These findings suggest high adult survival and yearling recruitment in all of those herd units.

Additional metrics to assess herd health are the observed ratio of yearlings to ewes and the ratio of lambs to ewes which indicate recruitment and fecundity (reproductive output depending on the age at which lambs are observed), respectively. Yearling to ewe ratios vary from 0.23 to 0.44 in 2014 (Table 4) which indicate positive or stable population growth assuming high adult survival. Lamb to ewe ratios vary from 0.33 to 0.8 in 2014 (Table 4) which are within the healthy range for these populations indicating good reproductive success.

Herd	Lamb:Ewe	Total Yearling:Ewe	
Olancha	0.33	0.44	
Langley	0.42	0.42	
Baxter	0.74	0.36	
Convict	0.62	0.23	
Gibbs	0.8	0.40	
Warren	0.72	0.27	

Table 4.Ratios ofjuvenile age classes toewesfrom 2014.Populations with poorminimumcountseitheryeararenotincluded.

Another way to assess lamb survival is to compare the total number of yearlings observed in each herd unit with the number of lambs observed there in the previous survey season. Observed lamb survival between 2013 and 2014 varied from 0.5 to 1.0 (Table 5). These values represent lamb survival between annual surveys, which occur months after lambs are born. Thus these estimates of lamb survival do not include survival rates of neonatal lambs. While a 50% survival rate may seem low compared to for iuvenile adult survival. it is not unusual age classes.

,			, 0
Herd	2013 Lambs	2014 Yearlings	Lamb Survival
Olancha	8	4	0.50
Baxter	24	14	0.58
Convict	5	3	0.60
Gibbs	7	4	0.57
Warren	3	3	1.00

Table 5. Lamb survival estimated by comparing the number of yearlings in 2013 to the number of lambs in 2012. All data are from minimum counts. Populations with incomplete minimum counts in either year are not included.

New Findings

Taboose Creek Occupation

For several years, the Recovery Program has suspected that a natural colonization of the Taboose Creek herd unit by Sawmill Canyon bighorn was underway (Stephenson et al. 2012). Observations made during this reporting period and collars deployed in these 2

herd units confirm that numerous rams make regular use of both areas. It is likely that a population of rams also resides permanently in the Taboose Creek herd unit, and several recent sightings of adult and yearling ewes strongly suggest that a reproductive population exists here as well. The Recovery Program will continue to make investigation of this herd unit a priority in 2015-2016.

New Habitat Use, Possible Range Expansions, and Long-Distance Movements

Deployed GPS collars provide insight into habitat use and long-distance movements by Sierra bighorn. In the Mt. Warren herd unit, a small ewe group has continued to use the Camiaca Peak area, where collared ewe S89 moved in November 2013. A summer survey also located 3 adult ewes and 2 lambs on Dore Peak, an area south of Lundy Canyon where no bighorn have been seen since avalanches during the heavy winter of 2010-2011 killed all animals known to use that habitat. It is possible that this small ewe group has persisted undetected in the area since 2010; an alternative explanation is that occupation of this area represents a recent range expansion or repossession for ewes in the Mt. Warren herd.

Studies of bighorn rams have often documented long-range movements, particularly during the rut (Geist 1971, Leslie and Douglas 1979, O'Brien et al. 2014). Deploying collars on Sierra bighorn rams allows us to document the significant distances that specific individuals travel. S311, a 9-year-old ram first captured in October 2014 in the Window Peak area of the Sawmill Canyon herd unit, traveled throughout the Sawmill Canyon herd unit and into the northern end of the Mt. Baxter herd unit before spending most of the winter on Cardinal Peak in the southern Taboose Creek herd unit (Figure 7).

These movements suggest the Sawmill Canyon, Mt. Baxter, and Taboose Creek herds function as a metapopulation with gene flow occurring between herds.



Figure 7. Movements of S311, a 9-year-old Sawmill Canyon ram, between October 2014 and March 2015.



Figure 8. Movements of Big Arroyo ewe S286 since her translocation in March 2014.

Habitat Exploration by Naïve Animals

Translocating Sierra bighorn often results in unanticipated movements by the naïve animals as they explore their new habitat. S286, a Wheeler Ridge ewe who was translocated to the Big Arroyo in March 2014, left that drainage on June 30, 2014 and traveled north, accompanied only by her lamb. She briefly crossed the Kern River north of Tyndall Creek, then crossed back to the Kern Ridge and eventually settled on Kern Point, where she has remained since late July 2014 (Figure 8). No other collared ewe has ever traveled to her location.

The 11 bighorn translocated to the Laurel Creek herd unit in March 2014 have since dispersed widely (Figure 9). Some traveled over 6 miles south to Coyote Peaks while others crossed the Kern River to the east, and 3 rams left the herd unit boundary for the Boreal



Figure 9. Movements of Laurel Creek animals after translocation in March 2015. Plateau, just west of the Mt. Langley herd unit. Three ewes, S377, S378, and S382, have remained east of the Kern River near the Hell-For-Sure drainage for several weeks. We will continue to monitor these animals over the 2015 summer, when the search for high-elevation habitat may draw them back within the Laurel Creek herd unit boundary.

By contrast, the animals translocated to the Cathedral Range herd unit in Yosemite National Park have all remained in the immediate vicinity of their release site, on the slopes above Washburn Lake.

Research Priorities

Genetic Research

Sierra bighorn are recognized for their genetic uniqueness as a separate subspecies; therefore, recovery efforts for this taxon are ultimately about conserving and enhancing this unique gene pool. Sierra bighorn survived epizootics caused by past domestic sheep grazing only in three herds in the southern Owens Valley, but they did not survive without genetic scars. They exhibit signatures of a genetic bottleneck and have the lowest genetic diversity measured for free-ranging native populations in the desert region. Genetic diversity in Sierra bighorn herds is sufficiently low that individuals at the lowest end of the heterozygosity (individual genetic diversity) spectrum may be less fit (Johnson et al. 2011). This presents a potential opportunity to increase genetic diversity in small and reintroduced populations to enhance population fitness and success.

Various authors have recommended that large numbers of bighorn sheep (more than 20) be used in reintroductions to maximize the representation of genetic diversity in new herds and to minimize founder effects (Fitzsimmons et al. 1997, Griffiths et al. 1982, and Wolf et al. 1996). However, Sierra bighorn translocation stock is both limited in the numbers of animals available and in the genetic diversity of those animals. With careful genetic planning including selective captures of individual bighorn, it might be possible to initiate highly diverse herds with fewer animals by maximizing genetic diversity in the founding gene pool.

To explore different genetic management options, we employed sampling experiments of existing data to examine the genetic consequences of three different approaches that might be used for founding populations: 1) all individuals selected for higher heterozygosity, 2) all individuals selected at random, and 3) all ewes selected randomly but rams selected individually for higher heterozygosity. In our sampling experiments, we measured average heterozygosity at 17 microsatellite loci and interpreted this as a direct measurement of genetic diversity. However, we did not look at allele structure or loss at the individual loci.

These sampling experiments revealed that (1) the first approach can produce founding gene pools with notably higher heterozygosity than any existing population, but that there are too few alleles remaining in the Sierra bighorn gene pool to support that level of genetic diversity over time (heterozygosity excess); (2) the third approach of selecting

only high heterozygosity rams provides a significant genetic improvement over random selection of bighorn, does not produce a large heterozygosity excess, and minimizes the number of sheep that need to be selectively recaptured; (3) genetic diversity is improved for this third approach if the rams are selected from multiple populations; and (4) random selection of ewes from a single population mating with selected rams resulted in genetic diversity similar to the remaining native populations. As a result of these findings, all recent reintroductions (Olancha Peak, Laurel Creek, Big Arroyo, and the Cathedral Range) initially translocated 7-10 pregnant females randomly captured from a single herd and 3-4 specific males selected for high individual heterozygosity from multiple source herds.

Greater selectivity in the individuals used to initiate a population should allow for fewer animals to represent variation in the gene pool. However, a downside of a smaller founding population is that matings between close relatives are more likely to occur, and such inbreeding will work against the advantages of the initial selectivity. The Olancha Peak herd was reintroduced in 2013 and augmented in 2014. This is the only recently-reintroduced herd where bighorn have been present long enough that rams now have the potential opportunity to breed their daughters. Of 6 lambs that were born in 2014 and survived to be sampled in late summer, genetic and observational studies found that 3 belonged to pregnant ewes moved there in 2014. Of the other 3, 2 are females that have the potential to be bred by their fathers. One of these was born very late, thus is very unlikely to breed in 2015 as a yearling. In 2015 we added 2 high heterozygosity rams to this population to dilute the probability of a father-daughter mating. We will consider this strategy in our other newly created populations as well.

It is at small population sizes that we can have the greatest influence on genetic population structure by adding high heterozygosity individuals. Within Sierra bighorn, the Mt. Gibbs herd unit stands out in showing clear signs of low genetic diversity, which is consistent with its demographic history and substantial isolation (Stephenson et al. 2012). This has raised the question of whether the population's growth rate might be improved by increasing genetic diversity through a genetic rescue by selective augmentation with high heterozygosity individuals. To increase the genetic diversity of this herd we have implemented two approaches, both of which used translocated ewes selected for high genetic diversity. First, in 2013, we augmented the existing ewe group (7 ewes) on Mt. Gibbs with 3 high heterozygosity ewes, two of which were pregnant. Second in 2015, we created a new deme of high heterozygosity ewes in the Alger Creek area on the south side of Mt. Wood, an area Mt. Gibbs rams have used regularly. This new deme was founded with 3 ewes translocated from the Sawmill Canyon herd and 2 ewes from the Mt. Langley herd, all of which were pregnant. This results in the current total 15 adult ewes of which 8 (>50%) are ewes selected for high genetic diversity.

Pine Creek Recreation Study

Over the last ten years, the Pine Creek area of the Wheeler Ridge herd unit has become an increasingly popular destination for hikers, sightseers, and rock climbers. Pine Creek Canyon is also routinely used as lambing habitat by Wheeler Ridge ewes. In 2014-2015 the Recovery Program began a study to develop a baseline estimate of current recreational use of this canyon. Tracking recreation over time will allow us to quantify this trend and detect any relationship between increased recreation and Sierra bighorn use of Pine Creek.

Home Range Analysis

Recovery Program staff used a dataset containing a decade's worth of GPS collar locations to define the home ranges of Sierra bighorn both at the individual and at the population level. They examined variation in home range size from year to year and in different seasons, as well as the relationship between home range size and population size. The results of this study may contribute to a better understanding of habitat selection and availability. Results will be summarized in next year's report.

Resource Selection Function

Species distribution models (SDMs) provide a measure of the importance of ecological variables that correlate with species occurrence. These models can provide a framework for the implementation of adaptive management in the recovery of Sierra bighorn. Model results can be applied to spatial data to produce maps representing the likelihood of species occurrence. In a study currently underway, we used one type of SDM, a resource selection function (RSF) generated by logistic regression, to examine how species rarity affects model predictions of the likelihood of occurrence.

This model and a winter RSF that accounts for altitudinal migration identified two large patches of bighorn habitat unrecognized by the Recovery Plan in remote geographic areas where there is a paucity of historic occurrence data (the Cathedral Range and Black Divide herd units) compared to more easily accessible areas east of the Sierra crest (Wehausen and Jones 2014). By quantifying habitat quality, these models will directly inform translocation efforts, allowing the Recovery Program to identify suitable areas for future Sierra bighorn reintroductions.

Public Outreach

Educating the Community

Community support is crucial to the success of conservation efforts for the recovery of Sierra bighorn. Because these animals are rare and occupy remote areas, most residents of the Eastern Sierra have never seen a Sierra bighorn in the wild and know very little about them. The Recovery Program partners with the Sierra Nevada Bighorn Sheep Foundation (SNBSF) to increase public awareness of this endangered subspecies and conservation work on its behalf.

The SNBSF continues to expand its educational programs. Since May 2014, the SNBSF has planned and carried out 23 public events, reaching over 1,500 people throughout the region. The events range from booths at local celebrations like Bishop Earth Day, CDFW's Trout Fest, and the Tri-County Fair, to school programs in which children

simulate the capture and processing of a toy bighorn sheep, entering the animal's measurements into a datasheet, fitting it with a radio collar, and using its heterozygosity score to determine its suitability for translocation.

In conjunction with the SNBSF, the Recovery Program also led 2 free public field trips in February and March 2015. Over 60 participants were given the opportunity to observe groups of Sierra bighorn on winter ranges, while Recovery Program staff members and SNBSF volunteers answered questions and provided historical and biological context.

Permanent Outreach Displays

The Migrating Mural, created by scientific illustrator Jane Kim, is a series of paintings depicting life-size Sierra bighorn on buildings along the Highway 395 corridor. Kim hopes the murals will bring public attention to the plight of Sierra bighorn and raise support for recovery efforts. The final mural in the series, painted on the Forest Service Visitor Center in Lee Vining, was completed in May 2014. Other Migrating Mural scenes appear at the Bishop Gun Club, Sage to Summit running store in Bishop, the Mt. Williamson Motel in Independence, and the Lone Pine Airport, spanning most of the north-south range of bighorn in the Sierra.



Figure 10. Detail from Jane Kim's Migrating Mural in Lee Vining; photograph courtesy of Jane Kim, *www.inkdwell.com*.

Future Recovery Actions

The Translocation Plan completed in 2015 outlines the augmentations and reintroductions the Recovery Program may carry out within the next 10 to 20 years. These translocations are a means of recreating the population distribution that characterized the subspecies before endangerment, while also increasing the genetic diversity and long-term viability of smaller herd units (Few et al. 2015). No translocations or augmentations are scheduled during the next reporting period.

Downlisting to threatened status will not occur until the risk of contact between wild bighorn and domestic sheep is eliminated. The Recovery Program will increase its focus on reducing the risk of contact between wild bighorn and domestic sheep. Program leaders will continue working to mitigate this risk in cooperation with land management agencies, landowners, and grazing permittees. Since its inception in 2000, the Recovery Program has helped to catalyze and document significant increases in the size and distribution of the Sierra bighorn population. The 2014-2015 reporting period witnessed the realization of a major objective: all 12 herd units included in recovery goals for the subspecies are now occupied. With additional translocations, continued population growth, and further steps taken to mitigate disease risk, Sierra bighorn may achieve Recovery Plan goals for downlisting to threatened status within the next 5 years.

Literature Cited

Few, A.P., J.M. Runcie, D.W. German, J.D. Wehausen, and T.R. Stephenson. 2013. 2012-2013 Annual Report of the Sierra Nevada Bighorn Sheep Recovery Program. California Department of Fish and Wildlife. 19pp.

Few, A.P., K. Knox, D. W. German, J. D. Wehausen, and T. R. Stephenson. 2015. 2015 Translocation Plan for Sierra Nevada Bighorn Sheep: A Focus on Strategic Planning. California Department of Fish and Wildlife. 54pp.

Geist, V. 1971. Mountain Sheep: A Study in Behavior and Evolution. The University of Chicago Press, Illinois, USA.

Johnson, H. E., L. S. Mills, T. R. Stephenson, and J. D. Wehausen. 2010. Populationspecific vital rate contributions influence management of an endangered ungulate. Ecological Applications 20: 1753-1765.

Kaplan, E. L. and P. Meier. 1958. Nonparametric estimation from incomplete observations. Journal of the American. Statistical Association 53: 457–481.

Lawrence, P. K., S. Shanthalingam, R. P. Dassanayake, R. Subramaniam, C. N. Herndon, D. P. Knowles, F. R. Rurangirwa, W. J. Foreyt, G. Wayman, A. M. Marciel, S. K. Highlander, and S. Srikumaran. 2010. Transmission of Mannheimia haemolytica from domestic sheep (Ovis aries) to bighorn sheep (Ovis canadensis): unequivocal demonstration with green fluorescent protein-tagged organisms. Journal of Wildlife Diseases 46:706-717, and erratum 46: 1346-1347.

Leslie, D. M., and C. L. Douglas. 1979. Desert bighorn sheep of the River Mountains, Nevada. Wildlife Monographs 66:1-56.

O'Brien, J. M., C. S. O'Brien, C. McCarthy, and T. E. Carpenter. Incorporating foray behavior into models estimating contact risk between bighorn sheep and areas occupied by domestic sheep. Wildlife Society Bulletin 38:321-331.

Runcie, J. M., A. P. Few, D. W. German, J. D. Wehausen, and T. R. Stephenson. 2014. 2013-2014 Annual Report of the Sierra Nevada Bighorn Sheep Recovery Program. California Department of Fish and Wildlife. 19pp.

Stephenson, T. R., J.D. Wehausen, A.P. Few, D.W. German, D.F. Jensen, D. Spitz, K. Knox, B.M. Pierce, J.L. Davis, J. Ostergard, and J. Fusaro. 2012. Annual Report of the Sierra Nevada Bighorn Sheep Recovery Program: A Decade in Review. California Department of Fish and Game. 57 pp.

Wehausen, J. D. and F. L. Jones. 2014. The historic distribution of bighorn sheep in the Sierra Nevada, California. California Fish and Game 100:417-435.

Wehausen, J. D., R. R. Ramey II, and S. T. Kelley. 2011. Domestic sheep, bighorn sheep, and respiratory disease: a review of experimental evidence. California Fish and Game 97: 7-24.

Wild Sheep Working Group. 2012. Recommendations for Domestic Sheep and Goat Management in Wild Sheep Habitat. Western Association of Fish and Wildlife Agencies.

U.S. Fish and Wildlife Service. 2007. Recovery Plan for the Sierra Nevada Bighorn Sheep. Sacramento, California. 199 pp.

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Epizootic Pneumonia of Bighorn Sheep following Experimental Exposure to *Mycoplasma ovipneumoniae*



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Abstract

Background: Bronchopneumonia is a population limiting disease of bighorn sheep (*Ovis canadensis*). The cause of this disease has been a subject of debate. Leukotoxin expressing *Mannheimia haemolytica* and *Bibersteinia trehalosi* produce acute pneumonia after experimental challenge but are infrequently isolated from animals in natural outbreaks. *Mycoplasma ovipneumoniae*, epidemiologically implicated in naturally occurring outbreaks, has received little experimental evaluation as a primary agent of bighorn sheep pneumonia.

Methodology/Principal Findings: In two experiments, bighorn sheep housed in multiple pens 7.6 to 12 m apart were exposed to *M. ovipneumoniae* by introduction of a single infected or challenged animal to a single pen. Respiratory disease was monitored by observation of clinical signs and confirmed by necropsy. Bacterial involvement in the pneumonic lungs was evaluated by conventional aerobic bacteriology and by culture-independent methods. In both experiments the challenge strain of *M. ovipneumoniae* was transmitted to all animals both within and between pens and all infected bighorn sheep developed bronchopneumonia. In six bighorn sheep in which the disease was allowed to run its course, three died with bronchopneumonia 34, 65, and 109 days after *M. ovipneumoniae* introduction. Diverse bacterial populations, predominantly including multiple obligate anaerobic species, were present in pneumonic lung tissues at necropsy.

Conclusions/Significance: Exposure to a single *M. ovipneumoniae* infected animal resulted in transmission of infection to all bighorn sheep both within the pen and in adjacent pens, and all infected sheep developed bronchopneumonia. The epidemiologic, pathologic and microbiologic findings in these experimental animals resembled those seen in naturally occurring pneumonia outbreaks in free ranging bighorn sheep.

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Introduction

Bighorn sheep are a North American species that has failed to recover from steep declines at the turn of the 20th century despite strict protections and intensive management, and two populations (Sierra Nevada and Peninsular) are currently classified as endangered [1]. Epizootic pneumonia is limiting bighorn sheep population restoration and as such, the etiology is of considerable interest. The first appearance of the disease in a population is typically in the form of epizootics that affect animals of all ages and is sometimes accompanied by high (>50%) mortality rates. Subsequently, epizootics affecting primarily lambs may occur for decades [2]. Various causes have been proposed for this disease, including lungworms (*Protostrongylus* sp.) [3–6], Pasteurellaceae, especially *Mannheimia (Pasteurella) haemolytica*, [7–12] and more recently, *Mycoplasma ovipneumoniae* [13–16]. In a recent comparative review of the evidence supporting each of these possible etiologies we concluded that M. ovipneumoniae was most strongly supported as the primary epizootic agent of bighorn sheep pneumonia [14]. However, the only two previous experimental challenge studies with M. ovipneumoniae either did not reproduce disease [13] or were confounded by challenges with other agents [16]. The objective of this study was to improve upon previous investigations to better assess the outcome of experimental introduction of M. ovipneumoniae to naïve bighorn sheep.

Methods

Ethics statement

This study was carried out in accordance with the recommendations in the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health and in conformance with United States Department of Agriculture animal research guidelines, under protocols #03854 and #04482 approved by the Washington State University (WSU) Institutional Animal Care and Use Committee. As described in those protocols, euthanasia was performed by intravenous injection of sodium pentobarbital for animals observed to be in severe distress associated with pneumonia during the study and prior to necropsy examination for surviving animals at the end of each experiment.

Experimental aims

Experiment 1 was conducted to investigate the transmission of *M. ovipneumoniae* to bighorn sheep and their subsequent development of disease, using an infected domestic sheep source. Experiment 2 was conducted to investigate experimental direct *M. ovipneumoniae* infection of a single bighorn sheep and the subsequent transmission of this agent to conspecifics. Both experiments were conducted in multiple pens separated by short distances, which allowed investigation of transmission to both commingled and non-commingled animals.

Experimental animals

All experimental animals originated from herds and flocks unexposed to *M. ovipneumoniae* as determined by repeated testing with both serology on blood serum and PCR on enriched nasal swab cultures (using the methods described later in the 'Microbiological testing' section). In Experiment 1, three hand-reared bighorn sheep (yearling rams BHS #82 and #89 and yearling ewe BHS #07) that originated from a captive flock at WSU and three purchased domestic sheep (adult ewes DS #00 and #01 and yearling ewe DS #LA) were co-housed in three 46 m² pens, with one domestic and one bighorn sheep per pen. Pens were separated by 7.6-12 m. Experiment 1 animals had all been commingled in a single pen for 104 days immediately prior to the beginning of this experiment, as previously described [15]. One of the four bighorn sheep used in that prior study had died of M. haemolytica pneumonia, while the other three, which had demonstrated no signs of respiratory disease in that study, were used in experiment 1. In Experiment 2, wild bighorn sheep captured from the Asotin Creek population in Hells Canyon were housed in two 700 m^2 pens, 7.6 m apart, with three animals per pen (Pen #1: adult ewe BHS #40, yearling ewe BHS #38, and yearling ram BHS #39; Pen #2: adult ewes BHS #41 and #42 and adult ram BHS #C). The study pens had either never previously housed domestic or bighorn sheep (pen 1 in experiment 1; both pens in experiment 2) or had been rested for greater than one year since their previous occupancy by any M. ovipneumoniae infected sheep (pens 2 and 3 in experiment 1) prior to these experiments.

Experimental design

Experiment 1. A domestic ewe (DS #00) was placed in isolation and experimentally infected with *M. ovipneumoniae*. The inoculum consisted of ceftiofur-treated (100 ug/ml, 2 hrs, 37° C; Pfizer, Florham Park, NJ) nasal wash fluids from a domestic sheep naturally colonized with *M. ovipneumoniae* [16]. Following ceftiofur treatment, no aerobic bacterial growth was observed from the nasal wash fluids cultured under conditions expected to permit growth of *M. haemolytica*, *B. trehalosi*, or *P. multocida* (Columbia blood agar with 5% sheep blood, 35°C, overnight, 5% CO₂). DS #00 was then challenged with the treated nasal wash fluid by infusion of 15 ml in each nares, 10 ml orally and 5 ml into each conjunctival sac. Subsequent nasal swab samples obtained on days 1, 2, 4 and 7 post-challenge were all PCR positive for *M. ovipneumoniae* using the method described later in the 'Microbiological testing' section confirming that the experimental infection

had been successful. On post challenge day 7, DS #00 was introduced into pen #1 with BHS #82. Following commingling, DS #00 and BHS #82 were restrained for collection of nasal swab samples on days 1, 2, 4, 7, 14, 21, 28, and subsequently at 30 day intervals until the experiment was terminated. Rectal temperatures were recorded from both sheep approximately twice each week. Sheep in pens #2 (BHS #89 and DS #01) and #3 (BHS #07 and DS #LA) were restrained for rectal temperature determination and collection of nasal swabs for microbiology at approximately monthly intervals. All pens were observed daily for clinical signs of respiratory disease. The experiment was conducted October 2009–January 2010.

Experiment 2. BHS #39 was inoculated with *M. ovipneu*moniae just prior to its release into pen #1 with non-inoculated BHS #38 and #40. Non-inoculated BHS #C, #41, and #42 were housed in pen #2 on the same day. The inoculum for BHS #39 was prepared as described for that used in experiment 1 but originated from a different domestic sheep source. In lieu of computation of colony forming units, which is not possible for M. ovipneumoniae due to inconsistent growth on plated media, viable M. ovipneumoniae counts in the inoculum were determined using most probable number (MPN) using a custom 3×4 format: Triplicate enrichment broth tubes were inoculated at each of four decimal dilutions $(10^{-2}-10^{-5})$ of the treated nasal wash fluid [17], incubated (72 hrs, 35C) then PCR was used to detect growth of viable M. ovipneumoniae. The treated fluid was determined to contain 930 MPN/ml (95% confidence interval, 230 to 3800 MPN). Two of the bighorn sheep (BHS #38 and #39) in pen 1 were recaptured by drive net on day 21 of the experiment for nasal swab sampling to detect M. ovipneumoniae infection; otherwise, no live animal sampling was conducted in experiment #2 to reduce the risk of traumatic injury of the wild bighorn sheep involved. The experiment was conducted December 2011-June 2012

Biosecurity. In both experiments, routine biosecurity measures included: 1) the pens containing the single M. ovipneumoniae-challenged animals (exposed pens) were located downwind of the prevailing wind direction from the pens containing no experimentally M. ovipneumoniae exposed animals (clean pens), 2) order of entry rules were established so that on any single day exposed pens were routinely entered by animal care staff for feeding and cleaning only after all work in clean pens had been completed, and 3) personal protective equipment (coveralls and boots) used in exposed pens were either not reused, or were sanitized prior to use in clean pens.

Clinical scores. Clinical score data were determined using the following cumulative point system: observed anorexia (1), nasal discharge (1), cough (2), dyspnea (1), head shaking (1), ear paresis (1) and weakness/incoordination (1).

Microbiological testing. Routine diagnostic testing performed by the Washington Animal Diagnostic Laboratory (fully accredited by the American Association of Veterinary Laboratory Diagnosticians) included detection of *M. ovipneumoniae*-specific and small ruminant lentivirus-specific antibodies in serum samples using competitive enzyme-linked immunosorbent assays (cELISA) [14,18,19], detection of *M. ovipneumoniae* colonization by broth enrichment of nasal swabs followed by *M. ovipneumoniae*-specific PCR testing of the broths [20,21], detection of Pasteurellaceae in pharyngeal swab samples by aerobic bacteriologic cultures, and detection of exposure to parainfluenza-3, border disease, and respiratory syncytial viruses by virus neutralization antibody assays applied to serum samples.

PCR tests specific for detection of *M. haemolytica*, *B. trehalosi*, and *P. multocida*, and *lktA* (the gene encoding the principal

virulence factor of *M. haemolytica* and *B. trehalosi*) were applied to DNA extracted from pneumonic lung tissues using previously described primers (Table 1) and methods with minor modifications. All reactions were conducted individually in 20 µL volumes containing 80–300 ng of template DNA. For M. haemolytica, B. trehalosi, lktA and P. multocida, reactions contained 0.5 units of HotStar Taq DNA polymerase (Qiagen), 2 µL 10x PCR buffer (Qiagen), 4 µL Q-solution (Qiagen), 40 µM of each dNTP (Invitrogen). The M. ovipneumoniae reaction used QIAGEN Multiplex PCR mix. Primers were used at final concentrations of $0.2 \,\mu M$ (M. haemolytica, B. trehalosi, P. multocida, and M. ovipneumoniae) or 0.5 µM (leukotoxin A). Each reaction included an initial activation and denaturation step (95°C, 15 min) and a final 72°C extension step (10 min for Mhgcp-2, lktA, lktA set-1, and LM primers; 9 min for KMT primers; 5 min for Btsod and Mhgcp primers). Cycling conditions were as follows: M. ovipneumoniae, 30 cycles of 95°C for 30 s, 58°C for 30 s, 72°C for 30 s; B. trehalosi and M. haemolytica (Mhgcp and Btsod primers), 35 cycles of 95°C for 30 s, 55°C for 30 s, 72°C for 40 s; P. multocida and lktA (lktA primers), 30 cycles of 95°C for 60 s, 55°C for 60 s, 72°C for 60 s; M. haemolytica (Mhgcp-2 primers), 40 cycles of 95°C for 30 s, 54°C for 30 s, 72°C for 30 s; lktA (lktA set-1 primers), 40 cycles of 95°C for 30 s, 52°C for 30 s, 72°C for 40 s. Leukotoxin expression was detected in Pasteurellaceae isolates by MTT dye reduction cytotoxicity assay as described previously [22].

The 16S–23S ribosomal operon intergenic spacer (IGS) regions of *M. ovipneumoniae* recovered from animals in these studies were PCR amplified (Table 1) and sequenced as previously described [23].

16S rDNA analyses to identify the predominant bacterial flora in pneumonic lung tissues. In previous studies, culture-independent evaluation of the microbial flora of lung tissues in naturally occurring bighorn sheep pneumonia revealed a polymicrobial flora late in the disease course [13,23]. For comparison, we applied the same methods to lung tissues of the experimentally challenged animals in this study. Note that more sensitive

detection of specific respiratory pathogens was provided by the PCR assays described earlier, whereas these 16S studies were designed instead to identify the numerically predominant bacteria in affected lungs. The library size used was based on the binary distribution to provide a 95% chance of detection of each taxon comprising 10% or more of the ribosomal operon frequency in the source tissue. Two 1 g samples of pneumonic lung tissues were aseptically collected from sites at least 10 cm apart, homogenized by stomaching, and DNA was extracted (DNeasy tissue kit; Qiagen, Valencia, CA) from 100 uL aliquots of each homogenate. 16S rDNA segments were PCR amplified and cloned as described [13]. Insert DNA was sequenced from 16 clones derived from each of the two homogenates from each animal, and each sequence was attributed to species (\geq 99% identity) or genus (\geq 97% identity) based on BLAST GenBank similarity [24].

Results

Experiment 1

M. ovipneumoniae infection of DS #00, introduced into pen 1 to start the experiment, was confirmed by positive nasal swab samples obtained on days 1, 4, and 7 after inoculation prior to its introduction into pen #1, and on days 1, 2, 4, 7, 14, 21, 28, 60 and 90 after its introduction into pen #1, confirming that the experimental colonization had been successful and maintained throughout experiment 1. M. ovipneumoniae was first detected in the bighorn sheep (BHS #82) commingled with DS #00 in pen #1 on day 28, and subsequent tests on days 60 and 90 were also positive. BHS #82 developed signs of respiratory disease including nasal discharge (onset day 37); coughing and fever (onset day 42); and lethargy and ear paresis (onset day 61) (Figure 1a). Signs of respiratory disease were observed in the bighorn sheep in pens #2(BHS #89) and #3 (BHS #07) beginning on days 62 and 67, respectively; these signs also included fever, lethargy, paroxysmal coughing, nasal discharge, head shaking, and drooping ears. No signs of respiratory disease were observed in the commingled domestic sheep at any time during the experiment. M.

Table 1. Primers and PCR reaction targets used in these experiments.

Pathogen/Virulence					
gene	Target	Primer Name	Sequence (5' \rightarrow 3')	Size (bp)	Reference
M. haemolytica	gcp	MhgcpF	AGA GGC CAA TCT GCA AAC CTC G	267	[33]
		MhgcpR	GTT CGT ATT GCC CAA CGC CG		
M. haemolytica	gcp	MhgcpF2	TGG GCA ATA CGA ACT ACT CGG G	227	[34]
		MhgcpR2	CTT TAA TCG TAT TCG CAG		
B. trehalosi	sodA	BtsodAF	GCC TGC GGA CAA ACG TGT TG	144	[33]
		BtsodAR	TTT CAA CAG AAC CAA AAT CAC GAA TG		
P. multocida	kmt1	KMT1T7	ATC CGC TAT TTA CCC AGT GG	460	[35]
		KMT1SP6	GCT GTA AAC GAA CTC GCC AC		
Pasteurellaceae leukotoxin	lktA	lktAF	TGT GGA TGC GTT TGA AGA AGG	1,145	[36]
		lktAR	ACT TGC TTT GAG GTG ATC CG		
M. haemolytica leukotoxin	lktA	IktAF set-1	CTT ACA TTT TAG CCC AAC GTG	497	[34]
		IktAR set-1	TAA ATT CGC AAG ATA ACG GG		
Mycoplasma ovipneumoniae	16s rDNA	LMF	TGA ACG GAA TAT GTT AGC TT	361	[20,21]
		LMR	GAC TTC ATC CTG CAC TCT GT		
Mycoplasma ovipneumoniae	16S-23S IGS	MolGSF	GGA ACA CCT CCT TTC TAC GG	Variable~490	[23]
		MolGSR	CCA AGG CAT CCA CCA AAT AC		

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ovipneumoniae was detected in nasal swab samples from all bighorn and domestic sheep in pens #2 and #3 when sampled on day 70. The bighorn sheep were euthanized for necropsy on days 93 (BHS #89) and 99 (BHS #82 and #07). At necropsy, significant abnormal findings were limited to the respiratory tract. Bronchopneumonia affecting 25–50% of the lung volume was observed in all three bighorn sheep (Figure 2). Histopathological examination revealed peribronchiolitis with large lymphoid cuffs, bronchiectasis with purulent exudates, pulmonary atelectasis, and hyperplastic bronchial epithelia lacking visible cilia (Figure 2).

Experiment 2

On day 21 following release of the inoculated bighorn into pen #1, *M. ovipneumoniae* was detected in the inoculated animal and one pen mate (BHS #38 and #39); the third animal (BHS #40) evaded capture and sampling on that day. The first signs of respiratory disease were observed in pen #1 animals on day 21 during drive net capture for sampling, apparently triggered by exertion (Figure 2a). On day 34, inoculated BHS #39 died in pen

#1. On day 49, signs of respiratory disease were first observed in the bighorn sheep in pen #2 (Figure 2b). On days 65 and 109, #41, and #42 in pen #2 died or were euthanized in extremis. The surviving three bighorn sheep exhibited varying degrees of respiratory disease: BHS #38 showed persistent respiratory disease, while BHS #40 and #C showed decreasing respiratory disease over time, which became minimal after days 161 and 154, respectively. On day 204, the three surviving bighorn sheep were euthanized for necropsy. At necropsy, significant abnormal findings were limited to the respiratory tract. All six bighorn sheep had bronchopneumonia, with consolidation of lung tissue volumes ranging from an estimated 5% (BHS #40) to 80–100% (BHS #41) (Figure 2). Histopathological examination revealed severe peribronchiolitis with large lymphoid cuffs as seen in experiment 1. Animals that died or were euthanized in extremis had an overlying necrotizing bronchiolitis (#39) or abscessing bronchiolitis with bronchiectasis (BHS #41, #42) (Figure 2).



Figure 1. Clinical signs exhibited by *M. ovipneumoniae* infected bighorn sheep. Clinical scores (3-day moving averages) of bighorn sheep following introduction of *M. ovipneumoniae*: A) Experiment 1, 3 separate pens; solid line, Pen 1, BHS #82; dashed line, Pen 2, BHS #89; dotted line, Pen 3, BHS #07; B) Experiment 2, Pen 1: solid line, BHS #39 (died day 34); dashed line, BHS #40; dotted line; BHS #38.; C) Experiment 2, Pen 2: solid line, BHS #41 (died day 65); dashed line, BHS #C. doi:10.1371/journal.pone.0110039.q001



Figure 2. Gross and histologic lesions in lungs of bighorn sheep experimentally infected with *M. ovipneumoniae*. Images of BHS #82 (A, B), BHS #39 (C, D), BHS #C (E, F) and BHS #42 (G, H). Original magnification of histologic images was 200X (B, D, H) or 100X (F). doi:10.1371/journal.pone.0110039.g002

Microbiology

All bighorn sheep in both experiments seroconverted to *M. ovipneumoniae* (Table 2). Most experimental animals had neutralizing antibody to parainfluenza-3 virus, but no significant changes in antibody titers were observed during the experimental period. Detectable antibody to other ovine respiratory viruses, including border disease virus, ovine progressive pneumonia virus, and respiratory syncytial virus was occasionally observed in single samples.

M. ovipneumoniae was detected at necropsy in both upper and lower respiratory tracts of all bighorn sheep except BHS #40 whose lung tissues were PCR negative and whose upper

respiratory samples were PCR indeterminate (Table 3). Aerobic cultures and/or PCR tests identified *B. trehalosi* from pneumonic lung tissues from all bighorn sheep in both experiments (Table 3). *B. trehalosi* isolates from BHS #82 and #07 carried *lktA* and expressed leukotoxin activity (Table 3). *P. multocida* and *M. haemolytica* were not detected in these animals by either aerobic culture or PCR.

Culture independent survey of bacteria in pneumonic bighorn sheep lung tissues

DNA sequences of cloned 16S rDNA revealed that the predominant bacterial species in pneumonic sections of lung were

Table 2. Antibody responses to M. ovipneumoniae and parainfluenza-3 (PI-3) virus.

			M. ovipneum	oniae ¹	PI-3 virus ²	
Experiment	ID	Pen	Pre ³	Post ³	Pre ³	Post ³
1	82	1	-8%	93%	512	512
1	89	2	-7%	88%	128	128
1	07	3	-1%	92%	256	512
2	38	1	-6%	74%	Neg	64
2	39	1	-13%	67%	Neg	<32
2	40	1	-23%	75%	64	512
2	41	2	-19%	82%	512	NT
2	42	2	-11%	82%	256	NT
2	С	2	-4%	66%	256	512

¹*M. ovipneumoniae* antibody detected by cELISA, expressed as percentage inhibition of the binding of an agent-specific monoclonal antibody [14,18]. ²PI-3 virus neutralizing antibody detected by virus neutralization [37].

³Pre samples in experiment 1 were obtained on the day that the *M. ovipneumoniae* colonized domestic sheep was introduced to pen 1 and in experiment 2 were obtained on the day that BHS #39 was inoculated with *M. ovipneumoniae*. 'Post' samples in both experiments were obtained at necropsy. Neg = No titer detected. NT = Not tested, due to inadequate specimen volume.

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diverse (Table 4). In experiment 1, *M. ovipneumoniae* was detected in the lung tissues of all animals. *B. trehalosi* also comprised substantial proportions of the pneumonic lung flora in two animals (BHS #82 and #07), while obligate anaerobic species, primarily Fusobacterium spp., predominated in the third animal (BHS #89). The flora identified in the pneumonic lungs of the animals in experiment 2 was also substantially comprised of mixed obligate anaerobes especially *Fusobacterium* spp. (Table 4).

Molecular epidemiology of respiratory pathogens. Consistent with epidemic transmission, *M. ovipneumoniae* strains recovered from all experimental sheep within each experiment shared identical IGS DNA sequences with the respective challenge inoculum (GenBank HQ615162 in experiment 1; KJ551511 in experiment 2).

Discussion

The most striking finding of these experiments was the high transmissibility of M. *ovipneumoniae* and the consistent development of pneumonia that followed infection of bighorn sheep. The bacterium was naturally transmitted from single experimentally inoculated animals (a domestic sheep in experiment 1 and a bighorn sheep in experiment 2) to all animals within and between pens up to 12 m distant. Eight of nine bighorn sheep exposed to M. *ovipneumoniae* developed severe bronchopneumonia and three died, while all the domestic sheep remained healthy.

Previous experimental challenge studies conducted with M. haemolytica or B. trehalosi in the absence of M. ovipneumoniae have not documented transmission. For example, Foreyt et al. [8]

Table 3. Microbiologic findings from pneumonic lung tissues, based on aerobic culture and species specific PCR.

Expt.	ID	Bacterial patho	gens identified in pneu	imonic lung	tissues	
		B. trehalosi	M. haemolytica	lktA	M. ovipneumoniae	Other ⁵
1	82	Cult, sodA ¹	Neg ²	Pos ³	165 ⁴	None
1	89	Cult, sodA	Neg	Neg ³	16S	Pasteurella sp.⁵
1	07	Cult, sodA	Neg	Pos	16S	Pasteurella sp.
2	38	Cult, sodA	Neg	Neg	16S	Pasteurella sp.
2	39	NT, sodA	NT, Neg ²	Neg	16S	NT ⁵
2	40	Cult	Neg	Neg	Neg ⁴	Trueperella pyogenes⁵
2	41	Cult, sodA	Neg	Neg	16S	None
2	42	Cult	Neg	Neg	16S	None
2	С	Cult	Neg	Neg	16S	Pasteurella sp.

¹Cult = *B. trehalosi* detected by bacterial culture; *sodA* = *B. trehalosi* detected by *sodA* species-specific PCR (Table 1); NT = Unable to test by bacterial culture (overgrowth by *Proteus* sp.).

²Neg = *M. haemolytica* not detected by either bacterial culture or by PCR with either *gcp* primer set (Table 1); NT = Unable to test by bacterial culture (overgrowth by *Proteus* sp.).

³Neg = Pasteurellaceae *lktA* not detected in DNA extracts from pneumonic lung tissues by two different *lktA* PCRs (Table 1) [34,36]. Pos = *lktA* detected in *B. trehalosi* isolates obtained from BHS #82 and #07 [36].

 $^{4}16S = M$. ovipneumoniae detected by PCR (Table 1) [20]; Neg = M. ovipneumoniae not detected by PCR.

⁵*Pasteurella* sp., *Trueperella pyogenes* = Bacteria isolated and identified by aerobic culture; *Pasteurella* sp. were determined not to be *B. trehalosi, M. haemolytica,* or *P. multocida*; NT = Unable to test by bacterial culture due to overgrowth by *Proteus* sp.

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xpt.	٩	Bacterial species ident	ified in pneumonic lung tis	sues			
		Btre ¹	Movi ¹	Fuso ¹	Prev ¹	Porphyro ¹	Other ¹
	82	20 (62.5) ²	8 (25)	0	3 (9.4)	0	1 (3.1)
	89	1 (3.1)	7 (21.9)	21 (65.6)	1 (3.1)	0	2 (6.3)
	07	16 (50.0)	12 (37.5)	0	0	0	4 (12.5)
	38	4 (7.1)	2 (3.6)	8 (14.3)	20 (35.7)	9 (16.1)	13 (23.2)
	υ	0	0	17 (30.4)	5 (8.9)	19 (33.9)	15 (26.8)
	39	2 (6.3)	0	24 (75.0)	0	0	6 (18.8)
	40	0	0	0	0	0	56 (100.0)
	41	1 (3.1)	0	21 (65.6)	5 (15.6)	0	5 (15.6)
	42	0	0	31 (96.9)	0	0	1 (3.1)

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M. ovipneumoniae-Induced Bighorn Sheep Pneumonia

reported a series of three experiments in which commingled bighorn sheep were either challenged with intra-tracheal *M. haemolytica* or given sterile BHI as controls. Four of the five control bighorn sheep survived without evidence of disease while commingled with eight *M. haemolytica*-challenged bighorn sheep, of which seven died of pneumonia [8]. Commingled bighorn sheep also remained healthy in several other studies where individual bighorn sheep died with apparent *M. haemolytica* bronchopneumonia (confirmed by isolation of this bacterium from lung tissues) [15,25,26].

In addition to high transmissibility, the time course of disease development and the predominant microbiology of the pneumonic lung tissues following experimental introduction of M. ovipneumoniae differed from that seen in previous bighorn sheep challenge experiments with other respiratory pathogens. Bighorn sheep directly challenged with leukotoxin positive *M. haemolytica* or *B.* trehalosi develop peracute bronchopneumonia and >90% die within a week of challenges with 10^5 cfu or more [16,27–30]. In contrast, disease following experimental M. ovipneumoniae exposures was considerably slower in onset (14-21 days post infection) and development (deaths occurring 34 to 109 days post infection; respiratory disease persisted up to 6 months postinfection); this slow time course closely resembles that documented previously in bighorn lamb pneumonia outbreaks [13]. After lethal M. haemolytica challenge, the agent is typically isolated from lung tissues in high numbers and pure cultures [15,25]; in contrast in naturally occurring pneumonia outbreaks M. ovipneumoniae may be predominant early in the disease course but 16S library analyses have been used to document its overgrowth by diverse other bacteria later in the disease course [14,23]. Although the numbers of animals in the experimental M. ovipneumoniae infection studies reported here are small, the results are consistent with the trend for early predominance of M. ovipneumoniae followed by overgrowth by diverse other bacterial later in the disease course (Tables 3 and 4) [13,14,23].

Our results also differ from our previous attempt to experimentally reproduce respiratory disease by challenge inoculation of 1-week-old bighorn lambs with M. ovipneumoniae, which produced minor lesions and seroconversion but no clinically significant respiratory disease [13]. However, laboratory passage of *M. ovipneumoniae* (as was performed in that experiment) has been reported to attenuate virulence in M. ovipneumoniae [31]. Challenge of bighorn sheep with un-passaged M. ovipneumoniae produced different results, as observed here in experiment #2. In another study [16], nasal washings from domestic sheep naturally colonized with M. ovipneumoniae or lung homogenates from a M. ovipneumoniae-infected bighorn sheep were used for challenge of bighorn sheep after ceftiofur treatment to eliminate detectable Pasteurellaceae. Consistent with increased virulence of unpassaged M. ovipneumoniae, infection and respiratory disease signs were observed in all four bighorn sheep, one of which died 19 days following challenge. The three surviving animals continued to exhibit respiratory disease signs for 42 days, at which time the experiment was terminated by challenge with M. haemolytica (using a dose documented to be rapidly fatal to bighorn sheep even in the absence of M. ovipneumoniae) [16]. As a result, the longer term effects of the mycoplasma infection were not determined in that study. Therefore, the experiments reported here are the first in which naïve bighorn sheep were exposed to un-passaged M. ovipneumoniae and then followed over a time period comparable with the naturally occurring disease course.

The possibility of viral agents contributing to the disease observed in this study cannot be completely ruled out, since the inoculum was derived from nasal washings from domestic sheep and no virucidal treatments were applied. However, a previous study using ultrafiltrates of bighorn sheep pneumonic lung tissues or nasal washings from domestic sheep failed to reproduce any respiratory disease in inoculated susceptible bighorn sheep [16]. In addition, serologic monitoring for the predominant domestic sheep respiratory viruses did not demonstrate seroconversion of the experimental animals in this study, as described in the Results and in Table 2. Therefore, the most parsimonious interpretation of the data presented here is that the disease observed resulted from *M. ovipneumoniae* infection and the sequelae of that infection.

The transmission of *M. ovipneumoniae* from pen-to-pen in these experiments strongly suggests that direct contact is not necessary for epizootic spread of pneumonia in bighorn sheep. Feeding, watering and other procedures involving animal care or research staff were designed to minimize the risk of human or fomitemediated transmission of the pathogen from pen to pen, although we recognize it is impossible to completely rule out this possibility. On the other hand, since aerosolized droplet transmission is recognized as a transmission route for the closely related bacterium, Mycoplasma hyopneumoniae (the cause of atypical pneumonia of swine) [32], it is plausible that a similar transmission mode occurs with M. ovipneumoniae. Infectious aerosols generated by coughing animals would likely contribute to the explosive nature of the pneumonia outbreaks observed following initial introduction of M. ovipneumoniae into naïve bighorn sheep populations.

In conclusion, we demonstrated that experimental *M. ovipneumoniae* infection of naïve bighorn sheep induces chronic, severe bronchopneumonia associated with multiple secondary bacterial infections and that this infection spread rapidly to animals both within the same pen and to animals in nearby pens. The significance of these findings would be clarified by parallel experiments specifically designed to determine transmissibility and associated disease outcomes in other agents associated with bighorn sheep pneumonia, particularly *M. haemolytica*, in the absence of *M. ovipneumoniae*. Furthermore, the case-fatality rates of *M. ovipneumoniae* infected animals described here contrasts

References

- Festa-Bianchet M (2008) Ovis canadensis. The IUCN Red List of Threatened Species. Available: http://www.iucnredlist.org/details/summary/15735/0. Accessed 2014. Jul 24.
- Cassirer EF, Plowright RK, Manlove KR, Cross PC, Dobson AP, et al. (2012) Spatio-temporal dynamics of pneumonia in bighorn sheep (*Ovis canadensis*). J Animal Ecol 82: 518–528.
- Marsh H (1938) Pneumonia in Rocky Mountain bighorn sheep. J Mammal 19: 214–219.
- Buechner HK (1960) The bighorn sheep in the United States, its past, present, and future. Wildl Monog 4: 3–174.
- Forrester DJ (1971) Bighorn sheep lungworm-pneumonia complex. In: Davis JW, Anderson RC, editors. Parasitic Diseases of Wild Mammals. Ames, IA: Iowa State University Press. 158–173.
- Demartini JC, Davies RB (1977) An epizootic of pneumonia in captive bighorn sheep infected with *Muellerius* sp. J Wildl Dis 13: 117–124.
- Miller MW (2001) Pasteurellosis. In: Williams ES, Barker, I K., editor. Infectious diseases of wild mammals. Ames IA USA: Iowa State University Press. 558.
- Foreyt WJ, Snipes KP, Kasten RW (1994) Fatal pneumonia following inoculation of healthy bighorn sheep with *Pasteurella haemolytica* from healthy domestic sheep. J Wildl Dis 30: 137–145.
- Kraabel BJ, Miller MW, Conlon JA, McNeil HJ (1998) Evaluation of a multivalent *Pasteurella haemolytica* vaccine in bighorn sheep: Protection from experimental challenge. J Wildl Dis 34: 325–333.
- Rudolph KM, Hunter DL, Foreyt WJ, Cassirer EF, Rimler RB, et al. (2003) Sharing of *Pasteurella* spp. between free-ranging bighorn sheep and feral goats. J Wildl Dis 39: 897–903.
- Rudolph KM, Hunter DL, Rimler RB, Cassirer EF, Foreyt WJ, et al. (2007) Microorganisms associated with a pneumonic epizootic in Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*). J Zoo Wildl Med 38: 548–558.
- Weiser GC, DeLong WJ, Paz JL, Shafii B, Price WJ, et al. (2003) Characterization of *Pasteurella multocida* associated with pneumonia in bighorn

with the nearly 100% mortality that follows experimental commingling of bighorn sheep with presumptively or documented M. ovipneumoniae-positive domestic sheep and suggests an important role for polymicrobial secondary infections in determining mortality rates, which could be investigated in future studies. Finally, M. ovipneumoniae was still detected in nasal swab samples of several surviving bighorn sheep that were euthanized at the completion of these studies, suggesting that survivors of naturally occurring pneumonia outbreaks may continue to carry and shed this agent in nasal secretions. Such carriage may provide a mechanism for the post-invasion disease epizootics in lambs described in free-ranging populations. If so, this presumptive carrier state requires further study to characterize the factors that determine its occurrence and persistence, as these may be critical for the development of effective management control measures for this devastating disease.

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Author Contributions

Conceived and designed the experiments: TEB EFC JLO S. Srikumaran WJF. Performed the experiments: TEB EFC JLO KAP KL S. Shanthalingam. Analyzed the data: TEB EFC KAP KL. Contributed reagents/materials/analysis tools: TEB EFC KAP KL S. Shanthalingam S. Srikumaran. Contributed to the writing of the manuscript: TEB EFC KAP S. Shanthalingam S. Srikumaran WJF.

- Besser TE, Cassirer EF, Potter KA, VanderSchalie J, Fischer A, et al. (2008) Association of Mycoplasma ovipneumoniae infection with population-limiting respiratory disease in free-ranging rocky mountain bighorn sheep (Ovis canadensis canadensis). J Clin Microbiol 46: 423–430.
- Besser TE, EF Cassirer, MA Highland, P Wolff, A Justice-Allen, et.al. (2012) Bighorn sheep pneumonia: Sorting out the cause of a polymicrobial disease. Prev Vet Med 108: 85–93.
- Besser TE, Cassirer EF, Yamada C, Potter KA, Herndon C, et al. (2012) Survival of Bighorn Sheep (*Ovis canadensis*) Commingled with Domestic Sheep (*Ovis aries*) in the Absence of *Mycoplasma ovipneumoniae*. J Wildl Dis 48: 168– 172.
- Dassanayake RP, Shanthalingam S, Herndon CN, Subramaniam R, Lawrence PK, et al. (2010) *Mycoplasma ovipneumoniae* can predispose bighorn sheep to fatal *Mannheimia haemolytica* pneumonia. Vet Microbiol 145: 354–359.
- Blodgett R (2010) Bacteriologic Analytical Manual Appendix 2: Most Probable Number from Serial Dilutions. Washington DC. Available: http://www.fda. gov/Food/FoodScience Research/LaboratoryMethods/ucm109656.htm. Accessed 2014 Jul 24.
- Ziegler JC, Lahmers KK, Barrington GM, Parish SM, Kilzer K, et al. (2014) Safety and Immunogenicity of a Mycoplasma ovipneumoniae Bacterin for Domestic Sheep (*Ovis aries*). PLoS One 9(4): e95698.
- Herrmann LM, Cheevers WP, Marshall KL, McGuire TC, Hutton MM, et al. (2003) Detection of serum antibodies to ovine progressive pneumonia virus in sheep by using a caprine arthritis-encephalitis virus competitive-inhibition enzyme-linked immunosorbent assay. Clin Diag Lab Immunol 10: 862–865.
- Lawrence PK, Shanthalingam S, Dassanayake RP, Subramaniam R, Herndon CN, et al. (2010) Transmission of *Mannheimia haemolytica* from domestic sheep (*Ovis aries*) to bighorn sheep (*Ovis canadensis*): unequivocal demonstration with green fluorescent protein-tagged organisms. J Wildl Dis 46: 706–717; erratum, J Wildl Dis 46: 1346.

sheep. J Wildl Dis 39: 536-544.

- McAuliffe L, Hatchell FM, Ayling RD, King AI, Nicholas RA (2003) Detection of *Mycoplasma ovipneumoniae* in *Pasteurella*-vaccinated sheep flocks with respiratory disease in England. Vet Rec 153: 687–688.
- Gentry MJ, Srikumaran S. (1991) Neutralizing monoclonal antibodies to *Pasteurella haemolytica leukotoxin affinity purify the toxin from crude culture* supernatants. Microb Pathog 10: 411–417.
- Besser TE, Highland M, Baker K, Anderson NJ, Ramsey JM (2012) Causes of pneumonia epizootics among bighorn sheep, western United States, 2008–2010. Emerg Infect Dis 18: 406–414.
- Petti CA (2007) Detection and identification of microorganisms by gene amplification and sequencing. Clin Infect Dis 44: 1108–1114.
- Foreyt WJ, Jenkins EJ, Appleyard GD (2009) Transmission of lungworms (*Muellerius capillaris*) from domestic goats to bighorn sheep on common pasture. J Wildl Dis 45: 272–278.
- Foreyt WJ, Lagerquist JE (1996) Experimental contact of bighorn sheep (Ovis canadensis) with horses and cattle, and comparison of neutrophil sensitivity to Pasteurella haemolytica cytotoxins. J Wildl Dis 32: 594–602.
- Onderka DK, Rawluk SA, Wishart WD (1988) Susceptibility of Rocky Mountain bighorn sheep and domestic sheep to pneumonia induced by bighorn and domestic livestock strains of *Pasteurella haemolytica*. Can J Vet Res 52: 439–444.
- Dassanayake RP, Shanthalingam S, Herndon CN, Lawrence PK, Cassirer EF, et al. (2009) Mannheimia haemolytica serotype A1 exhibits differential pathogenicity in two related species, Ovis canadensis and Ovis aries. Vet Microbiol 133: 366–371.
- Subramaniam R, Shanthalingam S, Bavananthasivam J, Kugadas A, Potter KA, et al. (2011) A multivalent *Mannheimia-Bibersteinia* vaccine protects bighorn

sheep against Mannheimia haemolytica challenge. Clin Vaccine Immunol 18: 1689–1694.

- Foreyt WJ, Snipes KP, Kasten RW (1994) Fatal pneumonia following inoculation of healthy bighorn sheep with *Pasteurella haemolytica* from healthy domestic sheep. J Wildl Dis 30: 137–145.
- Alley MR, Ionas G, Clarke JK (1999) Chronic non-progressive pneumonia of sheep in New Zealand - a review of the role of *Mycoplasma ovipneumoniae*. N Z Vet J 47: 155–160.
- 32. Desrosiers R (2011) Transmission of swine pathogens: different means, different needs. Anim Health Res Rev 12: 1–13.
- Dassanayake RP, Call DR, Sawant AA, Casavant NC, Weiser GC, et al. (2010) Bibersteinia trehalosi inhibits the growth of Mannheimia haemolytica by a proximity-dependent mechanism. Appl Environ Microbiol 76: 1008–1013.
- 34. Shanthalingam S, Goldy A, Bavananthasivam J, Subramaniam R, Batra SA, et al. (2014) PCR assay detects *Mannheimia haemolytica in culture-negative pneumonic lung tissues of bighorn sheep (Ovis canadensis)* from outbreaks in the Western USA, 2009–2010. J Wildl Dis 50: 1–10.
- Townsend KM, Frost AJ, Lee CW, Papadimitriou JM, Dawkins HJ (1998) Development of PCR assays for species- and type-specific identification of *Pasteurella multocida* isolates. J Clin Microbiol 36: 1096–1100.
- Fisher MA, Weiser GC, Hunter DL, Ward ACS (1999) Use of a polymerase chain reaction method to detect the leukotoxin gene IktA in biogroup and biovariant isolates of *Pasteurella haemolytica* and *P. trehalosi*. Am J Vet Res 60: 1402–1406.
- Rossi CR, Kiesel GK (1971) Microtiter tests for detecting antibody in bovine serum to parainfluenza 3 virus, infectious bovine rhinotracheitis virus, and bovine virus diarrhea virus. Appl Microbiol 22: 32–36.

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Phylogeographic and population genetic structure of bighorn sheep (*Ovis canadensis*) in North American deserts

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Fossil data are ambiguous regarding the evolutionary origin of contemporary desert bighorn sheep (Ovis canadensis subspecies). To address this uncertainty, we conducted phylogeographic and population genetic analyses on bighorn sheep subspecies found in southwestern North America. We analyzed 515 base pairs of mtDNA control region sequence and 39 microsatellites in 804 individuals from 58 locations. Phylogenetic analyses revealed 2 highly divergent clades concordant with Sierra Nevada (O. c. sierrae) and Rocky Mountain (O. c. canadensis) bighorn and showed that these 2 subspecies both diverged from desert bighorn prior to or during the Illinoian glaciation (\sim 315–94 thousand years ago [kya]). Desert bighorn comprised several more recently diverged haplogroups concordant with the putative Nelson (O. c. nelsoni), Mexican (O. c. mexicana), and Peninsular (O. c. cremnobates) subspecies. Corresponding estimates of effective splitting times (~17-3 kya), and haplogroup ages (~85-72 kya) placed the most likely timeframe for divergence among desert bighorn subspecies somewhere within the last glacial maximum. Median-joining haplotype network and Bayesian skyline analyses both indicated that desert bighorn collectively comprised a historically large and haplotype-diverse population, which subsequently lost much of its diversity through demographic decline. Using microsatellite data, discriminant analysis of principle components (DAPC) and Bayesian clustering analyses both indicated genetic structure concordant with the geographic distribution of 3 desert subspecies. Likewise, microsatellite and mitochondrial-based F_{ST} comparisons revealed significant fixation indices among the desert bighorn genetic clusters. We conclude these desert subspecies represent ancient lineages likely descended from separate Pleistocene refugial populations and should therefore be managed as distinct taxa to preserve maximal biodiversity.

Los datos de fósiles sobre el origen evolutivo de las ovejas del desierto (*Ovis canadensis* subespecies) contemporáneas son ambiguos. Para dilucidar esta incertidumbre, llevamos a cabo análisis filogeográficos y de genética de poblaciones entre cinco subespecies de ovejas del suroccidente de Norteamérica. Analizamos 515 pb de secuencia de la región control del ADN mitocondrial y 39 microsatélites en 804 ovejas de 58 localidades. Los análisis filogenéticos revelaron 2 clados altamente divergentes concordantes con ovejas de la Sierra Nevada (*O. c. sierrae*) y de las Montañas Rocosas (*O. c. canadensis*), y demostraron que estas dos subespecies divergieron antes o durante la glaciación de Illinois (315,000–94,000 años). Las ovejas del desierto formaron varios haplogrupos recientemente derivados concordantes con las subespecies de Nelson (*O. c. nelsoni*), México (*O. c. mexicana*) y peninsular (*O. c. cremnobates*). Las estimaciones correspondientes al tiempo de separación efectiva (17,000–3,000 años) y edades de haplogrupos (85,000–72,000 años) son los plazos más probables para las divergencias entre subespecies de ovejas del desierto dentro de la última glaciación máxima. Análisis de redes de haplotipos de unión de medias y análisis bayesianos de líneas de horizonte indicaron que las ovejas del desierto formaron una población históricamente grande y diversa en términos de haplotipos, que luego perdieron gran parte de su diversidad a través

de un descenso demográfico. Utilizando datos de microsatélites los análisis DAPC y TESS indicaron agrupamiento genético concordante con la distribución geográfica actual de las tres subespecies. Asimismo, comparaciones de $F_{\rm ST}$ con datos de microsatélites y mitocondriales revelaron índices de fijación significativos entre los grupos genéticos de ovejas del desierto. Concluimos que estas subespecies de ovejas del desierto representan linajes antiguos que probablemente descienden de poblaciones de distintos refugios del Pleistoceno, y que por lo tanto deben ser manejadas como taxones distintos para preservar su biodiversidad máxima.

Key words: desert bighorn sheep, desert southwest, divergence date, glacial refugia, haplotype, microsatellites, mtDNA, *Ovis canadensis*, phylogeography, subspecies

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Bighorn sheep (*Ovis canadensis* Shaw, 1804) are native to the deserts of southwestern North America (hereafter, desert southwest), as well as the adjacent and climatically distinct alpine zones of the Sierra Nevada and Rocky Mountain ranges. Once abundant, bighorn sheep suffered widespread local extinction following European settlement as a result of overharvest, livestock-transmitted disease, and habitat loss and fragmentation (Seton 1929; Buechner 1960; Valdez and Krausman 1999). Ongoing efforts to restore bighorn sheep throughout their native range, particularly in the desert southwest, have relied heavily on translocations (Rowland and Schmidt 1981; Bleich et al. 1990; Singer et al. 2000; Boyce et al. 2011). However, such actions require thorough understanding of both the taxonomy and phylogeographic structure among populations (Weeks et al. 2011).

Significant taxonomic revision of O. canadensis at the subspecific level has occurred during the past several decades, yet phylogenetic relationships have not been adequately tested with modern molecular methods. Currently recognized subspecies include California (O. c. californiana; not considered in this study), Rocky Mountain (O. c. canadensis), and Sierra Nevada (O. c. sierrae) bighorn, as well as disputed subspecies designations among desert populations. Reference texts (Wilson and Reader 2005) continue to use the morphology-based designations of Cowan (1940), recognizing 4 desert subspecies: Nelson (O. c. nelsoni), Mexican (O. c. mexicana), Peninsular (O. c. cremnobates), and Weems (O. c. weemsi) bighorn. However, subsequent morphometric studies questioned these subspecies as artifacts of small sample size and age-related size differences (Bradley and Baker 1967; Wehausen and Ramey 1993). Further, a restriction fragment length polymorphism (RFLP) study of mitochondrial DNA (mtDNA) failed to resolve these subspecies (Ramey 1995). As a result, Wehausen and Ramey (1993) proposed desert bighorn be synonymized to a single taxon (O. c. nelsoni).

Lack of a consistent taxonomy has created confusion among managers and conservation biologists. For instance, Peninsular bighorn sheep were designated threatened by the State of California in 1984 as *O. c. crembobates*. Since then, Peninsular bighorn have been provisionally synonymized with *O. c. nelsoni* (Wehausen and Ramey 1993) and were listed under the Endangered Species Act in 1999 (63 FR 13134), yet are protected as a distinct population segment. Ultimately, subspecies designations are valuable to conservation if they serve as commonly understood indicators of significant genetic variation and potential local adaptation that could be lost if mismanaged (i.e., translocated) as a single taxon. An updated genetic characterization of bighorn sheep occupying the desert southwest should therefore help inform taxonomy and management by examining how patterns of genetic variation compare with competing hypotheses regarding subspecies.

Achieving clarity regarding the phylogenetic history, and ultimately taxonomy, of desert bighorn sheep requires a basic understanding of the evolutionary history of the taxon. Unfortunately, the fossil record is somewhat ambiguous regarding the origin of contemporary desert bighorn in the desert southwest. Fossil evidence indicates Ovis continuously occupied at least 2 late Pleistocene glacial refugia in southern North America: 1 in the current Mojave Desert, established ~300 thousand years ago (kya), prior to the Illinoian glaciation (Jefferson 1991), and another in the north near Natural Trap Cave, Wyoming (Martin and Gilbert 1978; Wang 1988), established during the Sangamon interglacial (~100 kya). However, competing hypotheses regarding the origins of desert bighorn sheep relative to these refugial populations cannot be eliminated based on fossil geochronology (Geist 1985). The 1st hypothesis proposes that Ovis from the northern refugium spread south, ultimately joining or displacing sheep from the Mojave refugium to give rise to contemporary desert populations. The 2nd hypothesis proposes that the northern colonizers were outcompeted and replaced by Ovis expanding from the Mojave refugium. These hypotheses provide clear alternatives that are testable using phylogenetic methods. Predictions following from the 1st hypothesis include: 1) contemporary desert bighorn populations should exhibit haplotypes recently diverged from contemporary Rocky Mountain bighorn haplotypes-i.e., since the last glacial maximum (LGM); 2) these derived desert haplotypes should represent only a subset of the lineages (i.e., founder effect) reflected in the Rocky Mountain population, and 3) these northern-derived desert haplotypes potentially occur in association with more deeply divergent (pre-Illinoian) haplotypes originating from the Mojave refugium. Predictions following from the 2nd hypothesis include: 1) all haplotypes in contemporary desert bighorn populations belong to 1 or more lineages that are deeply divergent (pre-Illinoian) from those occurring in Rocky Mountain bighorn populations, and 2) the existence of more than 1 such lineage would provide evidence that multiple southern refugia contributed to colonization of the desert southwest.

In this study, we characterized the phylogeographic and genetic structure of bighorn sheep occupying the desert southwest. We utilized a large number of samples from previously under represented areas of the native range of desert bighorn sheep. For clarity, we utilized the disputed desert subspecies designations of Cowan (1940), as this taxonomy recognizes the greatest number of taxonomic units among which genetic variation could be compared. Our objectives were to 1) use mtDNA control region sequences and nuclear microsatellites to characterize phylogeographic and population genetic variation both within desert bighorn and in relation to the Sierra Nevada and Rocky Mountain subspecies, 2) estimate splitting times among subspecies to test fossil recordbased hypotheses regarding colonization of the desert southwest, 3) reconstruct historical demography to estimate the timeframe of population declines, and 4) use these results to evaluate genetic support for competing desert subspecies designations.

MATERIALS AND METHODS

Sample collection.—We used a total of 804 adult bighorn sheep (n = 437 F, 353 M, 14 unknown sex) captured by biologists from state agencies or harvested by hunters from 58 locations across the southwestern United States and northern Mexico, as well as 2 locations in Canada, during 1992-2013 (Fig. 1; Supporting Information S1). Desert bighorn samples (n = 655) were assigned to their geographic regions of origin, including the Peninsular Ranges, Transverse Ranges, Mojave, Sonoran, and Chihuahuan Deserts, Great Basin, and Colorado Plateau. This scheme allowed us to test the genetic evidence for competing subspecies designations within desert bighorn sheep without a priori assumptions regarding group membership. In addition to the desert bighorn sheep composing the core of our sample, we also included 52 endemic Sierra Nevada bighorn sheep, as well as 97 Rocky Mountain bighorn sheep from either Canada or (re)introduced populations in northern New Mexico and eastern Arizona (Fig. 1; Supporting Information S1). No samples of California or Weems bighorn sheep were available for inclusion in this study.

Laboratory methods.—Total genomic DNA was extracted from blood, muscle, or skin tissue using Qiagen DNeasy Blood and Tissue kits (Qiagen Inc., Valencia, California) following the manufacturer's protocol. Each sample was genotyped at 39 microsatellite loci described in Buchalski et al. (2015). Sex was confirmed via amplification of the Amelogenin marker described in Weikard et al. (2006). To estimate genotyping error, we randomly selected 30 samples, along with positive and negative controls, to blindly regenotype. We estimated the average error rate per locus as the ratio between the number of



Fig. 1.—Study area within the southwestern United States and northern Mexico, including 58 locations from which bighorn sheep (*Ovis canadensis*) subspecies were sampled. Significant geographic features are depicted as they relate to subspecies ranges. For locations, GMU refers to game management units as defined by the Arizona Game and Fish Department.

single-locus genotypes including at least 1 allelic mismatch and the number of replicated single-locus genotypes (Pompanon et al. 2005).

A fragment of the mitochondrial control region was amplified following the protocol described by Epps et al. (2005). Cycle sequencing was performed bidirectionally using BigDye 3.1 and an ABI 3730 Genetic Analyzer (Applied Biosystems, Foster City, California). Forward sequences were verified with the sequence of the reverse strand using Sequencher 5.1 (Gene Codes Corp.) and incomplete sequences, or those with discrepancies, were reamplified and resequenced. We aligned the sequences in MEGA 6 (Tamura et al. 2013) using the ClustalW algorithm (Thompson et al. 1994) under default settings, at which time we discovered a 75 base pair (bp) repetitive sequence (RS) localized in the left domain near the tRNA^{Pro} gene. All individuals examined had at least 2 copies of the RS, with a limited number ($\sim 5\%$) displaying 3 copies. We normalized the sequences by manually removing the extra RS from those haplotypes that had it and limited our analyses to the 515 bp fragment common to all individuals (see Supporting Information S2 for a full description). Sequences for each novel haplotype were deposited into GenBank (accession nos. KU363638-KU363690).

We used a basic local alignment search tool (BLAST— Altschul et al. 1997) to search the nucleotide database in GenBank for all unique haplotypes present in our data, finding 35 homologous sequences for desert bighorn sheep, including accession nos. AF076911–AF076917 (Boyce et al. 1999), AY903993–AY904017 (Epps et al. 2005), KP688366– KP688368 (Buchalski et al. 2015), and AY116621–AY116623 (unpublished sequences for 2 Mexican and 1 Weems bighorn). We downloaded the archived sequences, preserving the original haplotype names, for inclusion in our phylogenetic analyses.

Range-wide population genetic structure.--We used discriminant analysis of principle components (DAPC-Jombart et al. 2010) to identify population structure among microsatellite genotypes. This method entails no assumptions regarding the cause of structure (i.e., island model versus isolation-bydistance [IBD]) and, in contrast to other clustering approaches (i.e., Pritchard et al. 2000), does not assume Hardy-Weinberg or gametic equilibrium. Analysis was implemented in R 3.0.2 (R Development Core Team 2015) using the package adegenet 1.4-2 (Jombart 2008). The optimal number of genetic clusters (K), was estimated by conducting 10 independent runs of the find.clusters function with the diffNgroup option selected. The number of principal components as predictors for the discriminant analysis was set to 7 following alpha-score optimization (i.e., trade-off between power of discrimination and overfitting; Supporting Information S3). Scatterplots of microsatellite genotypes in relation to discriminant functions were created in adegenet.

We then used TESS 2.3.1 (Chen et al. 2007) to evaluate structure among microsatellite genotypes in a spatially explicit context. Program TESS accounts for spatial autocorrelation in allele frequencies due to IBD by treating sample location coordinates as prior information during estimation of admixture proportions. This allows for differentiation between clinal transitions and abrupt breaks (i.e., contact zones versus barriers) between discrete genetic groups or clusters (Durand et al. 2009; Francois and Durand 2010). We first ran the no-admixture model with 200,000 iterations, of which the initial 100,000 were excluded as burn-in, to test the number of clusters (K)from 2 to 10, with 10 replicates each. A plot of the deviance information criterion (DIC) against K was used to identify the most likely number of clusters. This value was then used in 100 replicate runs of the admixture model, using the same number of iterations as above. Individual cluster memberships from the 10 runs having the highest likelihoods were averaged using CLUMPP 1.1.2 (Jakobsson and Rosenberg 2007) and visualized using DISTRUCT 1.1 (Rosenberg 2004). Predictive maps of each genetic cluster were generated using custom R scripts provided with the TESS software download (http://www-timc. imag.fr/Olivier.Francois/TESS Plot.html).

We used nested hierarchical analysis of molecular variance (AMOVA-Excoffier et al. 1992) to examine the distribution of genetic variation associated with competing desert subspecies designations. In our 1st set of analyses, the Sierra Nevada and Rocky Mountain subspecies were compared to a varying number of groups within desert bighorn sheep. Desert bighorn grouping schemes included 1) the Peninsular, Nelson, and Mexican subspecies of Cowan (1940; K = 5), 2) Peninsular and Nelson bighorn pooled together as suggested by Wehausen and Ramey (1993; K = 4), and 3) all desert bighorn pooled together as implied by Ramey (1995; K = 3). To allow for lesser divergence within desert bighorn sheep in relation to the Sierra Nevada and Rocky Mountain subspecies, we conducted a 2nd set of analyses using desert bighorn only. We tested grouping schemes 1 and 2 based on the rationale above. This analytical design was applied to both microsatellite allele and control region haplotype frequency data in Arlequin 3.5.1.2 (Excoffier and Lischer 2010). Significance was determined from 10,000 permutations of the data.

We then evaluated pairwise differentiation between each genetic cluster identified above. Pairwise F_{ST} values based on microsatellite allele and control region haplotype frequencies were estimated following Weir and Cockerham (1984), as implemented in Arlequin. Ten thousand random permutations were used to test significance, and α for each test was adjusted for multiple comparisons using the modified false discovery rate (FDR) method (Benjamini and Yekutieli 2001).

We wished to quantify the spatial scale of IBD among desert bighorn sheep herds, while avoiding potential biases resulting from past translocations. Therefore, we identified native herds within our sample (n = 23) as those with no history of translocation (i.e., according to Bleich et al. 1990; Cox and Cummings 2005). Geographic distances among native herd locations were calculated in ArcGIS (ESRI, Redlands, California), ln transformed, and converted to a matrix. We then estimated group genetic distances as $F_{ST}/(1 - F_{ST})$ according to Slatkin (1995) for both microsatellite allele and control region haplotype frequencies in Arlequin. Correlations between genetic and geographic distances were determined using Mantel tests in the R package Ecodist (Goslee and Urban 2007). To better visualize the scale over which genetic marker frequencies were spatially autocorrelated, we created Mantel correlograms using distance class sizes of 20 km and the Vegan package (Oksanen et al. 2015). For all tests, correlations were determined using permutation tests with 1,000 randomizations.

Genetic diversity indices.-Indices of population genetic diversity were estimated for each genetic cluster identified above. We used Fisher's exact test (Guo and Thompson 1992) as implemented in Genepop 4.2 (Rousset 2008) to test for departures from Hardy-Weinberg proportions and genotypic linkage equilibrium using 10,000 dememorization steps, 20 batches, and 5,000 iterations per batch. Test results were adjusted for multiple pairwise comparisons using FDR correction. Estimates of the number of alleles per locus (N_{A}) , expected $(H_{\rm E})$ and observed $(H_{\rm O})$ heterozygosity, and the inbreeding coefficient (F_{1S}) were generated in GenAlex (Peakall and Smouse 2012). Allelic richness (A_r) was calculated using the methods of Mousadik and Petit (1996) as implemented in the PopGenReport package (Adamack and Gruber 2014) for R. The number of polymorphic sites, nucleotide diversity (π) , number of haplotypes (H_n) , and haplotype diversity (H_d) were calculated for mtDNA control region sequences using DNAsp 5.10 (Librado and Rozas 2009).

Phylogeographic analyses.—We constructed a phylogenetic tree of unique haplotype sequences in MEGA 6 using the maximum likelihood (ML) algorithm, with support at the nodes calculated from 1,000 bootstrap replicates. Evolutionary distances (i.e., branch lengths) were computed under the Hasegawa–Kishino–Yano (HKY) model of nucleotide substitution (Hasegawa et al. 1985), proportion of invariable sites, and gamma distribution shape (HKY+I+ Γ model), as this was determined to be the best-fitting model according to the Bayesian information criteria (BIC) in MEGA 6. All positions containing alignment gaps and missing data were eliminated from the data set for tree construction (complete deletion option). We used the Snow sheep (*Ovis nivicola*; GenBank accession no. DQ249894) indigenous to Asia as the outgroup.

Phylogenetic relationships among haplotypes were also inferred using median-joining network analysis in Network 4.6.1.3 (Bandelt et al. 1999). Within Network, we used the average number of mutations (rho) separating ancestral and descendent haplotypes (Forster et al. 1996; Saillard et al. 2000) to estimate haplogroup ages within desert bighorn, as well as the time to most recent common ancestor (TMRCA) between desert bighorn and both the Sierra Nevada and Rocky Mountain lineages.

To estimate effective splitting times between subspecies, we modeled the demographic history of bighorn by coalescent simulation in IMa2 (Hey 2010a, 2010b). We computed estimates and associated 95% highest posterior density (HPD) intervals, in terms of mutational accumulation under the HKY mutation model. We estimated only "effective" splitting times (i.e., as if no postdivergence gene flow occurred), rather than testing models that incorporated gene flow, because of the large number of pairwise comparisons and computational time that would

have been required. Therefore, if our assumptions regarding gene flow were incorrect, the resulting estimates would be conservative (i.e., erring toward more recent divergence). We performed replicate runs with different random number seeds for all comparisons to confirm consistency. Validity of results was evaluated based on unimodality of posterior distributions and their tendency to approach zero on both ends, stationarity of parameter estimates and model likelihoods, and the cumulative consistency of numerical estimates with one another and in relation to empirical estimates of net sequence divergence (Nei and Li 1979), which provided an intuitive qualitative check on simulation results.

We also constructed Bayesian skyline plots to infer changes in population size through time for each desert subspecies using BEAST 1.8.2 (Drummond et al. 2005; Drummond and Rambaut 2007). We used a HKY+ Γ model of nucleotide substitution with default (constant) settings and 10 skyline groups. Because our focus was on the intraspecific (evolutionarily recent) divergence among bighorn, we assumed a strict clock throughout (Brown and Yang 2011).

We translated mutation-scaled estimates of time into absolute estimates by multiplying by the expected number of years per mutation event. Previous estimates of mitochondrial mutation rates for Ovis spp. have varied due to different assumptions underlying the external calibrations. The divergence of bighorn sheep from other Ovis spp. was initially assumed to be 5.63 million years ago (My-Hiendleder et al. 1998), yet more recently was estimated to be as recent as 2.42 My (Rezaei et al. 2010), resulting in a 2.33-fold difference in the mutation rate implied for mtDNA. Although we used the control region in this study, cytochrome b (Cytb) has been found to mutate close to 2% per million years (Ma) for a range of large-bodied terrestrial mammals, including bovids (Nabholz et al. 2008). We reviewed the Cytb data available for Ovis spp. (Bunch et al. 2006; Rezaei et al. 2010), which suggested the more recent calibration resulted in a rate close to the expected 2% per Ma. The corresponding mutation rate if recalibrated to the more ancient date would be < 1% per Ma, which we found unrealistic. Therefore, we adopted the more recent date and recalibrated the control region estimates from Hiendleder et al. (1998). Specifically, we estimated the mutation rate and associated variance by averaging (and computing a confidence interval for) the 4 most recent Ovis nodes provided by Hiendleder et al. (2002, n = 4 from table 2, therein). These calculations resulted in an estimate of 6.1%, 95% CI 4.2-7.9% per Ma. Our use of this more recent calibration resulted in more conservative (recent) divergence estimates. All estimates and confidence limits presented here can be recalibrated to the lower (less conservative) rate by multiplying by 2.33 (the ratio of the 2 external calibration points, 5.63Ma/2.42 Ma).

RESULTS

Population genetic structure.—We obtained unique multilocus microsatellite genotypes for 804 individuals and observed agreement between 1,135 of the 1,170 single-locus genotypes
analyzed twice, indicating a genotyping error rate of 3%. The diffNgroups option for the DAPC differentiated microsatellite genotypes into 5 genetic clusters (K = 5) in 8 out of 10 runs. The scatterplot of individual genotypes using 4 discriminant functions indicated the Sierra Nevada and Rocky Mountain subspecies were highly discriminated from desert bighorn and one another, with strong separation visible along the first 2 principle component axes (Fig. 2a). The scatter plot also suggested the presence of hierarchical structure, with apparent substructure among desert bighorn. To further investigate this substructure, we conducted a 2nd DAPC using only desert bighorn genotypes (n = 655). The 3 clusters identified in the 1st DAPC were well discriminated along both axes with no overlap of 95% inertia ellipses (Fig. 2b). The TESS analysis further supported the results of DAPC. Mean DIC values indicated K = 5 as the best clustering option for our data (i.e., piecewise change in function shape at this value; Supporting Information S3). Individual admixture proportions (Fig. 2c) for each cluster indicated clear geographic structure among Sierra Nevada and Rocky Mountain bighorn, as well as desert clusters concordant with the subspecies designations of Cowan (1940), including 1) Peninsular bighorn from the Peninsular Ranges (n = 288), 2) Nelson bighorn from the Transverse Ranges, Mojave Desert, southern Great Basin, and Colorado Plateau (n = 180), and 3) Mexican bighorn from the Sonoran and Chihuahuan Deserts (n = 187; Fig. 2d).

Both TESS and DAPC indicated intermingled Nelson and Mexican bighorn genotypes associated with the northern Sonora Desert, north of the Bill Williams River in Arizona (i.e., location 40; Figs. 1 and 2). The TESS analysis also indicated low-level admixture between the Peninsular and Nelson genetic clusters in the southern Mojave Desert in California (locations 17-26; Fig. 2c). Interestingly, admixture proportions indicated an absence of introgression between Sierra Nevada genotypes and desert bighorn immediately to the east. As expected, Rocky Mountain genotypes occurred at sites of known (re)introduction for this subspecies, both within (eastern Arizona) and adjacent to (northern New Mexico) the native range of desert bighorn sheep (Figs. 2c and d). Admixture proportions indicated introgression of desert bighorn into the Rocky Mountain population in eastern Arizona (location 55), with no evidence of the reverse in adjacent desert bighorn herds (Fig. 2c).

The AMOVAs produced results similar to the DAPC, indicating significant variance among Sierra Nevada, Rocky Mountain, and desert bighorn, with substructure apparent in the latter. For the AMOVA including all samples, outcomes were similar for both mtDNA and microsatellite data (Table 1). Among-group variance was maximized at K = 3, with groups consisting of 1) Sierra Nevada, 2) Peninsular, Nelson, and Mexican, and 3) Rocky Mountain bighorn sheep—with significant (P < 0.001) among-group fixation indices (F_{CT}) of 0.22 for



Fig. 2.—(a) Scatterplot of the first 2 principal components of the DAPC suggests microsatellite genotypes form 5 genetic clusters, as well as hierarchical structure among bighorn sheep (*Ovis canadensis*) within the study area. Each point represents 1 individual and ellipses around clusters represent 95% confidence. (b) Scatterplot of the first 2 principal components of the DAPC used to identify genetic structure within desert bighorn only. (c) Posterior estimates of individual admixture proportions among genetic clusters (K = 5) as determined by TESS. Each bar represents an individual, and the height of the bar represents the relative probability of belonging to a given cluster. Sample locations are indicated above the chart, subspecies below. (d) Sample locations overlaid with predictive boundaries for each genetic cluster identified by TESS. Boundaries are based on simple kriging of the posterior probability of cluster membership at each location.

mtDNA and 0.16 for microsatellites. However, significant $F_{\rm CT}$ estimates for the alternative formulations of population structure (K = 4 pooling Peninsular and Nelson bighorn, and K = 5 considering each desert subspecies separately) suggested the presence of substructure. The desert bighorn only AMOVAs also supported the presence of substructure, with $F_{\rm CT}$ estimates significant (P < 0.001) and of similar magnitude at K = 2 and K = 3 for microsatellite and mtDNA data sets (Table 1).

Pairwise $F_{\rm ST}$ estimates based on mtDNA data indicated significant differentiation among all clusters (Table 2). We found the lowest estimates among the desert clusters (0.11–0.18), which is consistent with low discrimination as indicated by the DAPC scatterplot (Fig. 2a). Comparisons between the desert clusters and the Sierra Nevada (0.43–0.50) and Rocky Mountain subspecies (0.17–0.25) indicated higher genetic differentiation. This pattern was also reflected in the microsatellite data. Pairwise $F_{\rm ST}$ values among the desert clusters were lower (0.08–0.14; Table 2) than those comparisons to the

Sierra Nevada (0.19–0.26) or Rocky Mountain (0.15–0.25) subspecies.

The Mantel test based on microsatellite data found a strong positive correlation between (*ln*) geographic distance and genetic distance (r = 0.51; P < 0.001; Supporting Information S4), while the Mantel correlogram suggested genotype frequencies were spatially autocorrelated, with significant positive *r*-values between 0 and 60 km. The Mantel test using mtDNA data resulted in a lower correlation between geographic and genetic distance (r = 0.26; P = 0.034), and the correlogram indicated spatial autocorrelation in haplotype frequencies between 0 and 40 km.

Genetic diversity.—We observed substantial genetic diversity within each cluster identified (Table 3), with all 39 microsatellite loci polymorphic in each cluster. Average allelic richness ranged from 2.7 to 8.2 and observed heterozygosity was generally high, ranging from 0.37 to 0.58. We observed statistically significant deviations from HWE in all clusters except for the

Table 1.—Analysis of molecular variance results for different configurations of population genetic structure among 1) all bighorn samples and 2) desert bighorn samples only, using mtDNA and microsatellite data sets. The number of inferred genetic populations for each test is indicated by *K*. Letters (A–E) indicate membership of a subspecies to a genetic population under a specific test.

Subspecies			All sa	Desert samples						
	mtDNA				Microsatellites	5	mtI	DNA	Microsatellites	
	$\overline{K} = 3$	K = 4	<i>K</i> = 5	K = 3	K = 4	<i>K</i> = 5	K = 2	<i>K</i> = 3	K = 2	K = 3
Sierra Nevada	А	А	А	А	А	А				
Peninsular	В	В	В	В	В	В	В	В	В	В
Nelson	В	В	С	В	В	С	В	С	В	С
Mexican	В	С	D	В	С	D	С	D	С	D
Rocky Mountain	С	D	Е	С	D	Е				
$F_{\rm CT}^{\ a}$	0.22	0.15	0.16	0.16	0.12	0.13	0.11	0.08	0.06	0.08

^a All estimates were statistically significant at P < 0.001.

Table 2.—Pairwise F_{st} estimates based on 39 microsatellite loci (below diagonal) and 515 base pairs of mtDNA control region sequence (above diagonal) for bighorn sheep (*Ovis canadensis*) genetic clusters, approximating subspecies. All estimates were statistically significant following false detection rate (FDR) correction.

Genetic cluster	Sierra Nevada	Peninsular	Nelson	Mexican	Rocky Mountain
Sierra Nevada		0.50	0.43	0.43	0.57
Peninsular	0.26		0.18	0.16	0.25
Nelson	0.19	0.09		0.11	0.19
Mexican	0.26	0.14	0.08		0.17
Rocky Mountain	0.33	0.25	0.15	0.20	

Table 3.—Indices of genetic diversity (averages) for bighorn sheep (*Ovis canadensis*) genetic clusters, approximating subspecies, for both microsatellites (left) and mitochondrial DNA (right). The diversity indices used are as follows: *A*, alleles per locus; $A_{\rm R}$, allelic richness; $H_{\rm E}$, expected heterozygosity; $H_{\rm O}$, observed heterozygosity; $F_{\rm IS}$, inbreeding coefficient; $H_{\rm n}$, number of haplotypes; $H_{\rm d}$, haplotype diversity; π , nucleotide diversity.

Genetic cluster		Microsatellites							mtDNA				
	N	Α	A_{R}	$H_{\rm E}$	H_{0}	F _{IS}	n	H _n	$H_{\rm d}$	π			
Sierra Nevada	52	2.4	2.7	0.39	0.37	0.03	47	1	0	0			
Peninsular	187	4.7	4.9	0.54	0.50	0.09 ^a	175	10	0.76	0.0128			
Nelson	288	8.4	8.2	0.68	0.53	0.21ª	279	30	0.87	0.0126			
Mexican	180	6.2	6.3	0.60	0.53	0.13 ^a	170	25	0.91	0.0119			
Rocky Mountain	97	6.4	6.8	0.64	0.58	0.10ª	87	10	0.73	0.0073			

^a Deviation from Hardy–Weinberg equilibrium (homozygote excess) indicated by $P \le 0.001$.

Sierra Nevada subspecies, suggesting the presence of substructure in the remaining 4. This finding is not surprising given the spatial scale of our sampling, existing evidence of regional genetic structure among desert bighorn herds (Epps et al. 2010; Buchalski et al. 2015), and our results for IBD tests.

Normalization of the control region sequence data required the removal of RS 2 from 36% of Rocky Mountain, < 1% of desert, and 0% of Sierra Nevada samples. Thus, RS 2 was relatively common in Rocky Mountain bighorn as compared to the other subspecies. Data normalization resulted in 515 bp sequences with minimal missing data from 758 samples. Of the aligned nucleotide positions, 81 sites (16%) were variable and 70 sites (14%) were parsimony-informative. We discovered 74 distinct haplotypes, of which 24 were previously described in GenBank. We also identified 12 haplotypes in GenBank that were not present in our data and retained these for phylogenetic analyses. Accession numbers of all haplotypes analyzed are listed in Supporting Information S1. Haplotypes were frequently restricted to a single location or had localized distributions limited to neighboring mountain ranges. The number of mtDNA haplotypes corresponding to each genetic cluster ranged from 1 to 25 (Table 3). The Sierra Nevada sample exhibited only a single haplotype. Excluding this population, haplotype and nucleotide diversity were high ($H_d = 0.73 - 0.91$, $\pi = 0.0073 - 0.0128$).

Phylogeographic analyses.—Phylogenetic inference by building a ML tree indicated the presence of 3 distinct clades, 2 of which exhibited bootstrap support > 90% (Fig. 3a). The 3 clades corresponded approximately to Sierra Nevada, Rocky Mountain, and desert bighorn and composed a polytomy indicating no support for any specific divergence pattern. The ML tree also represented desert bighorn as a polyphyletic group. Clade 1 consisted of the single Sierra Nevada haplotype and desert bighorn haplotype MG3 (Fig. 3a, #1). Haplotype MG3 was found in 8 individuals from the Panamint Range and 1 individual from Eagle Crags, both in the northern Mojave Desert in California (Fig. 1; Supporting Information S1). Clade 2 consisted of Rocky Mountain haplotypes, both from within the native range for that subspecies (i.e., Alberta and British Columbia) and (re)introduced populations in Arizona and New Mexico. Clade 3 was not well supported statistically, but represented the most basal portion of the tree and consisted entirely of desert bighorn. Within Clade 3, subclades were largely concordant with the desert subspecies designations of Cowan (1940) and only occasionally polyphyletic. Finally, the haplotype for Weems bighorn sheep obtained from GenBank did not cluster with haplotypes from Peninsular bighorn sheep (Fig. 3a, #2), even though both are endemic to Baja California.

The unrooted, median-joining haplotype network also recognized 3 clades corresponding to Sierra Nevada, Rocky Mountain, and desert bighorn (Fig. 3b). We estimated TMRCA for the Rocky Mountain clade and desert bighorn at 680 ± 130 kya, and the Sierra Nevada clade and desert bighorn at 640±120 kya. In addition, we estimated TMRCA between the single Sierra Nevada haplotype and haplotype MG3 at 150 ± 60 kya. Within the desert clade (Supporting Information S5), the network was sparse with a center consisting of several inferred but unsampled haplotypes. There was little haplotype sharing among subspecies, and the geographic areas where haplotype sharing was observed (Fig. 4) coincided with zones of subspecies intergradation originally identified by Cowan (1940: 574), including the northern Sonoran Desert (locations 40 and 41), as well as the northern Peninsular Ranges (location 15). The network also indicated several endemic haplogroups within the Peninsular and Mexican subspecies with ages predating the LGM-103 to 56 kya for Peninsular bighorn and 160 to 9 kya for Mexican bighorn sheep (Supporting Information S5).

The IMa2 analyses estimated pairwise effective splitting times for Sierra Nevada, Rocky Mountain, and desert bighorn at the mid- to late Pleistocene (315-94 kya), although our pairwise estimates were incomplete (Table 4). Due to the presence of only a single haplotype in contemporary Sierra Nevada bighorn, and its close relationship to a desert bighorn haplotype, we did not estimate splitting times between these taxa (Table 4). Pairwise estimates among desert bighorn were considerably more recent (9-6 kya) than those with Rocky Mountain bighorn, with the exception of the Peninsular and Mexican populations (122 kya). Further, splitting time estimates from IMa2 generally increased with net sequence divergence following a saturating curve (Supporting Information S6), except for a single outlier representing the Mexican versus Penninsular bighorn comparison. One of the assumptions of IMa2 is that no intervening populations are missing from the analysis, which was clearly violated in this case and potentially responsible

Table 4.—IMa2 estimates of splitting times (× 1,000 years) based on control region sequences (above diagonal). The 95% highest posterior density of the estimates are indicated in parentheses. Average pairwise sequence divergence (Dxy) is indicated below the diagonal. Diagonal contains average sequence divergence within a taxon. Net sequence divergence (Da) is calculated by subtracting average within taxon sequence divergence from Dxy.

	Sierra Nevada	Peninsular	Nelson	Mexican	Rocky Mountain	Snow sheep
Sierra Nevada	0.0000				315 (114–532)	
Peninsular	0.0370	0.0127	6 (0-17)	122 (59–190) ^a	273 (67–442)	
Nelson	0.0350	0.0160	0.0141	9 (1-21)	94 (9–185)	
Mexican	0.0370	0.0160	0.0150	0.0119	299 (116-484)	
Rocky Mountain	0.0440	0.0360	0.0330	0.0370	0.0073	
Snow sheep	0.0580	0.0640	0.0630	0.0610	0.0710	

^a Inconsistency between the splitting time estimate and net sequence divergence.



Fig. 3.—(a) Rooted maximum likelihood tree based on 515 base pairs of the mtDNA control region illustrating 3 main bighorn sheep lineages. Branch lengths are scaled to evolutionary distances and bootstrap values > 50, based on 1,000 replicates, are shown next to the branches. Haplotype names correspond to those in Supporting Information S1 and colors to genetic clusters indicated in Fig. 2. #1—Desert haplotype representing ancient gene flow event or incomplete lineage sorting with Sierra Nevada bighorn. #2—Position of Weems bighorn haplotype obtained from GenBank. #3—For the purpose of illustration, frequencies for Hap 5 include the findings of Boyce et al. (1999) and Epps et al. (2010), to depict all published evidence of haplotype sharing between Peninsular and Nelson bighorn. (b) Unrooted median-joining network illustrating the 3 lineages. Branch lengths are proportional to the number of substitutions, and node sizes to the number of individuals represented.

for the unreasonably high estimate. We therefore conducted a 3 population analysis in IMa2, which constrained the splitting times among these 3 populations to be tree like (rooted to *O. nivicola* as an outgroup). These results estimated that Mexican bighorn split from Nelson and Penninsular bighorn 17 kya (95% HPD: 37–3 kya) and that the latter 2 populations separated 3 kya (95% HPD: 8–0.5 kya). Because our analyses assumed no gene flow since divergence, the effect of any subsequent gene flow would be to render our splitting time estimates too recent. Therefore, these estimates were conservative, particularly for desert subspecies where historical gene flow was most likely.

Estimates of historical demography via Bayesian skyline plots suggested Nelson bighorn had the largest historical population size, followed by Mexican bighorn, with Peninsular bighorn having the smallest historical size (Fig. 5). The Bayesian skyline plots were generally parallel for all 3 populations suggesting expansion during the Sangamon interglacial period, followed by large declines following the LGM. However, 95% highest posterior density intervals were insufficiently narrow to distinguish whether declines occurred during the late Pleistocene or Holocene (Supporting Information S7). Population decline apparently began the earliest and was the most pronounced (\sim 5×) in Nelson bighorn, whereas the Peninsular population appears to have declined more recently.

DISCUSSION

Genetic divergence among Sierra Nevada, Rocky Mountain, and desert bighorn sheep.—This study provides the most extensive characterization to date of genetic differentiation and structure among bighorn populations in the desert southwest. Phylogenetic analyses of mtDNA identified 2 well-supported clades associated with Sierra Nevada and Rocky Mountain bighorn. Desert bighorn haplotypes were basal to these clades, but were shallowly differentiated from one another. Population genetic analyses were consistent with this phylogenetic structure. The DAPC showed strong discrimination among all 3 major lineages (i.e., the 2 clades and desert bighorn) and the AMOVAs indicated among-group variance was maximized at K = 3.

The deep divergence among geographically endemic bighorn clades implied long-term isolation (Avise 2000). Rho estimates suggested that TMRCA of desert bighorn and both the Rocky Mountain and Sierra Nevada lineages dates prior to the Illinoian Glaciation. Further, our estimates of splitting times among these lineages suggest divergence during the late Pleistocene and appear comparable to other phylogenetic data for the subgenus *Pachyceros* (i.e., North American wild sheep, including *O. canadensis* and Dall sheep [*O. dalli*], as well as their Asian counterpart *O. nivicola*). Loehr et al. (2006) estimated divergence between Nelson and Rocky Mountain



Fig. 4.—Geographic distribution of mtDNA control region haplogroups among sampled herds of *Ovis canadensis* subspecies, shown as pie diagrams. Locations are numbered as in Fig. 1. For the purpose of illustration, haplotype frequencies for the San Jacinto population (15) include our results and the findings of Boyce et al. (1999), demonstrating a shared haplotype between the northern Peninsular Ranges and southern Mojave Desert.

bighorn at ~380 kya, which is generally consistent with our IMa2 estimates recalibrated to the 2.6% per Ma mutation rate (see "Materials and Methods"). Studies using *Cytb* and nuclear sequences estimated the divergence between *O. nivicola* and North American Pachyceriforms at 2.3–1.6 My, and the divergence between *O. canadensis* and *O. dalli* at 1.4–0.95 My (Bunch et al. 2006; Rezaei et al. 2010).

Our divergence estimates help to further resolve the origins of desert bighorn, as well as colonization of the desert southwest. The fossil record indicates Ovis continuously inhabited the Mojave region since ~300 kya (Jefferson 1991), as well as a more recent refugium located further north in Wyoming, with a fossil record of continuous Ovis presence since ~100 kya (Martin and Gilbert 1978; Wang 1988). Our data suggest these refugia were the result of separate colonization events from a Beringian source predating the Illinoian glaciation (i.e., on the order of 300 kya) during periods when ice-free corridors between Laurentide and Cordilleran ice sheets were present. Such a deep divergence between the Wyoming and Mojave refugial populations elevates the evolutionary significance of their relationships to contemporary desert bighorn. The geochronology of fossils suggests that bighorn first expanded from Wyoming into Nevada (beginning ~18 kya) and progressively further south, followed by later expansions from the Mojave refugium (~12 kya), rendering the fossil record somewhat ambiguous with respect to the origins of contemporary desert bighorn (Geist 1985). On the basis of phylogenetic positioning, our data clearly support a scenario where colonists from the Mojave refugium displaced the earlier northern colonists and strongly refute the possibility of northern colonists partially giving rise to contemporary desert bighorn.

Geist (1985) proposed that northern expansion from the Mojave refugium during the early Holocene (~12 kya) resulted in establishment of the Sierra Nevada subspecies (synonymous with California bighorn at the time of Geist's writing). Based on our findings, this seems unlikely. Net sequence divergence between the single Sierra Nevada haplotype and all 3 desert subspecies (~2.3%) corresponds to an estimated splitting time of approximately 125 kya (Supporting Information S6). Further, the Nelson bighorn haplotype that formed a clade with the single Sierra Nevada haplotype was sufficiently divergent to suggest the last contact between these 2 lineages predated the LGM (150 ± 60 kya). The polyphyletic nature of desert bighorn relative to Sierra Nevada bighorn could reflect either secondary contact between the lineages or incomplete lineage sorting. Despite the possibility of ancient gene flow, we found no evidence of contemporary gene flow between desert and Sierra Nevada bighorn based on microsatellite genotypes. Given that the Sierra Nevada Range is separated from desert bighorn occupied ranges by as little as 10 km in some areas, this finding suggests the possibility of nongeographic behavioral barriers or other forms of reproductive isolation between these subspecies.

Genetic relationships within desert bighorn sheep.—Our results indicated the desert subspecies defined by Cowan (1940;



Fig. 5.—Estimated changes in size $(N_e\mu)$ through time for 3 desert bighorn sheep populations based on Bayesian skyline reconstruction from mtDNA control region sequences. Plots illustrate recent declines in all populations ranging from the last glacial maximum (LGM) to the late Holocene (assuming 6.1% per Ma substitution rate). Estimates indicate that Nelson bighorn sheep, followed by Mexican bighorn sheep, had the historically largest population sizes, whereas Peninsular bighorn sheep had the smallest population which declined most recently.

excluding Weems bighorn sheep) diverged from one another more recently (Fig. 3a). Estimated splitting times based on the 3 population coalescent simulation suggested Mexican bighorn may have diverged as early as the late Pleistocene (37–3 kya), with the 2 other populations separating in the Holocene (8-0.5 kya). However, 2 observations suggest the possibility that splits among these subspecies could be considerably older. First, our assumption of no genetic exchange among desert subspecies since they diverged is conservative, and any actual gene flow would put estimates further back in time. Second, the haplotype network revealed several endemic haplogroups with ages significantly predating the LGM (Supporting Information S5). For Peninsular bighorn, all but 1 of its 13 haplotypes occurred in 3 endemic haplogroups, estimated on average to reflect derivation from their ancestral haplotypes ~85 kya (Fig. 3a). Mexican bighorn also showed isolation from Nelson bighorn populations, as the majority of its 25 haplotypes occurred in endemic haplogroups dating to a similar timeframe (~72 kya). All shared haplotypes occurred in areas recognized by Cowan (1940) as zones of intergradation between desert subspecies (i.e., the northern Peninsular Ranges and the northern Sonora Desert in the vicinity of the Bill Williams River; Fig. 4). Regardless of whether these shared haplotypes reflected ancient shared ancestry or recent gene flow, the matrilineal diversity of Peninsular and Mexican bighorn was significantly divergent from the Nelson subspecies.

Both the Bayesian skyline plots and haplotype network suggested that modern desert bighorn reflect a small fragmented subset of a once massive population. The network was sparse, with a large number of missing intermediate haplotypes. The Bayesian skyline analyses also suggested a large ancestral desert bighorn population that expanded during the Sangamon interglacial, followed by demographic decline since the LGM. Ramey's (1995) study using a much more slowly mutating mtDNA marker found a widespread desert haplotype, which sat at the center of a star-like phylogeny, consistent with a population expansion. Putting our findings and his findings together suggests an expansion across the southwest dating well before the Pleistocene-Holocene boundary as proposed by Geist (1985). Based on the estimated ages for several of the endemic desert haplogroups, we suggest Ovis persisted in multiple southern refugia during the LGM, as originally proposed by Ramey (1995), rather than a single Mojave refugium. Following deglaciation, changes in the distribution of habitat may have allowed for secondary contact among these populations, resulting in the more recent splitting time estimates we observed. Ultimately, all refugial populations experienced fragmentation and demographic decline during the Holocene.

Analyses of population genetic structure based on microsatellite and mtDNA also supported significant differentiation among desert subspecies. Both the DAPC and TESS analyses indicated genetic clustering concordant with Cowan's subspecies distributions. Likewise, AMOVA among desert subspecies produced significant fixation index estimates among groups ($F_{\rm CT}$), regardless of the underlying model of population structure. Microsatellite and mitochondrial DNA-based $F_{\rm ST}$ comparisons among the desert bighorn genetic clusters were statistically significant and indicated that desert bighorn do not form a single genetic population.

The TESS analysis indicated low-level admixture between the Peninsular and Nelson subspecies in the southern Mojave Desert (locations 17–26; Fig. 2c). This pattern of admixture was inconsistent with clinal variation indicative of an active contact zone (Durand et al. 2009), but rather appears to represent relict gene flow between the 2 lineages. We interpret this as evidence of secondary contact following postglacial expansion of the Peninsular and Nelson refugial populations. However, the degraded nature of the contact (i.e., low-level admixture versus a clinal transition) suggests a subsequent disruption of gene flow, possibly by contemporary anthropogenic barriers or range contraction of the Peninsular population during the last century. Quite importantly, the geographic location of these admixed genotypes matches the findings of previous morphometric analyses. Wehausen and Ramey (1993) used univariate and PC analyses to demonstrate major overlap in skull morphology characters between Peninsular and southern Mojave herds, both of which differed significantly from herds in the northern Mojave and Great Basin. This overlap was used to justify synonymizing Peninsular bighorn (O. c. cremnobates) with Nelson bighorn (O. c. nelsoni). Our genetic data suggest these morphological similarities may actually be the result of a relatively recent (i.e., Holocene) contact between the lineages. Further, TESS analyses showed no evidence of clinal variation between Nelson and Mexican bighorn, but rather intermingled genotypes in the northern Sonora Desert (i.e., location 40; Fig. 2). These findings suggest the Nelson and Mexican lineages may have only recently come into contact in eastern Arizona, possibly as a result of successful recovery and expansion. Additional sampling at a finer spatial scale would be necessary to precisely delineate the boundary between these 2 populations.

Mantel test and correlogram results indicated IBD was also a source of genetic structure among bighorn herds within desert subspecies. Lower correlation between genetic and geographic distances and the smaller spatial scale of genetic autocorrelation for the mtDNA relative to the nuclear markers was consistent with ewe philopatry (Krausman et al. 1999). This pattern of IBD indicates dispersal is negatively correlated with geographic distance between neighboring habitat patches (i.e., mountain ranges), reaching an asymptote at a distance beyond which dispersal is unlikely to occur (> 60 km). These findings agree with previous landscape genetics models for bighorn in the Mojave Desert that estimated the maximum effective dispersal distance of rams at 16.4 km-cost-units (corresponding to 16.4 km of flat terrain or 164 km of sloped terrain—Epps et al. 2007) and ewes at 10.0 km-cost-units (Creech et al. 2014). The scale of spatial autocorrelation we observed is reasonable for each marker type, considering that the distance between our sampling locations often covered both flat and mountainous terrain. Our results provide additional support for metapopulation structure in desert bighorn (Bleich et al. 1996), with genetic connectivity among mountain ranges occurring via a stepping-stone model of gene flow.

Genetic diversity of bighorn sheep populations.—Using the numerically largest and geographically broadest set of desert bighorn sheep samples analyzed to date, we found substantial genetic diversity throughout the native range. Observed heterozygosity and allelic richness were comparable or higher than other studies (Gutierrez-Espeleta et al. 2001; Epps et al. 2005, 2006; Buchalski et al. 2015) and suggest desert bighorn retained substantial range-wide genetic diversity despite demographic declines and loss of population connectivity. The federally endangered Sierra Nevada population had low genetic diversity, consistent with recent bottlenecks and small size. Low allelic richness and expected heterozygosity were comparable to the finding of Johnson et al. (2011), while mtDNA haplotype diversity (the presence of a single haplotype) had not previously been published for this population. Genetic diversity indices for the San Gabriel population in the Transverse Ranges (Fig. 1, location 16; $A_{\rm R} = 3.3$, $H_{\rm E} = 0.40$, $H_{\rm d} = 0$) were considerably lower than averages for Nelson bighorn ($A_{\rm R} = 8.2$, $H_{\rm E} = 0.68$, $H_{\rm d} = 0.87$) and were comparable to the Sierra Nevada population. Highway infrastructure associated with Los Angeles separates the San Gabriel population from others within the Transverse Ranges, suggesting that this population is largely isolated and may continue to lose genetic diversity via drift. Additional sampling to better characterize genetic diversity is necessary to fully evaluate the status of this population.

Conservation of desert bighorn sheep genetic diversity.—In this study, we provide evidence of genetic structure highly concordant with the desert subspecies proposed by Cowan (1940). However, full characterization of the phylogenetic history of desert bighorn would require additional analyses utilizing more conserved regions of the mitochondrial genome and potentially nuclear sequence data to more accurately estimate divergence dates. Ultimately, conflicts between subspecies designations based on morphological versus genetic data may prove difficult to resolve and are somewhat peripheral to the more practical challenge of identifying and conserving important biological diversity.

The 3 desert bighorn sheep lineages identified in this study occupy desert biomes that vary significantly in climate (Laity 2009), suggesting exposure to different selection regimes. Hence, local adaptation is expected to have shaped some of the genomic diversity among desert bighorn sheep. Functional differences among herds have been documented, which are assumed to have a genetic basis-including horn size and lambing period (Wehausen 1991, 2005). Identifying conservation units that recognize adaptive differences may prove essential for continued recovery, especially in response to increasing threats from disease outbreak and prolonged drought resulting from climate change. For example, evolutionary significant units (ESUs) place an emphasis on adaptive variation and evolutionary potential (Ryder 1986; Waples 1991; Moritz 1994; Crandall et al. 2000), with precedence for granting ESUs legal protection under the Endangered Species Act. We recommend the delineation of conservation units be guided by a landscape genomics approach (sensu—Funk et al. 2012), utilizing neutral loci and loci under selection to characterize adaptive differences among herds.

Translocations and reintroductions have been critical in helping bighorn sheep populations recover across western North America (Krausman 2000). While largely conducted to increase abundance and distribution, successful genetic management of bighorn sheep may also require translocations that increase heterozygosity and facilitate genetic rescue. Reintroduced herds typically have low genetic diversity resulting from founder events and subsequent drift (Hedrick et al. 2001; Whittaker et al. 2004; Hedrick 2014). While herd supplementation with unrelated animals can result in genetic rescue, both in terms of increased genetic diversity and higher fitness among hybrids (Hogg et al. 2006; Miller et al. 2012; Olson et al. 2012), outbreeding depression can also occur in crosses between populations within a species (i.e., between subspecies—Edmands 2007). Our data indicate desert subspecies became isolated during the LGM, or potentially earlier, in some cases with minimal secondary contact. For this reason, we feel translocations among Peninsular, Nelson, and Mexican bighorn are not advised. Our data suggest the maintenance of viable levels of genetic diversity should be attainable through translocations among herds within each of the 3 desert lineages. Whenever genetic rescue is contemplated, guidelines such as those proposed by Hedrick and Fredrickson (2010) should be consulted to evaluate the costs and benefits. In the absence of adequate data, managers should adopt the "local is best" translocation strategy, as proposed by Ramey (1995), as the most reliable means for preserving local adaptation.

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SUPPORTING INFORMATION

The Supporting Information documents are linked to this manuscript and are available at Journal of Mammalogy online (jmammal.oxfordjournals.org). The materials consist of data provided by the author that are published to benefit the reader. The posted materials are not copyedited. The contents of all supporting data are the sole responsibility of the authors. Questions or messages regarding errors should be addressed to the author.

Supporting Information S1.—Bighorn sheep (*Ovis canadensis*) samples used in this study organized by location.

Supporting Information S2.—Methods and rationale for alignment of mtDNA control region sequences.

Supporting Information S3.—(a) Spline interpolation of the optimal a-score (i.e., proportion of successful reassignments corrected for the number of retained PCs from the DAPC. (b) Mean values of the DIC, averaged over 10 runs, estimated by TESS for models with the number of genetic clusters (K) ranging from 2 to 10. Dashed lines were inserted to indicate a piecewise change in the function at K = 5. (c) Bayesian information criteria (BIC) for different numbers of genetic clusters. The chosen number of clusters (K = 5), based on 8 out of 10 independent runs of the *diffNgroups* algorithm in the package adegenet, is the minimum number after which the decrease in BIC becomes negligible.

Supporting Information S4.—Results of isolation-by-distance (IBD) tests among native herds (n = 23) of desert bighorn

sheep. Graphs are shown for microsatellites (top row) and mtDNA haplotypes (bottom row). Scatter plots of genetic distance ($F_{\rm ST}/(1 - F_{\rm ST})$ plotted against *ln* geographic distance (left) and Mantel correlograms illustrating the scale of spatial auto-correlation in allele/haplotype frequencies (right). For correlograms, closed squares indicate a significant Mantel correlation coefficient based on 1,000 permutations.

Supporting Information S5.—Unrooted median-joining network illustrating the phylogenetic relationships of mtDNA haplotypes within desert bighorn sheep. Haplotype names correspond to those in Supporting Information S1 and colors to genetic clusters indicated in Fig. 2. Node sizes scale to indicate the number of individuals within each haplotype. Shaded areas denote distinct haplogroups and dates represent estimated haplogroup ages (rho \pm *SD*). For the purpose of illustration, frequencies for haplotype 5 include our results and the findings of Boyce et al. (1999) and Epps et al. (2010), to depict all published evidence of haplotype sharing between Peninsular and Nelson bighorn.

Supporting Information S6.—Relationship between pairwise net sequence divergence (Da) and IMa2 estimates of divergence (in years before present), illustrating the outlier estimate for Peninsular versus Mexican bighorn (open circle).

Supporting Information S7.—Bayesian skyline plots generated from mtDNA control region sequences showing the historical demographic trends of Nelson (a), Mexican (b), and Peninsular (c) bighorn sheep in the desert southwest (assuming 6.1% per Ma substitution rate). The black line is the median size $(N_e\mu)$ estimate, and the gray lines represent the upper and lower 95% highest posterior density interval.

LITERATURE CITED

- ADAMACK, A. T., AND B. GRUBER. 2014. PopGenReport: simplifying basic population genetic analyses in R. Methods in Ecology and Evolution 5:384–387.
- ALTSCHUL, S. F., ET AL. 1997. Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. Nucleic Acids Research 25:3389–3402.
- AVISE, J. C. 2000. Phylogeography: the history and formation of species. Harvard University Press, Cambridge, Massachusetts.
- BANDELT, H. J., P. FORSTER, AND A. ROHL. 1999. Median-joining networks for inferring intraspecific phylogenies. Molecular Biology and Evolution 16:37–48.
- BENJAMINI, Y., AND D. YEKUTIELI. 2001. The control of the false discovery rate in multiple testing under dependency. Annals of Statistics 29:1165–1188.
- BLEICH, V. C., J. D. WEHAUSEN, K. R. JONES, AND R. A. WEAVER. 1990. Status of bighorn sheep in California, 1989 and translocations from 1971 through 1989. Desert Bighorn Council Transactions 34:24–26.
- BLEICH, V. C., J. D. WEHAUSEN, R. R. RAMEY, AND J. L. RECHEL. 1996. Metapopulation theory and mountain sheep: implications for conservation. Pp. 353–373 in Metapopulations and Wildlife Conservation (D. R. McCullough, ed.). Island Press, Washington, D.C.
- BOYCE, W. M., R. R. RAMEY, T. C. RODWELL, E. S. RUBIN, AND R. S. SINGER. 1999. Population subdivision among desert bighorn

sheep (*Ovis canadensis*) ewes revealed by mitochondrial DNA analysis. Molecular Ecology 8:99–106.

- BOYCE, W. M., M. E. WEISENBERGER, M. C. PENEDO, AND C. K. JOHNSON. 2011. Wildlife translocation: the conservation implications of pathogen exposure and genetic heterozygosity. BMC Ecology 11:5.
- BRADLEY, W. G., AND L. R. BAKER. 1967. Range of variation in Nelson bighorn sheep from the Desert Game Range and it taxonomic significance. Desert Bighorn Council Transactions 11:114–140.
- BROWN, R. P., AND Z. YANG. 2011. Rate variation and estimation of divergence times using strict and relaxed clocks. BMC Evolutionary Biology 11:271.
- BUCHALSKI, M. R., ET AL. 2015. Genetic population structure of Peninsular bighorn sheep (*Ovis canadensis nelsoni*) indicates substantial gene flow across US-Mexico border. Biological Conservation 184:218–228.
- BUECHNER, H. K. 1960. The bighorn sheep in the United States, its past, present, and future. Wildlife Monographs 4:1–174.
- BUNCH, T. D., C. WU, Y. P. ZHANG, AND S. WANG. 2006. Phylogenetic analysis of snow sheep (*Ovis nivicola*) and closely related taxa. Journal of Heredity 97:21–30.
- CHEN, C., E. DURAND, F. FORBES, AND O. FRANCOIS. 2007. Bayesian clustering algorithms ascertaining spatial population structure: a new computer program and a comparison study. Molecular Ecology Notes 7:747–756.
- COWAN, I. M. 1940. Distribution and variation in the native sheep of North America. American Midland Naturalist 24:505–580.
- COX, M., AND P. CUMMINGS. 2005. Status of desert bighorn sheep in Nevada, 2004–2005. Desert Bighorn Council Transactions 48:53–57.
- CRANDALL, K. A., O. R. P. BININDA-EMONDS, G. M. MACE, AND R. K. WAYNE. 2000. Considering evolutionary processes in conservation biology. Trends in Ecology & Evolution 15:290–295.
- CREECH, T. G., C. W. EPPS, R. J. MONELLO, AND J. D. WEHAUSEN. 2014. Using network theory to prioritize management in a desert bighorn sheep metapopulation. Landscape Ecology 29:605–619.
- DRUMMOND, A. J., AND A. RAMBAUT. 2007. BEAST: Bayesian evolutionary analysis by sampling trees. BMC Evolutionary Biology 7:8.
- DRUMMOND, A. J., A. RAMBAUT, B. SHAPIRO, AND O. G. PYBUS. 2005. Bayesian coalescent inference of past population dynamics from molecular sequences. Molecular Biology and Evolution 22:1185–1192.
- DURAND, E., F. JAY, O. E. GAGGIOTTI, AND O. FRANCOIS. 2009. Spatial inference of admixture proportions and secondary contact zones. Molecular Biology and Evolution 26:1963–1973.
- EDMANDS, S. 2007. Between a rock and a hard place: evaluating the relative risks of inbreeding and outbreeding for conservation and management. Molecular Ecology 16:463–475.
- EPPS, C. W., P. J. PALSBOLL, J. D. WEHAUSEN, G. K. RODERICK, AND D. R. MCCULLOUGH. 2006. Elevation and connectivity define genetic refugia for mountain sheep as climate warms. Molecular Ecology 15:4295–4302.
- EPPS, C. W., P. J. PALSBOLL, J. D. WEHAUSEN, G. K. RODERICK, R. R. RAMEY, AND D. R. MCCULLOUGH. 2005. Highways block gene flow and cause a rapid decline in genetic diversity of desert bighorn sheep. Ecology Letters 8:1029–1038.
- EPPS, C. W., J. D. WEHAUSEN, V. C. BLEICH, S. G. TORRES, AND J. S. BRASHARES. 2007. Optimizing dispersal and corridor models using landscape genetics. Journal of Applied Ecology 44:714–724.
- EPPS, C. W., J. D. WEHAUSEN, P. J. PALSBØLL, AND D. R. MCCULLOUGH. 2010. Using genetic tools to track desert bighorn sheep colonizations. Journal of Wildlife Management 74:522–531.

- EXCOFFIER, L., AND H. E. L. LISCHER. 2010. Arlequin suite ver 3.5: a new series of programs to perform population genetics analyses under Linux and Windows. Molecular Ecology Resources 10:564–567.
- EXCOFFIER, L., P. E. SMOUSE, AND J. M. QUATTRO. 1992. Analysis of molecular variance inferred from metric distances among DNA haplotypes: application to human mitochondrial DNA restriction data. Genetics 131:479–491.
- FORSTER, P., R. HARDING, A. TORRONI, AND H. J. BANDELT. 1996. Origin and evolution of Native American mtDNA variation: a reappraisal. American Journal of Human Genetics 59:935–945.
- FRANCOIS, O., AND E. DURAND. 2010. Spatially explicit Bayesian clustering models in population genetics. Molecular Ecology Resources 10:773–784.
- FUNK, W. C., J. K. MCKAY, P. A. HOHENLOHE, AND F. W. ALLENDORF. 2012. Harnessing genomics for delineating conservation units. Trends in Ecology & Evolution 27:489–496.
- GEIST, V. 1985. On Pleistocene bighorn sheep: some problems of adaptation, and relevance to today's American megafauna. Wildlife Society Bulletin 13:351–359.
- GOSLEE, S. C., AND D. L. URBAN. 2007. The ecodist package for dissimilarity-based analysis of ecological data. Journal of Statistical Software 22:1–19.
- GUO, S. W., AND E. A. THOMPSON. 1992. Performing the exact test of Hardy-Weinberg proportion for multiple alleles. Biometrics 48:361–372.
- GUTIERREZ-ESPELETA, G. A., P. W. HEDRICK, S. T. KALINOWSKI, D. GARRIGAN, AND W. M. BOYCE. 2001. Is the decline of desert bighorn sheep from infectious disease the result of low MHC variation? Heredity 86:439–450.
- HASEGAWA, M., H. KISHINO, AND T. A. YANO. 1985. Dating of the human-ape splitting by a molecular clock of mitochondrial DNA. Journal of Molecular Evolution 22:160–174.
- HEDRICK, P. W. 2014. Conservation genetics and the persistence and translocation of small populations: bighorn sheep populations as examples. Animal Conservation 17:106–114.
- HEDRICK, P. W., AND R. FREDRICKSON. 2010. Genetic rescue guidelines with examples from Mexican wolves and Florida panthers. Conservation Genetics 11:615–626.
- HEDRICK, P. W., G. A. GUTIERREZ-ESPELETA, AND R. N. LEE. 2001. Founder effect in an island population of bighorn sheep. Molecular Ecology 10:851–857.
- HEY, J. 2010a. The divergence of chimpanzee species and subspecies as revealed in multipopulation isolation-with-migration analyses. Molecular Biology and Evolution 27:921–933.
- Hey, J. 2010b. Isolation with migration models for more than two populations. Molecular Biology and Evolution 27:905–920.
- HIENDLEDER, S., B. KAUPE, R. WASSMUTH, AND A. JANKE. 2002. Molecular analysis of wild and domestic sheep questions current nomenclature and provides evidence for domestication from two different subspecies. Proceedings of the Royal Society B-Biological Sciences 269:893–904.
- HIENDLEDER, S., H. LEWALSKI, R. WASSMUTH, AND A. JANKE. 1998. The complete mitochondrial DNA sequence of the domestic sheep (*Ovis aries*) and comparison with the other major ovine haplotype. Journal of Molecular Evolution 47:441–448.
- HOGG, J. T., S. H. FORBES, B. M. STEELE, AND G. LUIKART. 2006. Genetic rescue of an insular population of large mammals. Proceedings of the Royal Society of London, B. Biological Sciences 273:1491–1499.
- JAKOBSSON, M., AND N. A. ROSENBERG. 2007. CLUMPP: a cluster matching and permutation program for dealing with label

switching and multimodality in analysis of population structure. Bioinformatics 23:1801–1806.

- JEFFERSON, G. T. 1991. Rancholabrean age vertebrates from the southeastern Mojave Desert, California. Pp. 27–40 in Crossing the borders: Quaternary studies in eastern California and southwestern Nevada (R. E. Reynolds, ed.). Mojave Desert Quaternary Research Center, San Bernardino County Museum Association, Redlands, California.
- JOHNSON, H. E., L. S. MILLS, J. D. WEHAUSEN, T. R. STEPHENSON, AND G. LUIKART. 2011. Translating effects of inbreeding depression on component vital rates to overall population growth in endangered bighorn sheep. Conservation Biology 25:1240–1249.
- JOMBART, T. 2008. adegenet: a R package for the multivariate analysis of genetic markers. Bioinformatics 24:1403–1405.
- JOMBART, T., S. DEVILLARD, AND F. BALLOUX. 2010. Discriminant analysis of principal components: a new method for the analysis of genetically structured populations. BMC Genetics 11:15.
- KRAUSMAN, P. R. 2000. An introduction to the restoration of bighorn sheep. Restoration Ecology 8:3–5.
- KRAUSMAN, P. R., A. V. SANDOVAL, AND R. C. ETCHBERGER. 1999. Natural history of desert bighorn sheep. Pp. 139–191 in Mountain Sheep of North America (R. Valdez and P. R. Krausman, eds.). University of Arizona Press, Tucson.
- LAITY, J. J. 2009. Deserts and desert environments. John Wiley & Sons, Hoboken, New Jersey, 360.
- LIBRADO, P., AND J. ROZAS. 2009. DnaSP v5: a software for comprehensive analysis of DNA polymorphism data. Bioinformatics 25:1451–1452.
- LOEHR, J., K. WORLEY, A. GRAPPUTO, A. CAREY, J. VEITCH, AND D. W. COLTMAN. 2006. Evidence for cryptic glacial refugia from North American mountain sheep mitochondrial DNA (vol 19, pg 19, 2006). Journal of Evolutionary Biology 19:2071–2071.
- MARTIN, L. D., AND B. M. GILBERT. 1978. Excavations at Natural Trap Cave. Transactions of the Nebraska Academy of Sciences and Affiliated Societies, Lincoln, Nebraska, 336.
- MILLER, J. M., J. POISSANT, J. T. HOGG, AND D. W. COLTMAN. 2012. Genomic consequences of genetic rescue in an insular population of bighorn sheep (*Ovis canadensis*). Molecular Ecology 21:1583–1596.
- MORITZ, C. 1994. Defining evolutionarily significant units for conservation. Trends in Ecology & Evolution 9:373–375.
- MOUSADIK, A., AND R. J. PETIT. 1996. High level of genetic differentiation for allelic richness among populations of the argan tree *Argania spinosa* (L) Skeels endemic to Morocco. Theoretical and Applied Genetics 92:832–839.
- NABHOLZ, B., S. GLEMIN, AND N. GALTIER. 2008. Strong variations of mitochondrial mutation rate across mammals the longevity hypothesis. Molecular Biology and Evolution 25:120–130.
- NEI, M., AND W. H. LI. 1979. Mathematical model for studying genetic variation in terms of restriction endonucleases. Proceedings of the National Academy of Sciences of the United States of America 76:5269–5273.
- OKSANEN, J., ET AL. 2015. Vegan: community ecology package. R package v.2.2.1 http://CRAN.R-project.org/package=vegan. Accessed 15 March 2015.
- OLSON, Z. H., D. G. WHITTAKER, AND O. E. RHODES. 2012. Evaluation of experimental genetic management in reintroduced bighorn sheep. Ecology and Evolution 2:429–443.
- PEAKALL, R., AND P. E. SMOUSE. 2012. GenAlEx 6.5: genetic analysis in Excel. Population genetic software for teaching and research-an update. Bioinformatics 28:2537–2539.

- POMPANON, F., A. BONIN, E. BELLEMAIN, AND P. TABERLET. 2005. Genotyping errors: causes, consequences and solutions. Nature Reviews Genetics 6:847–859.
- PRITCHARD, J. K., M. STEPHENS, AND P. DONNELLY. 2000. Inference of population structure using multilocus genotype data. Genetics 155:945–959.
- R DEVELOPMENT CORE TEAM. 2015. R: a language and environment for statistical computing, 2.13. R Foundation for Statistical Computing, Vienna, Austria.
- RAMEY, R. R. 1995. Mitochondrial DNA variation, population structure, and evolution of mountain sheep in the south-western United States and Mexico. Molecular Ecology 4:429–440.
- REZAEI, H. R., ET AL. 2010. Evolution and taxonomy of the wild species of the genus *Ovis* (Mammalia, Artiodactyla, Bovidae). Molecular Phylogenetics and Evolution 54:315–326.
- ROSENBERG, N. A. 2004. DISTRUCT: a program for the graphical display of population structure. Molecular Ecology Notes 4:137–138.
- ROUSSET, F. 2008. genepop'007: a complete re-implementation of the genepop software for Windows and Linux. Molecular Ecology Resources 8:103–106.
- ROWLAND, M. M., AND J. L. SCHMIDT. 1981. Transplanting desert bighorn sheep: a review. Desert Bighorn Council Transactions 25:21–28.
- RYDER, O. A. 1986. Species conservation and systematics: the dilemma of subspecies. Trends in Ecology & Evolution 1:9–10.
- SAILLARD, J., P. FORSTER, N. LYNNERUP, H. J. BANDELT, AND S. NORBY. 2000. mtDNA variation among Greenland Eskimos: the edge of the Beringian expansion. American Journal of Human Genetics 67:718–726.
- SETON, E. T. 1929. The bighorn. Pp. 519–573 in Lives of the game animals (E. T. Seton, ed.). Doubleday, Garden City, New York.
- SINGER, F. J., C. M. PAPOUCHIS, AND K. K. SYMONDS. 2000. Translocations as a tool for restoring populations of bighorn sheep. Restoration Ecology 8:6–13.
- SLATKIN, M. 1995. A measure of population subdivision based on microsatellite allele frequencies. Genetics 139:457–462.
- TAMURA, K., G. STECHER, D. PETERSON, A. FILIPSKI, AND S. KUMAR. 2013. MEGA6: molecular evolutionary genetics analysis version 6.0. Molecular Biology and Evolution 30:2725–2729.
- THOMPSON, J. D., D. G. HIGGINS, AND T. J. GIBSON. 1994. CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. Nucleic Acids Research 22:4673–4680.
- VALDEZ, R., AND P. R. KRAUSMAN. 1999. Description, distribution, and abundance of mountain sheep in North America. Pp. 1–21 in Mountain Sheep of North America (R. Valdez and P. R. Krausman, eds.). University of Arizona Press, Tucson.
- WANG, X. 1988. Systematics and population ecology of late Pleistocene bighorn sheep (*Ovis canadensis*) of Natural Trap Cave, Wyoming. Transactions of the Nebraska Academy of Sciences and Affiliated Societies, Lincoln, Nebraska, 194.
- WAPLES, R. S. 1991. Pacific salmon, *Oncorhynchus* spp., and the definition of "Species" under the Endangered Species Act. Marine Fisheries Review 53:11–22.
- WEEKS, A. R., ET AL. 2011. Assessing the benefits and risks of translocations in changing environments: a genetic perspective. Evolutionary Applications 4:709–725.
- WEHAUSEN, J. D. 1991. Some potentially adaptive characters of mountain sheep populations in the Owens Valley region. Pp. 209–230 in Natural history of eastern California and high-altitude research (C. A. Hall, V. Doyle-Jones, and B. Widawski, eds.). White Mountain Research Station, Los Angeles, California.

- WEHAUSEN, J. D. 2005. Nutrient predictability, birthing seasons, and lamb recruitment for desert bighorn sheep. Pp. 37–50 in Sweeney Granite Mountains Desert Research Center 1978–2003: a quarter century of research and teaching (J. Goerrissen and J. M. Andre, eds.). University of California Natural Reserve Program, Riverside.
- WEHAUSEN, J. D., AND R. R. RAMEY. 1993. A morphometric reevaluation of the Peninsular bighorn sheep subspecies. Desert Bighorn Council Transactions 37:1–10.
- WEIKARD, R., C. PITRA, AND C. KUHN. 2006. Amelogenin cross-amplification in the family bovidae and its application for sex determination. Molecular Reproduction and Development 73:1333–1337.

- WEIR, B. S., AND C. C. COCKERHAM. 1984. Estimating F-statistics for the analysis of population structure. Evolution 38:1358–1370.
- WHITTAKER, D. G., S. D. OSTERMANN, AND W. M. BOYCE. 2004. Genetic variability of reintroduced California bighorn sheep in Oregon. Journal of Wildlife Management 68:850–859.
- WILSON, D. E., AND D. M. READER. 2005. Mammal Species of the world. A taxonomic and geographic reference. 3rd ed. Johns Hopkins University Press, Baltimore, Maryland.

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Domestic sheep, bighorn sheep, and respiratory disease: a review of the experimental evidence

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Introduced infectious diseases pose a significant threat to wildlife populations and are exceptional conservation challenges, in part because they can precipitate much more rapid and devastating population declines than habitat encroachment. Pneumonia epizootics have played a major role in the dynamics and conservation challenges of bighorn sheep (Ovis canadensis) populations. A large proportion of native bighorn sheep populations south of Canada went extinct beginning in the second half of the 19th century. It has long been postulated, based on temporal and spatial correlations, that diseases transferred from domestic sheep (Ovis aries) played a major role in those losses. Although experimental research has repeatedly tested the hypothesis that domestic sheep carry strains of respiratory tract pathogens potentially fatal to bighorn sheep, debate continues over the role of domestic sheep in this disease process. In the context of a hierarchical set of hypotheses we review this experimental research that includes (1) contact trials involving bighorn sheep penned with domestic sheep and a variety of other native and domestic animal species; (2) inoculation experiments with no animal contact; (3) attempts to isolate and identify specific organisms responsible for pneumonia in bighorn sheep; and (4) vaccination experiments. Our review reveals that (1) experiments have repeatedly corroborated the hypothesis that bighorn sheep have a high probability of contracting fatal pneumonia following contact with domestic sheep; (2) low disease and mortality rates in numerous co-pasturing pen studies involving bighorn sheep and animals other than domestic sheep do not support the alternative explanation that the results of the co-pasturing studies involving domestic sheep were an artifact of captivity; (3) the identification of which organism(s) cause pneumonia in bighorn sheep following contact with domestic sheep remains unresolved, possibly because of disease complexity (multiple pathogens) and limitations of research tools applied; and (4) vaccination trials largely have failed to mitigate the spread of respiratory disease and appear to be an unrealistic solution to the problem. We discuss these findings relative to a variety of questions, misinterpretations, and implications for management decisions concerning bighorn sheep conservation.

Key words: Bighorn sheep, domestic sheep, Ovis aries, Ovis canadensis, Ovis dalli, pneumonia, respiratory disease, Pasteurella, Mannheimia

Introduced infectious diseases pose a tremendous threat to wildlife. This threat increases as animal population sizes decrease, thereby reducing the gene pool of potentially resistant individuals, further increasing the likelihood of endangerment and extinction. Examples of the devastating effects of introduced diseases on wildlife are legion (Daszak et al. 2000), and the history of bighorn sheep (Ovis canadensis) provides a well-studied example that includes conservation actions taken to protect populations and reverse declining population trends through reintroductions. The tremendous amount of money and effort spent to repatriate bighorn sheep and the intense policy disputes over conservation strategies make this species an excellent case study of wildlife conservation in the face of disease. In this paper we review the scientific evidence for the most widely-cited hypothesis concerning the cause of many pneumonia epizootics in bighorn sheep — that bighorn sheep have a high probability of contracting fatal respiratory disease after contact with domestic sheep (Ovis aries), hereafter referred to as the "contact hypothesis". The implications of this hypothesis relative to bighorn sheep conservation and related decisions by governmental agencies that permit domestic sheep grazing on their lands has made this a contentious issue. Decision makers cannot be expected to study the diverse literature on this subject, yet have to make informed decisions in the face of pressure from both sides of this issue. A detailed review of this literature is clearly needed to help decision makers assess the scientific merit of various claims, as well as to synthesize existing information. Given that the introduction of domesticated animals has been connected with emerging infectious diseases in other wildlife (Daszak et al. 2000), the lessons learned from disease research on bighorn sheep also may have broader applications.

The original distribution of the two native sheep species in western North America, bighorn sheep and Dall's sheep (*O. dalli*), included suitable habitat north to the Brooks Range in Alaska, south to Baja California and the northern reaches of mainland Mexico, and east as far as west Texas and badland and river break habitats immediately east of the Rocky Mountains in North and South Dakota and western Nebraska (Buechner 1960, Valdez and Krausman 1999). In a large portion of this habitat in Alaska and Canada, the distribution of native sheep remains essentially unchanged (Valdez and Krausman 1999). In contrast, across much of the southern range of bighorn sheep, many populations were extirpated, including all native populations in the states of Washington, Oregon, and neighboring regions of southwestern Idaho, northeastern California, and northwestern Nevada (Buechner 1960). The states of California and Nevada together lost an estimated total of 110 native populations (McQuivey 1978, Wehausen et al. 1987). Restoration efforts were initiated during the 20th century to counter continuing population losses,

and by 1990 more than 8,000 bighorn sheep had been moved in 592 translocation efforts, primarily to restock vacant habitat (Ramey 1993).

The large region where bighorn sheep extirpations have been so widespread coincides spatially with where domestic sheep have been grazed in North America, and temporally with the beginning of that grazing. While one cannot infer cause and effect from spatial and temporal correlations alone, it has long been hypothesized that diseases transferred from domestic sheep were a key factor in the widespread loss of bighorn sheep populations. For example, the principal cause of the first large-scale population losses in the 19th century was attributed to scabies introduced by domestic sheep, based largely on clinical evidence of scabies in bighorn sheep during die-offs, and the temporal association of these scabies outbreaks with the introduction of domestic sheep (Honess and Frost 1942, Jones 1950, Smith 1954, Buechner 1960). Further negative correlations between the presence of domestic sheep and the health of bighorn sheep populations have emerged in the 20th century. In Nevada McQuivey (1978) noted a negative correlation between past domestic sheep grazing and the persistence of native bighorn sheep populations, and considerable circumstantial evidence has accumulated suggesting the hypothesis that dieoffs of bighorn sheep frequently follow contact with domestic sheep (Goodson 1982, Martin et al. 1996, Singer et al. 2001, Coggins 2002, George et al. 2008). Where clinical evidence has been collected, pneumonia has been cited as the cause of death in those dieoffs (Goodson 1982, Martin et al. 1996).

METHODS

During the past three decades various aspects of the potential role of domestic sheep in respiratory disease of bighorn sheep have been researched. Because scientific progress is limited in part by how problems are analyzed to formulate hypotheses and deduce testable (falsifiable) predictions, in this review of those research results we attempt to isolate separate questions and hypotheses concerning disease transmission between domestic and wild sheep. In so doing, we formulate a hierarchical series of hypotheses that are refinements of the contact hypothesis. We also approach this review from the standpoint of opportunities for hypothesis falsification. Popper (1959) identified falsifiability of hypotheses as the fundamental criterion of valid scientific inquiry of questions of cause and effect. He also argued that "proof" falls outside of the realm of science; instead, acceptance of hypotheses and the strength of such corroboration is a function of the attempts at and opportunities for falsification. We consider these concepts as fundamental to the disease questions that we review.

RESULTS

Our separation of this review into different questions and hypotheses lead us to partition the findings into six categories that facilitate the discussion of these different topics.

Unplanned pen experiments.—The contact hypothesis has been tested numerous times in captive situations. One set of tests has been accidental in nature and, therefore, lacked experimental design. However, the information garnered from those captivity situations still served as tests of the contact hypothesis. One unplanned experiment

CALIFORNIA FISHAND GAME

occurred at Lava Beds National Monument, where in 1971 a population of bighorn sheep was established in a 5.4-km² enclosure (Blaisdell 1972). In 1980, nose-to-nose contact was observed through the enclosure fence between bighorn sheep and domestic sheep grazed on adjacent National Forest lands. Bighorn sheep began dying of pneumonia 2-3 weeks later and all 43 bighorn subsequently died (Foreyt and Jessup 1982). A second unplanned experiment involved bighorn sheep in Washington that had been in a 2.5-ha enclosure for 10 months when domestic sheep were added to the pen. Thirteen of 14 bighorn sheep subsequently died of pneumonia between 3 and 12 weeks after the introduction of the domestic sheep (Foreyt and Jessup 1982).

Planned pen experiments.—Following those unplanned experiments, 10 planned experiments specifically designed to test the contact hypothesis were carried out by three independent research groups using 1-6 captive bighorn sheep per trial. Four of those experiments used only domestic sheep (Onderka and Wishart 1988; Foreyt 1989, 1990, 1994), while contact in one (Foreyt 1994) involved mouflon sheep (*Ovis musimon*) and another five involved a mixed flock of domestic sheep and hybrids of argali (*Ovis ammon*) and mouflon sheep (Callan et al. 1991), the latter of which is the closest ancestor of domestic sheep (Ramey 2000, Hiendleder et al. 2002). The five trials involving hybrid sheep also included experimental treatments that attempted to control the resulting pneumonia in the bighorn sheep. Of the 23 bighorn sheep tested in those 10 trials, all died of respiratory disease following contact with domestic sheep, or were euthanized when close to death (Table 1). All domestic, mouflon, and hybrid sheep remained healthy.

Planned pen experiments with other species.—A couple of hypotheses might explain the planned pen results: (1) contact results in transmission of pathogens from domestic sheep to bighorn sheep that directly or indirectly lead to fatal pneumonia in the bighorn sheep (pathogen transmission hypothesis); or (2) the introduction of another species into the pen creates a negative psychological effect on the bighorn sheep, resulting in a compromised immune system leading to respiratory disease unrelated to the transmission of potential disease agents (stress hypothesis). Stress of behavioral origin similarly has frequently been hypothesized as an important factor in the livestock respiratory disease syndrome known as shipping fever (Hoerlein 1980, Yates 1982).

	Number of trials	Wild sheep tested	Wild sheep dying	Domestic sheep and other species used	Domestic sheep and other species dying
Unplanned Pen Trials with domestic sheep	2	57	56		
Planned Pen Trials with domestic sheep	10	23	23	167	0
Planned Pen Trials with other species	9	55	4	32	0
Unplanned Inoculation Trials	1	13	6		
Planned Inoculation Trials	6	15	14	9	0

TABLE 1.—Summary of data from contact trials of bighorn sheep co-pastured with other species and inoculation trials of bighorn sheep and Dall's sheep (planned inoculation trials only) that used *M. haemolytica* cultured from domestic sheep.

Winter 2011 DOMESTIC AND BIGHORN SHEEP RESPIRATORY DISEASE

Nine independent contact experiments by Foreyt (1992a, 1994), Foreyt and Lagerquist (1996), and Foreyt et al. (2009) involving bighorn sheep penned with (1) elk (Cervus elephus), white-tailed deer (Odocoileus virginianus), and mule deer (Odocoileus hemionus); (2) elk alone; (3) domestic goats (Capra hircus); (4) mountain goats (Oreamnos americanus); (5) llamas (Lama glama); (6) cattle (Bos taurus); and (7) horses (*Equus caballus*) serve as a test of the stress hypothesis (Table 2). Of 55 bighorn sheep tested in those experiments, only four died (Table 1). One was an old female whose death most likely was due to a tooth anomaly that adversely affected her feeding ability. The other deaths were a bighorn sheep in the experiment with steers that died of pneumonia (Foreyt and Lagerquist 1996) and two of seven bighorn co-pastured in one trial with domestic goats that died of pneumonia caused by Mannheimia haemolytica (Angen et al. 1999) biotype A, serotype 2 (Foreyt et al. 2009; Table 2). The significantly (P < 0.001; chi square test) lower proportion of bighorn sheep dying in pen trials that put bighorn sheep in contact with other species compared with experiments involving contact with sheep of Old World origin (Table 1) does not support the stress hypothesis. Instead, these findings suggest that the presence of other species in pens itself is unlikely to lead to bighorn sheep deaths and, furthermore, that species other than domestic sheep and their relatives are considerably less likely to transmit pathogens potentially fatal to bighorn sheep. This conclusion is consistent with a lack of circumstantial data linking most of these other species to bighorn sheep die-offs. Domestic goats appear to be the exception (Rudolph et al. 2003), and recent findings indicate that they also can carry other disease organisms with serious consequences for bighorn sheep (Jansen et al. 2006). However, the lack of disease transmission to bighorn sheep by the other species tested does not imply that they lack respiratory tract organisms pathogenic to bighorn sheep; instead, lack of disease may result from interspecific behavioral patterns that largely preclude contact and pathogen transmission.

 TABLE 2.—Details from contact experiments involving bighorn sheep co-pastured with domestic sheep (Dom. sheep), mouflon sheep, domestic goats (Dom. goats), white-tailed deer (W-T deer), mule deer, elk, mountain goats (Mt. Goats), llamas, horses, and cattle.

					С	o-pastureo	ł with:					
Bighorn used	Bighorn dead	Dom. sheep	Moufl on sheep	Dom. goats	W-T deer	Mule deer	Elk	Mt. goats	Llamas	Horses	Cattle	reference
2	2	2	-					-	-			Onderka and Wishart 1988
б	6	б	-	-	-	-	-	-	-	-	-	Foreyt 1989
2	2	2	-	-	-	-	-	-	-	-	-	Foreyt 1990
б	6	-	5	-	-	-	-	-	-	-	-	Foreyt 1994
2	2	2	-	-	-	-	-	-	-	-	-	Foreyt 1994
1	1	30^{a}	-	-	-	-	-	-	-	-	-	Callan et al. 1991
1	1	30^{a}	-	-	-	-	-	-	-	-	-	Callan et al. 1991
1	1	30^{a}	-	-	-	-	-	-	-	-		Callan et al. 1991
1	1	30^{a}	-	-	-	-	-	-	-	-	-	Callan et al. 1991
1	1	30^{a}	-	-	-	-	-	-	-	-	-	Callan et al. 1991
10	0	-	-	-	2	1	4	-	-	-	-	Foreyt 1992a
3	0	-	-	-	-	-	4	-	-	-	-	Foreyt 1992a
9	0	-	-	-	-	-	-	2	-	-	-	Foreyt 1994
9	0	-	-	-	-	-	-	-	3	-	-	Foreyt 1994
4	0	-	-	-	-	-	-	-	-	-	3	Foreyt 1994
5	1	-	-	-	-	-	-	-	-	-	3	Foreyt and Lagerquist 1990
6	1^{b}	-	-	-	-	-	-	-	-	3	-	Foreyt and Lagerquist 1990
2	0	-	-	3	-	-	-	-	-	-		Foreyt 1994
7	2	-	-	4	-	-	-	-		-		Forevt et al. 2009

mixture of domestic sheep and mouflon-argali hybrids

animal in poor condition at beginning of experiment; death likely caused by tooth anomalies and feeding difficulty.

CALIFORNIA FISHAND GAME

Inoculation experiments.—The pathogen transmission hypothesis can be further refined to the fatal strains hypothesis (Goodson 1982): that specific species, microbial strains, or viruses frequently carried by healthy domestic sheep are the cause of fatal pneumonia in bighorn sheep following contact between these species. This hypothesis has been tested by experiments in which captive bighorn sheep have been inoculated with bacteria cultured from the respiratory tracts of domestic sheep. Similar to the contact experiments, this has involved both accidental and planned experiments. The accidental experiment occurred when a lavage tube used to sample lung cells of domestic sheep was not fully sterilized before being used to obtain lung cultures from three captive bighorn sheep. All 10 bighorn sheep in this herd developed pneumonia, of which three died, as did three additional bighorn sheep added to the herd (Foreyt 1990).

The planned inoculation experiments comprise six independent trials carried out by two different research groups using *M. haemolytica* cultured from domestic sheep (Onderka et al. 1988, Foreyt et al. 1994, Foreyt and Silflow 1996). Of 13 bighorn sheep that were inoculated with those bacteria, 12 died of acute bronchopneumonia. Two groups of control bighorn sheep (five total) remained healthy, as did two groups of domestic sheep (nine total) that received the same inoculation doses as the bighorn sheep (Table 1). Two of these inoculation trials (Onderka et al. 1988, Foreyt and Silflow 1996) also included experiments in which the source of the *M. haemolytica* inoculum was cultured from healthy bighorn sheep. The three bighorn sheep used in those two trials showed no clinical signs of disease after the inoculations, and neither did seven domestic sheep similarly inoculated.

Foreyt et al. (1996) also carried out an inoculation trial of three Dall's sheep (*Ovis dalli dalli*). Two of these sheep received a *M. haemolytica* strain (A2) from domestic sheep that by inoculation trials was fatal to bighorn sheep, while the other received a strain not considered to be pathogenic. The sheep receiving the non-pathogenic strain remained healthy; the other two developed bronchopneumonia, from which one died, and one was euthanized prior to death.

Dassanayake et al. (2009) used 10 bighorn and 12 domestic sheep to test two forms of the *M. haemolytica* A1 strain in inoculation trials. Two bighorn and two domestic sheep were controls, while four of each species received the wild type A1 strain, and the other four received a mutant A1 form that lacked the leukotoxin gene (Murphy et al. 1995). One control domestic sheep died of causes unrelated to the experimental treatment. All other sheep survived without clinical pneumonia except the four bighorn sheep that received the wild type A1 strain, all of which died of acute bilateral pneumonia within 48 hours. These results appear to expand the list of strains fatal to bighorn sheep. However, the *M. haemolytica* A1 strain used was identified only as wild type with no information on its source (Dassanayake et al. 2009, Murphy et al. 1995).

Besser et al. (2008) tested the role of *Mycoplasma ovipneumoniae* alone in this disease process by inoculating two young bighorn lambs. Neither showed signs of clinical pneumonia.

Research to identify bacterial strains causing fatal pneumonia in bighorn sheep.—The results of the various contact and inoculation trials corroborate the pathogen transmission and fatal strains hypotheses. With sufficient diagnostic tools, it should theoretically be possible to identify the specific strain(s) of bacteria or other pathogens that cause fatal pneumonia in bighorn sheep. However, the goal of identifying all specific pathogens has proven elusive. Multiple bacterial species have been implicated as causing disease in bighorn sheep. While *M. haemolytica* has been cultured from many bighorn sheep dying of pneumonia following experimental contact with domestic sheep, especially the A2 strain, one set of experiments attributed the deaths instead to *Pasteurella multocida* (Callan et al. 1991). Additionally, some forms of *M. haemolytica* are now recognized as a separate species, *P. trehalosi* (Sneath and Stevens 1990). Traditional methods used to differentiate strains of *M. haemolytica* by biotypes and serotypes (Dunbar et al. 1990a, 1990b; Queen et al. 1994) have lacked adequate resolution. Previously unknown serotypes have been found in bighorn sheep (Dunbar et al. 1990a, Silflow et al. 1994, Sweeney et al. 1994, Ward et al. 1997), rendering these classification methods unsatisfactory for epidemiological investigations of this phenomenon (Jaworski et al. 1993).

To overcome limitations of traditional methods, additional diagnostic tools have been applied to *M. haemolytica* and *P. trehalosi* in attempts to develop more refined classifications that might better identify strains responsible for bighorn sheep deaths. These measures have included (1) binary classification as hemolytic or non-hemolytic (Wild and Miller 1991, 1994; Ward et al. 2002); (2) variation in surface proteins (Ward et al. 1990); (3) assays of toxicity relative to peripheral neutrophils (Silflow and Foreyt 1994, Silflow et al. 1994, Sweeney et al. 1994); (4) DNA fingerprinting to identify different genetic forms (Snipes et al. 1992, Jaworski et al. 1993; Foreyt et al. 1994, Ward et al. 1997, Weiser et al. 2003); and, (5) culture-independent PCR-based methods and sequencebased phylogenetic analyses of multiple genetic loci (Safaee et al. 2006, Kelley et al. 2006, Besser et al. 2008).

Silflow et al. (1989) found no differences between bighorn sheep and domestic sheep in a number of immune system measures involving phagocytes. In contrast, Silflow et al. (1993) identified a mechanism involving lysis of neutrophils by a cytotoxin produced by some *M. haemolytica* strains that might explain the high susceptibility of bighorn sheep to specific strains of *M. haemolytica*. Comparisons of neutrophil sensitivity to this cytotoxin for five native North American ungulates and domestic sheep found bighorn sheep, and especially Dall's sheep, to be notably more susceptible to neutrophil destruction than the other species tested (Silflow and Foreyt 1994, Silflow et al. 1994). While strains of *M. haemolytica* fatal to bighorn and Dall's sheep consistently showed high toxicity in cytotoxicity assays (Foreyt and Silflow 1996, Foreyt et al. 1996), other cytotoxic strains have not caused significant respiratory disease in bighorn sheep (Foreyt and Silflow 1996); thus, this cytotoxicity classification alone lacks adequate predictive power relative to respiratory disease in bighorn sheep. The same can be said of the other diagnostic methods. While DNA fingerprinting has been useful for investigating transmission of bacterial strains between different species and individuals (Ward et al. 1997), these methods also appear to lack predictive power relative to identifying strains that can cause fatal pneumonia in bighorn sheep.

One possible explanation for the failure of these diagnostic methods to consistently identify bacterial strains fatal to bighorn sheep is that the culturing methods they depend on do not identify most members of the microbial community sampled. Results from culture-independent PCR-based methods indicate that culture-based methods typically miss about 99% of microbial diversity in any given biological sample (Amann et al. 1995, Hugenholtz and Pace 1996, Tanner et al. 1999, Eckburg et al. 2005), including sheep respiratory tracts (Safaee et al. 2006, Besser et al. 2008). Furthermore, several studies have found evidence that horizontal gene transfer of the leukotoxin gene has occurred

CALIFORNIA FISH AND GAME

among *Mannheimia/Pasteurella* species sampled from different species and locales in both domestic (Davies et al. 2001, Davies et al. 2002) and wild sheep populations (Kelley et al. 2006). This same mechanism contributes to virulence in other bacteria, including shiga toxin, cholera toxin, and neurotoxins of *Clostridium botulinim* (Novick 2003). While Kelley et al. (2006) found that DNA sequences from *Mannheimia* and *Pasteurella* obtained from different host species and locales tend to form closely related clusters, horizontal gene transfer of leukotoxin and other virulence genes may explain a lack of correspondence between strains identified using traditional methods and their virulence. Evidence of extensive recombination of the toxin genes within *P. trehalosi and M. haemolytica* (Davies et al. 2001) suggests that presence of this gene in a population of *Mannheimia* or *Pasteurella* does not necessarily mean that it is virulent. Most recently, using culture-independent approaches, Besser et al. (2008) found evidence suggesting involvement of *Mycoplasma ovipneumoniae* in bighorn sheep respiratory disease.

Vaccination trials.—Vaccination of wild animals is logistically difficult at best in most situations and even more so for bighorn sheep because of the steep, craggy, relatively inaccessible habitat they often inhabit. Additionally, some vaccines require multiple doses to stimulate initial immune system response. Thus, vaccination is not a viable disease management option for most wild populations. Nevertheless, vaccination experiments have been carried out and might have applications to captive wild sheep and occasional free-ranging situations.

Ward et al. (1999) investigated immunologic responses of bighorn and domestic sheep to a vaccine against three strains of *M. haemolytica*. They found that the vaccine produced only a moderate and transient immunologic response. Miller et al. (1997) and Kraabel et al. (1998) tested a vaccine for three different *M. haemolytica* strains on captive bighorn sheep. The sheep were challenged with *P. trehalosi* cultured from lungs of free-ranging bighorn sheep during a pasteurellosis epizootic. Control and vaccinated bighorn both developed acute pneumonia, but vaccinated ones experienced lower mortality (30% vs. 80%).

For multiple years following pneumonia epizootics in bighorn sheep, it is common for most lambs of surviving females to die of pneumonia (Foreyt 1990, Coggins and Matthews 1992). Cassirer et al. (2001) conducted experiments with free-ranging and captive bighorn to test the efficacy of vaccines against *Mannheimia/Pasteurella* to reduce such lamb mortality, but vaccinated females had notably higher loss of lambs than nonvaccinated ewes.

Only two vaccination trials have used strains of *M. haemolytica* derived from domestic sheep as the post-vaccination challenge. Foreyt and Silflow (1996) inoculated two bighorn sheep twice with a non-lethal cytotoxic strain of *M. haemolytica* and six weeks later inoculated them with a lethal cytotoxic strain (A2) from domestic sheep. The non-lethal strain provided no significant protection, and both bighorn sheep died of bronchopneumonia. Foreyt (1992b) tested an experimental bacterin-toxoid vaccine for three *M. haemolytica* strains, using three treatment and three control bighorn sheep. After contact with domestic sheep, five of the six bighorn sheep, including the three vaccinated ones, died of pneumonia, with no evidence of any protection from the vaccine.

DISCUSSION

A variety of field observations spanning many decades led to the hypothesis that bighorn sheep have a high probability of developing fatal pneumonia following contact with domestic sheep. Subsequently, numerous independent experiments have tested this contact hypothesis, and the results have repeatedly corroborated it (Table 1, Table 2). There have been numerous opportunities to falsify the contact hypothesis under controlled conditions and none has done so. Many bighorn sheep have died in those experiments, and it seems unlikely that more such experiments will add further knowledge to the contact hypothesis.

The stress and pathogen transmission hypotheses were proposed as two basic mechanisms to explain the results of contact experiments; but only the pathogen transmission hypothesis was consistent with the experimental data. The pathogen transmission hypothesis and the more refined fatal strains hypothesis have been tested and corroborated by *M. haemolytica* inoculation experiments. While the realistic nature of the inoculation doses might be questioned, domestic sheep similarly inoculated remained healthy, as did control bighorn sheep; and similar inoculation doses of *M. haemolytica* strains cultured from bighorn sheep produced no clinical effects in either sheep species. These results are consistent with expectations from the fatal strains hypothesis and provided opportunities for falsification.

The effort to identify organisms causing pneumonia in bighorn sheep following contact with domestic sheep has not yielded simple answers; instead, this situation appears complex with many potentially pathogenic bacteria of multiple species identified. In part, this may reflect limitations of the technology applied to this question. New culture-independent methods are greatly expanding knowledge of microbial communities inhabiting animals (Eckburg et al. 2005) and are beginning to shed new light on disease transmission (Tanner et al. 1999). However, the hunt to identify organisms causing pneumonia in bighorn sheep appears to have been limited by a traditional search for specific bacterial species or strains. The expanding understanding of potential mechanisms underlying pathogenicity in other diseases, e.g., horizontal gene transfer (Schubert et al. 2009), may explain why such a traditional approach has not been successful for bighorn sheep. Indeed, even criteria for defining bacterial species remain unclear (Fraser et al. 2009).

Definitive identification of pathogens causing fatal pneumonia in bighorn sheep is a question of scientific interest that may ultimately have practical applications. However, the inability to definitively and consistently identify pathogens responsible for all bighorn sheep deaths following contact with domestic sheep does not have bearing on the question of whether such contact has a high probability of leading to deaths of bighorn sheep. These are different questions that frequently have been inappropriately intertwined. Shipping fever is a similar respiratory disease problem that costs the U. S. livestock industry many millions of dollars annually (Rehmtulla and Thomson 1981); yet, it also has not yielded a single causative disease agent despites decades of intensive research (Storz et al. 2000).

A glance backwards to the early days of human public health shows that stalling epidemics has not required complete knowledge of the disease mechanism or identification of the pathogen. Without any knowledge of the microbial cause of cholera, John Snow hypothesized that the source of the 1849 London epidemic as water from one well. He

CALIFORNIA FISHAND GAME

tested his hypothesis by removing the handle to the pump for that well, which provided corroboration when the epidemic ended abruptly (Glass 1986, Garrett 1994). His hypothesis was analogous to our contact hypothesis and his scientific conclusions did not require knowledge of the specific pathogen causing the disease. In fact, it was another three decades after Snow halted that particular epidemic before the cholera bacterium was established as the cause of that disease (Howard-Jones 1984). Because city planners refused to accept Snow's reasoning that water contaminated by sewage was the source of the epidemic, cholera outbreaks continued to plague London for decades (Garrett 1994). The resistance of some to the apparent role of domestic sheep in bighorn sheep pneumonia suggests a parallel situation.

One of the principal reasons some critics have cited for doubting the contact hypothesis is that Koch's postulates for establishing a causative relationship between a microbe and a disease have not been convincingly fulfilled. Among other things, Koch's postulates propose that to identify a microbial agent as the cause of a human disease, it is necessary to isolate the same organism from each case of the disease, and to produce that disease in an animal by inoculating it with that agent cultured from a diseased individual (Fredericks and Relman 1996). While the same postulates apply to animal diseases, Hanson (1988) concluded that the application of Koch's postulates to the study of wildlife diseases was a simplistic approach to a complex situation that had little meaning given current knowledge and technology, and this general concern has been echoed by others (Evans 1976, Fredericks and Relman 1996). Indeed, Koch himself later recognized that his postulates could not be satisfied in every case (Fredericks and Relman 1996). The respiratory disease relationship between domestic and bighorn sheep appears to epitomize that conclusion. By the definition of a disease implied by Koch's postulates, the disease phenomenon reviewed here may involve multiple disease processes involving multiple microbial species and strains. Additionally, a lesson from studies using cultureindependent PCR methods is that Koch's postulates can be applied to only a small fraction of potential pathogens that can be cultured for inoculation.

This review examined only the experimental evidence concerning whether domestic sheep are a likely source of respiratory pathogens potentially fatal to bighorn sheep. How any situation of potential contact between these species in the wild will play out is a complex question that involves a series of probabilistic events. First is the probability of contact between the two species. Second is the probability that pathogenic strains are transferred. Third is the probability that pathogen transmission will lead to pneumonia, a probability possibly influenced by the status of the immune system of the bighorn(s) receiving pathogenic strains relative to the dose received. Fourth is the process of pathogen transfer within an infected bighorn sheep population. Fifth is the probability of death of infected individuals, which will likely vary among populations due to multiple variables, including genetic constitution of the herd, nutrition, environmental stressors, and the virulence of pathogen(s). Because post die-off population dynamics are often influenced by survivors of such pneumonia epizootics that carry and transmit respiratory tract pathogens to lambs for years (Foreyt 1990, Coggins and Matthews 1992), there are questions of yet longer term interactions between herd immunity and pathogens. Below we touch on a few questions of this larger disease question.

Sheep in general are susceptible to pneumonia, and bighorn sheep appear particularly susceptible to this disease, exhibiting periodic pneumonia die-offs in the Rocky Mountain region (Buechner 1960, Stelfox 1971). While some of these epizootics can be traced to

contact with domestic sheep and subsequent inter-population migration of pathogens within metapopulations (Goodson 1982, Onderka and Wishart 1984, George et al. 2008), there is a large literature that we do not review documenting pneumonia outbreaks and die-offs in bighorn sheep populations with no known recent prior contact with domestic sheep (Goodson 1982, Martin et al. 1996). Researchers typically have attributed these latter pneumonia outbreaks to various environmental conditions likely to predispose wild sheep to respiratory disease (Festa-Bianchet 1988, Monello et al. 2001), but Hobbs and Miller (1992) suggested that such conditions might not be necessary. However, the lack of any documented pneumonia epizootics in the large expanse of wild sheep range in Canada and Alaska, where there has been almost (Heimer et al. 1992) no opportunity for direct or indirect contact with domestic sheep (Hoefs and Cowan 1979, Hoefs and Bayer 1984, Monello et al. 2001, Jenkins et al. 2007) is a pattern needing explanation. Among potential hypotheses is that bighorn sheep populations that have survived past pneumonia epizootics resulting from contact with domestic sheep continue to carry respiratory microbes from domestic sheep that (1) are lacking in Alaska and most of Canada; and, (2) render these bighorn sheep more susceptible to pneumonia when various environmental conditions converge to compromise immune systems and/or there is an evolutionary change in pathogen virulence.

The role of predisposing factors in outcomes of pneumonia epizootics of wild bighorn sheep populations stemming from recent contact with domestic sheep also is unclear. Results from pen experiments suggest that the virulence of pathogens transferred in such contact can overpower the immune system of bighorn sheep regardless of prior physical condition and diet quality; but, the applicability of experimental results to wild situations has nevertheless been questioned, and such epizootics in the wild do vary considerably in the proportion of the herd that dies (ca. 50-100%; Goodson 1982, Martin et al. 1996). While extensive replicated experiments on wild populations would be desirable to help clarify cause and effect, it is doubtful that such research will occur. Statistically it would be appropriate to have at least three treatment and three control populations. Given the value of the bighorn sheep resource and the implications of the existing data reviewed here, it is unlikely that any agency with jurisdiction over bighorn sheep would be willing to subject multiple healthy populations of bighorn sheep to the risk of a severe pneumonia epizootic resulting from such an experiment. Ethical questions also might arise. As scientific experiments, the pen trials we reviewed were carried out specifically to control as many confounding variables operating in wild populations as possible in order to best measure the effects of the variable of interest. In that regard those pen trials potentially yield more important information than might be obtained from experiments involving wild populations. Contact between domestic and bighorn sheep in the wild may not always produce the same consistency of results seen in controlled pen studies because of variables outlined above; however, it is well known in epidemiology that probabilities of disease transmission to susceptible hosts increase with repeated exposure (Frerichs 1995). Consequently, greater variation in observations from wild situations might be expected relative to results from pen studies. A prediction from the results of pen studies reviewed here might be that repeated opportunities for contact between domestic sheep and bighorn sheep eventually will lead to a pneumonia epizootic in the bighorn sheep. Aune et al. (1998) documented this for one bighorn sheep population in Montana.

CALIFORNIA FISH AND GAME

Relative to resource management decisions, the pertinent question is whether bighorn sheep have a high probability of developing fatal pneumonia following contact with domestic sheep. While desirable, it is not necessary to completely understand details of the disease process, or even identify responsible pathogens, to make appropriate management decisions. Relative to other judgments that must be made by resource management agencies, the potential effect on bighorn sheep of contact with domestic sheep appears remarkably clear cut. Where the health of any bighorn sheep populations is valued, the recommendation has been management actions that prevent contact with domestic sheep (Foreyt 1994, Foreyt et al. 1994). Such contact can occur in two ways: stray domestic sheep contacting bighorn sheep, or bighorn sheep contacting domestic sheep bands and spreading pathogenic microbes to other bighorn sheep. Keeping an adequate spatial buffer between bighorn sheep and domestic sheep has been considered the most reliable method to prevent contact between these species (Desert Bighorn Council Technical Staff 1990, Bureau of Land Management 1992, Schommer and Woolever 2001, Singer et al. 2001). However, this solution may not always be adequate because of distances bighorn sheep males sometimes travel, and politically is seldom simple to achieve. Depending on the situation, other approaches may be possible. Finding a management solution to this problem is dependent on the parties first agreeing that contact between domestic and bighorn sheep is a significant health threat for bighorn sheep. It is our hope that this review will help assure that such agreements will be based on a complete and critical review of pertinent scientific information that separates different falsifiable hypotheses, and thereby does not mix questions that should be addressed independently.

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LITERATURE CITED

- AMANN, R. I., W. LUDWIG, AND K. H. SCHLEIFER. 1995. Phylogenetic identification and *in situ* detection of microbial-cells without cultivation. Microbiological Reviews 59:143-169.
- ANGEN, O, R. MUTTERS, D. A. CAUGANT, J. E. OLSEN, AND M. BISGAARD. 1999. Taxonomic relationships of the [Pasteurella] haemolytica complex as evaluated by DNA-DNA hybridizations and 16s rRNA sequencing with proposal of Mannheimia haemolytica gen. nov., comb. nov., Mannheimia granulomatis comb. nov., Mannheimia glucosida sp. nov., Mannheimia ruminalis sp. nov. and Mannheimia varigena sp. nova. International Journal of Systematic Bacteriology 49:67-86.
- AUNE, K., N. ANDERSON, D. WORLEY, L. STACKHOUSE, J. HENDERSON, AND J. DANIEL. 1998. A comparison of population and health histories among seven Montana bighorn sheep populations. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 11:46-69.
- BESSER, T. E., E. F. CASSIRER, K. A. POTTER, J. VANDERSCHALIE, A. FISCHER, D. P. KNOWLES, D. R. HERNDON, F. R. RURANGIRWA, G. C. WEISER, AND S. SRIKUMARAN. 2008. Association of *Mycoplasma ovipneumoniae* infection with population-limiting

respiratory disease in free-ranging Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*). Journal of Clinical Microbiology 46:423-230.

- BLAISDELL, J. A. 1972. Progress report—Lava Beds bighorn re-establishment. Desert Bighorn Council Transactions 16:84-87.
- BUECHNER, H. K. 1960. The bighorn sheep in the United States, its past, present, and future. Wildlife Monographs 4:1-174.
- BUREAU OF LAND MANAGEMENT. 1992. Guidelines for domestic sheep management in bighorn sheep habitat. Instruction Memorandum No. 92-264. U.S. Department of the Interior, Washington, D.C., USA.
- CALLAN, R. J., T. D. BUNCH, G. W. WORKMAN, AND R. E. MOCK. 1991. Development of pneumonia in desert bighorn sheep after exposure to a flock of exotic wild and domestic sheep. Journal of the American Veterinary Medical Association 198:1052-1056.
- CASSIRER, E. F., K. M. RUDOLPH, P. FOWLER, V. L. COGGINS, D. L. HUNTER, AND M. W. MILLER. 2001. Evaluation of ewe vaccination as a tool for increasing bighorn lamb survival following pasteurellosis epizootics. Journal of Wildlife Diseases 37:49-57.
- COGGINS, V. L. 2002. Rocky Mountain bighorn sheep/domestic sheep and domestic goat interactions: a management prospective. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 13:165-174.
- COGGINS, V. L., AND P. E. MATTHEWS. 1992. Lamb survival and herd status of the Lostine bighorn herd following a *Pasteurella* die-off. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 8:147-154.
- DASSANAYAKE, R. P., S. SHANTHALINGAM, C. N. HERNDON, P. K. LAWRENCE, E. F. CASSIRER, K. A. POTTER, W. J. FOREYT, K. D. CLINKENBEARD, AND S. SRIKUMARAN. 2009. *Mannheimia haemolytica* serotype A1 exhibits differential pathogenicity in two related species, *Ovis canadensis* and *Ovis aries*. Veterinary Microbiology 133:366-71.
- DASZAK, P., A. E. CUNNINGHAM, AND A. D. HYATT. 2000. Emerging infectious diseases of wildlife—threats to biodiversity and human health. Science 287:443-449.
- DAVIES, R. L., S. CAMPBELL, AND T. S. WHITTAM. 2002. Mosaic structure and molecular evolution of the leukotoxin operon (lktCABD) in *Mannheimia (Pasteurella) haemolytica, Mannheimia glucosida*, and *Pasteurella trehalosi*. Journal of Bacteriology 184:266-277.
- DAVIES, R. L., T. S. WHITTAM, AND R. K. SELANDER. 2001. Sequence diversity and molecular evolution of the leukotoxin (lktA) gene in bovine and ovine strains of *Mannheimia* (*Pasteurella*) haemolytica. Journal of Bacteriology 183:1394-1404.
- DESERT BIGHORN COUNCIL TECHNICAL STAFF. 1990. Guidelines for management of domestic sheep in the vicinity of desert bighorn habitat. Desert Bighorn Council Transactions 34:33-35.
- DUNBAR, M. R., A. C. S. WARD, K. G. EYRE, AND M. BULGIN. 1990a. Serotypes of *Pasteurella haemolytica* in free ranging Rocky Mountain bighorn sheep. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 7:102-108.
- DUNBAR, M. R., A. C. S. WARD, AND G. POWER. 1990b. Isolation of *Pasteurella haemolytica* from tonsillar biopsies of Rocky Mountain bighorn sheep. Journal of Wildlife Diseases 26:210-213.

- ECKBURG, P. B., E. M. BIK, C. N. BERNSTEIN, E. PURDOM, L. DETHLEFSEN, M. SARGENT, S. R. GILL, K. E. NELSON, AND D. A. RELMAN. 2005. Diversity of the human intestinal microbial flora. Science 308:1635-1638.
- Evans, A. S. 1976. Causation and disease: the Henle-Koch postulates revisited. Yale Journal of Biology and Medicine 49:175-195.
- FESTA-BIANCHET, M. 1988. A pneumonia epizootic in bighorn sheep, with comments on preventive management. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 6:66-76.
- FOREYT, W. J. 1989. Fatal Pasteurella haemolytica pneumonia in bighorn sheep after direct contact with clinically normal domestic sheep. American Journal of Veterinary Research 50:341-344.
- FOREYT, W. J. 1990. Pneumonia in bighorn sheep: effects of *Pasteurella haemolytica* from domestic sheep and effects on survival and long-term reproduction. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 7:92-101.
- FOREYT, W. J. 1992a. Experimental contact association between sheep, elk, and deer with known *Pasteurella haemolytica* infections. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 8:213-218.
- FOREYT, W. J. 1992b. Failure of an experimental *Pasteurella haemolytica* vaccine to prevent respiratory disease and death in bighorn sheep after exposure to domestic sheep. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 8:155-163.
- FOREYT, W. J. 1994. Effects of controlled contact exposure between healthy bighorn sheep and llamas, domestic goats, mountain goats, cattle, domestic sheep, or mouflon sheep. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 9:7-14.
- FOREYT, W. J., AND D. A. JESSUP. 1982. Fatal pneumonia of bighorn sheep following association with domestic sheep. Journal of Wildlife Diseases 18:163-168.
- FOREYT, W. J., AND J. E. LAGERQUIST. 1996. Experimental contact of bighorn sheep (Ovis canadensis) with horses and cattle, and comparison of neutrophil sensitivity to *Pasteurella haemolytica* cytotoxins. Journal of Wildlife Diseases 32:594-602.
- FOREYT, W. J., AND R. M. SILFLOW. 1996. Attempted protection of bighorn sheep (*Ovis canadensis*) from pneumonia using a nonlethal cytotoxic strain of *Pasteurella haemolytica* Biotype A, Serotype¹¹. Journal of Wildlife Diseases 32:315-321.
- FOREYT, W. J., E. J. JENKINS, AND G. D. APPLEYARD. 2009. Transmission of lungworms (*Muellerius capillaris*) from domestic goats to bighorn sheep on common pasture. Journal of Wildlife Diseases 45:272-278.
- FOREYT, W. J., R. M. SILFLOW, AND J. E. LAGERQUIST. 1996. Susceptibility of Dall sheep (Ovis dalli dalli) to pneumonia caused by Pasteurella haemolytica. Journal of Wildlife Diseases 32:586-593.
- FOREYT, W. J., K. P. SNIPES, AND R. W. KASTEN. 1994. Fatal pneumonia following inoculation of healthy bighorn sheep with *Pasteurella haemolytica* from healthy domestic sheep. Journal of Wildlife Diseases 30:137-145.
- FRASER, C., E. J. ALM, M. F. POLZ, B. G. SPRATT, AND W. P. HANAGE. 2009. The bacterial species challenge: making sense of genetic and ecological diversity. Science 323:741-746.

- FREDERICKS, D. N., AND D. A. RELMAN. 1996. Sequence-based identification of microbial pathogens: a reconsideration of Koch's postulates. Clinical Microbiology Reviews 9:18-33.
- FRERICHS, R. R. 1995. HIV winners and losers. Epidemiology 6:329-331.
- GARRETT, L. 1994. The coming plague. Penguin Books, New York, New York, USA.
- GEORGE, J. L., D. J. MARTIN, P. M. LUKACS, AND M. W. MILLER. 2008. Epidemic pasteurellosis in a bighorn sheep population coinciding with the appearance of a domestic sheep. Journal of Wildlife Diseases 44:388-403.
- GLASS, R. I. 1986. New prospects for epidemiologic investigations. Science 234:951-955.
- GOODSON, N. J. 1982. Effects of domestic sheep grazing on bighorn sheep populations: a review. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 3:287-313.
- HANSON, R. P. 1988. Koch is dead. Journal of Wildlife Diseases 24:193-200.
- HEIMER, W. E., R. L. ZARNKE, AND F. J. MAUER. 1992. Pharyngeal microflora of Dall and domestic sheep in Alaska: management implication? Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 8:193-201.
- HIENDLEDER, S., B. KAUPE, R. WASSMUTH, AND A. JANKE. 2002. Molecular analysis of wild and domestic sheep questions current nomenclature and provides evidence for domestication from two different subspecies. Proceedings of the Royal Society London B 269:893-904.
- HOBBS, N. T., AND M. W. MILLER. 1992. Interactions between pathogens and hosts: simulations of pasteurellosis epizootics in bighorn sheep populations. Pages 997-1007 in D. R. McCullough and R. H. Barrett, editors. Wildlife 2001: populations. Elsevier Applied Science, Essex, England.
- HOEFS, M., AND M. B. BAYER. 1984. Demographic characteristics of an unhunted Dall sheep (*Ovis dalli dalli*) population in southwest Yukon, Canada. Canadian Journal of Zoology 61:1346-1357.
- HOEFS, M., AND I. M. COWAN. 1979. Ecological investigations of a population of Dall sheep (*Ovis dalli dalli* Nelson). Syesis 12 (supplement 1):1-81.
- HOERLEIN, A. B. 1980. Shipping fever. Pages 99-106 in H. E. Amstutz, editor. Bovine medicine and surgery. American Veterinary Publications, Inc., Santa Barbara, California, USA.
- HONESS, R. F., AND N. M. FROST. 1942. A Wyoming bighorn sheep study. Wyoming Game and Fish Department Bulletin 1:1-127.
- HOWARD-JONES, N. 1984. Robert Koch and the cholera vibrio: a centenary. British Medical Journal 288:379-381.
- HUGENHOLTZ, P., AND N. P PACE. 1996. Identifying microbial diversity in the natural environment: a molecular phylogenetic approach. Trends in Biotechnology 14:190-197.
- JANSEN, B. D., J. R. HEFFELFINGER, T. H. NOON, P. R. KRAUSMAN, AND J. C. DEVOS. 2006. Infectious keratoconjunctivitis in bighorn sheep, Silver Bell Mountains, Arizona, USA. Journal of Wildlife Diseases 42:407-411.
- JAWORSKI, M. D., A. C. S. WARD, D. L. HUNTER, AND I. V. WESLEY. 1993. Use of DNA analysis of *Pasteurella haemolytica* Biotype T isolates to monitor transmission in bighorn sheep (*Ovis canadensis canadensis*). Journal of Clinical Microbiology 31:831-835.

- JENKINS, E. J., A. M. VEITCH, S. L. KUTZ, T. K. BOLLINGER, T. M. CHIRINO-TREJO, B. T. ELKIN, K. H. WEST, E. P. HOBERG, AND L. POLLEY. 2007. Protostrongylid parasites and pneumonia in captive and wild thinhorn sheep (*Ovis dalli*). Journal of Wildlife Diseases 43:189-205.
- JONES, F. L. 1950. A survey of the Sierra Nevada bighorn. Sierra Club Bulletin 35:29-76.
- KELLEY, S. T., E. F. CASSIRER, G. C. WEISER, AND S. SAFAEE. 2006. Phylogenetic diversity of Pasteurellaceae and horizontal gene transfer of leukotoxin in wild and domestic sheep and domestic goats. Infection, Genetics, and Evolution 7:13-23.
- KRAABEL, B. J., M. W. MOLLER, J. A. CONLON, AND H. J. MCNEIL. 1998. Evaluation of a multivalent *Pasteurella haemolytica* vaccine in bighorn sheep: protection from experimental challenge. Journal of Wildlife Diseases 34:325-333.
- MARTIN, K. D., T. SCHOMMER, AND V. L. COGGINS. 1996. Literature review regarding the compatibility between bighorn and domestic sheep. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 10:72-77.
- McQUIVEY, R. P. 1978. The bighorn sheep of Nevada. Nevada Department of Fish and Game Biological Bulletin 6:1-81.
- MILLER, M. W., J. A. CONLON, H. J. MCNEIL, J. M. BULGIN, AND A. C. S. WARD. 1997. Evaluation of multivalent *Pasteurella haemolytica* vaccine in bighorn sheep: safety and serologic responses. Journal of Wildlife Diseases 33:738-748.
- MONELLO, R. J., D. L. MURRAY, AND E. F. CASSIRER. 2001. Ecological correlates of pneumonia epizootics in bighorn sheep herds. Canadian Journal of Zoology 79:1423-1432.
- MURPHY, G. L., L. C. WHITWORTH, K. D. CLINKENBEARD, AND P. A. CLINKENBEARD. 1995. Hemolytic activity of the *Pasteurella haemolytica* leukotoxin. Infection and Immunology 63:3209-3212.
- NOVICK, R. P. 2003. Mobile genetic elements and bacterial toxinoses: the superantigenencoding pathogenicity islands of *Staphylococcus arueus*. Plasmid 49:93-105.
- ONDERKA, D. K., AND W. D. WISHART. 1984. A major bighorn sheep die-off from pneumonia in southern Alberta. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 4:356-363.
- ONDERKA, D. K., AND W. D. WISHART. 1988. Experimental contact transmission of *Pasteurella haemolytica* from clinically normal domestic sheep causing pneumonia in Rocky Mountain bighorn sheep. Journal of Wildlife Diseases 24:663-667.
- ONDERKA, D. K., S. A. RAWLUK, AND W. D. WISHART. 1988. Susceptibility of Rocky Mountain bighorn sheep and domestic sheep to pneumonia induced by bighorn and domestic livestock strains of *Pasteurella haemolytica*. Canadian Journal of Veterinary Research 52:439-444.
- POPPER, K. 1959. The logic of scientific discovery. Harper and Row, New York, USA.
- QUEEN, C., A. C. S. WARD, AND D. L. HUNTER. 1994. Bacteria isolated from nasal and tonsillar samples of clinically healthy Rocky Mountain bighorn and domestic sheep. Journal of Wildlife Diseases 30:1-7.
- RAMEY II, R. R. 1993. Evolutionary genetics and systematics of North American mountain sheep. Ph.D. Thesis, Cornell University, Ithaca, New York, USA.
- RAMEY II, R. R. 2000. New perspectives on the evolutionary origins, historic phylogeography, and population structure of North American mountain sheep. Transactions of the North American Wild Sheep Conference 2:9-16.

- REHMTULLA, A. J., AND R. G. THOMSON. 1981. A review of the lesions in shipping fever of cattle. Canadian Veterinary Journal 22:1-8.
- RUDOLPH, K. M., D. L. HUNTER, W. J. FOREYT, E. F. CASSIRER, R. B. RINDER, AND A. C. S. WARD. 2003. Sharing of *Pasteurella* spp. between free-ranging bighorn sheep and feral goats. Journal of Wildlife Diseases 39:897-903.
- SAFAEE, S., G. C. WEISER, E. F. CASSIRER, R. R. RAMEY, AND S. T. KELLEY. 2006. Molecular survey of host microbial diversity in bighorn sheep. Journal of Wildlife Diseases 42:545-555.
- SCHOMMER, T. J., AND M. M. WOOLEVER. 2001. A process for finding management solutions to the incompatibility between domestic and bighorn sheep. USDA Forest Service Publication. Wallowa-Whitman National Forest. Baker City, Oregon, USA.
- SCHUBERT, S., P. DARLU, O. CLERMONT, A. WEISER, G. MAGISTRI, C. HOFFMANN, K. WEINERT, O. TENAILLON, I. MATIC, AND E. DENAMUR. 2009. Role of intraspecies recombination in the spread of pathogenicity islands within *Escherichia coli* species. PLos Pathogens 5(1): e1000257. doi:10.1371/journal.ppat.1000257.
- SILFLOW, R. M., AND W. J. FOREYT. 1994. Susceptibility of phagocytes from elk, deer, bighorn sheep, and domestic sheep to *Pasteurella haemolytica* cytotoxins. Journal of Wildlife Diseases 30:529-535.
- SILFOW, R. M., W. J. FOREYT, AND J. E. LAGERQUIST. 1994. Evaluation of the cytotoxicity of various isolates of *Pasteurella haemolytica* from bighorn sheep and other ungulate populations. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 9:1-6.
- SILFLOW, R. M., W. J. FOREYT, AND R. W. LEID. 1993. Pasteurella haemolytica cytotoxindependent killing of neutrophils from bighorn and domestic sheep. Journal of Wildlife Diseases 29:30-35.
- SILFLOW, R. M., W. J. FOREYT, S. M. TAYLOR, W. W. LAEGREID, H. D. LIGGIT, AND R. W. LEID. 1989. Comparison of pulmonary defense mechanisms in Rocky Mountain bighorn (*Ovis canadensis canadensis*) and domestic sheep. Journal of Wildlife Diseases 25:514-520.
- SINGER, F. J., L. C. ZEIGENFUSS, AND L. SPICER. 2001. Role of patch size, disease, and movement in rapid extinction of bighorn sheep. Conservation Biology 15:1347-1354.
- SMITH, D. R. 1954. The bighorn sheep in Idaho. Idaho Department of Fish and Game Wildlife Bulletin 1:1-154.
- SNEATH, P. H. A., AND M. STEVENS. 1990. Actinobacillus rossii sp. nov., Actinobacillus seminis sp. nov., nom. rev., Pasteurella bettii sp. nov., Pasteurella lymphangitidis sp. nov., Pasteurella mairi sp. nov., and Pasteurella trehalosi sp. nov. International Journal of Systematic Bacteriology 40:148-153.
- SNIPES, K. P., R. W. KASTEN, M. A. WILD, M. W. MILLER, D. A. JESSUP, R. L. SILFLOW, W. J. FOREYT, AND T. E. MILLER. 1992. Using ribosomal RNA gene restriction patterns in distinguishing isolates of *Pasteurella haemolytica* from bighorn sheep (*Ovis canadensis*). Journal of Wildlife Diseases 28:347-354.
- STELFOX, J. G. 1971. Bighorn sheep in the Canadian Rockies. A history 1800-1970. Canadian Field-Naturalist 85:101-122.
- STORZ, J., X. LIN, C. W. PURDY, V. N. CHOULJENKO, K. G. KOUSOULAS, F. M. ENRIGHT, W. C. GILMORE, R. E. BRIGGS, AND R. W. LOAN. 2000. Coronavirus and *Pasteurella*

infections in bovine shipping fever pneumonia and Evan's criteria for causation. Journal of Clinical Microbiology 38:3291-3298.

- SWEENEY, S. J., R. M. SILFLOW, AND W. J. FOREYT. 1994. Comparative leukotoxicities of *Pasteurella haemolytica* isolates from domestic sheep and free-ranging bighorn sheep (*Ovis canadensis*). Journal of Wildlife Diseases 30:523-528.
- TANNER M. A., D. SHOSKES, A. SHAHED, AND N. R. PACE. 1999. Prevalence of corynebacterial 16S rRNA sequences in patients with bacterial and "nonbacterial" prostatitis. Journal of Clinical Microbiology 37:1863-1870.
- VALDEZ, R., AND P. R. KRAUSMAN. 1999. Description, distribution, and abundance of mountain sheep in North America. Pages 1-22 in R. Valdez and P. R. Krausman, editors. Mountain sheep of North America. University of Arizona Press, Tucson, USA
- WARD, A. C. S., M. R. DUNDAR, D. L. HUNTER, R. H. HILLMAN, M. S. BULGIN, W. J. DELONG, AND E. R. SILVA. 1990. Pateurellaceae from bighorn and domestic sheep. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 7:109-117.
- WARD, A. C. S., D. L. HUNTER, M. D. JAWORSKI, P. J. BENOLKIN, M. P. DOBEL, J. B. JEFFRESS, AND G. A. TANNER. 1997. *Pasteurella* spp. in sympatric bighorn and domestic sheep. Journal of Wildlife Diseases 33:544-557.
- WARD, A. C. S., D. L. HUNTER, K. M. RUDOLPH, W. J. DELONG, J. M. BULGIN, L. M. COWAN, H. J. MCNEIL, AND M. W. MILLER. 1999. Immunologic responses of domestic and bighorn sheep to a multivalent *Pasteurella haemolytica* vaccine. Journal of Wildlife Diseases 35:285-296.
- WARD, A. C. S., G. C. WEISER, W. J. DEJONG, AND G. H. FRANK. 2002. Characterization of *Pasteurella* spp. isolated from healthy domestic pack goats and evaluation of the effects of a commercial *Pasteurella* vaccine. American Journal of Veterinary Research 63:119-123.
- WEHAUSEN, J. D., V. C. BLEICH, AND R. A. WEAVER. 1987. Mountain sheep in California: a historical perspective on 108 years of full protection. Transactions of the Western Section of the Wildlife Society 23:65-74.
- WEISER, G. C., W. J. DELONG, J. L. PAZ, B. SHAFII, W. J. PRICE, AND A. C. S. WARD. 2003. Characterization of *Pasteurella multocida* associated with pneumonia in bighorn sheep. Journal of Wildlife Diseases 39:536-544.
- WILD, M. A., AND M. W. MILLER. 1991. Detecting nonhemolytic Pasteurella haemolytica infections in healthy Rocky Mountain bighorn sheep (Ovis canadensis canadensis): influences of sample site and handling. Journal of Wildlife Diseases 27:53-60.
- WILD, M. A., AND M. W. MILLER. 1994. Effects of modified Cary and Blair medium on recovery of nonhemolytic *Pasteurella haemolytica* from Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*) pharyngeal swabs. Journal of Wildlife Diseases 30:16-19.
- YATES, W. D. G. 1982. A review of infectious bovine rhinotracheitis, shipping fever pneumonia and viral-bacterial synergism in respiratory disease of cattle. Canadian Journal of Comparative Medicine 46:225-263.

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Helen Nunn

From:	Shannon Kendall
Sent:	Monday, February 13, 2017 12:28 PM
То:	Fred Stump; John Peters; Bob Gardner; Larry Johnston
Cc:	Helen Nunn
Subject:	FW: Letter to the Mono County Board of Supervisors
Attachments:	13L-ST-2017-TA-0206 Management of Conway & Mattly Ranches for Grazing-
	Outgoing.pdf; Besser et al_2014.pdf; Cassaigne et al. 2010.pdf; Cassirer et al. 2013.pdf;
	Desert Bighorn Council_1990.pdf; Lawrence et al. 2010.pdf; USFWS_2014.pdf; USFWS_
	2015.pdf; USFWS 2016a.pdf; USFWS 2016b.pdf

Forwarded to you per request by Supervisor Corless. These materials will be included in the board agenda packet for 2/21.

Thanks, Shannon

From: Theisen, Shawna [mailto:shawna_theisen@fws.gov]
Sent: Monday, February 13, 2017 11:18 AM
To: Stacy Corless <<u>scorless@mono.ca.gov</u>>
Cc: Lee Carranza <<u>lee_carranza@fws.gov</u>>; Erin Nordin <<u>erin_nordin@fws.gov</u>>; Marcy Haworth
<<u>marcy_haworth@fws.gov</u>>
Subject: Letter to the Mono County Board of Supervisors

Hello Stacy,

Thank you for facilitating our participation in the Mono County Board of Supervisors' meeting pertaining to the future management of the Conway and Mattley grazing allotments. Please see the attached letter and a file containing scientific literature supporting the U.S. Fish and Wildlife Services' recommendations in the letter. If it would help to supply the board with the references on a CD, please let me know and I will bring several copies for the February 21st meeting.

I look forward to meeting you, Shawna

Shawna Theisen Assistant Field Supervisor Reno Fish and Wildlife Office (775) 861-6378 (desk) (775) 313-1910 (mobile)



United States Department of the Interior

Pacific Southwest Region FISH AND WILDLIFE SERVICE Reno Fish and Wildlife Office 1340 Financial Blvd., Suite 234 Reno, Nevada 89502 Ph: (775) 861-6300 ~ Fax: (775) 861-6301



February 13, 2017 File No. 2017-TA-0206

Board of Supervisors Mono County P.O. Box 715 Bridgeport, California 93517

Subject: Management of Conway and Mattly Ranches for Grazing

Dear Honorable Mono County Board of Supervisors:

Thank you for the opportunity to provide input to the Mono County Board of Supervisors as you consider the future management of Conway and Mattly Ranches. The U.S. Fish and Wildlife Service (Service) appreciates the amount of time and consideration Mono County (County) has devoted to this effort in order to try to accommodate all needs and interests on these County-leased properties. As we have expressed in previous letters, our primary concern is the continuation of domestic sheep grazing on the Conway and Mattly Ranches and the potential for disease transmission between domestic sheep (*Ovis aries*) and the state- and federally-listed endangered Sierra Nevada bighorn sheep (Sierra bighorn; *Ovis canadensis sierrae*) (Service 2014, 2015, 2016a, 2016b).

Bighorn sheep die-offs due to contact with domestic sheep have occurred across the western United States and British Columbia (Desert Bighorn Council 1990; Cassaigne *et al.* 2010). These events can have a long-term negative effect on population recovery due to declines from initial all-age die-offs, which can be followed by years of low recruitment from high lamb mortality (Cassirer *et al.* 2013). In some instances, it can be difficult to determine the exact cause of a dieoff event. However, experiments conducted by Lawrence *et al.* (2010) and Besser *et al.* (2014) demonstrate that bacteria associated with fatal pneumonia in bighorn sheep can be transferred between domestic sheep and bighorn sheep. It is the Service's opinion that research on disease transmission between domestic sheep and bighorn sheep, and bighorn sheep die-offs that have resulted from contact between these two closely-related species, provides strong evidence that disease transmission between domestic sheep and bighorn sheep can and does occur and is often, if not always, fatal.

Board of Supervisors

Sierra Nevada bighorn sheep are protected by the Endangered Species Act (ESA), 16 United States Code, Sections 1531-1544. The Service's responsibilities include administering the ESA. Section 1538, also known as Section 9, prohibits the taking of any endangered species. "Take" is defined in the statute, 16 USC 1532(19), as "to harass, harm, pursue, hunt, shoot, wound, kill, trap, capture, or collect or attempt to engage in any such conduct."

It is our opinion that disease transmission, and a resulting disease outbreak due to contact between Sierra bighorn and domestic sheep would represent take. Disease transmission between domestic sheep and Sierra bighorn could go undetected and ultimately, Sierra bighorn could transmit disease to their respective herd units and potentially throughout the entire population. Unauthorized take of Sierra bighorn as a result of disease transmission from domestic sheep grazed on County authorized lease(s) would be a violation of the ESA, Pursuant to 16 USC 1540, criminal penalties for such violations of the ESA can result in fines up to \$100,000 per individual, \$200,000 per organization and/or up to one year in prison. Civil penalties can result in fines up to \$25,000 for each violation.

We appreciate your staff reaching out to the Service and the California Department of Fish and Wildlife (CDFW) to discuss measures to protect both Sierra bighorn as well as the Bi-State population of Greater sage-grouse (*Centrocercus urophasianus*). However, current management practices combined with protective measures will not eliminate the risk of contact between domestic sheep and Sierra bighorn. Grazing domestic sheep on these properties will inevitably result in the transmission of disease to Sierra bighorn and thus put the lease and leasor at risk of causing take under the ESA. For these reasons, the Service and CDFW are reluctant to augment the Mt. Warren herd unit, one of the herd units located in close proximity to these County-leased properties, due to the potential for contact between Sierra bighorn and domestic sheep. Consequently, the Service believes that domestic sheep grazing on these properties is not compatible with the recovery of Sierra bighorn.

The only way to eliminate the risk of disease transmission is to maintain spatial separation (*i.e.*, distance) between domestic sheep and Sierra bighorn. The Service appreciates the opportunity to work with the County to identify alternative management strategies for Conway and Mattly Ranches that do not perpetuate the risk of disease transmission. One such strategy is to graze livestock other than domestic sheep or goats (which can also transmit disease to bighorn sheep) that will not pose a risk of disease transmission to Sierra bighorn. Another option would be to consider managing the properties as wildlife areas.

Thank you for your commitment to maintaining the conservation values of these properties. We believe that we have a robust and productive partnership with Mono County, as evidenced by of our mutual efforts to protect and conserve the Bi-State population of the Greater sage-grouse. We are hopeful that we can extend our partnership to the recovery of the Sierra bighorn, and believe that eliminating the risk of disease transmission from County-leased properties is central to that objective. We are excited by a future in which Mono County, the Service, CDFW and

Board of Supervisors

File No. 2017-TA-0206

other partners in Sierra Nevada bighorn sheep conservation are actively working together to pursue, and achieve, recovery of this species such that it no longer requires the protections of the ESA. If you have any questions regarding this letter or would like to discuss this matter further, please contact me or Shawna Theisen, Assistant Field Supervisor, at (775) 861-6300.

Sincerely,

ed

Field Supervisor

Attachments: References

Board of Supervisors

References

- Besser, T.E., E. F. Cassirer, K.A. Potter, K. Lahmers, J.L. Oaks, S. Shanthalingam, S. Srikumaran, and W.J. Foreyt. 2014. Epizootic Pneumonia of bighorn sheep following experimental exposure to Mycoplasma ovipneumoniae. PLos ONE 9(10): e110039.
- Cassaigne, I., R. A. Medellín, and J. A. Guasco. 2010. Mortality during epizootics in bighorn sheep: effects of initial population size and cause. Journal of Wildlife Diseases 46(3): 763-771.
- Cassirer, E. F., R. K. Plowright, K. R. Manlove, P. C. Cross, A.P. Dobson, K. A. Potter, and P. J. Hudson. Spatio-temporal dynamics of pneumonia in bighorn sheep. 2013. Journal of Animal Ecology 82: 518-528.
- Desert Bighorn Council. 1990. Guidelines for management of domestic sheep in the vicinity of desert bighorn habitat. Desert Bighorn Council 1990 Transactions. 3 pp.
- Lawrence, P.K., S. Shanthalingam, R.P. Dassanayake, R. Subramaniam, C.N. Herndon, D.P. Knowles, F.R. Rurangirwa, W.J. Foreyt, G. Wayman, A.M. Marciel, S.K. Highlander, and S. Srikumaran. 2010. Transmission of Mannheimia haemolytica from domestic sheep (Ovis aries) to bighorn sheep (Ovis canadensis): unequivocal demonstration with green fluorescent protein-tagged organisms. Journal of Wildlife Disease 46(3): 706-717.
- [Service] U.S. Fish and Wildlife Service. 2014. Letter to Mono County Board of Supervisors about the Mono County draft grant feed of conservation easement Conway and Mattly Ranches. Dated June 17, 2014. 27 pp.
- [Service] U.S. Fish and Wildlife Service. 2015. Letter to Mono County Community Development about Mono County General Plan Update and other associated documents. Dated October 16, 2015. 7 pp.
- [Service] U.S. Fish and Wildlife Service. 2016a. Letter to Mono County Board of Supervisors on the draft Conway Ranch Strategic Facility Plan. Dated June 17, 2016. 7 pp.
- [Service] U.S. Fish and Wildlife Service. 2016b. Letter to Mono County Board of Supervisors about the Conway/Mattly Ranch agenda item for the December 13, 2016 Board of Supervisors meeting. December 12, 2016.

Epizootic Pneumonia of Bighorn Sheep following Experimental Exposure to *Mycoplasma ovipneumoniae*



Thomas E. Besser^{1,2*}, E. Frances Cassirer³, Kathleen A. Potter^{1,2}, Kevin Lahmers¹, J. Lindsay Oaks^{1,2}, Sudarvili Shanthalingam¹, Subramaniam Srikumaran¹, William J. Foreyt¹

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Abstract

Background: Bronchopneumonia is a population limiting disease of bighorn sheep (*Ovis canadensis*). The cause of this disease has been a subject of debate. Leukotoxin expressing *Mannheimia haemolytica* and *Bibersteinia trehalosi* produce acute pneumonia after experimental challenge but are infrequently isolated from animals in natural outbreaks. *Mycoplasma ovipneumoniae*, epidemiologically implicated in naturally occurring outbreaks, has received little experimental evaluation as a primary agent of bighorn sheep pneumonia.

Methodology/Principal Findings: In two experiments, bighorn sheep housed in multiple pens 7.6 to 12 m apart were exposed to *M. ovipneumoniae* by introduction of a single infected or challenged animal to a single pen. Respiratory disease was monitored by observation of clinical signs and confirmed by necropsy. Bacterial involvement in the pneumonic lungs was evaluated by conventional aerobic bacteriology and by culture-independent methods. In both experiments the challenge strain of *M. ovipneumoniae* was transmitted to all animals both within and between pens and all infected bighorn sheep developed bronchopneumonia. In six bighorn sheep in which the disease was allowed to run its course, three died with bronchopneumonia 34, 65, and 109 days after *M. ovipneumoniae* introduction. Diverse bacterial populations, predominantly including multiple obligate anaerobic species, were present in pneumonic lung tissues at necropsy.

Conclusions/Significance: Exposure to a single *M. ovipneumoniae* infected animal resulted in transmission of infection to all bighorn sheep both within the pen and in adjacent pens, and all infected sheep developed bronchopneumonia. The epidemiologic, pathologic and microbiologic findings in these experimental animals resembled those seen in naturally occurring pneumonia outbreaks in free ranging bighorn sheep.

Citation: Besser TE, Cassirer EF, Potter KA, Lahmers K, Oaks JL, et al. (2014) Epizootic Pneumonia of Bighorn Sheep following Experimental Exposure to Mycoplasma ovipneumoniae. PLoS ONE 9(10): e110039. doi:10.1371/journal.pone.0110039

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Data Availability: The authors confirm that all data underlying the findings are fully available without restriction. All relevant data are within the paper.

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Competing Interests: The authors have declared that no competing interests exist.

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Introduction

Bighorn sheep are a North American species that has failed to recover from steep declines at the turn of the 20th century despite strict protections and intensive management, and two populations (Sierra Nevada and Peninsular) are currently classified as endangered [1]. Epizootic pneumonia is limiting bighorn sheep population restoration and as such, the etiology is of considerable interest. The first appearance of the disease in a population is typically in the form of epizootics that affect animals of all ages and is sometimes accompanied by high (>50%) mortality rates. Subsequently, epizootics affecting primarily lambs may occur for decades [2]. Various causes have been proposed for this disease, including lungworms (*Protostrongylus* sp.) [3–6], Pasteurellaceae, especially *Mannheimia (Pasteurella) haemolytica*, [7–12] and more recently, *Mycoplasma ovipneumoniae* [13–16]. In a recent comparative review of the evidence supporting each of these possible etiologies we concluded that M. ovipneumoniae was most strongly supported as the primary epizootic agent of bighorn sheep pneumonia [14]. However, the only two previous experimental challenge studies with M. ovipneumoniae either did not reproduce disease [13] or were confounded by challenges with other agents [16]. The objective of this study was to improve upon previous investigations to better assess the outcome of experimental introduction of M. ovipneumoniae to naïve bighorn sheep.

Methods

Ethics statement

This study was carried out in accordance with the recommendations in the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health and in conformance with United States Department of Agriculture animal research guidelines, under protocols #03854 and #04482 approved by the Washington State University (WSU) Institutional Animal Care and Use Committee. As described in those protocols, euthanasia was performed by intravenous injection of sodium pentobarbital for animals observed to be in severe distress associated with pneumonia during the study and prior to necropsy examination for surviving animals at the end of each experiment.

Experimental aims

Experiment 1 was conducted to investigate the transmission of *M. ovipneumoniae* to bighorn sheep and their subsequent development of disease, using an infected domestic sheep source. Experiment 2 was conducted to investigate experimental direct *M. ovipneumoniae* infection of a single bighorn sheep and the subsequent transmission of this agent to conspecifics. Both experiments were conducted in multiple pens separated by short distances, which allowed investigation of transmission to both commingled and non-commingled animals.

Experimental animals

All experimental animals originated from herds and flocks unexposed to *M. ovipneumoniae* as determined by repeated testing with both serology on blood serum and PCR on enriched nasal swab cultures (using the methods described later in the 'Microbiological testing' section). In Experiment 1, three hand-reared bighorn sheep (yearling rams BHS #82 and #89 and yearling ewe BHS #07) that originated from a captive flock at WSU and three purchased domestic sheep (adult ewes DS #00 and #01 and yearling ewe DS #LA) were co-housed in three 46 m² pens, with one domestic and one bighorn sheep per pen. Pens were separated by 7.6-12 m. Experiment 1 animals had all been commingled in a single pen for 104 days immediately prior to the beginning of this experiment, as previously described [15]. One of the four bighorn sheep used in that prior study had died of M. haemolytica pneumonia, while the other three, which had demonstrated no signs of respiratory disease in that study, were used in experiment 1. In Experiment 2, wild bighorn sheep captured from the Asotin Creek population in Hells Canyon were housed in two 700 m^2 pens, 7.6 m apart, with three animals per pen (Pen #1: adult ewe BHS #40, yearling ewe BHS #38, and yearling ram BHS #39; Pen #2: adult ewes BHS #41 and #42 and adult ram BHS #C). The study pens had either never previously housed domestic or bighorn sheep (pen 1 in experiment 1; both pens in experiment 2) or had been rested for greater than one year since their previous occupancy by any M. ovipneumoniae infected sheep (pens 2 and 3 in experiment 1) prior to these experiments.

Experimental design

Experiment 1. A domestic ewe (DS #00) was placed in isolation and experimentally infected with *M. ovipneumoniae*. The inoculum consisted of ceftiofur-treated (100 ug/ml, 2 hrs, 37° C; Pfizer, Florham Park, NJ) nasal wash fluids from a domestic sheep naturally colonized with *M. ovipneumoniae* [16]. Following ceftiofur treatment, no aerobic bacterial growth was observed from the nasal wash fluids cultured under conditions expected to permit growth of *M. haemolytica*, *B. trehalosi*, or *P. multocida* (Columbia blood agar with 5% sheep blood, 35° C, overnight, 5% CO₂). DS #00 was then challenged with the treated nasal wash fluid by infusion of 15 ml in each nares, 10 ml orally and 5 ml into each conjunctival sac. Subsequent nasal swab samples obtained on days 1, 2, 4 and 7 post-challenge were all PCR positive for *M. ovipneumoniae* using the method described later in the 'Microbiological testing' section confirming that the experimental infection

had been successful. On post challenge day 7, DS #00 was introduced into pen #1 with BHS #82. Following commingling, DS #00 and BHS #82 were restrained for collection of nasal swab samples on days 1, 2, 4, 7, 14, 21, 28, and subsequently at 30 day intervals until the experiment was terminated. Rectal temperatures were recorded from both sheep approximately twice each week. Sheep in pens #2 (BHS #89 and DS #01) and #3 (BHS #07 and DS #LA) were restrained for rectal temperature determination and collection of nasal swabs for microbiology at approximately monthly intervals. All pens were observed daily for clinical signs of respiratory disease. The experiment was conducted October 2009–January 2010.

Experiment 2. BHS #39 was inoculated with *M. ovipneu*moniae just prior to its release into pen #1 with non-inoculated BHS #38 and #40. Non-inoculated BHS #C, #41, and #42 were housed in pen #2 on the same day. The inoculum for BHS #39 was prepared as described for that used in experiment 1 but originated from a different domestic sheep source. In lieu of computation of colony forming units, which is not possible for M. ovipneumoniae due to inconsistent growth on plated media, viable M. ovipneumoniae counts in the inoculum were determined using most probable number (MPN) using a custom 3×4 format: Triplicate enrichment broth tubes were inoculated at each of four decimal dilutions $(10^{-2}-10^{-5})$ of the treated nasal wash fluid [17], incubated (72 hrs, 35C) then PCR was used to detect growth of viable M. ovipneumoniae. The treated fluid was determined to contain 930 MPN/ml (95% confidence interval, 230 to 3800 MPN). Two of the bighorn sheep (BHS #38 and #39) in pen 1 were recaptured by drive net on day 21 of the experiment for nasal swab sampling to detect M. ovipneumoniae infection; otherwise, no live animal sampling was conducted in experiment #2 to reduce the risk of traumatic injury of the wild bighorn sheep involved. The experiment was conducted December 2011-June 2012

Biosecurity. In both experiments, routine biosecurity measures included: 1) the pens containing the single M. ovipneumoniae-challenged animals (exposed pens) were located downwind of the prevailing wind direction from the pens containing no experimentally M. ovipneumoniae exposed animals (clean pens), 2) order of entry rules were established so that on any single day exposed pens were routinely entered by animal care staff for feeding and cleaning only after all work in clean pens had been completed, and 3) personal protective equipment (coveralls and boots) used in exposed pens were either not reused, or were sanitized prior to use in clean pens.

Clinical scores. Clinical score data were determined using the following cumulative point system: observed anorexia (1), nasal discharge (1), cough (2), dyspnea (1), head shaking (1), ear paresis (1) and weakness/incoordination (1).

Microbiological testing. Routine diagnostic testing performed by the Washington Animal Diagnostic Laboratory (fully accredited by the American Association of Veterinary Laboratory Diagnosticians) included detection of *M. ovipneumoniae*-specific and small ruminant lentivirus-specific antibodies in serum samples using competitive enzyme-linked immunosorbent assays (cELISA) [14,18,19], detection of *M. ovipneumoniae* colonization by broth enrichment of nasal swabs followed by *M. ovipneumoniae*-specific PCR testing of the broths [20,21], detection of Pasteurellaceae in pharyngeal swab samples by aerobic bacteriologic cultures, and detection of exposure to parainfluenza-3, border disease, and respiratory syncytial viruses by virus neutralization antibody assays applied to serum samples.

PCR tests specific for detection of *M. haemolytica*, *B. trehalosi*, and *P. multocida*, and *lktA* (the gene encoding the principal
virulence factor of *M. haemolytica* and *B. trehalosi*) were applied to DNA extracted from pneumonic lung tissues using previously described primers (Table 1) and methods with minor modifications. All reactions were conducted individually in 20 µL volumes containing 80–300 ng of template DNA. For M. haemolytica, B. trehalosi, lktA and P. multocida, reactions contained 0.5 units of HotStar Taq DNA polymerase (Qiagen), 2 µL 10x PCR buffer (Qiagen), 4 µL Q-solution (Qiagen), 40 µM of each dNTP (Invitrogen). The M. ovipneumoniae reaction used QIAGEN Multiplex PCR mix. Primers were used at final concentrations of $0.2 \,\mu M$ (M. haemolytica, B. trehalosi, P. multocida, and M. ovipneumoniae) or 0.5 µM (leukotoxin A). Each reaction included an initial activation and denaturation step (95°C, 15 min) and a final 72°C extension step (10 min for Mhgcp-2, lktA, lktA set-1, and LM primers; 9 min for KMT primers; 5 min for Btsod and Mhgcp primers). Cycling conditions were as follows: M. ovipneumoniae, 30 cycles of 95°C for 30 s, 58°C for 30 s, 72°C for 30 s; B. trehalosi and M. haemolytica (Mhgcp and Btsod primers), 35 cycles of 95°C for 30 s, 55°C for 30 s, 72°C for 40 s; P. multocida and lktA (lktA primers), 30 cycles of 95°C for 60 s, 55°C for 60 s, 72°C for 60 s; M. haemolytica (Mhgcp-2 primers), 40 cycles of 95°C for 30 s, 54°C for 30 s, 72°C for 30 s; lktA (lktA set-1 primers), 40 cycles of 95°C for 30 s, 52°C for 30 s, 72°C for 40 s. Leukotoxin expression was detected in Pasteurellaceae isolates by MTT dye reduction cytotoxicity assay as described previously [22].

The 16S–23S ribosomal operon intergenic spacer (IGS) regions of *M. ovipneumoniae* recovered from animals in these studies were PCR amplified (Table 1) and sequenced as previously described [23].

16S rDNA analyses to identify the predominant bacterial flora in pneumonic lung tissues. In previous studies, culture-independent evaluation of the microbial flora of lung tissues in naturally occurring bighorn sheep pneumonia revealed a polymicrobial flora late in the disease course [13,23]. For comparison, we applied the same methods to lung tissues of the experimentally challenged animals in this study. Note that more sensitive

detection of specific respiratory pathogens was provided by the PCR assays described earlier, whereas these 16S studies were designed instead to identify the numerically predominant bacteria in affected lungs. The library size used was based on the binary distribution to provide a 95% chance of detection of each taxon comprising 10% or more of the ribosomal operon frequency in the source tissue. Two 1 g samples of pneumonic lung tissues were aseptically collected from sites at least 10 cm apart, homogenized by stomaching, and DNA was extracted (DNeasy tissue kit; Qiagen, Valencia, CA) from 100 uL aliquots of each homogenate. 16S rDNA segments were PCR amplified and cloned as described [13]. Insert DNA was sequenced from 16 clones derived from each of the two homogenates from each animal, and each sequence was attributed to species (\geq 99% identity) or genus (\geq 97% identity) based on BLAST GenBank similarity [24].

Results

Experiment 1

M. ovipneumoniae infection of DS #00, introduced into pen 1 to start the experiment, was confirmed by positive nasal swab samples obtained on days 1, 4, and 7 after inoculation prior to its introduction into pen #1, and on days 1, 2, 4, 7, 14, 21, 28, 60 and 90 after its introduction into pen #1, confirming that the experimental colonization had been successful and maintained throughout experiment 1. M. ovipneumoniae was first detected in the bighorn sheep (BHS #82) commingled with DS #00 in pen #1 on day 28, and subsequent tests on days 60 and 90 were also positive. BHS #82 developed signs of respiratory disease including nasal discharge (onset day 37); coughing and fever (onset day 42); and lethargy and ear paresis (onset day 61) (Figure 1a). Signs of respiratory disease were observed in the bighorn sheep in pens #2(BHS #89) and #3 (BHS #07) beginning on days 62 and 67, respectively; these signs also included fever, lethargy, paroxysmal coughing, nasal discharge, head shaking, and drooping ears. No signs of respiratory disease were observed in the commingled domestic sheep at any time during the experiment. M.

Table 1. Primers and PCR reaction targets used in these experiments.

Pathogen/Virulence	Taunat	Duine en Norre		Cine (ha)	Deferreres
gene	Target	Primer Name	Sequence $(5^\circ \rightarrow 3^\circ)$	Size (bp)	Reference
M. haemolytica	gcp	MhgcpF	AGA GGC CAA TCT GCA AAC CTC G	267	[33]
		MhgcpR	GTT CGT ATT GCC CAA CGC CG		
M. haemolytica	gcp	MhgcpF2	TGG GCA ATA CGA ACT ACT CGG G	227	[34]
		MhgcpR2	CTT TAA TCG TAT TCG CAG		
B. trehalosi	sodA	BtsodAF	GCC TGC GGA CAA ACG TGT TG	144	[33]
		BtsodAR	TTT CAA CAG AAC CAA AAT CAC GAA TG		
P. multocida	kmt1	KMT1T7	ATC CGC TAT TTA CCC AGT GG	460	[35]
		KMT1SP6	GCT GTA AAC GAA CTC GCC AC		
Pasteurellaceae leukotoxin	lktA	lktAF	TGT GGA TGC GTT TGA AGA AGG	1,145	[36]
		lktAR	ACT TGC TTT GAG GTG ATC CG		
<i>M. haemolytica</i> leukotoxin	lktA	IktAF set-1	CTT ACA TTT TAG CCC AAC GTG	497	[34]
		IktAR set-1	TAA ATT CGC AAG ATA ACG GG		
Mycoplasma ovipneumoniae	16s rDNA	LMF	TGA ACG GAA TAT GTT AGC TT	361	[20,21]
		LMR	GAC TTC ATC CTG CAC TCT GT		
Mycoplasma ovipneumoniae	165–235 IGS	MolGSF	GGA ACA CCT CCT TTC TAC GG	Variable~490	[23]
		MolGSR	CCA AGG CAT CCA CCA AAT AC		

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ovipneumoniae was detected in nasal swab samples from all bighorn and domestic sheep in pens #2 and #3 when sampled on day 70. The bighorn sheep were euthanized for necropsy on days 93 (BHS #89) and 99 (BHS #82 and #07). At necropsy, significant abnormal findings were limited to the respiratory tract. Bronchopneumonia affecting 25–50% of the lung volume was observed in all three bighorn sheep (Figure 2). Histopathological examination revealed peribronchiolitis with large lymphoid cuffs, bronchiectasis with purulent exudates, pulmonary atelectasis, and hyperplastic bronchial epithelia lacking visible cilia (Figure 2).

Experiment 2

On day 21 following release of the inoculated bighorn into pen #1, *M. ovipneumoniae* was detected in the inoculated animal and one pen mate (BHS #38 and #39); the third animal (BHS #40) evaded capture and sampling on that day. The first signs of respiratory disease were observed in pen #1 animals on day 21 during drive net capture for sampling, apparently triggered by exertion (Figure 2a). On day 34, inoculated BHS #39 died in pen

#1. On day 49, signs of respiratory disease were first observed in the bighorn sheep in pen #2 (Figure 2b). On days 65 and 109, #41, and #42 in pen #2 died or were euthanized in extremis. The surviving three bighorn sheep exhibited varying degrees of respiratory disease: BHS #38 showed persistent respiratory disease, while BHS #40 and #C showed decreasing respiratory disease over time, which became minimal after days 161 and 154, respectively. On day 204, the three surviving bighorn sheep were euthanized for necropsy. At necropsy, significant abnormal findings were limited to the respiratory tract. All six bighorn sheep had bronchopneumonia, with consolidation of lung tissue volumes ranging from an estimated 5% (BHS #40) to 80–100% (BHS #41) (Figure 2). Histopathological examination revealed severe peribronchiolitis with large lymphoid cuffs as seen in experiment 1. Animals that died or were euthanized in extremis had an overlying necrotizing bronchiolitis (#39) or abscessing bronchiolitis with bronchiectasis (BHS #41, #42) (Figure 2).



Figure 1. Clinical signs exhibited by *M. ovipneumoniae* **infected bighorn sheep.** Clinical scores (3-day moving averages) of bighorn sheep following introduction of *M. ovipneumoniae*: A) Experiment 1, 3 separate pens; solid line, Pen 1, BHS #82; dashed line, Pen 2, BHS #89; dotted line, Pen 3, BHS #07; B) Experiment 2, Pen 1: solid line, BHS #39 (died day 34); dashed line, BHS #40; dotted line; BHS #38.; C) Experiment 2, Pen 2: solid line, BHS #41 (died day 65); dashed line, BHS #C. doi:10.1371/journal.pone.0110039.q001



Figure 2. Gross and histologic lesions in lungs of bighorn sheep experimentally infected with *M. ovipneumoniae.* Images of BHS #82 (A, B), BHS #39 (C, D), BHS #C (E, F) and BHS #42 (G, H). Original magnification of histologic images was 200X (B, D, H) or 100X (F). doi:10.1371/journal.pone.0110039.g002

Microbiology

All bighorn sheep in both experiments seroconverted to *M. ovipneumoniae* (Table 2). Most experimental animals had neutralizing antibody to parainfluenza-3 virus, but no significant changes in antibody titers were observed during the experimental period. Detectable antibody to other ovine respiratory viruses, including border disease virus, ovine progressive pneumonia virus, and respiratory syncytial virus was occasionally observed in single samples.

M. ovipneumoniae was detected at necropsy in both upper and lower respiratory tracts of all bighorn sheep except BHS #40 whose lung tissues were PCR negative and whose upper

respiratory samples were PCR indeterminate (Table 3). Aerobic cultures and/or PCR tests identified *B. trehalosi* from pneumonic lung tissues from all bighorn sheep in both experiments (Table 3). *B. trehalosi* isolates from BHS #82 and #07 carried *lktA* and expressed leukotoxin activity (Table 3). *P. multocida* and *M. haemolytica* were not detected in these animals by either aerobic culture or PCR.

Culture independent survey of bacteria in pneumonic bighorn sheep lung tissues

DNA sequences of cloned 16S rDNA revealed that the predominant bacterial species in pneumonic sections of lung were

Table 2. Antibody responses to M. ovipneumoniae and parainfluenza-3 (PI-3) virus.

			M. ovipneum	oniae ¹	PI-3 virus ²	
Experiment	ID	Pen	Pre ³	Post ³	Pre ³	Post ³
1	82	1	-8%	93%	512	512
1	89	2	-7%	88%	128	128
1	07	3	-1%	92%	256	512
2	38	1	-6%	74%	Neg	64
2	39	1	-13%	67%	Neg	<32
2	40	1	-23%	75%	64	512
2	41	2	-19%	82%	512	NT
2	42	2	-11%	82%	256	NT
2	С	2	-4%	66%	256	512

¹*M. ovipneumoniae* antibody detected by cELISA, expressed as percentage inhibition of the binding of an agent-specific monoclonal antibody [14,18]. ²PI-3 virus neutralizing antibody detected by virus neutralization [37].

³Pre samples in experiment 1 were obtained on the day that the *M. ovipneumoniae* colonized domestic sheep was introduced to pen 1 and in experiment 2 were obtained on the day that BHS #39 was inoculated with *M. ovipneumoniae*. 'Post' samples in both experiments were obtained at necropsy. Neg = No titer detected. NT = Not tested, due to inadequate specimen volume.

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diverse (Table 4). In experiment 1, *M. ovipneumoniae* was detected in the lung tissues of all animals. *B. trehalosi* also comprised substantial proportions of the pneumonic lung flora in two animals (BHS #82 and #07), while obligate anaerobic species, primarily Fusobacterium spp., predominated in the third animal (BHS #89). The flora identified in the pneumonic lungs of the animals in experiment 2 was also substantially comprised of mixed obligate anaerobes especially *Fusobacterium* spp. (Table 4).

Molecular epidemiology of respiratory pathogens. Consistent with epidemic transmission, *M. ovipneumoniae* strains recovered from all experimental sheep within each experiment shared identical IGS DNA sequences with the respective challenge inoculum (GenBank HQ615162 in experiment 1; KJ551511 in experiment 2).

Discussion

The most striking finding of these experiments was the high transmissibility of *M. ovipneumoniae* and the consistent development of pneumonia that followed infection of bighorn sheep. The bacterium was naturally transmitted from single experimentally inoculated animals (a domestic sheep in experiment 1 and a bighorn sheep in experiment 2) to all animals within and between pens up to 12 m distant. Eight of nine bighorn sheep exposed to *M. ovipneumoniae* developed severe bronchopneumonia and three died, while all the domestic sheep remained healthy.

Previous experimental challenge studies conducted with M. haemolytica or B. trehalosi in the absence of M. ovipneumoniae have not documented transmission. For example, Foreyt et al. [8]

Table 3. Microbiologic findings from pneumonic lung tissues, based on aerobic culture and species specific PCR.

Expt.	ID	Bacterial patho	gens identified in pneu			
		B. trehalosi	M. haemolytica	lktA	M. ovipneumoniae	Other ⁵
1	82	Cult, sodA ¹	Neg ²	Pos ³	165 ⁴	None
1	89	Cult, sodA	Neg	Neg ³	16S	Pasteurella sp. ⁵
1	07	Cult, sodA	Neg	Pos	16S	Pasteurella sp.
2	38	Cult, sodA	Neg	Neg	16S	Pasteurella sp.
2	39	NT, sodA	NT, Neg ²	Neg	16S	NT ⁵
2	40	Cult	Neg	Neg	Neg ⁴	Trueperella pyogenes ⁵
2	41	Cult, sodA	Neg	Neg	16S	None
2	42	Cult	Neg	Neg	16S	None
2	С	Cult	Neg	Neg	16S	Pasteurella sp.

¹Cult = *B. trehalosi* detected by bacterial culture; *sodA* = *B. trehalosi* detected by *sodA* species-specific PCR (Table 1); NT = Unable to test by bacterial culture (overgrowth by *Proteus* sp.).

²Neg = *M. haemolytica* not detected by either bacterial culture or by PCR with either *gcp* primer set (Table 1); NT = Unable to test by bacterial culture (overgrowth by *Proteus* sp.).

³Neg = Pasteurellaceae *lktA* not detected in DNA extracts from pneumonic lung tissues by two different *lktA* PCRs (Table 1) [34,36]. Pos = *lktA* detected in *B. trehalosi* isolates obtained from BHS #82 and #07 [36].

 $^{4}16S = M$. ovipneumoniae detected by PCR (Table 1) [20]; Neg = M. ovipneumoniae not detected by PCR.

⁵*Pasteurella* sp., *Trueperella pyogenes* = Bacteria isolated and identified by aerobic culture; *Pasteurella* sp. were determined not to be *B. trehalosi, M. haemolytica,* or *P. multocida*; NT = Unable to test by bacterial culture due to overgrowth by *Proteus* sp.

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xpt.	٩	Bacterial species identi	fied in pneumonic lung tis	sues			
		Btre ¹	Movi ¹	Fuso ¹	Prev ¹	Porphyro ¹	Other ¹
	82	20 (62.5) ²	8 (25)	0	3 (9.4)	0	1 (3.1)
	89	1 (3.1)	7 (21.9)	21 (65.6)	1 (3.1)	0	2 (6.3)
	07	16 (50.0)	12 (37.5)	0	0	0	4 (12.5)
	38	4 (7.1)	2 (3.6)	8 (14.3)	20 (35.7)	9 (16.1)	13 (23.2)
	U	0	0	17 (30.4)	5 (8.9)	19 (33.9)	15 (26.8)
	39	2 (6.3)	0	24 (75.0)	0	0	6 (18.8)
	40	0	0	0	0	0	56 (100.0)
	41	1 (3.1)	0	21 (65.6)	5 (15.6)	0	5 (15.6)
	42	0	0	31 (96.9)	0	0	1 (3.1)

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M. ovipneumoniae-Induced Bighorn Sheep Pneumonia

reported a series of three experiments in which commingled bighorn sheep were either challenged with intra-tracheal *M. haemolytica* or given sterile BHI as controls. Four of the five control bighorn sheep survived without evidence of disease while commingled with eight *M. haemolytica*-challenged bighorn sheep, of which seven died of pneumonia [8]. Commingled bighorn sheep also remained healthy in several other studies where individual bighorn sheep died with apparent *M. haemolytica* bronchopneumonia (confirmed by isolation of this bacterium from lung tissues) [15,25,26].

In addition to high transmissibility, the time course of disease development and the predominant microbiology of the pneumonic lung tissues following experimental introduction of M. ovipneumoniae differed from that seen in previous bighorn sheep challenge experiments with other respiratory pathogens. Bighorn sheep directly challenged with leukotoxin positive M. haemolytica or B. trehalosi develop peracute bronchopneumonia and >90% die within a week of challenges with 10^5 cfu or more [16,27–30]. In contrast, disease following experimental M. ovipneumoniae exposures was considerably slower in onset (14-21 days post infection) and development (deaths occurring 34 to 109 days post infection; respiratory disease persisted up to 6 months postinfection); this slow time course closely resembles that documented previously in bighorn lamb pneumonia outbreaks [13]. After lethal M. haemolytica challenge, the agent is typically isolated from lung tissues in high numbers and pure cultures [15,25]; in contrast in naturally occurring pneumonia outbreaks M. ovipneumoniae may be predominant early in the disease course but 16S library analyses have been used to document its overgrowth by diverse other bacteria later in the disease course [14,23]. Although the numbers of animals in the experimental M. ovipneumoniae infection studies reported here are small, the results are consistent with the trend for early predominance of M. ovipneumoniae followed by overgrowth by diverse other bacterial later in the disease course (Tables 3 and 4) [13,14,23].

Our results also differ from our previous attempt to experimentally reproduce respiratory disease by challenge inoculation of 1-week-old bighorn lambs with M. ovipneumoniae, which produced minor lesions and seroconversion but no clinically significant respiratory disease [13]. However, laboratory passage of *M. ovipneumoniae* (as was performed in that experiment) has been reported to attenuate virulence in M. ovipneumoniae [31]. Challenge of bighorn sheep with un-passaged M. ovipneumoniae produced different results, as observed here in experiment #2. In another study [16], nasal washings from domestic sheep naturally colonized with M. ovipneumoniae or lung homogenates from a M. ovipneumoniae-infected bighorn sheep were used for challenge of bighorn sheep after ceftiofur treatment to eliminate detectable Pasteurellaceae. Consistent with increased virulence of unpassaged M. ovipneumoniae, infection and respiratory disease signs were observed in all four bighorn sheep, one of which died 19 days following challenge. The three surviving animals continued to exhibit respiratory disease signs for 42 days, at which time the experiment was terminated by challenge with M. haemolytica (using a dose documented to be rapidly fatal to bighorn sheep even in the absence of M. ovipneumoniae) [16]. As a result, the longer term effects of the mycoplasma infection were not determined in that study. Therefore, the experiments reported here are the first in which naïve bighorn sheep were exposed to un-passaged M. ovipneumoniae and then followed over a time period comparable with the naturally occurring disease course.

The possibility of viral agents contributing to the disease observed in this study cannot be completely ruled out, since the inoculum was derived from nasal washings from domestic sheep and no virucidal treatments were applied. However, a previous study using ultrafiltrates of bighorn sheep pneumonic lung tissues or nasal washings from domestic sheep failed to reproduce any respiratory disease in inoculated susceptible bighorn sheep [16]. In addition, serologic monitoring for the predominant domestic sheep respiratory viruses did not demonstrate seroconversion of the experimental animals in this study, as described in the Results and in Table 2. Therefore, the most parsimonious interpretation of the data presented here is that the disease observed resulted from *M. ovipneumoniae* infection and the sequelae of that infection.

The transmission of *M. ovipneumoniae* from pen-to-pen in these experiments strongly suggests that direct contact is not necessary for epizootic spread of pneumonia in bighorn sheep. Feeding, watering and other procedures involving animal care or research staff were designed to minimize the risk of human or fomitemediated transmission of the pathogen from pen to pen, although we recognize it is impossible to completely rule out this possibility. On the other hand, since aerosolized droplet transmission is recognized as a transmission route for the closely related bacterium, Mycoplasma hyopneumoniae (the cause of atypical pneumonia of swine) [32], it is plausible that a similar transmission mode occurs with M. ovipneumoniae. Infectious aerosols generated by coughing animals would likely contribute to the explosive nature of the pneumonia outbreaks observed following initial introduction of M. ovipneumoniae into naïve bighorn sheep populations.

In conclusion, we demonstrated that experimental *M. ovipneumoniae* infection of naïve bighorn sheep induces chronic, severe bronchopneumonia associated with multiple secondary bacterial infections and that this infection spread rapidly to animals both within the same pen and to animals in nearby pens. The significance of these findings would be clarified by parallel experiments specifically designed to determine transmissibility and associated disease outcomes in other agents associated with bighorn sheep pneumonia, particularly *M. haemolytica*, in the absence of *M. ovipneumoniae*. Furthermore, the case-fatality rates of *M. ovipneumoniae* infected animals described here contrasts

References

- Festa-Bianchet M (2008) Ovis canadensis. The IUCN Red List of Threatened Species. Available: http://www.iucnredlist.org/details/summary/15735/0. Accessed 2014. Jul 24.
- Cassirer EF, Plowright RK, Manlove KR, Cross PC, Dobson AP, et al. (2012) Spatio-temporal dynamics of pneumonia in bighorn sheep (*Ovis canadensis*). J Animal Ecol 82: 518–528.
- Marsh H (1938) Pneumonia in Rocky Mountain bighorn sheep. J Mammal 19: 214–219.
- Buechner HK (1960) The bighorn sheep in the United States, its past, present, and future. Wildl Monog 4: 3–174.
- Forrester DJ (1971) Bighorn sheep lungworm-pneumonia complex. In: Davis JW, Anderson RC, editors. Parasitic Diseases of Wild Mammals. Ames, IA: Iowa State University Press. 158–173.
- Demartini JC, Davies RB (1977) An epizootic of pneumonia in captive bighorn sheep infected with *Muellerius* sp. J Wildl Dis 13: 117–124.
- Miller MW (2001) Pasteurellosis. In: Williams ES, Barker, I K., editor. Infectious diseases of wild mammals. Ames IA USA: Iowa State University Press. 558.
- Foreyt WJ, Snipes KP, Kasten RW (1994) Fatal pneumonia following inoculation of healthy bighorn sheep with *Pasteurella haemolytica* from healthy domestic sheep. J Wildl Dis 30: 137–145.
- Kraabel BJ, Miller MW, Conlon JA, McNeil HJ (1998) Evaluation of a multivalent *Pasteurella haemolytica* vaccine in bighorn sheep: Protection from experimental challenge. J Wildl Dis 34: 325–333.
- Rudolph KM, Hunter DL, Foreyt WJ, Cassirer EF, Rimler RB, et al. (2003) Sharing of *Pasteurella* spp. between free-ranging bighorn sheep and feral goats. J Wildl Dis 39: 897–903.
- Rudolph KM, Hunter DL, Rimler RB, Cassirer EF, Foreyt WJ, et al. (2007) Microorganisms associated with a pneumonic epizootic in Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*). J Zoo Wildl Med 38: 548–558.
- bignorn sneep (*Urus canadensus canadensus*). J Zoo Wuld Med 36: 546–556.
 Weiser GC, DeLong WJ, Paz JL, Shafii B, Price WJ, et al. (2003) Characterization of *Pasteurella anullocida* associated with pneumonia in bighorn

with the nearly 100% mortality that follows experimental commingling of bighorn sheep with presumptively or documented M. ovipneumoniae-positive domestic sheep and suggests an important role for polymicrobial secondary infections in determining mortality rates, which could be investigated in future studies. Finally, M. ovipneumoniae was still detected in nasal swab samples of several surviving bighorn sheep that were euthanized at the completion of these studies, suggesting that survivors of naturally occurring pneumonia outbreaks may continue to carry and shed this agent in nasal secretions. Such carriage may provide a mechanism for the post-invasion disease epizootics in lambs described in free-ranging populations. If so, this presumptive carrier state requires further study to characterize the factors that determine its occurrence and persistence, as these may be critical for the development of effective management control measures for this devastating disease.

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Author Contributions

Conceived and designed the experiments: TEB EFC JLO S. Srikumaran WJF. Performed the experiments: TEB EFC JLO KAP KL S. Shanthalingam. Analyzed the data: TEB EFC KAP KL. Contributed reagents/materials/analysis tools: TEB EFC KAP KL S. Shanthalingam S. Srikumaran. Contributed to the writing of the manuscript: TEB EFC KAP S. Shanthalingam S. Srikumaran WJF.

- Besser TE, Cassirer EF, Potter KA, VanderSchalie J, Fischer A, et al. (2008) Association of Mycoplasma ovipneumoniae infection with population-limiting respiratory disease in free-ranging rocky mountain bighorn sheep (Ovis canadensis canadensis). J Clin Microbiol 46: 423–430.
- Besser TE, EF Cassirer, MA Highland, P Wolff, A Justice-Allen, et.al. (2012) Bighorn sheep pneumonia: Sorting out the cause of a polymicrobial disease. Prev Vet Med 108: 85–93.
- Besser TE, Cassirer EF, Yamada C, Potter KA, Herndon C, et al. (2012) Survival of Bighorn Sheep (*Ovis canadensis*) Commingled with Domestic Sheep (*Ovis aries*) in the Absence of *Mycoplasma ovipneumoniae*. J Wildl Dis 48: 168– 172.
- Dassanayake RP, Shanthalingam S, Herndon CN, Subramaniam R, Lawrence PK, et al. (2010) *Mycoplasma ovipneumoniae* can predispose bighorn sheep to fatal *Mannheimia haemolytica* pneumonia. Vet Microbiol 145: 354–359.
- Blodgett R (2010) Bacteriologic Analytical Manual Appendix 2: Most Probable Number from Serial Dilutions. Washington DC. Available: http://www.fda. gov/Food/FoodScience Research/LaboratoryMethods/ucm109656.htm. Accessed 2014 Jul 24.
- Ziegler JC, Lahmers KK, Barrington GM, Parish SM, Kilzer K, et al. (2014) Safety and Immunogenicity of a Mycoplasma ovipneumoniae Bacterin for Domestic Sheep (*Ovis aries*). PLoS One 9(4): e95698.
- Herrmann LM, Cheevers WP, Marshall KL, McGuire TC, Hutton MM, et al. (2003) Detection of serum antibodies to ovine progressive pneumonia virus in sheep by using a caprine arthritis-encephalitis virus competitive-inhibition enzyme-linked immunosorbent assay. Clin Diag Lab Immunol 10: 862–865.
- Lawrence PK, Shanthalingam S, Dassanayake RP, Subramaniam R, Herndon CN, et al. (2010) Transmission of *Mannheimia haemolytica* from domestic sheep (*Ovis aries*) to bighorn sheep (*Ovis canadensis*): unequivocal demonstration with green fluorescent protein-tagged organisms. J Wildl Dis 46: 706–717; erratum, J Wildl Dis 46: 1346.

sheep. J Wildl Dis 39: 536-544.

- McAuliffe L, Hatchell FM, Ayling RD, King AI, Nicholas RA (2003) Detection of *Mycoplasma ovipneumoniae* in *Pasteurella*-vaccinated sheep flocks with respiratory disease in England. Vet Rec 153: 687–688.
- Gentry MJ, Srikumaran S. (1991) Neutralizing monoclonal antibodies to *Pasteurella haemolytica leukotoxin affinity purify the toxin from crude culture* supernatants. Microb Pathog 10: 411–417.
- Besser TE, Highland M, Baker K, Anderson NJ, Ramsey JM (2012) Causes of pneumonia epizootics among bighorn sheep, western United States, 2008–2010. Emerg Infect Dis 18: 406–414.
- Petti CA (2007) Detection and identification of microorganisms by gene amplification and sequencing. Clin Infect Dis 44: 1108–1114.
- Foreyt WJ, Jenkins EJ, Appleyard GD (2009) Transmission of lungworms (*Muellerius capillaris*) from domestic goats to bighorn sheep on common pasture. J Wildl Dis 45: 272–278.
- Foreyt WJ, Lagerquist JE (1996) Experimental contact of bighorn sheep (Ovis canadensis) with horses and cattle, and comparison of neutrophil sensitivity to Pasteurella haemolytica cytotoxins. J Wildl Dis 32: 594–602.
- Onderka DK, Rawluk SA, Wishart WD (1988) Susceptibility of Rocky Mountain bighorn sheep and domestic sheep to pneumonia induced by bighorn and domestic livestock strains of *Pasteurella haemolytica*. Can J Vet Res 52: 439–444.
- Dassanayake RP, Shanthalingam S, Herndon CN, Lawrence PK, Cassirer EF, et al. (2009) Mannheimia haemolytica serotype A1 exhibits differential pathogenicity in two related species, Ovis canadensis and Ovis aries. Vet Microbiol 133: 366–371.
- Subramaniam R, Shanthalingam S, Bavananthasivam J, Kugadas A, Potter KA, et al. (2011) A multivalent *Mannheimia-Bibersteinia* vaccine protects bighorn

sheep against Mannheimia haemolytica challenge. Clin Vaccine Immunol 18: 1689–1694.

- Foreyt WJ, Snipes KP, Kasten RW (1994) Fatal pneumonia following inoculation of healthy bighorn sheep with *Pasteurella haemolytica* from healthy domestic sheep. J Wildl Dis 30: 137–145.
- Alley MR, Ionas G, Clarke JK (1999) Chronic non-progressive pneumonia of sheep in New Zealand - a review of the role of *Mycoplasma ovipneumoniae*. N Z Vet J 47: 155–160.
- 32. Desrosiers R (2011) Transmission of swine pathogens: different means, different needs. Anim Health Res Rev 12: 1–13.
- Dassanayake RP, Call DR, Sawant AA, Casavant NC, Weiser GC, et al. (2010) Bibersteinia trehalosi inhibits the growth of Mannheimia haemolytica by a proximity-dependent mechanism. Appl Environ Microbiol 76: 1008–1013.
- 34. Shanthalingam S, Goldy A, Bavananthasivam J, Subramaniam R, Batra SA, et al. (2014) PCR assay detects *Mannheimia haemolytica in culture-negative pneumonic lung tissues of bighorn sheep (Ovis canadensis)* from outbreaks in the Western USA, 2009–2010. J Wildl Dis 50: 1–10.
- Townsend KM, Frost AJ, Lee CW, Papadimitriou JM, Dawkins HJ (1998) Development of PCR assays for species- and type-specific identification of *Pasteurella multocida* isolates. J Clin Microbiol 36: 1096–1100.
- 36. Fisher MA, Weiser GC, Hunter DL, Ward ACS (1999) Use of a polymerase chain reaction method to detect the leukotoxin gene IktA in biogroup and biovariant isolates of *Pasteurella haemolytica* and *P. trehalosi*. Am J Vet Res 60: 1402–1406.
- Rossi CR, Kiesel GK (1971) Microtiter tests for detecting antibody in bovine serum to parainfluenza 3 virus, infectious bovine rhinotracheitis virus, and bovine virus diarrhea virus. Appl Microbiol 22: 32–36.



MORTALITY DURING EPIZOOTICS IN BIGHORN SHEEP: EFFECTS OF INITIAL POPULATION SIZE AND CAUSE

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MORTALITY DURING EPIZOOTICS IN BIGHORN SHEEP: EFFECTS OF INITIAL POPULATION SIZE AND CAUSE

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ABSTRACT: One of the most severe threats to bighorn sheep (*Ovis canadensis*) populations is disease. With the objective of projecting possible epizootic consequences to bighorn sheep population dynamics, we examined 23 epizootic mortality episodes from presumably known causes that occurred in the United States and Canada from 1942 to 2005. These outbreaks were correlated with population size using regression models. Epizootic origins were documented by considering contact with a "new" pathogen for the bighorn sheep population or pneumonic processes, presumably triggered by stress. We suggest mortality rates are negatively related to population size in a logarithmic function, and offer a model to estimate the percentage of disease-related mortalities for a given population size of bighorn sheep. From a disease dynamics perspective, we suggest a minimum population of 188 bighorn sheep would be required to insure long-term persistence in the presence of epizootic disease.

Key words: Diseases, epizootics, Ovis canadensis, population size.

INTRODUCTION

Bighorn sheep populations have decreased significantly in recent decades due mainly to habitat fragmentation and degradation, poaching, disease, urban development, and human recreational activities (Valdez and Krausman, 1999). The total population of bighorn sheep in Mexico (Ovis canadnesis mexicana, Ovis canadnesis cremnobates, and Ovis canadnesis weemsi) is estimated between 5,500 and 8,800 animals (Medellin et al., 2005) distributed in Sonora, Baja California, and Tiburon Island. Bighorn sheep were extirpated from Nuevo León in the 1930s and from Chihuahua (Heffelfinger and Marquez-Muñoz, 2005) and Coahuila around the 1970s (Espinosa et al. 2006). Although conservation efforts, including reintroduction programs, are occurring in Chihuahua (Cassaigne, pers. obs.) and Coahuila (McKinney and Delgadillo-Villalobos, 2005; Sandoval and Espinosa-Treviño, 2001), the vast majority of the Mexican populations comprise only a few dozen individuals (Dirección General de Vida Silvestre, unpubl. data).

Although there is a general understand-

ing of the role diseases play in the survival of populations, in recent years this aspect has gained importance in the study, management, and conservation of wildlife. Disease has been considered the primary cause of many bighorn sheep population extinctions (Gross et al., 2000). Bighorn sheep are more susceptible than other sheep to a variety of pathogens that have been related to pneumonic epizootics with mortality rates of 25% to 100% (Onderka and Wishart, 1982; Jessup, 1985; Festa-Bianchet, 1988; Sandoval, 1988; Miller et al., 1991). The presence of domestic animals, especially domestic sheep (Ovis *aries*), adjacent to or in the same habitat as bighorn sheep increases the risk of transmission of pathogens that can be fatal for bighorn sheep (Ough and De Vos, 1986; Ramey et al., 2000). Additionally, animals that are restricted to small habitats or habitat fragments increase the possibility of retransmission of some diseases (Risenhoover et al., 1988) by remaining in contact with the sources of infection.

In addition to this increased susceptibility, several factors, including human

México D.F. C.P. 0138

activities, the presence of domestic and feral livestock, climate change, and population isolation that can lead to local overgrazing, can provoke chronic stress, which decreases immune response (Pruett, 2003; Kemenya and Schedlowskib, 2007). Stress is a key factor that can increase the risk of an epizootic outbreak. Finally, the forced isolation in which many bighorn sheep populations exist (Allen, 1980) promotes inbreeding depression. Many researchers have suggested that detrimental characteristics associated with this process, such as the loss of evolutionary adaptability and the increase of disease can substantially increase risk of local extinction (O'Brien and Everman, 1988; Mills and Smouse, 1994; Saccheri et al., 1998).

Considering these factors, the need to understand the possible impact of epizootics among bighorn sheep populations of differing sizes is apparent. Specifically, we should be asking whether smaller populations have greater mortality rates than larger populations, and whether this places them at greater risk of extinction. Smaller populations may experience more direct contact among individuals, resulting in faster transmission rates of pathogens. Also, being gregarious, bighorn sheep might also become more stressed when living in reduced numbers.

This study was designed to determine the existence, type, and level of association between population size and mortality rate during epizootics in bighorn sheep populations. In addition, we estimate and suggest a minimum viable population size that considers disease, and evaluate possible risks of extinction of bighorn sheep populations related to epizootic events.

MATERIALS AND METHODS

We compiled reports from the literature documenting disease outbreaks that lasted 1– 15 yr in bighorn sheep populations of known size in North America. To define an epizootic event, we used 30% as the defining mortality level. Among documented epizootics of freeranging bighorn sheep populations that occurred in the USA and Canada, we used only those in which the initial population size, mortality rate, and presumed cause were reported. Mortality rates reported did not identify specific age or gender segments of the population. The time line we considered for the epizootic mortalities was as reported by the authors from the time when mortalities were first observed until the population was presumably no longer decreasing. From this analysis we identified two different potential origins of epizootics. The first were epizootics that originated from a suspected contact with a new or unknown pathogen to which bighorn sheep had no natural defenses. These included some Pasteurella and Mannheimia serotypes from domestic sheep and were considered as introduction of a new pathogen. Secondly, we considered epizootics that originated from pathogens that were most likely present in the population but disease may have been triggered by external stress. These were considered as stress-induced.

Scabies has been related to several epizootics but has also been detected in populations with no attributed mortalities (Sandoval, 1980; Welsh and Bunch, 1982; Boyce and Weisenberger, 2005). Therefore, we classified these epizootics based exclusively on the associated factors reported by the authors. If stress factors were described, we considered the epizootic as stress-induced, but if no other factor besides scabies was reported, we categorized it to be of new-pathogen origin. Epizootics where the possible origin seems to be related to both of the cited factors along with the presence of livestock were considered to be of mixed origin, and were considered only for the general analysis. Mortality percentages were graphed by the initial population size and a logarithmic regression line was fitted. This model was used because it stabilizes when a population is projected to infinity.

To calculate an estimated mortality rate for a specific population size, we converted the logarithmic model to a linear model to develop a more accurate estimation. The formulas obtained were applied to different hypothetical population sizes. Although the minimum viable population size for bighorn sheep is controversial, most researchers recommend a founder population of 41 to 125 animals (Berger, 1990; Ehrenfeld, 1994; Gross et al., 2000; Singer et al., 2000a, b, 2001). Management of the founder population may assist populations below that range to persist for the long term. However, for this analysis we considered 50 animals as the minimum size for which a bighorn population would be able to recover after an epizootic event.

RESULTS

Since the 1880s, at least 36" epizootic episodes have been documented in bighorn sheep populations from the USA and Canada. From these we analyzed 23 that had the information necessary for our analysis (Table 1). From the 23 episodes, 13 originated from a new pathogen. Of these, 84% were pneumonias derived from contact with domestic sheep, 8% were suspected to have resulted from contact with domestic sheep, and 8% were scabies where the population apparently had not been exposed previously. We found eight cases of epizootics triggered by stress. Of these, three (37%)were related to changes in weather (and two [25%] of these three were complicated with scabies), 3 (37%) were related to close human activities including capture events, and the remaining two (25%) were related to multiple factors such as the presence of cattle, human activities, population peaks, or extreme weather conditions (e.g., prolonged droughts, extreme low temperatures). Two cases were considered of mixed origin (Aravaipa Canyon [Mouton et al., 1991] 1989 and Hells Canyon ram 1995 [Cassirer et al., 1996]) and were counted only in the total epizootics analysis. In the case of the San Andres epizootic (Sandoval, 1980), we considered only the first years of the epizootic (1976–78), in which the most severe decline was observed. This event might be analyzed in the future as a case of continuing stress plus the presence of psoroptic scabies, which, after a period of more than 20 yr (1976–97) resulted in the decline of a population of more than 200 sheep to a single ewe (Boyce and Weisenberger, 2005). The remaining epizootics were considered in their total period of decline (≤ 15 yr). All but two of the studies examined reported the duration of mortalities at ≤ 5 yr.

In the total epizootics analysis a negative logarithmic relationship was found

between population size before the epizootic (initial size) and the mortality in the epizootic (Fig. 1; $r^2 = 0.4286$, SE=16.03, P < 0.01). When dividing epizootics by their origin, we found no relationship for the new-pathogen origin but a negative logarithmic relationship for the stressinduced origin (Fig. 2; $r^2=0.8055$, SE= 14.9, P < 0.01). In epizootics originated by stress, mortality rate was more predictable than when we considered total epizootics. For the estimated mortality rate related to a certain hypothetical population size, we used the equation obtained from the conversion of the logarithmic function to a linear model. Total epizootics estimations were based on the equation: $y=85.0890+(-0.06293x); r^2=0.4283; SE=$ 4.84. Stress-induced epizootics estimations were based on the equation y= $83.6310 + (-0.07055x); r^2 = 0.7839; SE =$ 5.139.

To have a high probability of persisting, populations should consist of at least 173 animals to survive a stress-induced epizootic (Table 3) and 188 animals to survive a general epizootic (Table 2).

DISCUSSION

The relationship between percentage of mortality and population size suggests that future minimum viable population size (MVP) for bighorn sheep should be greater than conventionally reported to account for the high risk of disease. Usually MVP considers only genetic, demographic, and environmental factors (Primack, 2001), and disease is considered as natural and predictable. Bighorn sheep are more susceptible than other ovine species to certain pathogens. The population impact of an epizootic may be great enough to affect population persistence through reduced recruitment and continuing mortality that may occur for 3–5 yr (Jessup, 1985; Gross et al., 2000). Such impacts could result in the local extinction of a population. Gross et al. (2000) demonstrated that disease was the most

Epizootic date, place	Initial population/ mortality (%) ^b	Associated disease/possible cause	Origin of epizootic	Reference
1881–85. Wyoming	U/U	Scabies	Unknown	Lange, 1980
1880–90. Montana	U/U	Scabies	Unknown	Lange, 1980
1870–80. Idaho	U/U	Scabies	Unknown	Goodson, 1982
1870–79. California	U/U	Scabies	Unknown	Lange, 1980
1900–20. Bock Creek. Montana	U/U	Not determined	Unknown	Goodson, 1982
1917–30, Rocky Mountain National Park, Colorado	U/U	Pneumonia	Unknown	Goodson, 1982
1916–22, Utah	U/U	Scabies	Unknown	Goodson, 1982
1925, Sun River, Montana	U/70	Not determined	Unknown	Goodson, 1982
1931, Colorado	U/U	Scabies	Unknown	Lange, 1980
1936, Oregon	U/U	Scabies	Unknown	Lange, 1980
1939, Kootenay National Park, British Columbia	U/U	Pneumonia	Unknown	Goodson, 1982
1942–50, Thompson Falls, Montana ^a	50/100	Contact with domestic sheep	New pathogen	Goodson, 1982
1950, Dinosaur National Monument, Colorado	U/100	Not determined	Unknown	Goodson, 1982
1965–70, Upper Rock Creek, Mon- tana ^a	150/100	Pneumonia/contact with domestic sheep	New pathogen	Goodson, 1982
1965, Bull River, British Columbia ^a	250/97	Pneumonia/contact with domestic sheep	New pathogen	Goodson, 1982
1955–70, Big Hatchet, New Mexico ^a	125-150/84	Drought and other factors	Stress factors	Watts, 1979
1971, Black Gap Wildlife Manage- ment Area, Texas ^a	20/90	Pneumonia/stress when being released	Stress factors	Kilpatric, 1982
1976–78, San Andres National Wild- life Refuge, New Mexico ^a	200/67	Scabies/changes in weather	Stress factors	Sandoval, 1980
1980–81, Black Mountains, California and Nevada ^a	511/38	Scabies/drought, high population density	Stress factors	Welsh and Bunch, 1982
1980–81, Waterton Canyon, Colo- rado ^a	77/77	Pneumonia/human activities	Stress factors	Bailey, 1986
1981–82, Macquire Creek, British Columbia, Canada ^a	50/52	Pneumonia/contact with domestic sheep.	New pathogen	Goodson, 1982
1980, Lava Beds National Monument, California ^a	42/76	Pneumonia/capture stress	Stress factors	Blaisdell, 1982
1981, Mormon Mountains, Nevada ^a	600/50	Pneumonia/contact with domestic sheep	New pathogen	Jessup, 1981
1979–81, Methow Game Range, Washington ^a	14/93	Pneumonia/contact with domestic sheep	New pathogen	Foreyt and Jessup, 1982
1982, Wigwam, British Columbia, Canada ^a	300/50	Pneumonia/contact with domestic sheep	New pathogen	Goodson, 1982
1988, Warner Mountains, California ^a	65/100	Pneumonia/contact with domestic sheep	New pathogen	Weaver, 1989
1981, Latir Parks, New Mexico ^a	36/100	Pneumonia/contact with domestic sheep	New pathogen	Sandoval, 1988
1985, Sheep River Wildlife Sanctuary, Alberta $^{\rm a}$	250/54	Apparent pneumonia	Stress factors	Festa-Bianchet, 1988
1986, Lostine, Wallowa Mountains, Oregon ^a	97/70	Pneumonia/contact with domestic sheep	New pathogen	Coggins and Matthews, 1992
1988, Southeast Washington ^a	80/62	Scabies/contact with transplanted Rocky Mountain bighorn	New pathogen	Foreyt et al., 1990

TABLE 1. Epizootics and mortalities reported in bighorn sheep (Ovis canadensis) in the USA and Canada.

Epizootic date, place	Initial population/ mortality (%) ^b	Associated disease/possible cause	Origin of epizootic	Reference
1989, Aravaipa Canyon, Arizona ^a	195/59	Blue Tongue-EHD ^b / drought, cattle	Mixed origins	Mouton et al. 1991
1990–91, Whiskey Mountains, Dubois, Wyoming ^a	600-900/30-40	Pneumonia/cold temperatures	Stress factors	Ryder et al., 1992
1992–93, East Range, Nevada	U/U	Not determined	Unknown	Martin et al., 1996
1992–93, Desatoya Range, Nevada	U/U	Pneumonia	Unknown	Martin et al., 1996
1995, Hells Canyon, Washington and Oregon	700/50–75	Pneumonia/presence of cattle, goats, domestic sheep	Mixed origins	Cassirer et al., 1996
1997–2000, Kenosha and Tarryall Mountains, Colorado	250/50	Contact with domestic sheep	New pathogen	George et al., 2008
2005, Custer State Park, South Dakota ^a	200/75	Contact with domestic sheep	New pathogen	Freeman, 2006

TABLE	1.	Continued.

^a Epizootic analyzed for this study.

^b U = unknown; EHD = epizootic hemorrhagic disease.

important factor influencing bighorn sheep population dynamics and in a similar study, Singer et al. (2001) suggested 292±82 animals as the minimum population size that would be able to recover from an epizootic. In the state of Sonora (excluding Tiburon Island) there are 46 bighorn ranges, which have been divided into seven wildlife management system units (SUMAS; Dirección General de Vida Silvestre, unpubl. data). These SUMAS are connected bighorn sheep ranges where these populations may have contact. Considering that populations have genetic flow between them, at least three of these units do not have populations above 188 animals. Our results indicate that 188 is the minimum population size that would not be at risk of extinction following an epizootic event.

Causes of bighorn population extinctions often can be associated with additional factors that are independent of stress or disease. Predation, for example, may be important, especially to smaller populations. Our analyses were based on historical epizootics and these complex factors are present today. The information derived from these historic events were based on authors' knowledge of the initial population sizes and remaining numbers after the epizootic event, as well as the possible associated cause. Some of this information may not be as accurate as recent estimates. However, we consider that their observations were reliable for estimating a general mortality rate of the populations being studied.

Additional factors beyond population size should be considered. Current estimates of bighorn sheep populations in Mexico frequently are based on aerial survey data. Complex aspects such as population dynamics, probability of contact with domestic animals, inbreeding level, genetic flow among populations, and suitable habitat patch sizes are largely unknown. Many bighorn populations in Mexico are isolated and the loss of genetic variability can reduce population fitness through decreased reproductive ability and reduced immunologic capacity (Munson, 1993). These effects can increase mortality during an epizootic, increasing



FIGURE 1. Negative logarithmic relationship found between initial population size and mortality in the analysis of total bighorn sheep epizootics (1942–2005).

the probability of population extinction. Although there are studies of the health status of bighorn sheep populations, there is insufficient information from serologic or mortality studies of Mexican bighorn sheep populations to fully understand the pathogens potentially associated with disease-related declines in these populations. The potential for disease transmission following translocations among resident populations is a factor rarely considered in Mexico. In many Wildlife Management and Utilization Units in Mexico, bighorn sheep are kept in proximity to cattle (2-5 km) and in some cases they are separated only by fences (Cassaigne, pers. obs.). Even though there is no direct contact between bighorn sheep and domestic animals, many diseases can be indirectly transmitted by vectors. Goats have been observed near two important bighorn sheep areas in Sonora State and may be associated with population declines in those areas (Lee, 2004). Goats or



FIGURE 2. Negative logarithmic relationship found between initial population size and mortality in bighorn sheep epizootics induced by stress factors (1942–2005).

TABLE 2. Relationship between initial population size and the predicted mortality caused by epizootic events (see Materials and Methods for explanation of predictive model). For long-term persistence following an epizootic event (estimated remaining population \geq 50), a minimum initial population size of \geq 188 animals is required.

Initial population	Expected mortality (%)	Estimated remaining
20	80	4
50	82	9
150	75	37
188	73	50
200	72	56
250	69	77
300	69	102
500	53	235

cattle are present near bighorn sheep habitat and, in addition to increased potential for disease transmission, interactions with goats or cattle may also increase stress. Bissonette and Steinkamp (1996) reported avoidance of habitat by bighorn sheep when livestock were present. During an epizootic in Sierra del Viejo, Sonora, Mexico, the bighorn population decreased from 126 sheep in 1993 to 17 in 2003 (Lee, 2004). Specific causes for this decline are unknown and need to be understood to support management decisions related to Mexico's bighorn sheep recovery program.

TABLE 3. Relationship between initial population size and predicted mortality caused by epizootic events originating from stress factors (see Materials and Methods for explanation of predictive model). For long-term persistence following an epizootic event, a minimum population size of 173 animals is required.

Initial population	Expected mortality (%)	Estimated remaining
20	82	3
50	80	10
150	73	40
173	71	50
200	69	62
250	66	85
300	62	114
500	48	260

Our suggested disease-based MVP size of 188 animals does not imply that smaller populations cannot survive after facing an epizootic event, but populations below that number may have lower probabilities of recovery and long-term persistence, and would probably require more intensive and costly management. On the other hand, populations above 188 animals also could become extinct, since additional factors such as inbreeding, habitat patch sizes, fragmentation, predation, and environmental conditions can increase mortality during an epizootic. Minimizing stress factors and avoidance of close contact with domestic sheep would decrease the probability of an epizootic; however, no bighorn sheep population can be considered entirely without risk.

Based on 2004 bighorn sheep population estimates by the wildlife department (Dirección General de Vida Silvestre, unpubl. data) in Sonora, our minimum population numbers suggests that only 42% of existing bighorn sheep would persist in the long term. The situation in the USA and Canada could be similar due to the fragmentation and isolation of many populations (Valdez and Krausman, 1999).

Information related to disease and population dynamics is needed to conserve and recover bighorn sheep populations in Mexico. Results from this study suggest that individual populations should be managed to exceed 170 animals but the causes and population factors (e.g., genetic variability, stress associated with livestock contact, increased disease transmission on shared habitats with domestic animals) associated with epizootics need to be better defined to continue to refine and understand disease risks as they relate to long-term bighorn sheep management.

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LITERATURE CITED

- ALLEN, R. W. 1980. Natural mortality and debility. In The desert bighorn. Its life, history, ecology, and management, G. Monson and L. Sumner (eds.). University of Arizona Press, Tucson, Arizona. pp. 172–185.
- BAILEY, J. A. 1986. The increase and dieoff of Waterton Canyon bighorn sheep: Biology, management and dismanagement. Proceedings of the Northern Wild Sheep and Goat Council 5: 325–340.
- BERGER, J. 1990. Persistence of different-sized populations: An empirical assessment of rapid extinctions in bighorn sheep. Conservation Biology 4: 91–98.
- BISSONETTE, J. A., AND M. J. STEINKAMP. 1996. Bighorn sheep response to ephemeral habitat fragmentation by cattle. Great Basin Naturalist 56: 319–325.
- BLAISDELL, J. A. 1982. Lava beds wrap-up, what did we learn? Desert Bighorn Council Transactions 26: 32–33.
- BOYCE, W. M., AND M. E. WEISENBERGER. 2005. The rise and fall of psoroptic scabies in bighorn sheep in the San andres Mountains, New Mexico. Journal of Wildlife Diseases 41: 525– 531.
- CASSIRER, F. E., L. OLDENBURG, V. COGGINS, P. FOWLER, K. RUDOLPH, D. HUNTER, AND W. J. FOREYT. 1996. Overview and preliminary analysis of a bighorn sheep dieoff, Hells Canyon 1995– 96. Proceedings of the Northern Wild Sheep and Goat Council 10: 78–86.
- Coccins, V. L., AND P. MATTHEWS. 1992. Lamb survival and herd status of the Lostine bighorn herd following a *Pasteurella* die-off. Proceedings of the Northern Wild Sheep and Goat Council 8: 147–154.
- EHRENFELD, D. 1994. Persistence and population size in mountain sheep: Why different interpretations? Conservation Biology 8: 617–618.
- ESPINOSA, T. A., A. SANDOVAL, AND B. A. CONTRERAS. 2006. Historical distribution of desert bighorn sheep (*Ovis canadensis mexicana*) in Coahuila, Mexico. Southwestern Naturalist 51: 282–288.
- FESTA-BIANCHET, M. 1988. A pneumonia epizootic in bighorn sheep, with comments on preventive management. Proceedings of the Northern Wild Sheep and Goat Council 6: 66–76.
- FOREYT, W. J., AND D. A. JESSUP. 1982. Fatal pneumonia of bighorn sheep following association with domestic sheep. Journal of Wildlife Diseases 18: 163–168.
 - —, V. L. Coggins, and P. Fowler. 1990. Psoroptic scabies in bighorn sheep in Washing-

ton and Oregon. Proceedings of Northern Wild Sheep and Goat Council 7: 135–142.

- FREEMAN, G. 2006. New threats and old standbys. North Dakota Outdoors. February, 2006. pp 8– 13. http://gf.nd.gov/multimedia/ndoutdoors/issues/ 2006/feb/docs/wildlife-diseases.pdf. Accessed April 2010.
- GEORGE, J. L., D. J. MARTIN, P. M. LUKACS, AND M. W. MILLER. 2008. Epidemic pasteurellosis in a bighorn sheep population coinciding with the appearance of a domestic sheep. Journal of Wildlife Diseases 44: 399–403.
- GOODSON, N. J. 1982. Effects of domestic sheep grazing on bighorn sheep populations: A review. Proceedings of the Northern Wild Sheep and Goat Council 3: 287–313.
- GROSS, J. E., F. J. SINGER, AND M. MOSES. 2000. Effects of disease, dispersal, and area on bighorn sheep restoration. Restoration Ecology 8: 25–37.
- HEFFELFINGER, J. R., AND E. MARQUEZ-MUNOZ. 2005. Historical occurrence and distribution of desert bighorn sheep in Chihuahua, Mexico. Desert Bighorn Council Transactions 48: 28–38.
- JESSUP, D. A. 1981. Pneumonia in bighorn sheep: Effects on populations. California-Nevada Wildlife Society Transactions 1981: 72–78.
- ———. 1985. Diseases of domestic livestock which threaten bighorn sheep populations. Desert Bighorn Council Transactions 29: 29–33.
- KEMENYA, M. E., AND M. SCHEDLOWSKIB. 2007. Understanding the interaction between psychosocial stress and immune-related diseases: A stepwise progression. Brain Behavior and Immunity 21: 1009–1018.
- KILPATRIC, J. 1982. Status of desert bighorn sheep in Texas—1982. Desert Bighorn Council Transactions 26: 102–104.
- LANGE, R. E., JR. 1980. Psoroptic scabies in wildlife in the United States and Canada. Desert Bighorn Council Transactions 24: 18–20.
- LEE, R. 2004. A 10-year view of wild sheep management in Sonora, Mexico. Report prepared for Dirección General de Vida Silvestre, Secretaría de Medio Ambiente y Recursos Naturales, Mexico, D. F., Mexico, 3 pp.
- MARTIN, K. D., T. SCHOMMER, AND V. L. COGGINS. 1996. Literature review regarding the compatibility between bighorn and domestic sheep. Proceedings of the Northern Wild Sheep and Goat Council 10: 72–77.
- MCKINNEY, B. R., AND J. DELGADILLO-VILLALOBOS. 2005. Desert bighorn sheep reintroduction in Maderas del Carmen, Coahuila, Mexico. Desert Bighorn Council Transactions 48: 46–49.
- MEDELLIN, R. A., C. MANTEROLA, M. VALDEZ, D. G. HEWITT, D. DOAN-CRIDER, AND T. E. FULBRIGHT. 2005. History, ecology, and conservation of the pronghorn antelope, bighorn sheep, and black bear in Mexico. *In* Biodiversity conservation in Northern Mexico, J. L. Cartron and G. Ceballos

(eds.). University of New Mexico Press, Albuquerque, New Mexico. pp. 387–404.

- MILLER, M. W., T. HOBBS, AND E. S. WILLIAMS. 1991. Spontaneous pasteurellosis in captive Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*): Clinical, laboratory, and epizootiological investigations. Journal of Wildlife Diseases 27: 534–542.
- MILLS, L. S., AND P. E. SMOUSE. 1994. Demographic consequences of inbreeding in remnant populations. American Naturalist 144: 412–431.
- MOUTON, R. J., R. M. LEE, AND R. J. OLDING. 1991. A desert bighorn sheep decline in Aravaipa Canyon, Arizona. Desert Bighorn Council Transactions 35: 18–19.
- MUNSON, L. 1993. Inbreeding and disease in captive wild animals. *In* Zoo and wild animal Medicine. Current therapy 3. 3rd Edition, M. E. Fowler (ed.). W. B. Saunders Company Ltd., Philadelphia, Pennsylvania. pp. 73–79.
- O'BRIEN, S. J., AND J. F. EVERMAN. 1988. Interactive influence of infectious disease and genetic diversity in natural populations. Trends in Ecology and Evolution 3: 254–259.
- ONDERKA, D. K., AND W. D. WISHART. 1982. Experimental contact transmission of *Pasteurella* haemolytica from clinically normal domestic sheep causing pneumonia in Rocky Mountain bighorn sheep. Journal of Wildlife Diseases 24: 663–667.
- OUGH, W. D., AND J. C. DE VOS. 1986. Intermountain travel corridors and their management implications for bighorn sheep. Desert Bighorn Council Transactions 28: 32–36.
- PRIMACK, R. 2001. Problemas de las poblaciones pequeñas. In Fundamentos de conservación biológica. Perspectivas latinoamericanas, R. Primack, R. Roíz, P. Feinsinger, R. Dirzo and F. Massardo (eds.). Fondo de Cultura Económica, México D.F., pp. 363–383.
- PRUETT, S. B. 2003. Stress and the immune system. Pathophysiology 9: 133–153.
- RAMEY, R. R., G. LUIKART, AND F. J. SINGER. 2000. Genetic bottlenecks resulting from restoration efforts: The case of bighorn sheep in Badlands National Park. Restoration Ecology 8: 85–90.
- RISENHOOVER, K. L., J. A. BAILEY, AND L. A. WAKELYN. 1988. Assessing the Rocky Mountain bighorn sheep management problem. Wildlife Society Bulletin 16: 346–352.
- RYDER, T. J., E. S. WILLIAMS, K. MILLS, K. H. BOWLES, AND T. E. THORNE. 1992. Effects of pneumonia on population size and lamb recruitment in Whiskey Mountain bighorn sheep. Proceedings of the Northern Wild Sheep and Goat Council 8: 136–146.
- SACCHERI, I., M. KUUSARI, M. KANKARE, P. VIKMAN, W. FORTELIUS, AND I. HANSKI. 1998. Inbreeding and extinction in a butterfly population. Nature 392: 491–494.

- SANDOVAL, A. V. 1980. Management of a psoroptic scabies epizootic in bighorn sheep (*Ovis canadensis mexicana*) in New Mexico. Desert Bighorn Council Transactions 24: 21–28.
 - 1988. Bighorn sheep die-off following association with domestic sheep: Case history. Desert Bighorn Council Transactions 32: 36–38.
 — AND A. ESPINOSA-TREVIÑO. 2001. Status of the bighorn sheep management programs in Coa-
 - huila, Mexico. Desert Bighorn Council Transactions 45: 53–61.
- SINGER, F. J., C. M. PAPOUCHISA, AND K. K. SYMONDS. 2000a. Translocation as a tool for restoring populations of bighorn sheep. Restoration Ecology 8: 6–13.
 - —, V. C. BLEICH, AND M. A. GUDORF. 2000b. Restoration of bighorn sheep metapopulations in and near Western national parks. Restoration Ecology 8: 14–24.
 - —, L. C. ZEIGENFUSS, AND L. SPICER. 2001. Role of patch size, disease, and movement in rapid

extinction of bighorn sheep. Conservation Biology 15: 1347–1354.

- VALDEZ, R., AND P. R. KRAUSMAN. 1999. Description, distribution, and abundance of mountain sheep in North America. *In* Mountain sheep of North America, R. Valdez and P. R. Krausman (eds.). University of Arizona Press, Tucson, Arizona. pp. 3–19.
- WATTS, T. J. 1979. Status of the Big Hatchet desert sheep population, New Mexico. Desert Bighorn Council Transactions 23: 92–94.
- WEAVER, R. A. 1989. Status of bighorn sheep in California. 1988. Desert Bighorn Council Transactions 33: 11–13.
- WELSH, G. W., AND T. D. BUNCH. 1982. Three-year observation of psoroptic scabies in desert bighorn sheep from northwestern Arizona. Desert Bighorn Council Transactions 26: 42–44.

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Spatio-temporal dynamics of pneumonia in bighorn sheep

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Summary

1. Bighorn sheep mortality related to pneumonia is a primary factor limiting population recovery across western North America, but management has been constrained by an incomplete understanding of the disease. We analysed patterns of pneumonia-caused mortality over 14 years in 16 interconnected bighorn sheep populations to gain insights into underlying disease processes.

2. We observed four age-structured classes of annual pneumonia mortality patterns: all-age, lamb-only, secondary all-age and adult-only. Although there was considerable variability within classes, overall they differed in persistence within and impact on populations. Years with pneumonia-induced mortality occurring simultaneously across age classes (i.e. all-age) appeared to be a consequence of pathogen invasion into a naïve population and resulted in immediate population declines. Subsequently, low recruitment due to frequent high mortality outbreaks in lambs, probably due to association with chronically infected ewes, posed a significant obstacle to population recovery. Secondary all-age events occurred in previously exposed populations when outbreaks in lambs were followed by lower rates of pneumonia-induced mortality in adults. Infrequent pneumonia events restricted to adults were usually of short duration with low mortality.

3. Acute pneumonia-induced mortality in adults was concentrated in fall and early winter around the breeding season when rams are more mobile and the sexes commingle. In contrast, mortality restricted to lambs peaked in summer when ewes and lambs were concentrated in nursery groups.

4. We detected weak synchrony in adult pneumonia between adjacent populations, but found no evidence for landscape-scale extrinsic variables as drivers of disease.

5. We demonstrate that there was a >60% probability of a disease event each year following pneumonia invasion into bighorn sheep populations. Healthy years also occurred periodically, and understanding the factors driving these apparent fade-out events may be the key to managing this disease. Our data and modelling indicate that pneumonia can have greater impacts on bighorn sheep populations than previously reported, and we present hypotheses about processes involved for testing in future investigations and management.

Key-words: bacterial pneumonia, livestock-wildlife interface, Markov model, time series

Introduction

Over the past 20 years, considerable advances have been made in understanding the spatio-temporal patterns of disease persistence and fade-out following invasion into susceptible host populations. Infections that generate

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rapid mortality such as Ebola virus, burn through susceptible populations until there are no more hosts and effectively die out (Sanchez et al. 2001). Infections with a strong immunizing effect, such as measles in England and Wales, persist in populations and exhibit biannual epidemic peaks that coincide with the birth and aggregation of sufficient susceptibles (Bjørnstad & Grenfell 2008). The dynamics of strong immunizing or fatal infections can leave a distinct spatio-temporal signature, although an infection that results in predictable disease in one instance, may appear almost chaotic in another setting; for example, contrast the dynamics of measles in the UK and Niger (Ferrari et al. 2008). Describing these spatiotemporal patterns can reveal underlying processes and this approach can be especially important in understanding infections that have recently invaded a population where the transmission routes or aetiological agents are not clear (Cleaveland et al. 2007). In this article, we examine the spatio-temporal dynamics of pneumonia in bighorn sheep, where the disease has been described for at least 80 years (Rush 1927), but debate continues about the identities and roles of causal agents, and disease remains an important factor limiting recovery of populations.

Bighorn sheep (Ovis canadensis) are social, sexually dimorphic ungulates. The species commonly occurs in spatially structured, demographically independent, interconnected populations in steep, rugged terrain. Males and females pursue different life-history strategies (Bleich et al. 1996; Rubin, Boyce & Caswell-Chen 2002). Interactions between the sexes are concentrated around the breeding season which is relatively short in northern latitudes and high altitudes (Bunnell 1982; Thompson & Turner 1982; Bleich, Bowyer & Wehausen 1997; Valdez & Krausman 1999). Seasonal breeding also governs contact patterns between age classes, and each year a pulse of neonates is reared in female-juvenile nursery groups. Outside the breeding season, mature males and females generally occur in male-only, female-only or female-offspring associations. Males are more mobile and more likely than females to contact conspecific hosts in adjacent populations, or potential disease reservoirs such as domestic sheep (Bleich, Bowyer & Wehausen 1997; Rubin et al. 1998; DeCesare & Pletscher 2006).

Pneumonia is a significant factor limiting the distribution and abundance of bighorn sheep (Gross, Singer & Moses 2000; Cassirer & Sinclair 2007; Boyce *et al.* 2011). The disease is associated with infection by directly transmitted bacteria, principally thought to be *Mycoplasma ovipneumoniae* and *Mannheimia haemolytica*, but, as is often the case with pneumonia, the precise aetiology remains unclear (Foreyt, Snipes & Kasten 1994; Besser *et al.* 2008, 2012b; Dassanayake *et al.* 2009, 2010). Initially, infection probably originates in domestic sheep, but once it has spilled over into bighorn sheep populations it is most likely maintained in the population and spread by bighorn sheep. Bighorn sheep appear highly susceptible to infection from domestic sheep: nearly all (98%) of a total of 90 bighorn sheep that were co-pastured with domestic sheep in 11 experimental commingling studies conducted between 1979 and 2009 died of pneumonia within 100 days, while the domestic sheep remained healthy (summarized in Besser et al. (2012a). Although these captive experimental results support field observations by naturalists and field biologists (Grinnell 1928; Shillenger 1937; Goodson 1982; George et al. 2008), they do not replicate the range of demographic variation in pneumonia events observed under natural conditions. Pneumonia described in free-ranging bighorn sheep populations includes acute die-offs with wide ranges in all-age mortality (10-90%), chronic or sporadic low levels of adult mortality, and annual or sporadic epizootics with high mortality rates restricted to juveniles from 1 to many (>20) years following all-age outbreaks (Rush 1927; Jorgenson et al. 1997; Aune et al. 1998; Enk, Picton & Williams 2001; Hnilicka et al. 2002). The aim of this paper was to use empirical data to describe these mortality patterns in detail and to develop hypotheses about the underlying processes involved. Indeed, a lack of data has so far constrained models of pneumonia dynamics in bighorn sheep (Hobbs & Miller 1992; Gross, Singer & Moses 2000; Clifford et al. 2009; Cahn et al. 2011). Our objective was to develop an understanding of the disease that will ultimately aid in identifying and assessing intervention options.

Materials and methods

STUDY AREA

We studied bighorn sheep in a 22 732 km² area encompassing Hells Canyon of the Snake River in the Blue Mountain and Columbia Plateau ecoregions of Idaho, Oregon and Washington (-117.875°, 46.500° to -116.250°, 44.750°, Fig. 1). Bighorn sheep occupy three climate zones within this diverse area from lowest to highest elevation: Snake River, Blue Mountains and Wallowa Mountains. The low elevation Snake River canyon is warm and dry with temperatures averaging 17.6 °C at Lewiston, ID. Average annual precipitation of 31.4 cm occurs fairly evenly year-round except during the months of July and August. The adjacent uplands including the Blue Mountains in Washington, are cooler and wetter with average temperatures of 10 °C in Pomeroy, Washington (WA) and average annual precipitation of 61 cm at Asotin, WA and 66 cm in Pomerov. The upper elevations in the Wallowa and Seven Devils mountains receive annual precipitation of up to 205 cm, over two-thirds of which occurs as snow. Temperature averages 7 °C at the base of the Wallowa Mountains in Enterprise, OR and annual precipitation averages 76 cm. Seasonal temperature patterns in all three climate zones are similar, with highs in July and August and lows in December and January (Johnson & Simon 1987; Western 2008)

Bighorn sheep are native to Hells Canyon, but were extirpated by 1945, probably through a combination of unregulated hunting, competition with livestock for forage and diseases introduced from domestic sheep (Smith 1954; Johnson 1980; Coggins & Matthews 1996). From 1971 to 1995, wildlife agencies in Idaho, Oregon and Washington translocated a total



Fig. 1. Distribution of the 16 bighorn sheep populations in the Hells Canyon metapopulation, Idaho, Oregon and Washington. AS = Asotin; WE = Wenaha; BB = Black Butte; BC = Big Canyon; BRC = Bear Creek; IM = Imnaha; LHC = Lower Hells Canyon; LO = Lostine; MU = Muir; MV = Mountain View; MY = Myers Creek; RB = Redbird; SC = Upper Saddle Creek; SM = Sheep Mountain; UHCID = Upper Hells Canyon, Idaho; UHCOR = Upper Hells Canyon, Oregon.

of 329 bighorn sheep into Hells Canyon and moved 79 within the metapopulation, establishing 12 interconnected populations prior to our study (Figure S1). Another four populations were established and one population supplemented with translocations 1997-2005, during our study. Populations were delineated by movement patterns of females (Rubin et al. 1998). Females rarely move between populations whereas males may move seasonally or disperse among populations. Periodic pneumonia outbreaks were documented prior to this study, although monitoring was sporadic and most pneumonia events were documented following reports of sick and dying sheep. Over the same time period, domestic sheep grazing declined dramatically. However, reduced numbers of domestic sheep and goats continue to graze intermittently on public and private lands. Active management is ongoing to prevent contact between species: 22 bighorn sheep, five domestic goats and three domestic sheep were removed from areas where there was risk of contact during the study, nonetheless, some potential for disease transmission from domestic sheep and goats existed for all bighorn sheep populations throughout the study.

MONITORING

In 1995 and 1996, all-age pneumonia outbreaks occurred in five populations in the northern part of the project area (Cassirer *et al.* 1996). In 1997, we started monitoring movements and survival of radio-collared bighorn sheep in three of these populations (Redbird, Black Butte and Wenaha) as part of an unsuccessful vaccination trial to improve lamb survival (Cassirer *et al.* 2001). We collared animals in additional populations in 1998, 1999, 2000, 2006 and 2010 including animals that were translocated and, as animals left the study due to death or were censored due to radio failure, we replaced them by collaring new individuals.

State wildlife agencies have conducted periodic ground and aerial surveys since initial reintroductions in 1971. Between 1997 and 2010, annual helicopter surveys were conducted between February and April. Visibility of sheep is high (87%), as determined by detection of radio-collared animals (Idaho Fish and Game data) and population estimates were derived by combining helicopter counts with observations from ground and observations from fixed-wing monitoring of radio-collared animals. Most lambs were born in May and we conducted our population analyses on a biological year, May–April. Annual exponential rate of population increase was calculated as $r = \ln(N_t/N_{t-1})$. During this period, 735–900 bighorn sheep were estimated to occur within the metapopulation. Estimated population sizes ranged from 5 to 190, with a median of 35.

We calculated annual adult survival by sex as the proportion alive in May that survived to the following May in populations with at least five radio-collared animals. Summer lamb survival was the proportion of known offspring of radio-collared ewes that survived until October (approximately to weaning). We classified a female as having a lamb when she was observed alone with, or nursing a lamb. We assumed lambs were dead when the female was no longer associating with a lamb. We located dead lambs through visual observation. We defined recruitment as the ratio of lambs to ewes recorded in the annual February–April surveys.

We located radio-collared sheep at least bi-weekly from the ground or from fixed-wing aircraft. We located females up to several times per week during lamb-rearing to monitor productivity and lamb survival. Radiocollars were equipped with a motion-sensitive switch. When no movement was detected for 4 h, the switch was activated and we conducted an investigation on site and collected the entire carcass or tissue samples for analysis at the Washington Animal Disease and Diagnostic Laboratory (WADDL), Washington State University, Pullman. On the basis of site investigations and necropsy results, we classified causes of death as disease, predation, accident or injury, human-caused or unknown. We censored animals that died within 30 days of capture and animals translocated to Hells Canyon did not enter the study until the start of the biological year following translocation (2–4 months following release).

PATHOLOGY

We based diagnoses of pneumonia on gross and histological examination of lung tissue at WADDL. Gross features used to diagnose pneumonia included consolidation, presence of lung adhesions, abscesses, bronchiectasis or pleuritis. Affected areas of the lung were characterized by tissue colour, consistency and ability to float in formalin. Histological features of acute pneumonia included fibrin and oedema, increased presence of pulmonary macrophages, neutrophils, necrotic neutrophils,

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necrosis, haemorrhage and bacterial colonies in lung tissue. Chronic pneumonia was characterized by the presence of fibrosis, abscesses or bronchiectasis. Bronchiolar epithelial hyperplasia and peribronchiolar lymphocytic infiltrates in the absence of fibrosis or abscessation was designated as subacute pneumonia. Severity (mild, moderate or severe) was based primarily on the percentage of both right and left lung fields affected on gross examination. Severity assessed by histopathology was based on the total percentage of affected tissue on individual sections of lung. Five to 15% total affected lung or tissue was considered mild, >15–50% was moderate and > 50% was severe.

HEALTH STATUS

We used confirmed and suspected (for lambs) pneumonia-caused mortalities to characterize the seasonality, duration and intensity of four types of pneumonia events by population and year: (i) all-age pneumonia, (ii) secondary all-age pneumonia, (ii) adults only, and (iv) lambs only. We classified a population-year as healthy if animals were radiocollared in the population, but we did not detect any pneumonia in adults or detect or suspect pneumonia in lambs as described in the results.

ANALYSIS

We used Mann–Whitney's U test and Wilcoxon's Rank Sum (Siegel & Castellan 1988) to compare median survival rates of adults and juveniles and population growth by health class due to lack of normality in the data (Shapiro-Wilks test P < 0.0001). We analysed seasonal patterns in lamb survival to weaning by translocation status and climate zone with Kaplan–Meier estimates and log-rank tests (Kaplan & Meier 1958).

We fit Bayesian survival models to analyse the effect of pneumonia on the daily mortality risk from birth to 140 days in lambs. Starting at day 0 (birth), we used a piecewise-constant hazard approach where the instantaneous daily mortality hazard, h(a), was assumed to be constant for each day. Daily hazard estimates were smoothed using a first order conditional autoregressive approach, $h(a) = \exp(\beta + y(a))$, where β is a global intercept with an improper flat prior distribution and y (a) was specified using the car.normal function in WinBUGS assuming a Uniform(0,10) hyperprior on σ , and τ , the car.normal precision parameter, set equal to $\frac{1}{\sigma^2}$ (Besag, York & Mollie 1991; Heisey et al. 2010). We used Markov chain Monte Carlo methods to generate separate posterior distributions for daily mortality hazards by health class (pneumonia or healthy). We ran three Markov chains for 100 000 iterations, discarded the first 50 000 steps, and thinned the remaining steps so that our posterior included every 10th iteration. The Markov chains readily converged (Gelman-Rubin statistic ≤ 1.13 for healthy years, and ≤ 1.02 for pneumonia-years). Further details are provided in Appendix S1.

To identify significant seasonal clustering in adult pneumonia mortalities, we fit a logistic regression model to a series of seasons. The response was a binomial equal to the proportion of adult pneumonia mortalities occurring in that season weighted by month, and the predictor was a binary season indicator for 'summer' or 'winter'. We varied the months categorized as summer by starting with the lamb-rearing months, May-August, and classifying all other months as 'winter' and systematically extended the endpoints of the summer season. We present the grouping that showed the greatest difference between seasons.

In populations where we documented pneumonia during the study (we excluded the healthy Asotin and Upper Saddle Creek populations), we used health status in the current year (a categorical predictor taking on separate values for all-age pneumonia, adult-only pneumonia, lamb-only pneumonia or healthy, with healthy as the baseline) as a predictor for future pneumonia (coded as 0 if the next year was healthy, and 1 otherwise). To test for differences among translocated and resident populations, logistic regression models were of the form, $\frac{\pi(x)}{1-\pi(x)} = e^{\alpha_i + \beta X_i}$ where e^{α_i} is the odds of pneumonia this year given last year's health status and $e^{\beta X_i}$ is the multiplicative adjustment to these odds accounting for the population's translocation status, X_i (an indicator taking on the value 0 for resident populations and 1 for translocated populations). We used Firth's bias-reduction technique for complete separation (Firth 1993) because we always observed pneumonia the year following all-age pneumonia.

We estimated annual transition probabilities between pneumonia classes for populations that had experienced epizootics by building a matrix from the frequency of transitions between classes during the study. Since the transition matrix was regular and irreducible (any state could potentially transition to any other state), we derived the stationary distribution by repeatedly multiplying the probability transition matrix by itself until row values converged (*c.* 15 iterations) (Taylor & Karlin 1998).

To assess the evidence for spatial synchrony of pneumonia, we used logistic regression to evaluate the influence of pneumonia status in neighbouring populations on a population's odds of pneumonia. We calculated centroids of 95% contours of fixed kernel home ranges of radiolocations of resident animals by population in Hawth's Tools (Beyer 2004) and ArcMap 9.3 (ESRI 2008). We defined a population's neighbours to be all populations with centroids within a designated Euclidean distance (from 10 to 70 km) of the population of interest. Pneumonia in neighbours was a categorical predictor that took on the value 1 if any neighbouring population had pneumonia in the year of interest, and 0 otherwise. We included years when pneumonia was known to be present in the neighbourhood, even if some neighbours were not sampled. We recognize that our probability of detecting pneumonia was less than 1, so we excluded data points (range from 26 to 53% of points at each distance category) where no pneumonia was detected in neighbours, but not all neighbours were sampled. Since a population's pneumonia status in year t-1 altered its pneumonia odds in year t, we included last year's pneumonia status in both the population of interest and the neighbouring populations as predictors in the models. To evaluate the effect of translocations, we added an indicator variable for translocated populations in the neighbourhood.

Data were analysed in the R statistical computing environment (R Development Core Team, 2008) through the lme4 (Bates, Maechler & Dai 2008) and logistf (Pioner *et al.* 2006) packages. The lamb mortality hazard model was fit in WinBUGS version 1.4 (Lunn *et al.* 2000) through R version 2.13.0 using the R2WinBUGS package (Sturtz, Ligges & Gelman 2005).

Results

PNEUMONIA IN ADULTS

Between 1997 and 2010, 477 bighorn sheep were radiocollared (313F, 164M) in 14 populations (Fig. 1) and monitored for a total of 141 population-years (1–14 years per population). On average, 117 radio-collared adults (range 35–146) were monitored each year, with a median of 24% (range 5–100%) of adults collared in each study population (Table S1). This included 339 resident sheep monitored for 1220 sheep-years. Another 104 sheep translocated to Hells Canyon from presumably healthy populations in British Columbia, Alberta and Montana 1997–2002, and 34 sheep that were moved within the Hells Canyon metapopulation 1999–2005 were monitored for a total of 459 sheep-years. The translocations established the Big Canyon, Muir Creek, and Myers Creek and Saddle Creek populations, and supplemented existing populations at Asotin, Upper Hells Canyon Oregon, Lostine and Bear Creek (Table S1 and Figure S1).

We determined a cause of death for 179 of 264 radiocollared bighorn sheep (94M, 170F) that died and 53 (30%) were diagnosed with bacterial pneumonia (17M, 36F). We also found 12 (8M, 4F) unmarked dead adult sheep that were diagnosed with bacterial pneumonia. Pneumonia-caused mortality of radio-collared sheep was 27% (28 of 104) of translocated animals and 7% of radio-collared resident animals (25 of 339, $\chi^2 = 28.87$, 1 d.f., P < 0.01).

PNEUMONIA IN LAMBS

We submitted 129 unmarked dead lambs from 14 populations for necropsy and euthanized 11 live lambs in four populations. We determined a cause of death for 104 lambs and 92 (88%) were diagnosed with pneumonia including 9 of 11 euthanized lambs (Besser *et al.* 2008). Although juveniles of all ages died from pneumonia, most mortality was prior to weaning, between 4 and 14 weeks of age (Fig. 2). We found no differences in the summer survival distribution functions of lambs in years with pneumonia among the Snake River, Blue Mountains and Wallowa Mountains climate zones ($\chi^2 = 0.1$, 2 d.f.,



Fig. 2. Daily mortality hazard from 0 to 140 days of lambs born to radio-collared ewes in population-years where no pneumonia was documented (in black, 267 lambs) and where pneumonia was diagnosed (in red, 262 lambs). Solid line is the smoothed daily hazard, dark and light bars represent 50% and 95% credible intervals from a conditional autoregressive model. The 95% credible intervals for lamb hazards in pneumonia and healthy population-years did not overlap between the ages of 27 and 101 days.

P = 0.97) or between lambs of translocated and resident ewes ($\chi^2 = 1.5$, 1 d.f., P = 0.23).

Due to the difficulty of detecting freshly dead unmarked lambs in a large, relatively inaccessible and rugged landscape, we assigned a class of 'suspected pneumonia' in lambs based on (1) the distinct temporal signature of documented pneumonia-induced mortality in 37 lamb-only or secondary all-age population-years (Fig. 2); and (2) observations of clinical signs of pneumonia including lethargy, coughing, nasal discharge and discovery of intact dead lambs that were too autolysed for diagnosis. We were conservative in assigning the suspected class of pneumonia to lambs. Median summer lamb survival and recruitment (lamb : ewe ratio) were higher or did not differ in population-years with documented vs. suspected pneumonia (Fig. 3).

HISTOPATHOLOGY

Lung lesions observed at necropsy included acute fibrinous bronchopneumonia and pleuritis, sub-acute bronchointerstitial pneumonia with lymphocytic cuffing of airways and bronchiolar hyperplasia, and chronic pneumonia with fibrosis and abscessation. Acute lesions were observed in approximately half of the mortalities regardless of age class (30 of 65 adults and 33 of 66 lambs). Chronic lesions were present in about half (33) of the adult mortalities compared with about a quarter of the lambs (15). Subacute lesions were more common in lambs (n = 18, 27%) than in adults (n = 2, 3%).

SEASONAL PATTERNS

There was no difference between sexes in monthly patterns of pneumonia-caused adult mortality ($\chi^2 = 6.77$, d.f. = 11, P = 0.82). In both sexes, the odds of pneumonia-caused mortalities were almost three times higher between October and February than during the rest of the year (odds ratio 2.85, 95% CI 1.7–4.8, P < 0.0001). The seasonal pattern was driven by mortalities with acute lesions (odds ratio 4.29, 95% CI 1.7–10.9, P = 0.002). Deaths of animals with chronic lesions were more evenly distributed across seasons (odds ratio 1.9, 95% CI 1.0-4.0, P = 0.05). No acute pneumonia was detected in adults between May and July, the period when most (80%) pneumonia mortalities were detected in lambs. Peak pneumonia mortalities in lambs at 1-3 months of age corresponded to the period when ewes congregated in nursery groups and mortalities associated with pneumonia in adults peaked during the breeding season when mixed sex group sizes were largest (Fig. 4).

TEMPORAL AND SPATIAL PATTERNS

Pneumonia was detected or suspected in 33–77% of the study populations each year. Two populations remained healthy throughout the study: Asotin and Saddle

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Fig. 3. Summer lamb survival and recruitment in healthy, adultonly, all-age and lamb pneumonia-years vs. suspected all-age or lamb pneumonia-years. The horizontal line denotes the median, the box encloses 50% of the observations and the whiskers show the 2.5th and 97.5th percentiles. Median summer lamb survival and recruitment did not differ significantly between allage pneumonia population-years when pneumonia was detected in both adults and lambs and population-years when pneumonia was detected in adults and suspected in lambs ($W \le 48$, P > 0.10).

Creek (Fig. 5). Survival and population growth patterns differed significantly among age-structured health classes, indicating that pneumonia was a dominant and additive source of mortality (Table 1).

Pneumonia restricted to lambs (lamb-only) was the most frequent class of pneumonia observed, and populations usually remained stable (Table 1). Pneumonia in both adults and lambs simultaneously (all-age) occurred in translocated populations in biological years 2000, 2002



Fig. 4. Seasonal patterns of pneumonia and life-history events. (a) Monthly distribution of pneumonia mortalities detected in adults and lambs. (b) Median group sizes of groups with lambs and ewe-mature ram groups by month.

and 2003. This accounted for 68% (19 of 28) of the pneumonia mortalities in translocated animals and resulted in immediate population declines. Secondary all-age pneumonia events occurred in both resident and translocated sheep in populations that had previously experienced allage outbreaks. These events were characterized by summer pneumonia outbreaks in lambs followed by lower rates of pneumonia-induced mortality in adults. Pneumonia in adults only was an infrequent, usually low mortality event (Table 1).

We observed high survival and stable to increasing populations in population-years classified as healthy, even in populations with a previous history of pneumonia. However, once pneumonia invaded a population, healthy periods were usually of short duration (median 1 year, range 1–3 years, Table 1, Fig. 5).

Euclidian distance Median between population centroids was 67 km with a range from 1 (populations separated by the Snake River) to 156 km (Fig. 5). We detected no significant differences in probability of relative to distance neighbouring pneumonia to populations with pneumonia. There was a slight, but insignificant increase in probability of adult or all-age pneumonia-years in populations centred 20 km or less apart ($\beta_{NeighbuorPN} = 0.97$, SE = 0.76, P = 0.20) and no spatial correlation of pneumonia in lambs (Figure S2). Adding a 1-year lag or an indicator for the presence of translocated populations in the neighbourhood did not alter this result (P > 0.32).

We found a significant predictive effect of current pneumonia class on health status of the population the follow-



Fig. 5. Fourteen year time series of pneumonia classes in 16 populations in the Hells Canyon bighorn sheep metapopulation 1995–2010. Black circles represent documented pneumonia in adults, small grey dots represent documented or suspected pneumonia in lambs, open squares indicate no pneumonia detected or suspected. White background with no symbols indicates no data. Grey background indicates years prior to establishment of population through translocation. Vertical lines to the left of the plot connect population centroids at three distance scales. All population centroids were within 156 km or less. AS = Asotin; WE = Wenaha; MV = Mountain View; BB = Black Butte; RB = Redbird; LHC = Lower Hells Canyon, Oregon; IM = Imnaha; BC = Big Canyon; MU = Muir; MY = Myers Creek; SC = Upper Saddle Creek; UHCOR = Upper Hells Canyon, Oregon; UHCID = Upper Hells Canyon, Idaho; SM = Sheep Mountain; LO = Lostine; BRC = Bear Creek.

ing year. Continued pneumonia, usually in lambs, was most likely following all-age and secondary all-age (98%) or lamb-only pneumonia-years (83%). The probability of a pneumonia-year following adult-only and healthy years was similar (63% and 62%, respectively, P = 0.98), and pneumonia was significantly less likely after healthy years than all-age or lamb pneumonia-years ($P \le 0.05$, Table 2).

We used the observed frequency of transitions between health classes to develop a transition matrix (Table 2) with Markov properties: there were a finite number of health classes (or states), health class in the current year was dependent on health in the previous year, and any health class could transition to any other health class. Thus, we could predict the stationary distribution of health classes. Assuming transition probabilities among health classes remain constant, pneumonia is predicted in 81% of populations annually: lamb-only pneumonia 57%, all-age and secondary all-age pneumonia combined 17%, adult-only pneumonia 7%. To further illustrate the dynamics of pneumonia-induced mortality, we combined the stationary distribution with mortality and transition rates (Tables 1 and 2) for a visual representation of the impact of disease over time (Fig 6).

Discussion

Analysis of a 14-year time series of pneumonia in 16 interconnected bighorn sheep populations revealed that

age-structured classes of pneumonia and healthy years had markedly different demographic impacts on populations. All-age pneumonia was consistently associated with population declines, but ultimately, lambs carried the greatest burden of disease. Rates of pneumonia-induced mortality in lambs can vary significantly by population and year, but on average, pneumonia in lambs had an even greater impact than previously reported (Clifford *et al.* 2009; Cahn *et al.* 2011). Recurring annual pneumonia epizootics in lambs may pose the greatest threat to population recovery, and when accompanied by high adult survival, the true consequences of disease may not be realized until senescent adults die and are not replaced.

While pathogen invasion, reinvasion, persistence and fade-out can't be confirmed in the absence of known disease agents, we can evaluate evidence for these processes to develop hypotheses for future investigation. High initial all-age mortality, when compared with subsequent adult mortality in translocated and resident populations is consistent with invasion of pathogens into groups of apparently naïve individuals. Pneumonia in lambs after all-age events must be due to infection from carrier ewes as lambs have little contact with other potential sources of pathogens prior to weaning (Festa-Bianchet 1991; Bleich, Bowyer & Wehausen 1997). Lamb pneumonia outbreaks have also been described in captivity with similar conclusions (Foreyt 1990; Ward *et al.* 1992; Cassirer *et al.* 2001). Pneumonia in lambs is thus a good indication of

Outbreak class	и	n pneumonia cases confirmed	Consecutive years	Population growth (r)	Ewe survival ^a	Ram survival ^a	Summer lamb survival	Spring lamb:ewe ratio
All-age	б	28 Adult	1 (1, 1)	-0.34(-0.42, -0.28)	0.50 (0.29, 0.57)	NA	0.71 (0.29, 0.88)	0.13 (0.06, 0.17)
Secondary all-age	16	(20 Adult, 27 Lamb)	1 (1, 1)	$0 \ (-0.17, \ 0.03)$	$0.82 \ (0.60, 1)$	0.75 (0.67, 1)	$0.10^{\rm b}$ (0, 0.69)	0.05^{b}
Adult only	11	14	1 (1, 2)	0.03 (-0.17, 0.16)	0.83(0.67, 1)	$0.74 \ (0.67, \ 0.83)$	0.75(0.36, 1)	$0.42 \ (0.21, \ 0.79)$
Lamb only	62	65	2 (1, 4)	0 (-0.69, 0.22)	0.93 (0.56, 1)	$0.83 \ (0.60, 1)$	$0.20^{\rm b}$ (0, 0.67)	$0.14^{\rm b}$ (0, 0.60)
Healthy	49	0	1 (1, 13)	0.12 (-0.03, 0.37)	0.93 (0.71, 1)	0.90(0.71, 1)	$0.83 \ (0.33, 1)$	$0.47 \ (0.13, \ 0.73)$
Lamb only Healthy	62 49	65 0	2(1, 4) 2(1, 13)	$\begin{array}{c} 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 $	0.93 (0.56, 1) 0.93 (0.71, 1)	0.83 (0.60, 1) 0.90 (0.71, 1)	$0.20^{\rm b}$ (0, 0.67) 0.83 (0.33, 1)	

Fable 1. Demographic characteristics of health classes in 14 Hells Canyon bighorn sheep populations, 1997–2010. Data reported as median (range). Years where no adults were radiocollared were

Survival in years with documented pneumonia in lambs (n = 8 secondary all-age, n = 33 lamb-only)

infection and pathogen shedding in ewes. The absence of pneumonia-induced mortality or clear symptoms in these ewes during outbreaks in lambs confirms that they have either developed resistance or perhaps tolerance of the pathogen(s) that are lethal to their offspring (Råberg, Graham & Read 2009). Reasons for more frequent fadeout following years with pneumonia restricted to adults remains unclear, but could be explained by differences in pathogens, host immunity or transmission rates.

Our study confirms previously reported accounts of seasonality of pneumonia deaths in bighorn sheep, a pattern commonly observed in infectious diseases of humans and wildlife (Spraker et al. 1984; Aune et al. 1998: Enk. Picton & Williams 2001: Altizer et al. 2006: Cassirer & Sinclair 2007). Age-specific seasonal patterns in pneumonia mortality corresponded to breeding and lamb-rearing: life-history events that are accompanied by especially intensive and concentrated social interactions. The distinct seasonality of adult pneumonia mortality observed in wild populations is not observed in captive experimental bighorn and domestic sheep commingling trials where bighorn sheep die of pneumonia regardless of season. Seasonal physiological or environmental factors are therefore probably less important in precipitating pneumonia epizootics than the timing of pathogen introduction, pathogen virulence and exposure to infections (contact rates). The lack of synchrony of disease events across populations and the absence of an effect of climate on lamb survival during pneumonia-years also suggest that weather or other landscape-scale extrinsic variables (Grenfell et al. 1998; Cattadori, Haydon & Hudson 2005), are unlikely to be important drivers of pneumonia in Hells Canyon.

In lambs, most pneumonia-induced mortality occurred between 1 and 3 months of age, a period that coincided with aggregation in nursery groups. Lamb-to-lamb contact may be an important route of infection as happens in many directly transmitted human 'childhood diseases'; thus, the synchrony in parturition and subsequent concentration of ewes during lamb-rearing which is typical in northern latitudes, could contribute to the timing and high rates of mortality. This period also coincides with the age when passively acquired immunity is probably waning in lambs (Rajala & Castrén 1995), which would further promote transmission and mortality.

By analysing long-term monitoring data to elucidate disease processes from patterns of mortality, we have diverged from studies of bighorn sheep pneumonia that focus on identifying the primary causal agent. The benefits of such a study were that we were able to examine demographic patterns at comparatively large spatial and temporal scales, allowing us to make inferences about processes such as disease introduction, persistence and fade-out. However, the weakness in our approach is an inability to track a known pathogen and directly measure transmission (i.e. infection may occur long before mortality); no opportunity to verify pathogen absence during healthy years;

Table 2. Temporal pattern of pneumonia within affected populations: annual probabilities of transition among health states and annual
probability of any pneumonia. Populations that remained healthy throughout the study, population-years before the initial observation
of pneumonia, and years where no adults were radiocollared were excluded from analyses (Fig. 5 and Table S2)

		Transition st	ate			Probability of any pneumonia	
Initial state	п	All-age ^a	Healthy	Adult	Lamb	<i>P</i> -value relative to healthy state)	
Healthy	24	0.13	0.33	0.08	0.46	0.62 (0.4, 0.8)	
All-age ^a	17	0.18	0.00	0.06	0.72	0.97 (0.8, 1; P < 0.01)	
Adult	11	0.18	0.36	0.09	0.36	0.63 (0.3, 0.9; P = 0.98)	
Lamb	54	0.15	0.19	0.07	0.59	$0.82 \ (0.6, \ 0.9; \ P = 0.05)$	

^aAll-age and secondary all-age classes combined.



Fig. 6. Long-term patterns of pneumonia mortality in bighorn sheep populations experiencing epizootics, Hells Canyon 1997–2010. Stationary distribution of four age-structured population health classes (all-age includes secondary all-age) and the probabilities of staying within a class or transitioning out. Pneumonia classes are circles scaled by relative frequency multiplied by median death rates of ewes (black) and/or lambs (grey) in the class. The healthy class is not scaled. The thickness of arrows between classes is proportional to transition probabilities (Table 2).

and no possibility to monitor genetic variation in the pathogen over time. Given these limitations, as well as the usual constraints of marking and monitoring animals in the field, a primary concern is an imperfect detection probability for pneumonia, which could lead to overestimating healthy population-years. However, the likelihood of detecting pneumonia was not correlated with the intensity of monitoring as measured by the proportion of the population that was radiocollared (median in suspected and detected pneumonia-years = 0.22; in healthy years = 0.28, U = 3331.5, 1 d.f., P = 0.09, Tables S1 and S2), or the frequency of locations (median locations per animal per year in suspected and detected pneumonia-years = 30; in healthy years = 32; U = 2427.5, 1 d.f., P = 0.45). Therefore, there was no bias towards monitoring populations with pneumonia and, despite potentially misclassifying some lower mortality pneumonia events, we still detected significant differences in population dynamics between several different classes of pneumonia and healthy years. Survival and population growth were also similar in years classified as healthy in populations with and without a history of pneumonia, suggesting that healthy years, with true absence of disease-related mortality (but not necessarily true absence of infection), did occur, even in populations with previous pneumonia, and these classifications are useful and appropriate for describing the system.

Our observations concur with many of the results of previous studies, but also raise questions about disease models that assume all-age pneumonia outbreaks followed by lamb mortality at a constant or declining rate for a period of usually 1-6 years (Gross, Singer & Moses 2000; Clifford et al. 2009; Cahn et al. 2011). We observed that pneumonia persisted within populations (or was periodically reintroduced) consistently longer than previous models have assumed, and, as indicated by the Markov model stationary frequency distribution, continued to affect allage classes, not just lambs. The consequence is that allage pneumonia events can result in sporadic or chronic, long-term reduction of survival of both adult and juvenile age classes. The disparity between our findings and previous studies may be due to the greater sampling intensity, duration and spatial scale of our study. Furthermore, whereas initial invasion associated with high rates of mortality is fairly easy to detect, the end of an epizootic is not always clear. Previously published models assume that low mortality or healthy years represent the pathogen extinction and the end of the epizootic. However, if disease in a long-lived animal like bighorn sheep is accompanied by latent periods and low rates of mortality in chronically infected animals, absence of mortality may not reflect absence of pathogens. Long-term dynamics could be a function of changes in immune status in individuals and include stochastic events common to small populations, such as dispersal, colonization, recruitment, death, intermittent pathogen shedding or lambing status of asymptomatic carriers.

By analysing long-term patterns, we have generated hypotheses about the disease processes associated with pneumonia epizootics in bighorn sheep. As with other diseases with high levels of heterogeneity, these processes are probably affected by a number of factors, including previous exposure of hosts, pathogen dose or virulence, and spatial structuring and contact rates in host popula-

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tions (Grassly & Fraser 2008; Salkeld et al. 2010; Wendland et al. 2010; Jesse & Heesterbeek 2011). On the basis of the patterns we observed, the disease appears to be an infection that, in some ways is similar to measles and other immunizing diseases in humans in that it spreads through all-age classes during invasion, but subsequently mainly affects susceptible juveniles. However, in contrast with measles, pathogens apparently persist, occasionally causing fatal pneumonia in previously exposed adults, and the variable lung lesions and associated bacteriology suggest a polymicrobial aetiology, thus secondary pathogens may play a role in severity and recurrence (Besser et al. 2012b). The course of the disease may also be affected by the timing of pathogen invasions relative to contact rates associated with seasonal breeding and parturition. The importance of between-population transmission and recurrent infection from domestic sheep deserves additional investigation as do the conditions that lead to disease and pathogen fade-out.

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References

- Altizer, S., Dobson, A., Hosseini, P., Hudson, P., Pascual, M. & Rohani, P. (2006) Seasonality and the dynamics of infectious disease. *Ecology Letters*, 9, 467–484.
- Aune, K.E., Anderson, N., Worley, D., Stackhouse, L., Henderson, J. & Daniel, J.E. (1998) A comparison of population and health histories among seven Montana bighorn sheep populations. *Proceedings Northern Wild Sheep and Goat Council*, **11**, 46–69.
- Bates, D., Maechler, M. & Dai, B. (2008) lme4: Linear mixed-effects models using S4 classes. R package version 0.999375-28. http://lme4.r-forge. r-project.org/
- Besag, J.P., York, J. & Mollie, A. (1991) Bayesian image-restoration, with two applications in spatial statistics. *Annals of the Institute of Statistical Mathematics*, 43, 1–20.
- Besser, T.E., Cassirer, E.F., Potter, K.A., VanderSchalie, J., Fischer, A., Knowles, D.P., Herndon, D.R., Rurangirwa, F.R., Weiser, G.C. & Srikumaran, S. (2008) Association of *Mycoplasma ovipneumoniae* infection with population-limiting respiratory disease in free-ranging Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*). Journal of Clinical Microbiology, 46, 423–430.
- Besser, T.E., Cassirer, E.F., Yamada, C., Potter, K.A., Herndon, C.N., Foreyt, W.J., Knowles, D.P. & Srikumaran, S. (2012a) Survival of bighorn sheep (*Ovis canadensis*) commingled with domestic sheep (*Ovis*

aries) in the absence of Mycoplasma ovipneumoniae. Journal of Wildlife Diseases, 48, 168-172.

- Besser, T.E., Highland, M., Baker, K., Cassirer, E.F., Anderson, N.J., Ramsey, J.M., Mansfield, K.M., Bruning, D., Wolff, P., Smith, J.B. & Jenks, J.A. (2012b) Causes of pneumonia epizootics among bighorn sheep, western United States, 2008–2010. *Emerging Infectious Diseases*, 18, http://dx.doi.org/10.3201/eid1803.111554.
- Beyer, H.L. (2004) Hawth's Analysis Tools for ArcGIS. http://www.spatialecology.com/htools.
- Bjørnstad, O. & Grenfell, B. (2008) Hazards, spatial transmission and timing of outbreaks in epidemic metapopulations. *Environmental and Ecological Statistics*, 15, 265–277.
- Bleich, V.C., Bowyer, R.T. & Wehausen, J.D. (1997) Sexual segregation in mountain sheep: resources or predation? *Wildlife Monographs*, 134, 1– 50.
- Bleich, V.C., Wehausen, J.D., Ramey III, R.R. & Rechel, J.L. (1996) Metapopulation Theory and Mountain Sheep: Implications for Conservation. Island Press, Washington, DC, USA.
- Boyce, W.M., Weisenberger, M.E., Penedo, M.C. & Johnson, C.K. (2011) Wildlife translocation: the conservation implications of pathogen exposure and genetic heterozygosity. *BMC Ecology*, 11, 5.
- Bunnell, F.L. (1982) The lambing period of mountain sheep synthesis, hypotheses, and tests. *Canadian Journal of Zoology-Revue Canadienne* De Zoologie, **60**, 1–14.
- Cahn, M.L., Conner, M.M., Schmitz, O.J., Stephenson, T.R., Wehausen, J.D. & Johnson, H.E. (2011) Disease, population viability, and recovery of endangered Sierra Nevada bighorn sheep. *Journal of Wildlife Man*agement, **75**, 1753–1766.
- Cassirer, E.F. & Sinclair, A.R.E. (2007) Dynamics of pneumonia in a bighorn sheep metapopulation. *Journal of Wildlife Management*, **71**, 1080– 1088.
- Cassirer, E.F., Oldenburg, L.E., Coggins, V.L., Fowler, P., Rudolph, K.M., Hunter, D.L. & Foreyt, W.J. (1996) Overview and preliminary analysis of a bighorn sheep dieoff, Hells Canyon 1995-96. *Biennial Symposium Northern Wild Sheep and Goat Council*, **10**, 78–86.
- Cassirer, E.F., Rudolph, K.M., Fowler, P., Coggins, V.L., Hunter, D.L. & Miller, M.W. (2001) Evaluation of ewe vaccination as a tool for increasing bighorn lamb survival following pasteurellosis epizootics. *Journal of Wildlife Diseases*, 37, 49–57.
- Cattadori, I.M., Haydon, D.T. & Hudson, P.J. (2005) Parasites and climate synchronize red grouse populations. *Nature*, 433, 737–741.
- Cleaveland, S., Mlengeya, T., Kaare, M., Haydon, D., Lembo, T., Laurenson, M.K. & Packer, C. (2007) The conservation relevance of epidemiological research into carnivore viral diseases in the Serengeti. *Conservation Biology*, **21**, 612–622.
- Clifford, D.L., Schumaker, B.A., Stephenson, T.R., Bleich, V.C., Cahn, M.L., Gonzales, B.J., Boyce, W.M. & Mazet, J.A.K. (2009) Assessing disease risk at the wildlife-livestock interface: a study of Sierra Nevada bighorn sheep. *Biological Conservation*, 142, 2559–2568.
- Coggins, V.L. & Matthews, P.E. (1996) Rocky mountain bighorn sheep in Oregon, history and present status. *Biennial Symposium Northern Wild Sheep and Goat Council*, 10, 87–92.
- Dassanayake, R.P., Shanthalingam, S., Herndon, C.N., Lawrence, P.K., Cassirer, E.F., Potter, K.A., Foreyt, W.J., Clinkenbeard, K.D. & Srikumaran, S. (2009) *Mannheimia haemolytica* serotype A1 exhibits differential pathogenicity in two related species, *Ovis canadensis* and *Ovis aries. Veterinary microbiology*, **133**, 366–371.
- Dassanayake, R.P., Shanthalingam, S., Herndon, C.N., Subramaniam, R., Lawrence, P.K., Bavananthasivam, J., Cassirer, E.F., Haldorson, G.J., Foreyt, W.J., Rurangirwa, F.R., Knowles, D.P., Besser, T.E. & Srikumaran, S. (2010) *Mycoplasma ovipneumoniae* can predispose bighorn sheep to fatal *Mannheimia haemolytica* pneumonia. *Veterinary microbiology*, **145**, 354–359.
- DeCesare, N.J. & Pletscher, D.H. (2006) Movements, connectivity, and resource selection of Rocky Mountain bighorn sheep. *Journal of Mam*malogy, 87, 531–538.
- Enk, T.A., Picton, H.D. & Williams, J.S. (2001) Factors limiting a bighorn sheep population in Montana following a dieoff. *Northwest Science*, 75, 280–291.
- ESRI. (2008) ArcMap 9.3, Service Pack 1. http://www.esri.com. Redlands, CA.
- Ferrari, M.J., Grais, R.F., Bharti, N., Conlan, A.J.K., Bjørnstad, O.N., Wolfson, L.J., Guerin, P.J., Djibo, A. & Grenfell, B.T. (2008) The dynamics of measles in sub-Saharan Africa. *Nature*, **451**, 679– 684.

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528 E. Frances Cassirer et al.

- Festa-Bianchet, M. (1991) The social system of bighorn sheep: grouping patterns, kinship and female dominance rank. *Animal Behaviour*, 42, 71–82.
- Firth, D. (1993) Bias reduction of maximum likelihood estimates. *Biomet*rika, 80, 27–38.
- Foreyt, W.J. (1990) Pneumonia in bighorn sheep: effects of *Pasteurella haemolytica* from domestic sheep and effects on survival and long-term reproduction. *Biennial Symposium Northern Wild Sheep and Goat Council*, 7, 92–101.
- Foreyt, W.J., Snipes, K.P. & Kasten, R.W. (1994) Fatal pneumonia following inoculation of healthy bighorn sheep with *Pasteurella haemolytica* from healthy domestic sheep. *Journal of Wildlife Diseases*, **30**, 137–145.
- George, J.L., Martin, D.J., Lukacs, P.M. & Miller, M.W. (2008) Epidemic pasteurellosis in a bighorn sheep population coinciding with the appearance of a domestic sheep. *Journal of Wildlife Diseases*, 44, 388–403.
- Goodson, N.J. (1982) Effects of domestic sheep grazing on bighorn sheep populations: a review. *Biennial Symposium Northern Wild Sheep and Goat Council*, 3, 287–313.
- Grassly, N.C. & Fraser, C. (2008) Mathematical models of infectious disease transmission. *Nature Reviews Microbiology*, 6, 477–487.
- Grenfell, B.T., Wilson, K., Finkenstadt, B.F., Coulson, T.N., Murray, S., Albon, S.D., Pemberton, J.M., Clutton-Brock, T.H. & Crawley, M.J. (1998) Noise and determinism in synchronized sheep dynamics. *Nature*, **394**, 674–677.
- Grinnell, G.B. (1928) Mountain sheep. Journal of Mammalogy, 9, 1-9.
- Gross, J.E., Singer, F.J. & Moses, M.E. (2000) Effects of disease, dispersal, and area on bighorn sheep restoration. *Restoration Ecology*, 8, 25–37.
- Heisey, D.M., E., O.E., Cross, P.C., Joly, D.O., Langenberg, J.A. & Miller, M.W. (2010) Linking process to pattern: estimating spatiotemporal dynamics of a wildlife epidemic from cross-sectional data. *Ecological Monographs*, 80, 221–240.
- Hnilicka, P.A., Mionczynski, J., Mincher, B.J., Hinschberger, M., Oberlie, S., Thompson, C.B., Yates, B. & Siermer, D.D. (2002) Bighorn sheep lamb survival, trace minerals, rainfall, and air pollution: are there any connections? *Biennial Symposium Northern Wild Sheep and Goat Council*, **13**, 70–94.
- Hobbs, N.T. & Miller, M.W. (1992) Interactions between pathogens and hosts: simulation of pasteurellosis epizootics in bighorn sheep populations. *Wildlife 2001: Populations*, (eds D.R. McCullough & R.H. Barrett), pp. 997–1007. Elsevier, London
- Jesse, M. & Heesterbeek, H. (2011) Divide and conquer? Persistence of infectious agents in spatial metapopulations of hosts. *Journal of Theoretical Biology*, 275, 12–20.
- Johnson, R.L. (1980) Re-introduction of bighorn sheep in Washington. Biennial Symposium Northern Wild Sheep and Goat Council, 2, 106–112.
- Johnson, C.G. & Simon, S.A. (1987) Plant associations of the Wallowa-Snake province. U.S. Forest Service Pacific Northwest Region Report R6-ECOL-TP-255B-86, Baker City, Oregon, USA.
- Jorgenson, J.T., Festa-Bianchet, M., Gaillard, J.-M. & Wishart, W.D. (1997) Effects of age, sex, disease, and density on survival of bighorn sheep. *Ecology*, 78, 1019–1032.
- Kaplan, E.L. & Meier, P. (1958) Nonparametric estimation from incomplete observations. *Journal of the American Statistical Association*, 1, 53, 457–481.
- Lunn, D.J., Thomas, A., Best, N. & Spiegelhalter, D. (2000) WinBUGS -A Bayesian modelling framework: concepts, structure, and extensibility. *Statistics and Computing*, 10, 325–337.
- Pioner, M., Dunkler, D., H., S. & Heinze, G. (2006) Logistf:Firth's bias reduced logistic regression. R package version 1.06., http://www.meduniwien.ac.at/msi/biometrie/programme/fl/index.html.
- R Development Core Team. (2008) R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.
- Råberg, L., Graham, A.L. & Read, A.F. (2009) Decomposing health: tolerance and resistance to parasites in animals. *Philosophical Transactions* of the Royal Society B: Biological Sciences, 364, 37–49.
- Rajala, P. & Castrén, H. (1995) Serum Immunoglobulin concentrations and health of dairy calves in two management systems from birth to 12 weeks of age. *Journal of Dairy Science*, 78, 2737–2744.
- Rubin, E.S., Boyce, W.M. & Caswell-Chen, E.R. (2002) Modeling demographic processes in an endangered population of bighorn sheep. *Jour*nal of Wildlife Management, 66, 796–810.

- Rubin, E.S., Boyce, W.M., Jorgenson, M.C., Torres, S.G., Hayes, C.L., O'Brien, C.S. & Jessup, D.A. (1998) Distribution and abundance of bighorn sheep in the Peninsular Ranges, California. *Wildlife Society Bulletin*, 26, 539–551.
- Rush, W.M. (1927) Notes on diseases in wild game mammals: mountain sheep. *Journal of Mammalogy*, 8, 163–165.
- Salkeld, D.J., Salathe, M., Stapp, P. & Jones, J.H. (2010) Plague outbreaks in prairie dog populations explained by percolation thresholds of alternate host abundance. *Proceedings of the National Academy of Sciences of the United States of America*, 107, 14247–14250.
- Sanchez, A., Khan, A.S., Zaki, S.R., Nabel, G.J., Ksiazek, T.G. & Peters, C. J. (2001) Filoviridae: Marburg and Ebola viruses. *Fields Virology* (eds D.M. Knipe & P.M. Howley), pp. 1279–1304. Lippincott Williams & Wilkins, Philadelphia, USA.
- Shillenger, J.E. (1937) Disease relationship between domestic animals and wildlife. Transactions North American Wildlife and Natural Resources Conference, 2, 298–302.
- Siegel, S. & Castellan, N.J.J. (1988) Nonparametric statistics for the behavioral sciences, 2nd edn. McGraw-Hill, New York.
- Smith, D.R. (1954) The bighorn sheep in Idaho: its status, life history, and management. Wildlife Bulletin No. 1, Federal Aid to Wildlife Restoration Act., Idaho Project 99R, pp. 154. Idaho Department of Fish and Game, Boise, Idaho, USA.
- Spraker, T.R., Hibler, C.P., Schoonveld, G.G. & Adney, W.S. (1984) Pathologic changes and microorganisms found In bighorn sheep during a stress-related die-off. *Journal of Wildlife Diseases*, 20, 319–327.
- Sturtz, S., Ligges, U. & Gelman, A. (2005) R2WinBUGS: a package for running Win BUGS from R. Journal of Statistical Software, 12, 1–16.
- Taylor, H.M. & Karlin, S. (1998) An introduction to stochastic modeling, 3rd edn. Academic Press, San Diego, CA.
- Thompson, R.W. & Turner, J.C. (1982) Temporal geographic variation in the lambing season of bighorn sheep. *Canadian Journal of Zoology-Revue Canadienne De Zoologie*, **60**, 1781–1793.
- Valdez, R. & Krausman, P.R. (1999) Mountain sheep of North America. University of Arizona Press, Tucson.
- Ward, A.C.S., Hunter, D.L., Jaworski, M.D., Lane, M.V., Zaugg, J.L. & Bulgin, M.S. (1992) Naturally occurring pneumonia in caesarian-derived Rocky Mountain bighorn sheep lambs. *Biennial Symposium Northern Wild Sheep and Goat Council*, 8, 164–173.
- Wendland, L.D., Wooding, J., White, C.L., Demcovitz, D., Littell, R., Berish, J.D., Ozgul, A., Oli, M.K., Klein, P.A., Christman, M.C. & Brown, M.B. (2010) Social behavior drives the dynamics of respiratory disease in threatened tortoises. *Ecology*, **91**, 1257–1262.
- Western, R.C.C. (2008) Historical Climate information. Desert Research Institute, Reno, NV, http://www.wrcc.dri.edu/.

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Supporting Information

Additional Supporting Information may be found in the online version of this article.

Appendix S1. Lamb mortality hazard analysis.

Figure S1. Demographic histories of Hells Canyon bighorn sheep populations.

Figure S2. Spatial synchrony of pneumonia-years across populations.

Table S1. Number and proportion of adult bighorn sheep radiocollared in each population by year and translocation status.

Table S2. Health classification of population-years (141) used to estimate demographic characteristics and transition probabilities of pneumonia and healthy years in Hells Canyon, 1997–2010.

GUIDELINES FOR MANAGEMENT OF DOMESTIC SHEEP IN THE VICINITY OF DESERT BIGHORN HABITAT

Technical Staff Desert Bighorn Council

The Bureau of Land Management (BLM) requested that the Technical Staff (Tech Staff) of the Desert Bighorn Council (DBC) prepare management guidelines for domestic sheep in the vicinity of desert bighorn habitat. Desert bighorn habitat includes all geographicareas that would provide for the life requisites of desert bighorn sheep, as defined by state wildlife and/or land management agencies. This request followed a meeting of BLM biologists concerned with problems resulting from interactions between bighorn sheep (Ovis canadensis ssp.) and domestic sheep (0. aries).

The Tech Staff understands that 2 additional factors should be considered. First, the BLM has prepared, or is preparing, land use planning documents in several western states (Nev., Ariz., Colo., and Ut.) that would allow reintroduction of desert bighorns (*O*. c. nelsoni, 0. c. mexicana, and 0. c. cremnobates) into suitable historic habitat. Several potential bighorn reintroductions in Nevada have been contested by the livestock industry; e.g., woolgrowers and cattlemen. They contend that bighorn reintroductions will seriously hamper their ability to graze livestock of their choice on public lands. Second, in 1989, the BLM issued a "Rangewide Plan for Managing Habitat of Desert Bighorn Sheep on Public Lands," which states "Livestock grazing on desert bighorn habitats will be managed via land-use or activity plans to mitigate impacts to desert bighorns and their habitats to ensure objectives for desert bighorn are achieved."

The DBC is comprised of state fish and game and federal agency biologists, private research organizations, academia, and the public. The 4 primary objectives of the DBC are to: provide for the exchange of information on the needs and management of desert bighorns; stimulate and coordinate studies in all phases of the life history, ecology, management and protection, recreational, and economic uses of desert bighorns; provide a clearinghouse for information among all agencies, organizations, and individuals professionally engaged in work on the desert bighorn; and function in a professional advisory capacity, where appropriate, on local, national, and international questions involving the management and protection of desert bighorn.

The DBC's Tech Staff is comprised of 7 elected members. One of the functions of the Tech Staff is to answer requests from agencies and organizations such as the BLM, regarding desert bighorn management.

This document describes problems associated with domestic sheep and bighorn interactions, with emphasis on diseases. Recommendations are then provided to minimize interaction, especially physical contact between domestic and bighorn sheep.

The Tech Staff appreciates the opportunity to consider the problems and develop these guidelines, with the underlying goal of eliminating domestic sheep and bighorn conflicts on public lands.

BACKGROUND

Current bighorn numbers are <2% of what they were prior to the coming of European man and his livestock and firearms (Wagner 1978). Following enormous population declines in the late 1800s and early 1900s, bighorn populations did not recover, in contrast to other wildlife species such as mule deer (Odocoileus hemionus) and elk (Cervus elaphus). Bighorns have demonstrated much less tolerance than other na-

tive North American ungulates to poor range conditions, interspecific competition, overhunting, and stress caused by loss of habitat. Furthermore, they have shown a much greater susceptibility to diseases (Goodson 1982).

Bighorns have died from a wide variety of diseases that they have contracted from domestic sheep. These include scabies (a major cause of mortality in the 1800s and as late as the 1970s in New Mexico), chronic frontal sinusitis, internal nematode parasites (worms), pneumophilic bacteria, footrot, parainfluenzaIII, bluetongue, and soremouth (contagious echthyma) (Jessup 1985). Documented bighorn die-offs were recorded as early as the mid-1800s and have continued up to the present (Jessup 1985, Goodson 1982, Foreyt and Jessup 1982, Sandoval 1988, Weaver 1988). Die-off documentation covers not only desert bighorns, but also California bighorns (*O. c. californiana*) and Rocky Mountain bighorns (*O. c. canadensis*). Bighorn die-offs have occurred in every state in the western United States.

In broad perspective, when there has been contact between apparently healthy bighorns and domestic sheep, the bighorns die within a few days to a few weeks. While many diseases or stress factors may be involved, bighorns exposed to domestic sheep almost invariably die from pneumonia.

Little is known about the actual mechanism(s) that lead to the demise of bighorns after they have come into contact with domestic sheep. In all of the cases of bighorn die-offs following direct contact with domestic sheep or overlap of grazing in bighorn ranges, 2 things are apparent.

- There is a preponderance of evidence (Table 1) strongly linking the presence of domestic sheep with the subsequent loss of part or all of the affected bighorn population. Of the 25 documented cases (Table 1) 4 of the situations were in controlled laboratory experiments in 3 states, and 2 were in situations where bighorns were penned in large paddocks.
- 2. The effects have all been I way-bighorns have died, while domestic sheep never have suffered ill effects because of coming into contact with bighorn. The prevailing theory on why this has occurred can be summed up as follows: New World sheep (bighorns) are so susceptible to diseases of Old World sheep (domestics) because the bighorns did not co-evolve with the above-listed diseases, as did domestic sheep. Bighorns have not developed effective immunity against these diseases. Domestic sheep are inoculated or, through natural selection over hundreds of years, have developed a resistance against some of these diseases, but carry blood titers for most of them. When there is contact between bighorns and domestic sheep, the bighorns have little defense. This theory is analogous to the accepted explanation for the transmission of human diseases carried to the Native Americans by Europeans. The Native American populations had no immunity to Old World diseases and suffered many documented die-offs.

RECOMMENDATIONS

The DBC Tech Staff has reviewed the bighorn sheep problem and developed recommendations for eliminating domestic and bighorn sheep conflicts on public lands. They consist of 1 general recommendation and 4 specific recommendations dealing with buffer strips, livestock supervision, trailing, and reintroductions. Each recommendation is preceded by a statement of the issue, followed by a justification.

General Recommendation

Issue.—Desert bighorn that come into contact with domestic sheep die as a result of the contact.

Recommendation. – Domestic sheep in the vicinity of desert bighorn ranges should be managed so that desert bighorn never come into contact with domestic sheep nor the disease organisms that domestic sheep carry.

Justification.—Evidence (Table 1) indicates that contact with domestic sheep is almost invariably lethal to desert bighorn. The recommendations that follow deal with methods to minimize interaction, especially physical contact between domestic and bighorn sheep.

34 GUIDELINES

Table I. Bighorn declines and die-offs resulting from contracts with domestic sheep.

Location	Cause of die-off	Results	Year(s)	Source
Sun River, Mont.		≥70 died	1910-35	Goodson (1982)
Upper Rock Ck., Mont.		All died	1965-70s	Goodson (1982)
Thompson Falls, Mont.		All died	1940-60	Goodson (1982)
Kootenay National Park, B.C., Can.	Pneumonia		1939	Goodson (1982)
Bull River, B.C., Can.	Pneumonia	96% died	1965	Bandy (1968) in Goodson (1982)
MacQuire Creek, B.C., Can.	Pneumonia		1981-82	Davidson in Goodson (1982)
Lava Beds National Monument, Calif.ª	Pneumonia	All died	1980	Blaisdell (1982)
Mormon Mts., Nev.	Pneumonia	50% died	1980	Jessup (1981)
Dinosaur National Monument, Colo.		All died	1950	Barmore (1962) in Goodson (1982)
Rock Creek, Mont.		8 left	1900-20	Goodson (1982)
Rocky Mtn. National Park, Colo.	Pneumonia	All died	1917-30	Packard (1939a, 1939b) in Goodson (1982)
Methow Game Range, Wash. ^a	Pneumonia	13 of 14 died	1979-81	Foreyt and Jessup (1982)
Warner Mt., Calif.	Pneumonia	All died	1988	Weaver (1988)
Oregon	Scabies		1936	Lange (1980)
California	Scabies		1870-79, 1898	Jones (1900) in Lange (1980)
Grey Bull River, Wyo.			1881	Honess and Frost (1942) in Lange (1980)
Wyo., Mont.			1885	Hornaday (1901 in Lange (1980)
Colo.	Scabies		1859-31	Packard (1946) in Lange (1980)
Rocky Mtn. National Park, Colo.	Scabies		1878-1903	Lange (1980)
Latir Parks, N.M.	Pneumonia	All died	1978-82	Sandoval (1988)
Utah St. Univ., Utah ^b	Pneumonia	All died	1970s	Spillett in Goodson (1982)
Univ. B.C., Can. ^b	Pneumonia	All died	1970s	Hebert in Goodson (1982)
Colorado St. Univ., Colo. ^b	Pneumonia	All died	1970s	Hibler in Goodson (1982)
Utah St. Univ., Utah ^b	Pneumonia	_4 of 5 died	1988	T. D. Bunch (Utah State Univ., pers. commun.)

^aLarge pen or paddock.

^bUniversity controlled conditions.

Specific Recommendation 1: Buffer Strips

Issue.—Desert bighorn and domestic sheep must be spatially separated to minimize the possibility of these 2 species coming into contact. No domestic sheep grazing should be authorized or allowed within

buffer strips \geq 13.5 km wide surrounding desert bighorn habitat, except where topographic features or other barriers prevent any interaction.

Justification.-Armentrout and Brigham (1988) recommended a 13.5krn-wide separation strip as optimum, based on 9 cited literature sources. Bighorn and domestic sheep separation distances cited in the literature range from 3.2 to 32 km. The California Department of Fish and Game (1983), in its discussion of conflicting land uses, recommended that domestic sheep grazing be eliminated within 3.2 km of bighorn habitat where feasible. The 3.2-km buffer strip also is included in the Mina Habitat Management Plan in Nevada (U.S. Dep. Interior, BLM 1988a) in ≥ 1 land-use plan in the Boise, Idaho BLM District (Goodson 1982); and in the Winnemucca, Nevada BLM 1978 grazing Environmental Impact Statement for the Sonoma-Gerlach Resource Area. A 9.6-kmwide buffer strip was recommended in the Lahontan Resource Management Plan (RMP) and the Stillwater Habitat Management Plan in Nevada (U.S. Dep. Interior, BLM 1985, 1986b). The widest recommended buffer (32 km) was used in Arizona. A 32-km buffer was agreed upon in the original Memorandum of Understanding (MOU) between the BLM and Arizona Game and Fish Department. However, when the master MOU was redrafted in 1976, the section relating to domestic sheep grazing in bighorn habitat was not included (Gallizioli 1980). Situations involving potential bighorn and domestic sheep conflicts in Arizona now are handled on a case-by-case basis.

The reason for the 32-km buffer strip was concern over the chronic frontal sinusitis in desert bighorn. This disease occurs when bot fly (*Oestrous* ovis) larvae enter the sinus cavities of bighorns, grow too large to get out, and die, thus infecting the bighorn (Bunch 1978). Sinus cavities in desert bighorns are much larger than those in domestic sheep. The major unanswered question asked by biologists in the 1970s was "what is the range of the bot fly?" Although the U.S. Department of

Agriculture has investigated this question, there is no definitive answer, as it depends upon variables such as temperature, precipitation, and wind. The 32-km buffer strip, however, was felt to be adequate (Gallizioli 1980).

Another problem when considering buffer strips is that young (3-4 yr old) desert bighorn, especially rams, tend to travel extensively (≤ 64 km). Extensive travel by bighorns increases the potential for nose-to-nose contact with domestic sheep. Nose-to-nose contact and resultant transmission of disease(s) was blamed for the catastrophic loss of penned bighorns at the Lava Beds National Monument, California in 1980 (Blaisdell 1982) and in the total population loss of transplanted bighorns in the Warner Mountains, California, in 1988 (Weaver 1988).

Considering all the evidence presented above and cited in Armentrout and Brigham (1988), the Tech Staff feels that buffer strips of \geq 13.5-km are needed to minimize the potential of disease transmission, including chronic frontal sinusitis, and to avert nose-to-nose contact between wandering bighorns and domestic sheep.

Specific Recommendation 2: Livestock Supervision

Issue.—Domestic sheep must be closely and carefully herded to prevent them from straying into desert bighorn range.

Recommendation. — Domestic sheep that are trailed or grazed outside the 13.5-km buffer, but in the vicinity of desert bighorn ranges, should be closely supervised by competent, capable, and informed herders.

Justification.—There is virtually no practical way to control movements of young bighorns, but control of domestic sheep is possible. The key to minimizing impacts by domestic sheep upon bighorns is very close supervision of domestic bands by herding, both while trailing and grazing. Both the Warner Mountains and Lava Beds bighorn die-offs were attributed to stray domestic sheep. Had domestic sheep herding been more intensive, neither of these catastrophes probably would have occurred.

Sheep herders and their control of domestic sheep bands vary considerably. Many herders come to the United States from other countries, especially South America. Many have never herded sheep before their amval in the U.S. Permittees who graze domestic sheep on public lands should ensure that their herders are competent and capable and that herders understand the potential problems that may be caused by straying domestic sheep.

The Tech Staff recognizes that the **BLM's** grazing regulations may need modification to further implement this recommendation. Existing regulations provide that the authorized officer can require herders. The regulations also could be strengthened to allow impoundment of stray domestic sheep, whenever they are found in occupied bighorn habitat. This recommendation could be partially implemented by directives requiring that BLM area managers, range conservationists, and wildlife biologists meet with the permittees and their herders to explain the importance of close supervision by the herders and what could result if domestic sheep are allowed to stray.

Specific Recommendation 3: Trailing

Issue.—Domestic sheep being trailed near desert bighorn range are likely to transmit diseases to bighorns, especially when ewes are in estrus.

Recommendation.—Domestic sheep should be trucked rather than trailed, when trailing would bring sheep closer than 13.5 km to bighorn range. Trailing should never occur when domestic ewes are in estrus.

Justification.—Many domestic sheep are still trailed between grazing allotments. The Tech Staff recommends that domestic sheep be trucked whenever possible to minimize possible contact with bighorns. Close supervision by herders is essential. The time of trailing also is important. When domestic ewes are in estrus, they will attract bighorn rams from distances > 3.2 km. The Tech Staff recommends, therefore, that domestic sheep not be trailed closer than 13.5 km to occupied bighorn habitat. Domestic sheep also should not be trailed when ewes are in estrus, to reduce potential for bighorn sheep contact. This prescription should be included in BLM grazing regulations as part of the supervision and husbandry requirements.

Specific Recommendation 4: Reintroduction

Issue.—Ranges formerly occupied by domestic sheep can harbor diseases detrimental to desert bighorn.

Recommendation.—Bighorn sheep should not be reintroduced into areas where domestic sheep have grazed during the previous 4 years.

Justification.—Our concern involves bighorn reintroductions into habitatsformerly occupied by domestic sheep. The Tech Staff does not advocate the co-use of bighorn habitat by both bighorn and domestic sheep. Two diseases that could be transmitted to bighorn after domestic sheep have been removed are footrot and soremouth (Jessup 1985, Kistner 1982). Both of these diseases can lie in the soil and, when conditions are right, be transmitted to bighorns. The soremouth virus can remain viable in the soil for 10 to 20 years (Jessup 1985, Lance 1980).

SUMMARY

The DBC Tech Staff herein has identified some of the problems associated with bighorn and domestic sheep interactions, and has recommended procedures that should eliminate or reduce contact between domestic and desert bighorn sheep. These recommendations include: no nose-to-nose contact between bighorn and domestic sheep; a minimum of a 13.5-km-wide buffer strip between ranges used by domestic sheep and bighorns; trucking of domestic sheep in preference to trailing, and no trailing when domestic ewes are in estrus; and no bighorn reintroductions onto areas that have been grazed by domestic sheep during the previous 4 years.

REFERENCES CITED

- Armentrout, D. J. and W. R. Brigham. 1988. Habitat suitability rating system for desert bighorn sheep in the Basin and Range Province. U.S. Dep. Interior, Bur. Land Manage. Tech. Note 384. 18pp.
- Blaisdell, J. A. 1982. Lava beds wrap-up-what did we learn? Desert Bighorn Counc. Trans. 26:32–33.
- Bunch, T. D., S. R. Paul, and H. Crutchen. 1978. Chronic sinusitis in the desert bighorn (Ovis canadensisnelsoni). Desert Bighorn Counc. Trans. 22:16–20.
- California. 1983. A plan for bighorn sheep in California. Calif. Dep. Fish and Game. 11pp.
- Foreyt, W. J. and D. A. Jessup. 1982. Fatal pneumonia of bighorn sheep following association with domestic sheep. J. Wildl. Diseases 18:163–168.
- Gallizioli, S. 1980. Memo from the Arizona Game and Fish Dep. to Clair M. Whitlock, BLM State Director, Arizona. **3pp**.
- Goodson, N. 1982. Effects of domestic sheep grazing on bighorn sheep populations: a review. Northern Sheep and Goat Counc. 3:287–313.
- Jessup, D. A. 1981. Pneumonia in bighorn sheep: effects on populations. Transactions of Cal-Neva Wildlife (Annual meeting of the Western Section of the Wildlife Society and California-Nevada Chapter of the American Fisheries Society). 72–78pp.
- ——. 1985. Diseases of domestic livestock which threaten bighorn sheep populations. Desert Bighorn Counc. Trans. 29:29–33.
- Kistner, T. P. 1982. Letter to Josh Warburton, BLM, Bums, Oregon. 11pp.
- Lance, W. E. 1980. Implications of contagious echthyma in bighorn sheep. Northern Sheep and Goat Counc. 2:16–18.
- Lange, R. E., Jr. 1980. Psoroptic scabies in wildlife in the United States and Canada. Desert Bighorn Counc. Trans. 24:18–20.
- Sandoval, A. V. 1988. Bighorn sheep die-off following association with domestic sheep : case history. Desert Bighorn Counc. 32:36–37.
- U.S. Dep. Interior, BLM. 1985. Lahontan resource management plan, Carson City District, Nevada. 150pp.
- ——. 1988a. Mina habitat management plan, Carson City District, Nevada. 60pp.
- **——**. 1988*b*. Stillwaterhabitat management plan, Carson City District, Nevada. 40pp.
- Wagner, F. H. 1978. Western rangeland: troubled American resource. Trans. North Amer. Wildl. Conf. 43:453–461.
- Weaver, R. A. 1988. Status of bighorn sheep in California, 1987. Desert Bighorn Counc. Trans. 31:20.

TRANSMISSION OF *MANNHEIMIA HAEMOLYTICA* FROM DOMESTIC SHEEP (*OVIS ARIES*) TO BIGHORN SHEEP (*OVIS CANADENSIS*): UNEQUIVOCAL DEMONSTRATION WITH GREEN FLUORESCENT PROTEIN-TAGGED ORGANISMS

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Previous studies demonstrated that bighorn sheep (Ovis canadensis) died of ABSTRACT: pneumonia when commingled with domestic sheep (Ovis aries) but did not conclusively prove that the responsible pathogens were transmitted from domestic to bighorn sheep. The objective of this study was to determine, unambiguously, whether Mannheimia haemolytica can be transmitted from domestic to bighorn sheep when they commingle. Four isolates of M. haemolytica were obtained from the pharynx of two of four domestic sheep and tagged with a plasmid carrying the genes for green fluorescent protein (GFP) and ampicillin resistance (AP^R). Four domestic sheep, colonized with the tagged bacteria, were kept about 10 m apart from four bighorn sheep for 1 mo with no clinical signs of pneumonia observed in the bighorn sheep during that period. The domestic and bighorn sheep were then allowed to have fence-line contact for 2 mo. During that period, three bighorn sheep acquired the tagged bacteria from the domestic sheep. At the end of the 2 mo of fence-line contact, the animals were allowed to commingle. All four bighorn sheep died 2 days to 9 days following commingling. The lungs from all four bighorn sheep showed gross and histopathologic lesions characteristic of M. haemolytica pneumonia. Tagged M. haemolytica were isolated from all four bighorn sheep, as confirmed by growth in ampicillin-containing culture medium, PCR-amplification of genes encoding GFP and Ap^{R} , and immunofluorescent staining of GFP. These results unequivocally demonstrate transmission of *M. haemolytica* from domestic to bighorn sheep, resulting in pneumonia and death of bighorn sheep.

Key words: Bighorn sheep, domestic sheep, green fluorescent protein, Mannheimia haemolytica, Ovis canadensis, pneumonia, transmission.

INTRODUCTION

The large decline in the bighorn sheep (*Ovis canadensis*) population in North America, from an estimated two million at the beginning of the 19th century to fewer than 70,000 now (2009) (Buechner, 1960; Valdez and Krausman, 1999), has been attributed in part to diseases, particularly pneumonia caused by bacteria of the genera *Mannheimia*, *Bibersteinia*, and *Pasteurella* (Coggins, 1988; Miller, 2001). Bighorn sheep are much-more susceptible to pneumonia than are domestic sheep (*Ovis aries*; Foreyt, 1994). Since the early 1980s, there

have been anecdotal field reports of bighorn deaths due to pneumonia following contact with domestic sheep (Foreyt and Jessup, 1982; Coggins, 1988; George et al., 2008).

Bacteria of the genera *Mannheimia*, *Bibersteinia*, and *Pasteurella* are commensal bacteria in the pharynx and nasal cavities of domestic and bighorn sheep (Ward et al., 1990). Experimental inoculation of some of the isolates from domestic sheep—isolates which do not readily cause disease in the domestic sheep—have resulted in fatal pneumonia in bighorn sheep (Onderka et al., 1988; Foreyt et al., 1994). In five experimental

commingling studies conducted by three investigators, 41 of 43 bighorn sheep died following contact with domestic sheep (Onderka and Wishart, 1988; Foreyt, 1989, 1990; Callan et al., 1991). These findings appeared to confirm earlier reports of the death of bighorn sheep after contact with domestic sheep, thus incriminating domestic sheep in the induction of fatal pneumonia in bighorn sheep. Although Mannheimia (Pasteurella) haemolytica, Bibersteinia (Pasteurella) trehalosi, and Pasteurella multocida were isolated from the dead bighorn sheep, these studies did not demonstrate that these organisms were transmitted from the domestic sheep to the bighorn sheep. In some of these studies, the bacteria that were isolated from the dead bighorn sheep were not shown to be present in the domestic sheep. It is possible that the bacteria responsible for the death of the bighorn sheep were not carried by the domestic sheep. It is also conceivable that these bacteria were present in the domestic sheep, but were not isolated, because nasal swabs rather than pharyngeal swabs were obtained or because adequate numbers of bacterial colonies from the initial isolation were not picked up for further characterization. Even the isolation of bacteria belonging to the same species, serotype, or biotype, from the domestic sheep and bighorn sheep did not demonstrate that the organism was transmitted from domestic sheep.

Our objective was to determine, unambiguously, whether a respiratory pathogen can be transmitted from domestic sheep to bighorn sheep. Multiple genera, species, and serotypes of bacteria can colonize the nasal cavities and the pharynx of a single animal (Ward et al., 1997). *Mannheimia haemolytica*, *B. trehalosi*, and *P. multocida* are commonly isolated from pneumonic lungs of bighorn sheep, (Jaworski et al., 1998; Kelley et al., 2007; George et al., 2008). *Mannheimia haemolytica* consistently causes severe bronchopneumonia and the rapid death of bighorn sheep under experimental conditions (Onderka et al., 1988; Foreyt et al., 1994; Dassanayake et al., 2009). Therefore, we selected *M. haemolytica* for this study. We obtained four *M. haemolytica* isolates from the nasopharynx of domestic sheep and tagged them with a plasmid encoding genes for green fluorescent protein (GFP), and for beta-lactamase (Bla), which confers ampicillin resistance (Ap^R). The four domestic sheep were colonized with the tagged bacteria and allowed to commingle with bighorn sheep to determine whether there was transmission of the GFP-tagged bacteria.

MATERIALS AND METHODS

Screening of animals for respiratory pathogens

Experimental protocols were reviewed and approved by the Institutional Animal Care and Use Committee (IACUC) at Washington State University.

Four, clinically normal domestic sheep from the same flock were selected for the study. Nasal and pharyngeal swabs, from two groups of four domestic sheep and four bighorn sheep, were collected twice at 1- to 2-wk intervals. The swabs were collected from the domestic sheep at the beginning of the study (61 wk and 63 wk prior to the beginning of the transmission study) to obtain M. haemolytica isolates for tagging with GFP and Ap^R. The bighorn sheep were sampled 42 days and 35 days prior to the beginning of the transmission study. The swabs were analyzed for the presence of ovine respiratory disease (ORD) pathogens by protocols routinely used at Washington Animal Disease Diagnostic Laboratory (WADDL; Pullman, Washington, USA). The pathogens screened for included the bacteria M. haemolytica, B. trehalosi, and Mycoplasma ovipneumoniae and the viruses respiratory syncytial virus (RSV), parainfluenza 3 virus (PI-3), bovine herpesvirus1 (BHV-1), and bovine viral diarrhea virus (BVDV).

Isolation of viruses from nasopharyngeal swabs and lungs

The bovine turbinate (BT) cell line was used for viral propagation because these cells were known to support the growth of all the above viruses. Swabs in universal viral transport medium (BD Biosciences, Sparks, Maryland, USA) were vortexed, and the medium was plated onto BT cells in minimal essential medium (MEM) supplemented with 10% fetal bovine serum (FBS; free of antibodies to known respiratory viruses) and antibiotics (penicillin-streptomycin 100 IU/ml; gentamicin 50 μ g/ml; and fungizone 25 μ g/ml). Inoculated cell cultures were incubated at 37 C in a humidified atmosphere of 5% CO₂. The BT cells were observed daily for cytopathic effect.

Isolation of *M. ovipneumoniae* and *M. haemolytica* from nasopharyngeal swabs and lungs

Swabs from each animal were streaked onto blood agar plates and kept at 37 C overnight under aerobic and anaerobic growth conditions. The bacterial colony morphology on brain-heart infusion (BHI) sheep blood agar and triple sugar iron (TSI) medium; Gram staining; the ability to hydrolyze arabinose, trehalose, indole, nitrate, xylose, and catalase; and oxidase activity were used to differentiate *M. haemolytica* from *B. trehalosi* and *P. multocida* isolates. *Mycoplasma ovipneumoniae* was isolated by growth on pleuropneumonia-like organism broth and selective agar plates according to a previously described protocol (Besser et al., 2008).

Serotyping of M. haemolytica isolates

Mannheimia haemolytica strains were serotyped using serotype-specific rabbit antisera obtained from Glynn Frank (National Animal Disease Center, Ames, Iowa, USA). Cells from a single colony of overnight growth on a sheep blood agar plate were swirled for 30 sec in 30 μ l of serum on a glass microscope slide. Agglutination was observed under a dissecting microscope. Serotype-specific antisera for the following serotypes were tested: A1, A2, A5, A6, A7, A8, A9, A10, A11, A12, A13, A14, and A16.

Polymerase chain reaction (PCR) detection of *M. haemolytica*

The PCR assay specific for *M. haemolytica* has been described (Dassanayake et al., 2010). A portion of the gene encoding *M. haemolytica* O-sialoglycoprotein endopeptidase (*gcp*; Genbank accession number AY83967) was amplified by PCR using primers MhgcpF: 5'-AGA GGC CAA TCT GCA AAC CTC G-3' and reverse primer MhgcpR: 5'-GTT CGT ATT GCC CAA CGC CG-3'. PCRs were carried out in a final, 50- μ l volume with GoTaq[®] PCR SuperMix (Promega Inc., Madison, Wisconsin, USA) with 0.2 μ M each primer and 2 μ l bacterial culture. The PCR cycling conditions consisted of an initial denaturation at 95 C for

5 min followed by 35 cycles of denaturation at 95 C for 30 sec, annealing at 55 C for 30 sec, and extension at 72 C for 40 sec, and a final elongation at 72 C for 5 min. The PCR products were visualized after electrophoresis in 1.0% agarose gels run at 7.0 V/cm and staining with ethidium bromide.

PCR detection of M. ovipneumoniae

Both standard PCR and real-time PCR (RT-PCR) were used. Standard PCR amplification conditions were essentially the same as previously described (Besser et al., 2008). Real-time PCR was developed in-house at WADDL using the following primers: Movip F: 5'-GGG GTG CGC AAC ATT AGT TA-3'; Movip R: 5'-CTT ACT GCT GCC TCC CGT AG-3'; and Movip (Probe): 5'-6-FAM-TTA GCG GGG CCA AGA GGC TGT A-BHQ-1-3' derived from GenBank sequences EU290066 and NR_ 025989 of M. ovipneumoniae. The RT-PCR was run in an ABI 7500 Fast Thermocycler (Applied Biosystems, Carlsbad, California, USA) with the following cycling parameters: Stage 1: 1 hold at 50 C for 2 min (optics off) 95 C for 600 sec (optics off); Stage 2: 45 repeat cycles of 95 C for 15 sec (optics off) to denature and 61 C for 60 sec for annealing and extension (optics on). Test samples were read on the FAM wavelength. Those with a cycle threshold below 40.0 on the FAM channel were classed as positive for M. ovipneumoniae.

Tagging of *M. haemolytica* isolates with a plasmid carrying the genes encoding GFP and Ap^{R}

Plasmid pAM2425 was constructed by cloning the *gfp* gene from plasmid pAG408 into an M. haemolytica shuttle vector, pAM2355 (Marciel, 2001). Briefly, the ClaI/ *Eco*RI fragment of pAG408 was cloned into a pBluescript KS II+ plasmid carrying the leukotoxin C promoter, then the P_{lktC}::gfp fusion was amplified using M13 universal forward (5'-GTĀ AAA CGĀ CGG CCA GT-3') and modified reverse (5'-GGG ATA TCT AGA AGC TTA ACA GCT ATG ACC ATG ATT ACG-3', HindIII site italicized) primers, and then cloned as a HindIII/XbaI fragment into the Bla-resistant vector pAM2355 to create pAM2425 (Fig. 1). All constructions were performed in Escherichia coli XL1-Blue (Stratagene, La Jolla, California, USA) as described (Fedorova and Highlander, 1997). Plasmid DNA was purified using the Qiagen miniprep kit (Qiagen, Valencia, California, USA), and the four M. haemolytica isolates from the domestic sheep were transformed with plasmid pAM2425, by electroporation, as described by Craig et al. (1989). One-hundred



FIGURE 1. Schematic representation of the plasmid pAM2425 carrying *gfp* and *bla* genes. Plasmid pAM2425 was constructed by cloning the *gfp* gene from plasmid pAG408 into a *Mannheimia haemolytica* shuttle vector pAM2355, as described in materials and methods.

nanograms of plasmid DNA were added to each cuvette, which contained 100 µl electrocompetent cells. An electrical pulse of 15-20 kilovolt, 400 ohm, 25 µfarad was applied and, immediately, 1 ml BHI/SOC medium (BHI broth; 2.5 mM KCl; 10 mM MgSO₄; 10 mM MgCl₂; 20 mM glucose) was added and the mixture was incubated at 37 C for 3-4 hr to allow expression of markers. Onehundred-microliter aliquots were spread onto sheep blood agar plates containing 20 µg/ml ampicillin (Bioline, Randolph, Massachusetts, USA) and plates were incubated overnight at 37 C. Ampicillin-resistant colonies containing pAM2425 were identified by colony PCR using *gfp* and *bla* gene-specific primers, respectively (gfp forward 5'-ATG AGT AAA GGA GAA GAA CT-3' and reverse 5'-GTA TAG TTC ATC CAT GCC ATG-3' and bla forward 5'-ATG TTA AAT AAG TTA AAA ATC-3' and reverse 5'-TTA GTT GAG CTG TAA AGT ATG AAA TAC-3'), in a 25-µl mastermix reaction containing GoTaq, as directed by the manufacturer (Promega Corp.) with slight modification. The PCR cycling conditions consisted of an initial denaturation at 95 C for 5 min, followed by 30 cycles of denaturation at 94 C for 30 sec, annealing at 55 C for 30 sec, extension at 72 C for 1 min, and a final elongation at 72 C for 10 min.

Leukotoxin production by *M. haemolytica* isolates before and after tagging with GFP and Ap^{R}

Leukotoxin production by the *M. haemolytica* isolates was confirmed by subjecting culture supernatant fluid to MTT dye reduction cytotoxicity assay as described by Gentry and Srikumaran (1991). The percent cytotoxicity was calculated as follows: % cytotoxicity = [1-(OD of toxin-treated cells/OD of toxinuntreated cells)]×100.

Colonization of domestic sheep with tagged *M. haemolytica*

Bacteria were cultured overnight at 37 C in BHI agar supplemented with 5% sheep blood (Remel, Lenexa, Kansas, USA). Tagged M. *haemolytica* was cultured on plates containing BHI supplemented with 20 µg/ml ampicillin (Bioline). To prepare the inoculum, the bacteria were cultured in BHI broth at 37 C for 2-3 hr followed by growth in Roswell Park Memorial Institute (RPMI) 1640 medium, without phenol red (GIBCO), under the same conditions. The bacterial suspension was diluted in RPMI 1640 to obtain the desired concentration (colony-forming units [CFU]/ ml; Petras et al., 1995). Using an atomizer, about 10⁹ CFU of tagged *M. haemolytica* in 5 ml of phosphate-buffered saline (PBS) were sprayed intranasally into all four domestic sheep from which they were originally isolated. Nasal and pharyngeal swabs were collected 2 wk following inoculation to confirm the presence of tagged bacteria by colony PCR, as described above. A serotype-2 strain of *M. haemolytica*, isolated several years ago from a domestic sheep (Foreyt et al., 1994), also was tagged with the plasmid carrying the *gfp* and *bla* genes. This strain failed to colonize the pharynx of the four domestic sheep and was not used further.

Domestic sheep-bighorn sheep contact experiments

On day 0, the four domestic sheep and the four bighorn sheep were placed in two identical pens (about 20×3 m) separated by another pen $(20 \times 10 \text{ m})$, and animals were monitored for clinical signs. After 1 mo, the bighorn sheep were moved into the middle pen so that they had fence-line contact with domestic sheep. For the next 2 mo, the animals were observed for clinical signs of pneumonia, and nasal and pharyngeal swabs were collected twice (days 51 and 60) for detection of the presence of tagged *M. haemolytica.* After 2 mo in fence-line contact, the domestic sheep and bighorn sheep were allowed to commingle in the middle pen (20×10 m).

Clinical assessment and necropsy

The bighorn sheep were observed once a day for clinical signs including anorexia,

Bacteria recovered, sample $1/sample 2^b$ Animal Sample Mh Bt^d Paste Movi^f site no. +/-1 Р +/+^g -/-+/+ -/--/-Ν -/-+/-2 Р -/+ -/-+/-+/+ Ν +/+ -/--/--/-3 Р -/+ -/-+/-+/-Ν -/+ -/--/--/-5Р +/-+/--/-+/-_/_ +/-N -/--/-

TABLE 1. Microbial profile of the nasopharynx of domestic sheep before commingling.

^a Site of sample collection: P = pharynx; N = nasal cavity.
 ^b Sample 1/sample 2 = Swabs collected at two different dates.

^c Mh = Mannheimia haemolytica.

^d Bt = *Bibersteinia trehalosi*.

^e Past = Pasteurella species.

^f Movi = Mycoplasma ovipneumoniae.

 $^{g}(-) = Absent or not detected; (+) = present.$

lethargy, cough, dyspnea, and nasal discharge. When the animals began to show clinical signs of pneumonia, they were observed more frequently. Animals that died during the experiment were necropsied within 6 hr. Lungs were removed from each animal and carefully examined for lesions of pneumonia. The degree of involvement of the lung lobes was estimated as percent pneumonic scores (percent of lung that appeared pneumonic on visual examination). Pleuritis was noted as present or absent. Representative samples of pneumonic and normal lung tissue were prepared for both bacteriologic and histopathologic examination (Odugbo et al., 2004). Animals that showed severe signs of pneumonia were euthanized by intravenous administration of pentobarbital and then necropsied in the same manner as those found dead.

Detection of tagged M. haemolytica

Colony PCR: Swabs were directly streaked onto sheep blood agar plates containing $20 \mu g/$ ml ampicillin and the plates were incubated overnight at 37 C. The following day, 5–10 representative colonies from each plate were picked and subjected to colony PCR assay, performed as described above, to confirm the presence of *gfp* and *bla* genes.

Imunoflurorescence labeling of GFP-tagged M. haemolytica: To detect GFP by immunofluorescence, bacterial cells were fixed in 2% paraformaldehyde for 10 min, washed with PBS, and incubated with 100 μ l of FITCconjugated rabbit polyclonal antibodies specific for GFP (Abcam, Cambridge, Massachusetts, USA) for 30 min at 4 C. The cells were washed with PBS and mounted onto microscopic slides and visualized using a fluorescence microscope.

RESULTS

Microbial flora of the upper respiratory tract before commingling

Microbial isolation revealed that all four domestic sheep carried *Pasteurellaceae* in the nasopharynx (Table 1). All four also yielded *M. haemolytica* from nasopharyngeal samples, at least once, prior to commingling (Table 1). All four domestic sheep were culture-positive for *M. ovipneumoniae* but were negative for the respiratory viruses RSV, PI-3, BVDV, and BHV-1.

Prior to beginning the study, the four bighorn sheep were negative for viruses and for *M. ovipneumoniae* by culture (Table 2). However, three of the bighorn sheep yielded *M. haemolytica* from nasopharyngeal swabs and all four had *B. trehalosi* in their pharynx (Table 2).

Characteristics of the *M. haemolytica* isolates from domestic sheep selected for tagging

Four *M. haemolytica* isolates obtained from two of the domestic sheep were designated as numbers 7, 10, 15, and 16. These isolates were determined to be *M. haemolytica* by cultural and biochemical characteristics and were confirmed by *M. haemolytica*-specific PCR assays. Serotype analysis with antisera specific for all known serotypes (A1, A2, A5, A6, A7, A8, A9, A10, A11, A12, A13, A14, and A16) revealed that isolate 7 belonged to serotype 9, while the other three were untypable. All of these isolates produced leukotoxin in culture (Fig. 2).

Mannheimia haemolytica isolates from domestic sheep get tagged with the plasmid carrying the *gfp* and *bla* genes

Growth of tagged *M. haemolytica* isolates on ampicillin plates suggested that
	Sample	Bacteria recovered before commingling (sample 1/sample 2) ^b			Sample	Bacteria recovered after commingling (postmortem)			
Animal	site ^a	$\mathrm{Mh^{c}}$	Bt^d	Movi ^e	site	Mh	Bt	Movi (culture)	Movi (PCR)
Y13	Р	$-/-^{\mathrm{f}}$	+/+	_/_	Р	+	_	_	_
	Ν	_/_	_/_	_/_	Ν	+	+	_	_
					L	+	+	_	_
Y15	Р	—/+	+/	_/_	Р	_	+	+	+
	Ν	+/+	_/_	_/_	Ν	+	_	_	_
					L	+	_	—	_
Y16	Р	+/	+/+	_/_	Р	_	+	_	_
	Ν	_/_	_/_	_/_	Ν	+	+	_	_
					L	+	+	_	_
Y47	Р	_/_	+/+	_/_	Р	nd^{g}	nd	nd	_
	Ν	—/+	_/_	_/_	Ν	nd	nd	nd	_
					L	+	_	_	+

TABLE 2. Microbial profile of the nasopharynx of bighorn sheep before and after their commingling with domestic sheep. Bacteria were recovered via culture, except that *Mycoplasma ovipneumoniae* was also detected postmortem using polymerase chain reaction (PCR) assay.

 a Site of sample collection: P = pharynx; N = nasal cavity; L = lung.

 $^{\rm b}$ Sample 1/sample 2 = Swabs collected on two different dates.

^c Mh = Mannheimia haemolytica.

^d Bt = *Bibersteinia trehalosi*.

^e Movi = Mycoplasma ovipneumoniae.

f(-) = Absent or not detected; (+) = present.

 g nd = not done.

the bacteria were successfully tagged with GFP and Ap^{R} . PCR using *gfp*- and *bla*specific primers confirmed the presence of gfp (Fig. 3A) and bla (Fig. 3C) in all four isolates. Immunofluorescence assays using FITC-labeled anti-GFP antibodies further confirmed the expression of GFP in these isolates (Fig. 4A). Cytotoxicity assays of the culture supernatant fluid, before and after the tagging, revealed that the leukotoxin production was not affected by the presence of extrachromosomal plasmid (Fig. 2). In a separate experiment, two bighorn sheep inoculated intratracheally with 5×10^9 CFU of the *M. haemoly*tica isolates tagged with GFP/Ap^R plasmid developed pneumonia and died within 2 days postinoculation, indicating that organisms tagged with the GFP/Ap^R plasmid were pathogenic.

GFP- and Ap^R-tagged *M. haemolytica* effectively colonize the nasopharynx of domestic sheep

Three inoculations using a cocktail of all four, tagged *M. haemolytica* isolates re-

sulted in colonization of the nasopharynx of three of the four domestic sheep. The colonization was detected by analyzing nasal and pharyngeal swabs for two consecutive weeks postinoculation (data not shown). The PCR amplification of gfp and bla genes confirmed the presence of the plasmid-tagged *M. haemolytica* in all of the three domestic sheep. All of the four domestic sheep continued to remain clinically normal after inoculation with tagged *M. haemolytica*.

Domestic sheep transmit GFP- and Ap^R -tagged *M. haemolytica* to bighorn sheep

The domestic sheep and bighorn sheep were separated by about 10 m in individual pens during the first month. During that time, no symptoms of respiratory disease were observed in either domestic sheep or bighorn sheep. Three bighorn sheep (Y13, Y15, and Y47) yielded tagged *M. haemolytica* from samples collected on days 51, 60, or both (21 days, 30 days, or both after fence-line contact began), as



FIGURE 2. Leukotoxin production by Mannheimia haemolytica isolates before and after tagging with the plasmid carrying gfp and bla. Culture supernatant fluids from the *M. haemolytica* isolates numbers 7, 10, 15, and 16, before and after tagging with the plasmid carrying gfp and bla, were subjected to the MTT-dye reduction cytotoxicity assay. The percent cytotoxicity was calculated as follows: % cytotoxicity = $[1-(OD \text{ of toxin-treated cells/OD of$ $toxin-untreated cells}] \times 100$. The open and shaded bars represent % cytotoxicity of culture supernatant fluids from the respective isolates, before and after tagging, respectively. Results shown are the means of three independent experiments. The error bars indicate standard deviations of the means.

revealed by gfp and bla gene-specific PCR. One of these bighorn sheep (Y15) developed coughing on day 83, 32 days following the first evidence of tagged *M. haemolytica* infection, but none of the animals died. On day 92 (2 days postcommingling), one bighorn sheep (Y15) died. The remaining animals at this time were lethargic and showed intermittent coughing. On day 95 (5 days postcommingling), two more bighorn sheep (Y13 and Y16) died, and on day 99 (9 days postcommingling), the remaining bighorn sheep (Y47) exhibited severe clinical signs of pneumonia and was euthanized.

Induction of pneumonia in, and death of, bighorn sheep are caused by *M. haemolytica* transmitted by the domestic sheep

Postmortem examinations revealed that all four bighorn sheep had acute, bilateral, fibrinohemorrhagic pneumonia that was equally distributed on both sides (Fig. 5A).



FIGURE 3. Detection of gfp and bla in *Mannheimia haemolytica* isolates by polymerase chain reaction (PCR) amplification. The *M. haemolytica* isolates tagged with the plasmid carrying gfp and bla, and the *M. haemolytica* isolates recovered from the lungs of the four dead bighorn sheep, were tested for the presence of gfp and bla by PCR analysis using primers described under materials and methods. Panels A and B represent PCR amplification of gfp. Panels C and D represent PCR amplification of bla. Ut=the untagged *M. haemolytica* (pool of all 4 isolates); Pl=plasmid pAM2425 used as positive control in PCR to indicate the presence of gfp and bla; numbers 16, 15, 10, and 7 represent the tagged isolates and the numbers Y47, Y16, Y15, and Y13 represent *M. haemolytica* isolated from the lungs of bighorn sheep numbers Y47, Y16, Y15, and Y13 at necropsy. MW=molecular weight markers. Results of one representative experiment out of three are shown.



FIGURE 4. Detection of expression of GFP by immunofluorescence staining. The Mannheimia haemolytica isolates tagged with the plasmid carrying gfp and bla, and the M. haemolytica isolates recovered from the lungs of the four dead bighorn sheep, were tested for the expression of GFP by immunofluorescence staining with FITC-conjugated rabbit anti-GFP antibodies. All four tagged isolates (7, 10, 15, and 16), and isolates recovered from the lungs of all four dead bighorn sheep (Y13, Y15, Y16, and Y47), were positive for fluorescence expression. Fluorescence exhibited by one representative tagged isolate (Panel A1), and one representative isolate recovered from the lungs of the dead bighorn sheep (Panel B1), are shown. Panel A2 and B2 represent untagged M. haemolytica used as the negative control.

Estimated percent pneumonic involvement ranged from 70-95% in both the lungs. Fibrinous pleuritis was present in all four bighorn sheep. Although the lungs from the different bighorn sheep varied in severity in gross lesions, they were histologically very similar. In affected areas of the lungs, alveolar spaces and bronchioles were filled with edema, fibrin, red blood cells, and dense collections of primarily macrophages and neutrophils (Fig. 5B). The inflammatory cells showed degenerative changes and often had streaming nuclei ('oat cells'). Many alveolar walls, and occasional bronchiolar walls, were disrupted by necrosis and hemorrhage. When present, pleuritis was fibrinous.

Re-isolation of tagged *M. haemolytica* from pneumonic lungs of bighorn sheep

The swabs taken from lungs during necropsy were plated on BHI-agar plates which, upon incubation, showed the presence of colonies resistant to 20 µg/ml ampicillin. Further gfp gene- and bla gene-specific PCR confirmed the presence of tagged bacteria in the lungs (Fig. 3B, D). Immunofluorescence assays using FITC-labeled antiGFP antibodies further confirmed the expression of GFP in these isolates (Fig. 4B). None of the tagged isolates recovered from the lungs were typable with the antisera specific for the known serotypes of M. haemolytica (A1, A2, A5, A6, A7, A8, A9, A10, A11, A12, A13, A14, and A16).

DISCUSSION

Several anecdotal reports suggest that bighorn sheep die from pneumonia following contact with domestic sheep (Foreyt and Jessup, 1982; Coggins, 1988; George et al., 2008). Fatal pneumonia in bighorn sheep following experimental inoculation of *M. haemolytica* isolates from domestic sheep, isolates which did not cause disease in the domestic sheep, prompted researchers to perform commingling experiments to determine whether there was transmission of respiratory pathogens from domestic sheep to bighorn sheep (Onderka and Wishart, 1988; Foreyt, 1989, 1990; Callan et al., 1991). Although over 95% of the bighorn sheep in these studies died following contact with domestic sheep, there was not clear documentation of transmission of *M. haemolytica*, or of any other pathogen, from domestic sheep to bighorn sheep.

Whole genome sequencing, pulsed field gel electrophoresis, or amplified fragment length polymorphism, ribotyping, multilocus enzyme electrophoresis, and multilocus sequence typing are molecular tools that are available to compare bacterial pathogens isolated from domestic sheep and bighorn sheep. Whole genome se-



FIGURE 5. Representative gross lesions and histopathology of the lungs of the dead bighorn sheep. (A) Typical gross appearance of the lungs of the dead bighorn sheep. The lungs were removed from the carcass for examination, and the total area of gross lung consolidation was discerned by visual inspection and by palpation. In this case, the right cranial and middle, and the left middle lung lobes, are dark red and consolidated, and additional consolidation was evident from palpation; darkened areas in the photograph were subsequently determined to be areas of severe hemorrhage. Fibrin strands on the lung surface indicate pleuritis. (B) The typical histopathologic appearance of the lungs of the dead bighorn sheep. Lung tissue samples of bighorn sheep were aseptically removed and processed for histopathology. Alveolar septa are necrotic and replaced by fibrin and debris. Bronchioles and alveoli are filled with streaming mononuclear cells. H&E stain. $100 \times$.

quencing is an elaborate and expensive procedure. The other molecular methods are time-consuming and cannot identify bacterial isolates with 100% certainty (Pitt, 1999; Yakubu et al., 1999). We reasoned that tagging the bacterial isolates obtained from domestic sheep, recolonizing the nasopharynx of these animals with the tagged bacteria, and commingling them with bighorn sheep would circumvent these problems and provide an irrefutable method of determining whether bacterial pathogens can be transmitted from domestic sheep to bighorn sheep. We selected *M. haemolytica* for this study because of its documented ability to consistently induce pneumonia in, and death of, bighorn sheep (Onderka et al., 1988; Foreyt et al., 1994; Dassanayake et al., 2009). We employed two markers, the GFP and Ap^{R} , to enhance the validity of our findings. We also utilized two tests to detect each marker (PCR and immunofluorescence for GFP and growth on ampicillin-containing medium and PCR

for Ap^{R}). The growth of the tagged *M.* haemolytica in the presence of ampicillin, the PCR amplification of the genes gfpand bla, and the immunofluorescence staining with anti-GFP antibodies clearly indicated that the four isolates of *M.* haemolytica obtained from the domestic sheep were tagged with the markers (Fig. 3A, C, 4A). These three parameters were used to clearly document the successful colonization of the pharynx of domestic sheep by the tagged *M.* haemolytica and, more importantly, to identify the tagged organisms isolated from the dead bighorn sheep (Fig. 3B, D, 4B).

Tagged-isolate 7 typed as serotype 9 while the other three (numbers 10, 15, and 16) were untypable. However, none of the isolates recovered from the lungs of the four dead bighorn sheep typed as serotype 9. This could be because the tagged-isolate 7 did not colonize the nasopharynx of domestic sheep; because it colonized the domestic sheep but was not shed in adequate amounts to be acquired by the bighorn sheep; or because it was acquired by the bighorn sheep but not recovered by us because it was present in the lungs in lower numbers than the other isolates at the time of sampling. Nevertheless, transmission from domestic sheep to bighorn sheep clearly occurred because other tagged isolates of *M. haemolytica* were recovered from the lungs of every bighorn sheep.

Our finding that three out of the four bighorn sheep acquired the tagged M. haemolytica within 1 mo of fence-line contact indicates that such contact was adequate for transmission of these organisms to occur. Death of the first bighorn sheep occurred about 1 mo after tagged M. haemolytica was first detected in that animal. This lag period may have been necessary for the transmitted M. haemo*lytica* to colonize and proliferate to the threshold number of organisms required to induce pneumonia and death in bighorn sheep. It is conceivable that the bighorn sheep that acquired the tagged M. hae*molytica* during the fence-line contact would have died even without commingling with the domestic sheep. This notion is supported by the fact that one bighorn died only 2 days after commingling with the domestic sheep. However, in order to determine with certainty whether fenceline contact is adequate for induction of pneumonia and death of bighorn sheep, the experiment would need to be performed with a longer period of fence-line contact.

It is also possible that another pathogen(s) was necessary to predispose the bighorn sheep to pneumonia by *M*. *haemolytica* infection. The bighorn sheep were not positive for *M. ovipneumoniae* before commingling with the domestic sheep. Lung tissue from one of the dead bighorn sheep was positive for *M. ovipneumoniae* by standard and RT-PCR (Table 2), and *M. ovipneumoniae* was detected in the nasopharynx of a second dead bighorn sheep by culture and PCR, which raises the possibility that these

organisms, along with the tagged M. haemolytica, were transmitted from the domestic sheep to the bighorn sheep. It is possible that during the lag period, M. ovipneumoniae colonized the upper respiratory tract of at least two bighorn sheep and predisposed them to the tagged M. haemolytica, but whether M. ovipneumoniae played any role in the other two bighorn sheep seems even less certain, based on available data (Table 2). In domestic sheep, M. ovipneumoniae has been shown to render the cilia on the epithelial cells of the upper respiratory tract dysfunctional (Jones et al., 1985; Niang et al., 1998). Previous studies have shown that *M. ovipneumoniae* does not kill bighorn sheep (Besser et al., 2008) but can predispose them to *M. haemolytica* infection (Dassanayake et al., 2010). However, it is not likely that *M. ovipneumoniae* is a necessary predisposing factor for fatal infection of bighorn sheep by every strain of *M. haemolytica* because, in an earlier study, intranasal inoculation with M. haemolytica resulted in the death of 75% of inoculated bighorn sheep (n=4) within 48 hr (unpubl. data). The *M. haemolytica* used in that study was a serotype 2 strain, which is known to be virulent in bighorn sheep (Foreyt et al., 1994). Therefore, we believe that only less-virulent strains of M. haemolytica may require M. ovipneumoniae or another predisposing agent. Studies are currently underway to elucidate the role of M. ovipneumoniae in the development of pneumonia in bighorn sheep following contact with domestic sheep. In summary, this study irrefutably demonstrated the transmission of *M. haemolytica* from domestic sheep to bighorn sheep and the resulting pneumonia and death of bighorn sheep.

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LITERATURE CITED

- BESSER, T. E., E. F. CASSIRER, K. A. POTTER, J. VANDERSCHALLE, A. FISCHER, D. P. KNOWLES, D. R. HERNDON, F. R. RURANGIRWA, G. C. WEISER, AND S. SRIKUMARAN. 2008. Association of Mycoplasma ovipneumoniae infection with population-limiting respiratory disease in free-ranging Rocky Mountain bighorn sheep (Ovis canadensis canadensis). Journal of Clinical Microbiology 46: 423–430.
- BUECHNER, H. K. 1960. The bighorn sheep in the United States, its past, present, and future. Wildlife Monograph 4: 3–174.
- CALLAN, R. J., T. D. BUNCH, G. W. WORKMAN, AND R. E. MOCK. 1991. Development of pneumonia in desert bighorn sheep after exposure to a flock of exotic wild and domestic sheep. Journal of the American Veterinary Medical Association 198: 1052–1056.
- CoccINS, V. L. 1988. The Lostine Rocky Mountain bighorn sheep die-offs and domestic sheep. Proceedings of the Biennial Symposium of Northern Sheep and Goat Council 6: 57–64.
- CRAIG, F. F., J. G. COOTE, R. PARTON, J. H. FREER, AND N. J. GILMOUR. 1989. A plasmid which can be transferred between *Escherichia coli* and *Pasteurella haemolytica* by electroporation and conjugation. Journal of General Microbiology 135: 2885–2890.
- DASSANAYAKE, R. P., S. SHANTHALINGAM, C. N. HERNDON, P. K. LAWRENCE, E. F. CASSIRER, K. A. POTTER, W. J. FOREYT, K. D. CLINKENBEARD, AND S. SRIKUMARAN. 2009. Mannheimia haemolytica serotype A1 exhibits differential pathogenicity in two related species, Ovis canadensis and Ovis aries. Veterinary Microbiology 133: 366–371.
 - —, D. R. CALL, A. A. SAWANT, N. C. CASAVANT, G. C. WEISER, D. P. KNOWLES, AND S. SRIKU-MARAN. 2010. *Bibersteinia trehalosi* inhibits growth of *Mannheimia haemolytica* by a proximity-dependent mechanism. Applied and Environmental Microbiology 76: 1008–1013.
 - —, SHANTHALINGAM, S., C. N. HERNDON, R. SUBRAMANIAM, P. K. LAWRENCE, J. BAVANANTHASI-VAM, E. F. CASSIRER, G. J. HALDORSON, W. J. FOREYT, F. R. RURANGIRWA, D. P. KNOWLES, T. E. BESSER, AND S. SRIKUMARAN. 2010. Mycoplasma ovipneumoniae can predispose bighorn sheep to fatal Mannheimia haemolytica pneumonia. Veterinary Microbiology (Epub ahead of print). doi: 10.1016/J.vetmic.2010.04.011.
- FEDOROVA, N. D., AND S. K. HIGHLANDER. 1997. Plasmids for heterologous expression in *Pasteurella haemolytica*. Infection and Immunity 186: 207–211.

- FOREYT, W. J. 1989. Fatal Pasteurella haemolytica pneumonia in bighorn sheep after direct contact with clinically normal domestic sheep. American Journal of Veterinary Research 50: 341–344.
 - —. 1990. Pneumonia in bighorn sheep: Effects of *Pasteurella haemolytica* from domestic sheep and effects on survival and long term reproduction. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 7: 92–101.
 - . 1994. Effects of controlled contact exposure between healthy bighorn sheep and llamas, domestic goats, mountain goats, cattle, domestic sheep, or mouflon sheep. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 9: 7–14.
 - —, AND D. A. JESSUP. 1982. Fatal pneumonia of bighorn sheep following association with domestic sheep. Journal of Wildlife Diseases 18: 163– 168.
- —, K. P. SNIPES, AND R. W. KASTEN. 1994. Fatal pneumonia following inoculation of healthy bighorn sheep with *Pasteurella haemolytica* from healthy domestic sheep. Journal of Wildlife Diseases 30: 137–145.
- GENTRY, M. J., AND S. SRIKUMARAN. 1991. Neutralizing monoclonal antibodies to *Pasteurella haemolytica* leukotoxin affinity-purify the toxin from crude culture supernatants. Microbial Pathogenesis 10: 411–417.
- GEORGE, J. L., D. J. MARTIN, P. M. LUKACS, AND M. W. MILLER. 2008. Epidemic Pasteurellosis in a bighorn sheep population coinciding with the appearance of a domestic sheep. Journal of Wildlife Diseases 44: 388–403.
- JAWORSKI, M. D., D. L. HUNTER, AND A. C. S. WARD. 1998. Biovariants of isolates of *Pasteurella* from domestic and wild ruminants. Journal of Veterinary Diagnostic Investigations 10: 49–55.
- JONES, G. E., W. A. KEIR, AND J. S. GILMOUR. 1985. The pathogenicity of *Mycoplasma ovipneumoniae* and *Mycoplasma arginini* in ovine and caprine tracheal organ cultures. Journal of Comparative Pathology 95: 477–487.
- KELLEY, S. T., E. F. CASSIRER, G. C. WEISER, AND S. SAFAEE. 2007. Phylogenetic diversity of Pasteurellaceae and horizontal gene transfer of leukotoxin in wild and domestic sheep. Infection, Genetics and Evolution 7: 13–23.
- MARCIEL, A. M. 2001. Genetic tools for studying the transcription of *Mannheimia (Pasteurella) haemolytica*: Identification of cis-, trans-, and environmental factors modulating leukotoxin expression. PhD Thesis, Baylor College of Medicine, Houston, Texas, pp. 129–132.
- MILLER, M. W. 2001. Pasteurellosis. In Infectious diseases of wild mammals, 3rd Edition, E. S. Williams and I. K. Barker (eds.). Iowa State University Press, Ames, Iowa, pp. 330–339.
- NIANG, M., R. F. ROSENBUSCH, M. C. DEBEY, Y. NIYO,

J. J. ANDREWS, AND M. L. KAEBERLE. 1998. Field isolates of *Mycoplasma ovipneumoniae* exhibit distinct cytopathic effects in ovine tracheal organ cultures. Zentralbl Veterinary Medicine A 45: 29–40.

- Odugbo, M. O., L. E. Odama, J. U. UMOH, and L. H. LOMBIN. 2004. The comparative pathogenicity of strains of eight serovars and untypable strains of *Mannheimia haemolytica* in experimental pneumonia of sheep. Veterinary Research 35: 661– 669.
- ONDERKA, D. K., AND W. D. WISHART. 1988. Experimental contact transmission of *Pasteurella* haemolytica from clinically normal domestic sheep causing pneumonia in Rocky Mountain bighorn sheep. Journal of Wildlife Diseases 24: 663–667.
 - —, S. A. RAWLUK, AND W. D. WISHART. 1988. Susceptibility of Rocky Mountain bighorn sheep and domestic sheep to pneumonia induced by bighorn and domestic livestock strains of *Pasteurella haemolytica*. Canadian Journal of Veterinary Research 52: 439–444.
- PETRAS, S. F., M. CHIDAMBARAM, E. F. ILLYES, S. FROSHAUER, G. M. WEINSTOCK, AND C. P. REESE. 1995. Antigenic and virulence properties of

Pasteurella haemolytica leukotoxin mutants. Infection and Immunity 63: 1033–1039.

- PITT, T. L. 1999. Molecular typing in practice. Journal of Hospital Infection 43: S85–S88.
- VALDEZ, R., AND P. R. KRAUSMAN. 1999. Mountain sheep of North America. University of Arizona Press, Tucson, Arizona, pp. 19–20.
- WARD, A. C., M. R. DUNBAR, D. L. HUNTER, R. H. HILLMAN, M. S. BULGIN, W. J. DELONG, AND E. R. SILVA. 1990. Pasteurellaceae from bighorn and domestic sheep. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 7: 109–117.
- , D. L. HUNTER, M. D. JAWORSKI, P. J. BENOLKIN, M. P. DOBEL, J. B. JEFFRESS, AND G. A. TANNER. 1997. *Pasteurella* spp. in sympatric bighorn and domestic sheep. Journal of Wildlife Diseases 33: 544–547.
- YAKUBU, D. E., J. R. FARIBORZ, R. ABADI, AND T. H. PENNINGTON. 1999. Molecular typing methods for *Neisseria meningitidis*. Journal of Medical Microbiology 48: 1055–1064.
- Submitted for publication 19 October 2009. Accepted 16 February 2010.



United States Department of the Interior

Pacific Southwest Region FISH AND WILDLIFE SERVICE Nevada Fish and Wildlife Office 1340 Financial Blvd., Suite 234 Reno, Nevada 89502 Ph: (775) 861-6300 ~ Fax: (775) 861-6301



June 17, 2014

Board of Supervisors Mono County P. O. Box 715 Bridgeport, California 93517

Dear Mono County Board of Supervisors;

Subject:

Mono County Draft Grant Deed of Conservation Easement Conway and Mattly Ranches

We have reviewed the draft Grant Deed of Conservation Easement Conway and Mattly Ranches and associated documents. We commend Mono County's efforts to protect the natural, scenic, open space, habitat, and historic values (referred to as "Conservation Values") present on the Conway and Mattly Ranches (collectively referred to in the associated documents as "Conway Ranch"). We also support the development of a conservation easement between Mono County and the Eastern Sierra Land Trust to preserve, protect, and maintain these Conservation Values. However, we do have concerns regarding the activities and uses, operations, and management of these properties and potential effects to the federally endangered Sierra Nevada bighorn sheep (*Ovis canadensis sierrae;* Sierra bighorn), and the federally proposed Bi-State Distinct Population Segment of the Greater sage-grouse (*Centrocercus urophasianus*; sage-grouse) and its proposed critical habitat.

The U.S. Fish and Wildlife Service's (Service) responsibilities include administering the Endangered Species Act of 1973, as amended (Act), including sections 7, 9, and 10. Section 9 of the Act prohibits the taking of any federally listed endangered or threatened species. Section 3(19) of the Act defines take to mean to harass, harm, pursue, hunt, shoot, wound, kill, trap, capture, or collect, or to attempt to engage in any such conduct. Service regulations (50 CFR 17.3) define harm to include significant habitat modification or degradation which actually kills

Board of Supervisors, Mono County

or injures wildlife by significantly impairing essential behavioral patterns, including breeding, feeding, or sheltering. Harassment is defined by the Service as an intentional or negligent action that creates the likelihood of injury to wildlife by annoying it to such an extent as to significantly disrupt normal behavioral patterns which include, but are not limited to, breeding, feeding, or sheltering. The Act provides for civil and criminal penalties for the unlawful taking of listed species.

Exemptions to the prohibitions against take may be obtained through coordination with the Service in two ways. If the subject project is to be funded, authorized, or carried out by a Federal agency and may affect a listed species, the Federal agency must consult with the Service, pursuant to section 7(a)(2) of the Act. If a proposed project does not involve a Federal agency but may result in the take of a listed animal species, the project proponent should apply for an incidental take permit, pursuant to section 10(a)(1)(B) of the Act. At present, we believe that several activities proposed or already occurring on these properties have the potential to result in take of federally listed or proposed species. We offer these comments to call your attention to these activities, and the need for further discussion with our agency.

Domestic Sheep Grazing and Sierra Bighorn

Domestic sheep can transfer bacteria that cause pneumonia to bighorn sheep (Lawrence et al. 2010). The 2007 Sierra Nevada Bighorn Sheep Recovery Plan identifies disease transmission and associated die-offs as a potentially significant source of mortality for this species (Service 2007). Additionally, disease can affect bighorn sheep populations well-beyond the initial mortality event. Disease may persist in a population and cause high mortality of lambs, which results in low recruitment and limits population recovery (Cassirer et al. 2013). In recent years, between 2009 and 2010, a bighorn sheep die-off occurred in nine herds located across five western states and resulted in a mortality rate of between 5 and 95 percent, depending upon the herd (WAFWA 2010). Three of the nine herds had observed or suspected association with domestic sheep or goats and the other herds occupied range near allotments where domestic sheep to Sierra bighorn could result in the take of a listed species and also negatively affect its recovery.

In 2010, representatives from the Service, California Department of Fish and Wildlife (CDFW), and members of the Mono County Board of Supervisors met to discuss the Service's and CDFW's concerns regarding domestic sheep grazing on Conway Ranch and the risk of disease transmission to Sierra bighorn. We have attached the letter that was sent to Mr. Dan Lyster following this meeting (Attachment 1). As mentioned in the attached letter, we discussed findings from a disease risk assessment model that was developed by a subgroup of the Sierra bighorn recovery team to assess the disease risk associated with domestic sheep grazing allotments in the eastern Sierra Nevada. While the model specifically addresses domestic sheep grazing allotments on federal land, it can also be used to inform decisions about domestic sheep grazing on private property. The results of this model indicate that both the Conway and Mattly

Board of Supervisors, Mono County

Ranch properties are located within a predicted area of potential contact (Croft et al. 2009; Attachment 2). This predicted area of potential contact indicates that there is a high risk of Sierra bighorn coming into contact with domestic sheep.

It is unclear in the draft Conway Ranch Conservation Easement Management Plan as to when domestic sheep graze the Conway and Mattly Ranches. We ask that Mono County and the Eastern Sierra Land Trust clarify when domestic sheep will be grazed on these two properties. In 2012, we received a letter from the Los Angeles Department of Water and Power stating that, in late October, domestic sheep were observed trespassing on their property, which is adjacent to Mattly Ranch. The risk of contact between Sierra bighorn and domestic sheep increases during the Sierra bighorn breeding season (September through November) when rams are likely to make long-distance movements in search of females. Consequently, grazing this late in the season poses a serious risk to Sierra bighorn.

Greater Sage-Grouse

On October 28, 2013, the Service issued a proposed rule to list the Bi-State Distinct Population Segment of Greater sage-grouse (sage-grouse) as a threatened species and designate critical habitat. The Conway and Mattly Ranches are both located within proposed critical habitat for the sage-grouse. We encourage Mono County, as a partner in the Bi-State Local Area Working Group, to consider any potential impacts to sage-grouse and its habitat associated with the activities and uses, operations, and management of these properties.

Aquaculture

The draft Conway Ranch Conservation Easement Management Plan and associated documents do not indicate what fish species will be used for commercial fish-rearing. However, it is our understanding that, in the past, Mono County has expressed interest in rearing the federally threatened Lahontan cutthroat trout (*Oncorhynchus clarki henshawi*) on these properties. In the draft Grant Deed of Conservation Easement Conway and Mattly Ranches, it states that the construction of a fish-rearing raceway in the "Mattly Ranch Aquaculture Area" could occur if "a State or Federal wildlife agency identifies the development of a site as important to the recovery of endangered species and provide funding for a facility, operations and maintenance". The rearing of a federally threatened fish species on private property will require further discussions with the Service.

We appreciate the opportunity to review the draft Grant Deed of Conservation Easement Conway and Mattly Ranches Grant Deed and associated documents. We would like to coordinate with representatives from Mono County and the Eastern Sierra Land Trust to refine Board of Supervisors, Mono County

the draft Conway Ranch Conservation Easement Management Plan and identify measures that would meet the needs of the county, while also, protecting federally listed and proposed species. If you have any questions regarding this letter, please contact Erin Nordin at (760) 872-5020.

Sincerely,

Edward D. Koch State Supervisor

Enclosures

ccs:

Tony Dublino, Mono County, South county Office, Mammoth Lakes, California

- Dr. Tom Stephenson, Senior Environmental Scientist, California Department of Fish and Wildlife, Inland Deserts Region, Bishop, California
- Kay Ogden, Executive Director, Lands Director, Eastern Sierra Land Trust, Bishop, California

Aaron Johnson, Lands Director, Eastern Sierra Land Trust, Bishop, California

LITERATURE CITED

- Cassirer, E.F., R.K Plowright, K.R. Manlove, P.C. Cross, A.P. Dobson, K.A. Potter, and P.J. Hudson. Spatio-temporal dynamics of pneumonia in bighorn sheep. 2013. Journal of Animal Ecology 82:518–528.
- Croft, B., M. Haworth, M. Hennessy, R. Mazur, S. Nelson, R. Perloff, J. Robson, and T. Stephenson. 2009. Revised February 25, 2010. Application of the document entitled, a process for identifying and managing risk of contact between Sierra Nevada bighorn sheep and domestic sheep. April 2009. 19 pp.
- Lawrence, P.K., S. Shanthalingam, R.P. Dassanayake, R. Subramaniam, C.N. Herndon, D.P. Knowles, F.R. Rurangirwa, W.J. Foreyt, G. Wayman, A.M. Marciel, S.K. Highlander, and S. Srikumaran. 2010. Transmission of *Mannheimia haemolytica* from domestic shcep (*Ovis aries*) to bighorn sheep (*Ovis canadensis*): unequivocal demonstration with green fluorescent protein-tagged organisms. Journal of Wildlife Disease 46(3): 706-717.
- U.S. Fish and Wildlife Service. 2007. Recovery Plan for the Sierra Nevada bighorn sheep. Sacramento, California. 199 pp.
- Western Association of Fish and Wildlife Agencies (WAFWA). 2010. Summary of nine bighorn sheep herds that died across five western states. Wild Sheep Working Group. June 22, 2010. 2 pp.

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United States Department of the Interior

81440-2011-CPA-00456

Dan Lyster Mono County P. O. Box 2415 Mammoth Lakes, California 93546



Dear Mr. Lyster:

IN REPLY REFER TO:

We appreciated the opportunity to meet with you and discussion of the strength of the strengt of the strength sheep grazing at Conway Ranch and the risk of disease transmission to the federally endangered Sierra Nevada bighorn sheep (Ovis canadensis sierrae). This meeting was part of the U.S. Fish and Wildlife Service's effort to closely coordinate with you and other non-federal entities that authorize domestic sheep grazing in areas that have a potential for disease transmission between domestic sheep and Sierra Nevada bighorn sheep.

At the meeting, we presented findings from a risk assessment model that are relevant to domestic sheep grazing on Conway Ranch. As directed in the 2007 Recovery Plan for the Sierra Nevada Bighorn Sheep, the U.S. Fish and Wildlife Service established a subgroup of the recovery team to assess the disease risk associated with domestic sheep grazing allotments in the eastern Sierra Nevada. The model developed by the risk assessment team, and presented at our meeting, allows land managers to assess the relative risk of Sierra Nevada bighorn sheep rams entering areas where domestic sheep are grazed. The relative risk values, calculated by the model, provide information on the likelihood of a Sierra Nevada bighorn sheep ram entering an area based on habitat type and proximity to Sierra Nevada bighorn sheep populations. Based on these values, a land manager can determine which grazing allotments pose a higher risk of disease transmission. The risk assessment model indicates that Conway Ranch is located within an area of high risk; therefore, it is important to develop a land management plan for Conway Ranch that addresses the potential for disease transmission.

We would like to coordinate with you on development of a land management plan for Conway Ranch. We hope that the outcome of this process will provide benefits for Mono County and increased protection for Sierra Nevada bighorn sheep. As discussed at our meeting, we will continue to search for funding opportunities to aid in the development of a land management plan for Conway Ranch. In the meantime, we hope that you will continue to work with us on this endeavor. We will contact you at a future date to discuss this proposed plan in more detail.





United States Department of the Interior

FISH AND WILDLIFE SERVICE Ventura Fish and Wildlife Office 2493 Portola Road, Suite B Ventura, California 93003



IN REPLY REFER TO: 81440-2011-CPA-0045

December 9, 2010

Dan Lyster Mono County P. O. Box 2415 Mammoth Lakes, California 93546

Dear Mr. Lyster:

We appreciated the opportunity to meet with you and discuss our concerns regarding domestic sheep grazing at Conway Ranch and the risk of disease transmission to the federally endangered Sierra Nevada bighorn sheep (*Ovis canadensis sierrae*). This meeting was part of the U.S. Fish and Wildlife Service's effort to closely coordinate with you and other non-federal entities that authorize domestic sheep grazing in areas that have a potential for disease transmission between domestic sheep and Sierra Nevada bighorn sheep.

At the meeting, we presented findings from a risk assessment model that are relevant to domestic sheep grazing on Conway Ranch. As directed in the 2007 *Recovery Plan for the Sierra Nevada Bighorn Sheep*, the U.S. Fish and Wildlife Service established a subgroup of the recovery team to assess the disease risk associated with domestic sheep grazing allotments in the eastern Sierra Nevada. The model developed by the risk assessment team, and presented at our meeting, allows land managers to assess the relative risk of Sierra Nevada bighorn sheep rams entering areas where domestic sheep are grazed. The relative risk values, calculated by the model, provide information on the likelihood of a Sierra Nevada bighorn sheep ram entering an area based on habitat type and proximity to Sierra Nevada bighorn sheep populations. Based on these values, a land manager can determine which grazing allotments pose a higher risk of disease transmission. The risk assessment model indicates that Conway Ranch is located within an area of high risk; therefore, it is important to develop a land management plan for Conway Ranch that addresses the potential for disease transmission.

We would like to coordinate with you on development of a land management plan for Conway Ranch. We hope that the outcome of this process will provide benefits for Mono County and increased protection for Sierra Nevada bighorn sheep. As discussed at our meeting, we will continue to search for funding opportunities to aid in the development of a land management plan for Conway Ranch. In the meantime, we hope that you will continue to work with us on this endeavor. We will contact you at a future date to discuss this proposed plan in more detail.



Dan Lyster

Should you have any questions regarding this matter, contact Erin Shapiro at the Ventura Fish and Wildlife Office at (805) 644-1766, extension 369.

Sincerely,

5 α_{ij} Carl P. Benz

Assistant Field Supervisor

Application of the Document Entitled A Process for Identifying and Managing Risk of Contact between Sierra Nevada Bighorn Sheep and Domestic Sheep

Original April 3, 2009

Brian Croft¹, Marcy Haworth², MaryBeth Hennessy³, Rachel Mazur⁴, Steven Nelson⁵, Richard Perloff³, Joe Robson⁶, and Tom Stephenson⁷

Revised February 25, 2010

Brian Croft¹, Amy Fesnock⁸, Marcy Haworth², Rachel Mazur⁴, Leeann Murphy³, Steven Nelson⁵, Richard Perloff³, and Tom Stephenson⁷

INTRODUCTION

On February 12, 2009, representatives of the U.S. Forest Service (Inyo and Humboldt-Toiyabe National Forests), Bureau of Land Management (Bishop Field Office), U.S. Fish and Wildlife Service (Ventura and Nevada Fish and Wildlife Offices), and the California Department of Fish and Game (Bishop Field Office) held a meeting in Bishop, California, to discuss implementation of *A Process for Identifying and Managing Risk of Contact between Sierra Nevada Bighorn Sheep and Domestic Sheep* (Baumer *et al.* 2009; Risk Assessment). During this meeting, the land managers requested further assistance interpreting and applying the information provided in the Risk Assessment. They also expressed a need for guidance that would facilitate consistency in application of the Risk Assessment among the various agencies and jurisdictions. This guidance was requested within the context of recommendations 1, 2, and 8 provided in Section E of the *Recovery Plan for the Sierra Nevada Bighorn Sheep* (U.S. Fish and Wildlife Service 2007), which outlined a Recommended Strategy for Preventing Contact between Domestic Sheep or Goats and Sierra Nevada Bighorn Sheep.

Individuals representing the above-mentioned agencies met on February 19, 2009, to further discuss the information provided in the Risk Assessment and to develop recommendations for land managers to use when applying this information in their analyses of management options to prevent contact between domestic sheep and Sierra Nevada bighorn sheep. This document (Application Document) was developed as a result of this interagency coordination and provides recommendations specific to implementation of the Risk Assessment.

¹ U.S. Fish and Wildlife Service, Ventura Fish and Wildlife Office, Ventura, California; ²U.S. Fish and Wildlife Service, Nevada Fish and Wildlife Office, Reno, Nevada; ³ U.S. Forest Service, Inyo National Forest, Forest Headquarters, Bishop, California; ⁴ U.S. Forest Service, Humboldt-Toiyabe National Forest, Forest Headquarters, Sparks, Nevada; ⁵ Bureau of Land Management, Bishop Field Office, Bishop, California; ⁶ U.S. Forest Service, Inyo National Forest, White Mountain Ranger District, Bishop, California; ⁷ California; ⁷ California Department of Fish and Game, Bishop, California; ⁸Bureau of Land Management, California; State Office, Sacramento, California.

After completion of the 2009 domestic sheep grazing season and a meeting between the U.S. Forest Service's Regional Forester (Region 4) and the U.S. Fish and Wildlife Service's Regional Director (Region 8) on November 23, 2009, further management coordination was recommended among the federal and state agencies to address domestic sheep grazing in proximity to Sierra Nevada bighorn sheep. As a result, the original Application Document has been revised herein to further clarify how to apply the Risk Assessment.

All participants understand that the Risk Assessment is dynamic and that future updates, based on additional Sierra Nevada bighorn sheep movement data and other pertinent information, will likely change the results. This understanding is essential in the context of recovery for Sierra Nevada bighorn sheep. For recovery to occur, Sierra Nevada bighorn sheep populations must increase in numbers, and populations must be distributed among the 12 herd units identified as essential for conservation of the species in the final recovery plan. These herd units were identified in the Sierra Nevada bighorn sheep recovery plan based on historic bighorn sheep locations and the availability of summer and winter range (U.S. Fish and Wildlife Service 2007). Therefore, the likelihood of contact between Sierra Nevada bighorn sheep and domestic sheep is predicted to increase over time as recovery goals are met. The analyses used in the Risk Assessment and this Application Document focus on the potential for contact between Sierra Nevada bighorn sheep and domestic sheep is predicted to increase over time as recovery goals are met. The analyses used in the Risk Assessment and this Application Document focus on the potential for contact between Sierra Nevada bighorn sheep and domestic sheep in the eastern Sierra Nevada.

Section E of the recovery plan recommended a strategy for preventing contact between domestic sheep (and goats) and Sierra Nevada bighorn sheep. It also addressed the possibility that development and use of the Risk Assessment could alter our understanding of the risk of contact between domestic sheep and Sierra Nevada bighorn sheep. It recommended that land managers incorporate use of the Risk Assessment into allotment specific analyses of management options to prevent contact. The Risk Assessment and this revised Application Document provide land managers additional tools for analyzing the risk of contact on domestic sheep allotments. Application of these tools may change our understanding of the risk of contact on certain allotments from that described in Section E of the recovery plan. The Risk Assessment, this Application Document, and the recovery plan are guidance documents that land managers should use along with any additional information from the published literature or Sierra Nevada bighorn sheep monitoring efforts in their decision-making processes to prevent contact between domestic sheep and the federally-listed, endangered Sierra Nevada bighorn sheep.

APPLICATION OF THE RISK ASSESSMENT

A Process for Identifying and Managing Risk of Contact between Sierra Nevada Bighorn Sheep and Domestic Sheep (Baumer et al. 2009; Risk Assessment) identified the following five-step process for Risk Assessment implementation:

1. Determine the relative likelihood that a Sierra Nevada bighorn sheep will utilize habitat where domestic sheep are grazed;

- 2. Assess whether grazing domestic sheep in a specific allotment could result in contact with Sierra Nevada bighorn sheep;
- 3. Determine whether changes in the temporal (*e.g.*, seasonal closures) or spatial use of allotments would prevent contact between Sierra Nevada bighorn sheep and domestic sheep;
- 4. Determine whether implementing the grazing practices detailed in Section III of the Risk Assessment would prevent contact between Sierra Nevada bighorn sheep and domestic sheep; and
- 5. Monitor and verify whether grazing practices are being implemented and assess their effectiveness in reducing straying of domestic sheep.

Below, we describe a method for applying this five-step process on lands managed by the Inyo and Humboldt-Toiyabe National Forests and the Bureau of Land Management (Bishop Field Office). For more information on the five-step process, please refer to Section IV of the Risk Assessment.

Step 1 - Determine the relative likelihood that a Sierra Nevada bighorn sheep will utilize habitat where domestic sheep are grazed.

Please refer to the Risk Assessment (Section II pages 3-6; Attachment 6; Figure 2; Table 1) for a more complete description including the outcome of this previously completed step. Briefly, a geographic information system (GIS) based model was developed that incorporated Sierra Nevada bighorn sheep habitat suitability and proximity to herd use areas to determine the relative likelihood of Sierra Nevada bighorn sheep use of any particular location within the modeled area.

First, a GIS layer of *habitat suitability* was developed to indicate habitat preferences based on elevation, slope, aspect, hillshade, distance to escape terrain, terrain ruggedness, and vegetation (forest-nonforest). The final habitat suitability layer combines the habitat variables and incorporates each based on its importance. This GIS layer, with thousands of pixels (*i.e.*, geographic parcels) each representing the habitat suitability of 30 meter x 30 meter cells, was applied across the eastern Sierra landscape. In order to determine the relative resistance to bighorn sheep movement for a particular portion of the landscape, the habitat suitability layer was inverted to create a *resistance to movement* layer. In this layer, each pixel now represents the lack of habitat suitability for bighorn sheep at each location.

Second, to determine the relative likelihood of contact between bighorn sheep and domestic sheep at any location, a measure of the actual or potential presence of bighorn sheep was needed for each location. To do that, all known locations of radio-collared bighorn sheep, including GPS, ground, and aerial telemetry locations, were used to create home ranges for each herd unit. Core areas of these home ranges were then delineated using 50 percent kernels to create a *core home range* layer. Only those locations within cach corc home range were then used as source points for indentifying potential movement paths for bighorn sheep.

Finally, a cost-weighted distance function was applied to the core home range layer, utilizing the resistance to movement layer, to create a *cost distance layer* specific to Sierra Nevada bighorn sheep. This calculates the cost of travel from one cell to the next, initiated at the core home range locations. In the resulting composite cost distance layer, the value of each pixel represents the cumulative cost associated with travel to that point from a core home range location by a Sierra Nevada bighorn sheep.

Because bighorn sheep will not travel indefinitely, the model used a maximum dispersal limit of 60 kilometers from core home range locations (based on travel distances of GPS collared Sierra Nevada bighorn sheep rams) (Sierra Nevada Bighorn Sheep Recovery Program 2004). The cost-weighted distance was calculated from the core home range locations to the maximum dispersal limit.

There are a few recognized limitations to the GIS-based model, including: 1) not all Sierra Nevada bighorn sheep rams are collared, therefore, distance and location data points collected over the modeled time period are limited in number, 2) the vegetation layer used to map habitat suitability was limited in resolution, and 3) while the model was based on the best available data, it cannot predict every outcome.

In addition to the process described above, which is carried over and utilized in this Application Document, the Risk Assessment provided an additional step that averaged all of the pixel values within the boundary of a given allotment. This averaged value was meant to represent the relative likelihood that a Sierra Nevada bighorn sheep would utilize a given allotment. In the Risk Assessment, this value is referred to as the Mean Inverse Weighted Distance (MIWD). Table 1 of the Risk Assessment lists all the allotments analyzed with their MIWD values. Values closer to 1 indicate a high likelihood of use by a Sierra Nevada bighorn sheep. Values closer to 0 indicate a low likelihood of use by a Sierra Nevada bighorn sheep.

The use of MIWD, as discussed above, estimates the mean relative likelihood of Sierra Nevada bighorn sheep use of a given allotment. However, averaging pixel values to obtain a single value for an allotment ignores the variability of pixel values across the allotment. Therefore, an allotment could have portions that are highly conducive to bighorn sheep use, but the allotment's MIWD value would not reflect this because it is an average of all pixels values within the allotment.

In addition, the Risk Assessment does not provide a threshold above which a Sierra Nevada bighorn sheep's use of an allotment would be considered too high a risk for contact. Such a threshold has been requested by land managers to assist in making grazing management decisions. The Application Document does not use the mean relative likelihood of Sierra Nevada bighorn sheep use of a given allotment (*i.e.*, MIWD) in determining risk of contact, but provides an alternative method in Step 2.

Step 2 - Assess whether grazing domestic sheep in a specific allotment could result in contact with Sierra Nevada highorn sheep.

The Risk Assessment provided an equation for determining a relative risk of contact between Sierra Nevada bighorn sheep and domestic sheep for each allotment based on the allotment's mean relative likelihood that a bighorn sheep would occur there and the dates and length of time the allotment would be grazed (*i.e.*, Relative Risk = MIWD X (number of grazing days + Julian Date; Section II, page 5). The resulting value provides a relative value, but not a threshold value for the risk of contact for each allotment.

After further evaluation of this equation, a few issues also became evident about whether this is an appropriate way to represent risk. These issues include: 1) uncertainty about how the variables relate to one another and whether some are more important than others, 2) equation variables are expressed using different scales, 3) the likelihood that a Sierra Nevada bighorn sheep would occur in an allotment is based on the mean likelihood which ignores the variability of risk across an allotment, and 4) the timing of use of an allotment is more influential in the equation than the bighorn sheep location information.

The equation is, therefore, inappropriate to use in determining whether use of an allotment crosses a threshold of acceptable risk of contact between domestic sheep and Sierra Nevada bighorn sheep. It may be appropriate to use the equation to make modifications to grazing seasons to reduce risk of contact for allotments located further away from Sierra Nevada bighorn sheep core population areas that have a low likelihood of Sierra Nevada bighorn sheep use (see Step 4). For allotments with a high likelihood of Sierra Nevada bighorn sheep use, employing the equation to make modifications to grazing seasons to reduce risk of contact is not appropriate. As a result of these concerns, it is recommended that this equation's role in the determination of the risk of contact between domestic sheep and Sierra Nevada bighorn sheep be diminished.

A more direct approach to assessing the level of risk of contact between domestic sheep and Sierra Nevada bighorn sheep by allotment is to determine whether an allotment or portion of an allotment crosses a threshold of acceptable risk. There are two parts to this analysis. One part is to determine a threshold of acceptable risk and the other is to determine whether this threshold is crossed in an allotment or portion of an allotment.

To determine a threshold of acceptable risk, the final GIS layer (as described in Step 1) was overlain with locations where Sierra Nevada bighorn sheep are known to have occurred. To reduce error and ensure spatial accuracy, only those locations collected using GPS were used, resulting in 6,719 Sierra Nevada bighorn sheep ram locations (2002-2007). Data from these years was utilized because this was the most up to date data available at the time the model and the Risk Assessment were completed. It is important to use a multi-year data set to capture the full range of potential bighorn sheep movement patterns under various conditions. Use of a smaller range of years would reduce the amount of interannual variability that is captured and would provide a narrower view of potential Sierra Nevada bighorn sheep use than is likely to occur.

After the GIS layer was overlain with the Sierra Nevada bighorn sheep location data, the pixel value for each individual ram location in the data set was recorded to determine the full range of pixel values that correspond to Sierra Nevada bighorn sheep locations. All pixels in the GIS layer that have values within this range have a high likelihood of Sierra Nevada bighorn sheep use. By mapping the location of all pixels with values within this range, it is possible to highlight all locations in the eastern Sierra that have a high likelihood of bighorn sheep use instead of examining the relative likelihood as an average over an entire allotment. California Department of Fish and Game's 2002 to 2007 Sierra Nevada bighorn sheep monitoring efforts provided sufficient data for this analysis.

Through the analysis described above, it was determined that the values for all the known locations of collared rams within this dataset ranged from 0.833 to 1.0. The lowest value (0.833) for a known bighorn sheep location may, therefore, be used as a threshold value, above which bighorn sheep use is highly likely. If domestic sheep are grazed in locations (i.e., pixels) that have a high likelihood of Sierra Nevada bighorn sheep use, the potential for contact between domestic sheep and Sierra Nevada bighorn sheep exists and is predicted. Because the goal of the Risk Assessment is to prevent contact between the two species, values currently between 0.833 and 1.0 fall into a category of unacceptable risk. The locations of those areas that have a value between 0.833 and 1.0 and their relationship to domestic sheep grazing allotments are mapped in Figures 1 and 2. These predicted areas of potential contact (i.e., areas with high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833) allow for identification of portions of allotments that are above the 0.833 threshold. This is critical to making sure that land managers do not underestimate risk on some portions of a given allotment. At this time, if the relative likelihood of a Sierra Nevada bighorn sheep being in an allotment or portion of an allotment is 0.833 or greater, a high/unacceptable risk of contact between the two species exists for that allotment or portion of an allotment. If the value is less than 0.833, a low risk of contact between the two species exists for that allotment or portion of an allotment. Areas adjacent to these high contact risk areas should employ best management grazing practices to ensure that domestic sheep do not move into areas of high contact risk (see Step 4).

Since most Sierra Nevada bighorn sheep are not collared, the identified range of the relative likelihood of a Sierra Nevada bighorn sheep being in a location provides a scientific approach to management given the uncertainty associated with observing and tracking the movements of a subset of the population. The 0.833 threshold is based on known locations of GPS collared Sierra Nevada bighorn sheep. Sierra Nevada bighorn sheep location data based on VHF and observational data (some with locations that would expand range of values) were not included. It is also possible that uncollared Sierra Nevada bighorn sheep have moved farther, and into a wider range of habitats, than is demonstrated by the mapped areas of potential contact (*i.e.*, areas with high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833). Due to these restrictions and uncertainty, the value 0.833 is considered a conservative threshold for evaluating the relative risk of contact on an allotment.

The current threshold of 0.833 is based on the best available data at this time. Based on a preliminary review, additional Sierra Nevada bighorn sheep GPS location data collected from 2007 to 2009 (but not yet included in the model) remain within the 0.833 and above values of habitat modeled. Over time, new Sierra Nevada bighorn sheep location data could change the threshold. The value will be at least 0.833 (unless a dramatic loss of a herd occurred), but it will likely become lower as Sierra Nevada bighorn sheep increase in number and expand geographically as is necessary for recovery.

Distances between domestic sheep grazing areas and bighorn sheep locations have been considered by others when evaluating the risk of contact and disease transmission, and distance buffers to separate the two species have been previously recommended. Guidance developed by the Bureau of Land Management (1998) recommended buffer distances up to 9 miles around native wild sheep habitat unless topographic features or other barriers minimized the risk of contact. Singer et al. (2001) recommended bighorn sheep be restored to areas that are greater than 14.3 miles from domestic sheep grazing areas. A number of other documents address the concerns associated with domestic sheep grazing near Sierra Nevada or other bighorn sheep habitats. These documents discuss the need for buffers but do not recommend specific distances, or they suggest effective separation through spatial or temporal measures to reduce the risk of contact between the two species (Onderka et al. 1988, Sweeney et al. 1994, Ward et al. 1997, Schommer and Woolever 2001, Western Association of Fish and Wildlife Agencies 2007, George et al. 2008, Miller et al. 2008, Clifford et al. 2009). While a specific distance is not recommended in this Application Document to prevent contact, it is recognized that the proximity between these two species relates to the risk of contact.

The likelihood of contact plays an important role in the risk of disease transmission between domestic sheep and Sierra Nevada bighorn sheep in the Sierra Nevada. Contact may result in the possible introduction of new pathogens from domestic sheep to Sierra Nevada bighorn sheep that may cause pneumonia. There is concern that this could lead to the loss of entire bighorn sheep herds in the Sierra Nevada.

Clifford *et al.* (2009) developed a spatially explicit disease transmission model to quantitatively assess the risk of respiratory disease transmission from domestic sheep to Sierra Nevada bighorn sheep. This model was used to predict the impacts of a respiratory disease outbreak within and among populations in the Northern, Central, and Southern Recovery Units. We acknowledge several of the study's limitations. These include: 1) disease transmission data was from enclosures or experimental situations, 2) all available Sierra Nevada bighorn sheep location data was not included which could have increased home range size, 3) future Sierra Nevada bighorn sheep movements based on growth or exploration were not modeled, and 4) quantifying husbandry practices that may have increased contact risk, such as grazing estrous domestic sheep females, was not possible. Clifford *et al.* (2009) reported that the most frequently predicted levels of population mortality due to disease (33 to 76 percent in the North; 45 to 77 percent in the Central) were consistent with other estimates of mortality ranging from 28 percent to 80 percent reported in respiratory disease outbreaks of free-ranging bighorn sheep populations located elsewhere (with or without suspected contact with domestic sheep)

(Festa-Bianchet 1988, Ryder et al. 1992, Cassirer et al. 1996, Enk et al. 2001, George et al. 2008).

Because the current range wide population of this species is less than 400 individuals, the loss of an entire or almost entire Sicrra Nevada bighorn sheep herd(s) due to disease transmission would significantly impact recovery efforts for this species.

Step 3 - Determine whether changes in the temporal (*e.g.*, seasonal closures) or spatial use of allotments would prevent contact between Sierra Nevada bighorn sheep and domestic sheep.

Land managers should also consider the following criteria when evaluating whether an allotment or portion of an allotment identified as having a high/unacceptable risk of contact using Step 2 (*i.e.*, within the areas of potential contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833) may remain open and still ensure the prevention of contact between Sierra Nevada bighorn sheep and domestic sheep (*i.e.*, allotment specific criteria – see below).

If none of the criteria listed below applies to a given allotment, the allotment or portions of the allotment equal to or greater than the 0.833 threshold should not be grazed by domestic sheep to prevent contact between domestic sheep and Sierra Nevada bighorn sheep. If one or more of the criteria listed below apply to a given allotment, an allotment specific evaluation should be completed to determine whether the site-specific criteria provide sufficient barriers to Sierra Nevada bighorn sheep movement to prevent contact.

Criteria

- Allotments or portions of allotments within the predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833; Figures 1 and 2) that are separated from occupied Sierra Nevada bighorn sheep habitat by towns, highly developed recreation areas, or other human developments that inhibit bighorn sheep movement.
- 2) Allotments or portions of allotments within the predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833; Figures 1 and 2) that are separated from occupied Sierra Nevada bighorn sheep habitat by large contiguous forested areas that inhibit bighorn sheep movement.
- 3) Allotments or portions of allotments within the predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833; Figures 1 and 2) that are east of the U.S. Highway 395 management boundary recommended in Section II-E of the final *Recovery Plan for Sierra Nevada Bighorn Sheep* (U.S. Fish and Wildlife Service 2007).
- 4) Allotments or portions of allotments within the predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn

sheep use equal to or greater than 0.833; Figures 1 and 2) that are separated from occupied Sierra Nevada bighorn sheep habitat by major bodies of water (e.g., Mono Lake, Crowley Lake) that inhibit bighorn sheep movement.

For allotments or portions of allotments that meet one or more of the four criteria listed above, land managers may determine that full closure is not required to prevent contact between domestic sheep and Sierra Nevada bighorn sheep. After allotment specific analysis, the responsible land management agency may determine that a given allotment that meets one or more of the above criteria does not require seasonal or spatial restrictions to prevent contact. Conversely, allotment specific analysis may reveal that an allotment that meets one or more of the above criteria does require seasonal and/or spatial restrictions to prevent contact. In such a case, seasonal closure or closure of a portion of the allotment would be a suitable alternative to full closure if the agency determines that grazing under these restrictions would prevent contact between domestic sheep and Sierra Nevada bighorn sheep. Allotment specific seasonal closure periods should be determined by the responsible land management agency in cooperation with the affected permittee and in coordination and consultation, if appropriate, with the U.S. Fish and Wildlife Service.

In addition to the four criteria listed above, land managers may use the equation (Relative Risk = MIWD X (number of grazing days + Julian Date) provided in Section II of the Risk Assessment to quantify the predicted change in the relative risk that would occur as a result of temporal and/or spatial restrictions for a given allotment or allotment subunit. Use of the equation may provide a useful decision-making tool if the land manager determines that seasonal and/or spatial restrictions are needed, but decision makers must keep in mind the equation limitations discussed in Step 2.

If land managers determine, through the allotment specific analysis process described above, that contact between domestic sheep and Sierra Nevada bighorn sheep cannot be prevented on a given allotment, the allotment or portions of the allotment should not be grazed by domestic sheep.

Step 4 - Determine whether implementing the grazing practices detailed in Section III of the Risk Assessment would prevent contact between Sierra Nevada bighorn sheep and domestic sheep (*i.e.*, prevent domestic sheep from straying into areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact with Sierra Nevada bighorn sheep)).

The Risk Assessment indicates that land managers can alleviate some risk of contact between domestic sheep and Sierra Nevada bighorn sheep through implementation of best management grazing practices. However, when evaluating the risk of contact, both domestic sheep movements and Sierra Nevada bighorn sheep movements must be considered. Land managers should not regard the implementation of best management grazing practices as an appropriate means of preventing contact between domestic sheep and Sierra Nevada bighorn sheep on an allotment or portion of an allotment where the analysis described above has determined that there is a high/unacceptable risk of contact (*i.e.*, modeled areas of likely bighorn sheep use are equal to or greater than 0.833). These best management grazing practices, as detailed in Section III of the Risk Assessment, are designed primarily to keep domestic sheep from straying from the herd and/or allotment, to detect domestic sheep that have strayed from the herd and/or allotment, and to reduce predation on domestic sheep within an allotment. They are not designed to prevent Sierra Nevada bighorn sheep from entering an allotment and coming into contact with domestic sheep.

Herding and guard dogs have traditionally benefitted herders by keeping domestic sheep together and reducing predation. While they may have some potential to keep Sierra Nevada bighorn sheep from coming into contact with domestic sheep on an allotment, there is currently no scientific literature or other information that indicates or supports the assertion that herding or guard dogs can effectively prevent bighorn sheep from coming into contact with domestic same propriate as stand-alone mitigation for use on allotments or portions of allotments within the predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833) (Figures 1 and 2).

The appropriate use of best management grazing practices is to keep domestic sheep from straying out of open allotments where there is a low risk of contact and into areas where there is a high risk of contact with Sierra Nevada bighorn sheep. As described in the Risk Assessment, the only method to ensure the prevention of contact between domestic sheep and Sierra Nevada bighorn sheep is to avoid the use of overlapping ranges through temporal and/or spatial separation.

The allotment specific application of the best management grazing practices recommended in Section III of the Risk Assessment should be determined by the responsible land management agency in cooperation with the affected permittee and in coordination and consultation, if appropriate, with the U.S. Fish and Wildlife Service.

Step 5 - Monitor and verify whether grazing practices are being implemented and assess their effectiveness in reducing straying of domestic sheep.

Land managers should monitor and verify that livestock producers are appropriately implementing best management grazing practices as discussed in Section III of the Risk Assessment on those allotments where they are applied (Step 4). For those allotments where the best management grazing practices are required, land managers should compile all monitoring and reporting information from both permittees and agency personnel on an annual basis and evaluate whether these practices are being fully and effectively implemented.

RESULTS OF RISK ASSESSMENT APPLICATION

The predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833)

between Sierra Nevada bighorn sheep and domestic sheep (Figures 1 and 2), determined through application of Step 2, overlap 27 domestic sheep allotments or allotment subunits (Table 1, Figure 2). Of those, eight are vacant to prevent contact between domestic sheep and Sierra Nevada bighorn sheep. Five currently permitted allotments or allotment subunits are entirely within the predicted areas of potential contact (*i.e.*, areas with a high/ unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833) (Tamarack, Cameron Canyon, Dunderberg, Rock Creek – Hilton Unit, and Sherwin Deadman 1). Only a portion of the predicted areas of potential contact overlaps the remaining 13 allotments (Table 1, Figure 2). Table 1 identifies the allotments that overlap the predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833). Table 1, column 6, indicates the mean relative likelihood that a Sierra Nevada bighorn sheep would use a given allotment. Table 1, column 7, indicates the maximum relative likelihood that a Sierra Nevada bighorn sheep would use any point within a given allotment.

RECOMMENDATIONS

We recommend that the Humboldt-Toiyabe and Inyo National Forests and the Bureau of Land Management (Bishop Field Office) analyze all of their respective allotments that overlap the predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833) between domestic sheep and Sierra Nevada bighorn sheep (Table I, Figures 1 and 2) as outlined in Steps 2 and 3.

If the responsible land management agency determines that a given allotment, or portion of an allotment, does not meet one or more of the criteria identified in Step 3, we recommend that the allotment, or portion of the allotment, identified as having a high/unacceptable risk of contact not be grazed by domestic sheep to prevent contact between domestic sheep and Sierra Nevada bighorn sheep. There is uncertainty associated with determining when contact may occur and the likely adverse effects to Sierra Nevada bighorn sheep populations that could result from a contact related disease outbreak. Therefore, it is recommended that any currently permitted allotment, or portion of an allotment, identified as having a high risk/unacceptable risk of contact as a result of the above analysis, be closed on either a temporary non-use or emergency basis until the responsible land management agency determines whether permanent closure to domestic sheep grazing is warranted.

If the responsible agency determines that a given allotment meets one or more of the criteria identified in Step 3, we recommend that the agency determine whether temporal and/or spatial restrictions are necessary to prevent contact through the appropriate agency analysis and decision making process. In instances where domestic sheep could stray from an open allotment into an areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833), we recommend the implementation of best management grazing practices from Section III of the Risk Assessment as outlined in Steps 4 and 5.

The Risk Assessment, this revised Application Document, and the recovery plan are guidance documents that land managers should use along with any additional information from the published literature or Sierra Nevada bighorn sheep monitoring efforts in their decision-making process to prevent contact between domestic sheep and the federally-listed, endangered Sierra Nevada bighorn sheep. The decision tree provided below may be helpful when evaluating the allotments.

As stated in the Risk Assessment, the model will be updated by the California Department of Fish and Game in coordination with land management agencies as new information is collected on bighorn sheep movement and domestic sheep allotment management. The model should be rerun when new information (*e.g.*, changes in bighorn sheep distribution/movement, habitat conditions and/or domestic sheep grazing regimes) is available. At a minimum, we recommend that Sierra Nevada bighorn sheep location data be reviewed biennially and compared to the 0.833 threshold. If this new location data indicates a substantial shift of bighorn sheep habitat use and therefore changes the risk of contact, the model should be updated to incorporate this new data and new model run outputs made available to agencies. Model updates will be contingent on funding by state and federal agencies or other sources.

Decision Tree

A. Assess whether domestic sheep in an allotment could contact bighorn sheep

1. Does the allotment or portion of the allotment overlap modeled areas of likely bighorn sheep use equal to or greater than 0.833 as indicated in Table 1?

Yes. The allotment or portion of the allotment equal to or greater than 0.833 should not be grazed by domestic sheep and closure of the allotment should be considered, unless it meets one or more of the four criteria in Step 3.

No. Domestic sheep grazing may be permitted. Best management grazing practices should be implemented in areas where domestic sheep might stray off of a low risk allotment into a high risk allotment. Go to C.

B. Determine if the allotment meets one of the Step 3 criteria and whether changes in temporal or spatial use of the allotment would prevent contact

1. Does the allotment meet one or more of the criteria in Step 3?

Yes. The responsible land management agency may determine that full closure is not required to prevent contact. The equation in Step 2 may be used to determine what changes in temporal and/or spatial restrictions would reduce the risk of contact to a low level. Go to B 2. Also go to C.

No. The allotment or portion of the allotment that overlaps modeled areas of likely bighorn sheep use is equal to or greater than 0.833. The allotment or portion of the allotment should not be grazed by domestic sheep and closure of the allotment should be considered.

2. Will changes in temporal or spatial use of the allotment prevent contact?

Yes. Domestic sheep grazing may be permitted under temporal and/or spatial restrictions.

No. The allotment should not be grazed by domestic sheep.

C. Determine whether best management grazing practices would prevent contact

1. Will the use of best management grazing practices prevent contact?

Yes. The allotment or portion of an allotment is less than 0.833 and/or meets one or more of the Step 3 criteria. The allotment may be grazed by domestic sheep with implementation of best management grazing practices. Also go to D.

No. The allotment or portion of the allotment is equal to or greater than 0.833 and none of the Step 3 criteria apply. The allotment or portion of the allotment should not be grazed by domestic sheep and closure of the allotment should be considered.

D. Monitor and verify whether grazing practices are being implemented effectively

If domestic sheep grazing is permitted and grazing practices are implemented to keep domestic sheep from straying off of a low risk allotment into a high risk allotment, monitoring and reporting information should be compiled annually and evaluated.

TABLES AND FIGURES

Table 1. Allotments that overlap the predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact; modeled areas of likely bighorn sheep use equal to or greater than 0.833) between Sierra Nevada bighorn sheep and domestic sheep described in Step 2. The Allotment Mean Likelihood of Use values are taken from the Risk Assessment. The values represent the mean likelihood that a Sierra Nevada bighorn sheep will use an allotment. The Allotment Maximum Likelihood of Use values represent the maximum likelihood that a Sierra Nevada bighorn sheep will use a location within an allotment.

Allotmont Nome	Invision [®]	Status	Total Permitted	Permitted Julian Day	Allotment Mean Likelihood	Allotment Maximum Likelihood
Alger's Lake	INF	Vacant	NA ^b	NA NA	0.929	0.966
Bloody Canyon	INF	Vacant	NA	NA	0.936	0.974
Cameron Canyon	HTNF	Permitted	95	289	0.911	0.949
Dog Creek	BLM	Permitted	153	305	0.881	0.949
Dunderberg	HTNF	Permitted ^c	95	274	0.970	0.997
Green Creek	BLM	Permitted	153	305	0.852	0.911
Green Creek	HTNF	Vacant	NA	NA	0.929	0.960
Horse Meadow	INF	Vacant	NA	NA	0.937	0.963
Jordan Basin	HTNF	Vacant	NA	NA	0.971	0.999
June Lake (East)	INF	Permitted	62	244	0.800	0.864
June Lake (West)	INF	Vacant	NA	NA	0.836	0.907
Little Mormon	BLM	Permitted	153	305	0.805	0.864
Little Round Valley 1	BLM	Permitted	14	292	0.850	0.873
Little Round Valley 3	BLM	Permitted	14	292	0.831	0.840
McGee	INF	Permitted ^d	92	251	0.903	0.952
Rancheria Gulch	BLM	Permitted	153	305	0.784	0.932
Rickey (South)	HTNF	Permitted	NA	NA	0.827	0.848
Rock Creek 1 (West)	INF	Vacant	NA	NA	0.864	0.906
Rock Creek 2 (Highway)	INF	Vacant	ŇA	NA	0.865	0.885
Rock Creek 3 (East)	INF	Permitted	92	243	0.819	0.868
Rock Creek 4 (Hilton)	INF	Permitted	66	227	0.860	0.876
Sherwin/Deadman 1 (Mammoth)	INF	Permitted	87	274	0.915	0.931
Sherwin/Deadman 2 (Sawmill)	INF	Permitted	87	274	0.844	0.917
Sherwin/Deadman 4 (Smokey Bear)	INF	Permitted	87	274	0.794	0.882
Summers Meadow S&G	HTNF	Permitted	30	305	0.855	0.885
Tamarack	HTNF	Permitted	95	289	0.912	0.958
Volcanic Tablelands	BLM	Permitted	46	167	0.716	0.871

^a Bureau of Land Management (BLM), Humboldt-Toiyabe National Forest (HTNF), Inyo National Forest (INF). ^b Not applicable (NA).

c Not grazed in 2007-2009.

d Closed since finalization of original Application Document

Figure 1 – Red areas are the mapped locations that have Likelihood of Use values between 0.833 and 1 and are considered the predicted areas of potential contact (*i.e.*, areas with a high/unacceptable risk of contact). Green allotments are U.S. Forest Service domestic sheep allotments. Beige allotments are Bureau of Land Management domestic sheep allotments. Allotments are mapped over the Risk Assessment's Sierra Nevada bighorn sheep Likelihood of Use layer (*i.e.*, brown and yellow layer) and the Risk Assessment's Sierra Nevada bighorn sheep ram habitat suitability layer (*i.e.*, black and white layer).



Rickey South Green Creek (BLM) Little Mormon Summers Meadow Rancheria Gulch Tamarack **Cameron** Canyon June Lake (east) Green Creek (HAT Dog C Dunderberg Jordan Basin Horse Meadow Bloody Canyon Sherwin Deadman 4 Algers Lake June Lake (West) Little Round Valley Sherwin Deadman 2 Sherwin Deadman 1 McGee Rock Creek (Hilton) Little Round Valley 3 Volcanic Tablelands Rock Creek (west) **Rock Creek (Highway** Rock Creek (east)

Figure 2 – Labeled allotments overlap the predicted areas of potential contact in red (*i.e.*, areas with a high/unacceptable risk of contact) between Sierra Nevada bighorn sheep and domestic sheep.

LITERATURE CITED

- Baumer, A., N. East, J. Echenique, M. Haworth, M. Leinassar, C. Papouchis, T. Stephenson, D. Weaver, and G. Wilson. 2009. A process for identifying and managing risk of contact between Sierra Nevada bighorn sheep and domestic sheep. February 2009. 37 pp.
- Cassirer, E. F., L. E. Oldenburg, V. L. Coggins, P. Fowler, K. Rudolph, D. L. Hunter, and W. J. Foreyt. 1996. Overview and preliminary analysis of a bighorn sheep dieoff-Hells Canyon 1995-96. Proceedings of the Biennial Symposium Northern Wild Sheep and Goat Council 10:78-86.
- Clifford, D. L., B. A. Schumaker, T. R. Stephenson, V. C. Bleich, M. L. Cahn, B. J. Gonzales, W. M. Boyce, and J. A. K. Mazet. 2009. Assessing disease risk at the wildlife-livestock interface: a study of Sierra Nevada bighorn sheep. Biological Conservation 142:2559-2568.
- Enk, T. A., H. D. Picton, and J. S. Williams. 2001. Factors limiting a bighorn sheep population in Montana following a dieoff. Northwest Science 75:280-291.
- Festa-Bianchet, M. 1988. A pneumonia epizootic in bighorn sheep, with comments on preventive management. Biennial Symposium of the Northern Wild Sheep and Goat Council 6:66-76.
- George, J. L., D. J. Martin, P. M. Lukacs, and M. W. Miller. 2008. Epidemic pasteurellosis in a bighorn sheep population coinciding with the appearance of a domestic sheep. Journal of Wildlife Diseases 44:388-403.
- Miller, M. W., D. P. Knowles, and M. S. Bulgin. 2008. Pasteurellosis transmission risks between domestic and wild sheep. CAST Commentary QTA 2008-1. Council for Agricultural Science and Technology, Ames, Iowa. 8 pp.
- Onderka, D. K., S. A. Rawluk, and W. D. Wishart. 1988. Susceptibility of Rocky Mountain bighorn sheep and domestic sheep to pneumonia induced by bighorn and domestic livestock strains of *Pasteurella haemolytica*. Canadian Journal of Veterinary Research 52:439-444.
- Ryder, T. J., E. S. Williams, K. W. Mills, K. H. Bowles, and E. T. Thorne. 1992. Effect of pneumonia on population size and lamb recruitment in Whiskey Mountain bighorn sheep. Proceedings of the Biennial Symposium of the Northern Wild Sheep and Goat Council 8:136-146.
- Schommer, T., and M. Woolever. 2001. A process for finding management solutions to the incompatibility between domestic and bighorn sheep. Wallowa-Whitman National Forest, Baker City, Oregon. 20 pp., plus appendices.

- Sierra Nevada Bighorn Sheep Recovery Program. 2004. Sierra Nevada bighorn sheep recovery and domestic livestock: preliminary risk assessment of disease in the Eastern Sierra. California Department of Fish and Game, Bishop, California. 16 pp., plus appendices.
- Sweeney, S. J., R. M. Silflow, and W. J. Foreyt. 1994. Comparative leukotoxicities of *Pasteurella haemolytica* isolates from domestic sheep and free-ranging bighorn sheep (*Ovis canadensis*). Journal of Wildlife Discases 30(4):523-528.
- U.S. Fish and Wildlife Service. 2007. Recovery Plan for the Sierra Nevada Bighorn Sheep. Sacramento, California. xiv + 199 pp.
- Ward, A. C. S., D. L. Hunter, M. D. Jaworski, P. J. Benolkin, M. P. Dobel, J. B. Jeffress, and G. Tanner. 1997. *Pasteurella* spp. in sympatric bighorn and domestic sheep. Journal of Wildlife Diseases 33:544-557.
- Western Association of Fish and Wildlife Agencies. 2007. Recommendations for domestic sheep and goat management in wild sheep habitat. Wild Sheep Working Group, Initial Committee. 21 pp., plus appendices.



United States Department of the Interior

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October 16, 2015 File No. 2015- CPA-0104

Ms. Wendy Sugimura Mono County Community Development Post Office Box 347 Mammoth Lakes, California 93546

Subject: Mono County General Plan Update and Other Associated Documents

Dear Ms. Sugimura:

This letter is in response to Mono County's update of the Mono County General Plan and other associated documents. Specifically, Mono County is updating the Mono County General Plan, the Regional Transportation Plan, and three elements of the Integrated Waste Management Plan. This is considered a "project" as defined in the California Environmental Quality Act (CEQA) guidelines; therefore, a draft environmental impact report (EIR) has also been prepared to analyze the potential environmental effects associated with implementation of the project. The project includes goals, objectives, policies, actions, implementation programs, regulations and ordinances, and the repeal of a specific plan. We are providing our comments under the authorities of the Endangered Species Act of 1973, as amended (ESA; 16 U.S.C. 1531 *et seq.*), Migratory Bird Treaty Act of 1918, as amended (MBTA; 16 U.S.C. 703 *et seq.*), and other authorities of the Department of Interior.

Bi-State Distinct Population Segment of the Greater Sage-Grouse (*Centrocercus urophasianus*; sage-grouse)

On April 21, 2015, we announced the withdrawal of our proposed rule to list the Bi-State distinct population segment of sage-grouse as threatened under the ESA. Our decision was largely predicated on conservation commitments provided by our partners in the Bi-State area, primarily through the Bi-State Action Plan, and further by needed changes to existing regulatory mechanisms. Mono County has been an active partner in conservation of sage-grouse in the Bi-State area and plays an important role in both specific conservation of the species by reducing conflicts among multiple user groups. We are encouraged by the draft 2015 General Plan update and believe this regulatory mechanism will facilitate long-term conservation of sage-grouse in

Wendy Sugimura

the area. We recognize the General Plan tends to be a fairly general or conceptual document and we encourage you to remain cognizant of the important role the General Plan will play in conservation as you move toward implementation and interpretation of the document in future years with respect to evaluating site specific developments.

More specifically, we would like to commend you on several aspects of the General Plan that we believe will be instrumental in long-term conservation of the species through reducing site specific impacts as well as limiting landscape scale concerns such as fragmentation. As delineated in the Bi-State Action Plan, the County continues to move toward the elimination of the Benton Crossing Landfill. We consider this to be extremely important and applaud your resolve to manage this specific stressor. Additionally, we strongly support your decision to withdraw the previous Conway Ranch Management Plan and designate this area as open space. While we continue to have concerns over livestock grazing in the area (*see* Sierra Nevada Bighorn Sheep *section below*), we believe this decision is a strong move in the right direction. Finally, we consider many of the Land Use and Conservation/Open Space Elements intended to contain growth in existing communities; limit habitat disturbance outside of communities; preserve open space; and afford deference to sage-grouse habitat during discretionary permit review will greatly enhance the Bi-State area Partners' abilities to maintain individual sage-grouse populations and the connectivity among them.

Finally, we have several comments pertaining to specific items delineated in the proposed project. We request these comments be given careful consideration.

Common Ravens

Common ravens (*Corvus corax*) can have a significant impact on nesting success of sage-grouse and populations of the species can be artificially inflated in areas where anthropogenic foods subsidies are apparent. It appears that vehicle collisions with deer is a concern within the county and several actions are identified to minimize this safety issue. Assuming these actions will not be 100 percent effective, we encourage you to explore ways to collect and dispose of carcasses, thereby removing them as potential food sources for common ravens. Similarly, ranch operations will frequently dispose of dead animals by simply hauling them to a remote corner of the ranch property and leaving the carcasses exposed on the ground (i.e., "bone yards"). Actions (as allowed by law) that eliminate or restrict disposal of dead livestock in a manner that makes their carcasses available to common ravens should also be adopted.

Communication Towers

Cellular towers have been implicated to negatively affect sage-grouse populations and devices intended to limit perching and nesting by sage-grouse predators on these structures are generally ineffective. Several actions in the Project identify the need for additional cellular towers to facilitate communication and improve safety in the County. While we recognize this need, we encourage you to be deliberative in placement of this infrastructure and to the greatest extent possible restrict placement in sage-grouse habitat.
<u>Roads</u>

The Regional Transportation Plan identified an interest in improving State Route 270 and the Cottonwood Canyon Road in the Bodie Hills. While we are unsure at this time as to the benefit or cost of this specific action on sage-grouse, increased vehicle traffic in general has been shown to negatively affect sage-grouse populations. Therefore, we encourage you to fully consider and discuss with Bi-State partners any actions that would increase vehicle traffic in the Bodie Hills.

Non-native Grass

We have significant concerns with nonnative annual grasses, such as cheatgrass (*Bromus tectorum*) and red brome (*Bromus rubens*), due to the substantial negative impacts these species can have on native sagebrush (*Artemisia* sp.) habitats. We would strongly encourage Mono County, via the General Plan or other appropriate mechanism, to assist private land owners in addressing these invasive weed concerns.

<u>EIR</u>

We recommend fulfilling your commitment identified in the alternatives section of the EIR (p 6-14) to incorporate the additional proactive conservation measures identified in Alternative 3 into the proposed project at this time; including those actions described in Table 6-2 pertaining to enhancement of Bi-State grater sage-grouse habitat such as fence design and density; conservation easements; limiting the significance of impacts; and closing county roads during the breeding season.

Lahontan Cutthroat Trout (Oncorhynchus clarkii henshawi)

In the Draft Land Use Element section (p. II-133), Action 24.F.1.a. states that Witcher and Birch Creeks were identified by the California Department of Fish and Wildlife (CDFW) as locations for the reintroduction of Lahontan cutthroat trout. We would like to clarify that these two streams are located outside of the native range of Lahontan cutthroat trout and not considered streams needed for recovery of the species. The U.S. Fish and Wildlife Service (Service) works collaboratively with CDFW to introduce Lahontan cutthroat trout into locations that contribute to the recovery of the species and we have not had any discussions with CDFW regarding the introduction of Lahontan cutthroat trout to Witcher and Birch Creeks.

The following recovery waters on the Humboldt Toiyabe National Forest within the Walker River watershed contain all the self-sustaining stream populations of Lahontan cutthroat trout in Mono County: 1) Slinkard Creek, 2) Mill Creek, 3) Silver Creek, 4) Wolf Creek, 5) Murphy Creek, and 6) By-Day Creek. Another out-of –basin stream is O'Harrel Creek which is located northeast of Crowley Lake on the Inyo National Forest. In addition to these self-sustaining stream populations, Table 1 below identifies the lakes and stream segments that are stocked with Lahontan cutthroat trout by CDFW; however, waters stocked by Lahontan cutthroat trout can change.

County	Landowner	Waterbody Name
Mono	Inyo NF	June Lake
Mono	Inyo NF	McCleod Lake
Mono	Inyo NF	Silver Lake
Mono	Inyo NF,	Steelhead Lake
Mono	City of LA	Crowley Lake
Mono	Inyo NF	Gull Lake
Mono	unknown	WF Walker River section 2
Mono	HTNF	WF Walker River section 3
Mono	HTNF	EF Walker River below Bridgeport Res.
Mono	HTNF	Bridgeport Reservoir
Mono	HTNF	Kirmen Lake
Mono	HTNF	Lane Lake
Mono	HTNF	Roosevelt Lake

Table 1. Waterbodies stocked by CDFW with Lahontan cutthroat trout

Sierra Amphibians

We appreciate your recognition in Policy 2.A.13. that certain lakes and streams in the backcountry may be more appropriate for the conservation of sensitive, threatened or endangered species, such as the federally-listed as endangered Sierra Nevada yellow-legged frog (*Rana sierrae*) and federally-listed as threatened Yosemite toad (*Anaxyrus canorus*), as identified in Policy 2.A.13. Please note that critical habitat for Sierra Nevada yellow-legged frog and Yosemite toad are expected to publish in the Federal Register in the near future. As recovery efforts advance for this species, we look forward to working with the County.

Sierra Nevada Bighorn Sheep (Ovis canadensis sierrae; Sierra bighorn)

In the Draft Land Use Element section (pp. II-9, II-10, II-79, II-81), there are discussions about supporting historical uses in the Mono Basin, including domestic sheep grazing. Currently, domestic sheep grazing occurs on both the Conway and Mattly Ranches. These two properties are located less than 0.5 miles from the Mt. Warren herd unit. The Mt. Warren herd unit is identified as an "essential herd unit" or a herd unit that is most likely to support the recovery of the subspecies in the U.S. Fish and Wildlife Service's Recovery Plan for the Sierra Nevada Bighorn Sheep (2007).

On June 17, 2014, we sent a letter to the Mono County Board of Supervisors on the draft Grant Deed of Conservation Easement Conway and Mattly Ranches indicating our concerns regarding domestic sheep grazing on these two properties. As stated in that letter, the Conway and Mattly Ranch properties are located within a predicted area of potential contact which indicates that there is a high risk of Sierra bighorn coming into contact with domestic sheep. Disease transmitted by domestic sheep was one threat identified in the final rule to list Sierra bighorn as

endangered (65 FR 20) and is considered the primary factor leading to the decline and extirpation of bighorn sheep across western North America. Consequently, we remain concerned about the continued grazing of domestic sheep in areas adjacent to habitat occupied by Sierra bighorn.

The Sierra bighorn population has been steadily increasing since it was listed; however, disease events still post a threat to the subspecies and the effects can last for decades. Due to the potential impact to the Sierra bighorn population from domestic sheep grazing, we recommend including Sierra bighorn in Wildlife Resources section 4.5.2.6 (Section in original document should be corrected to 4.4.2.6) and discussing the potential impacts to the species. Including Sierra bighorn in this discussion would be consistent with Policy 24.F.3. (Protect wildlife and native plants, especially rare and endangered species) and Action 24.F.3.e. (Support the CDFW's continuing program to reintroduce native game species (bighorn sheep)), which were both identified in the Draft Land Use Element with Edits section of the General Plan Update (p. II-134).

We look forward to continuing discussion and collaboration toward solutions to this concern.

Sierra Nevada Red Fox (Vulpes vulpes necator)

We recommend updating information related to the Sierra Nevada red fox. As of October 8, 2015, the Sierra Nevada distinct population segment was determined to be warranted but precluded from listing under the ESA (80 FR 60990). As a result, the Service considers this subspecies to be a candidate species under the ESA. This population occurs near Sonora Pass in Mono, Tuolumne, and Alpine Counties. We recommend that the second paragraph and Table 4.4-7 under the Biological Resources section on pages 4.4-22 and 4.4-24, respectively, be modified to reflect this new information.

Migratory Birds

Migratory birds are a public trust resource of the Service and are protected by the Migratory Bird Treaty Act. You can find a list of species protected by the Migratory Bird Treaty Act at 50 Code of Federal Regulations 10.13. The Migratory Bird Treaty Act prohibits the "take" or possession of migratory birds; "take" under this law means to pursue, hunt, shoot, wound, kill, trap, capture, or collect, or attempts to do so (50 Code of Federal Regulations 10.12).

The Migratory Bird Treaty Act is a strict liability statute, meaning that proof of intent, knowledge, or negligence is not an element of a violation of this statute. The statute's language is clear that an action resulting in the "taking" of an individual of a protected species is a violation of this law. The Migratory Bird Treaty Act does not specifically authorize the incidental take of migratory birds; consequently, the Service does not issue permits authorizing such take.

The Service carries out its mission to protect migratory birds by fostering relationships with entities that have taken effective steps to avoid take, by encouraging others to implement measures to avoid take, and through investigations and enforcement when appropriate. We encourage you to work closely with the Service to identify available protective measures when developing project plans to safeguard wildlife and to implement those measures where applicable. Examples of recommended conservation measures can be found here: http://www.fws.gov/birds/management/project-assessment-tools-and-guidance/conservation-measures.php.

We appreciate the opportunity to provide comments on the updated General Plan. If you have any questions regarding this letter, please contact me at (775) 861-6300 or Erin Nordin, of my staff, at (760) 872-5020 or Erin_Nordin@fws.gov.

Sincerely,

Edward D. Koch Field Supervisor

References

U.S. Fish and Wildlife Service (Service). 2007. Recovery Plan for the Sierra Nevada Bighorn Sheep. Sacramento, California. xiv + 199 pages.



United States Department of the Interior

Pacific Southwest Region FISH AND WILDLIFE SERVICE Reno Fish and Wildlife Office 1340 Financial Blvd., Suite 234 Reno, Nevada 89502 Ph: (775) 861-6300 ~ Fax: (775) 861-6301



June 17, 2016 File No. 2016-CPA-0077

Board of Supervisors Mono County P. O. Box 715 Bridgeport, California 93517

Dear Mono County Board of Supervisors:

Subject: Comments on the April 2016 Draft Conway Ranch Strategic Facility Plan

Thank you for providing a public process in the development of a strategic plan to guide future activities, maintenance, and development of the Conway Ranch property. We commend Mono County's efforts to develop a plan that maintains and enhances the conservation, economic, recreation and cultural values that are present on Conway and Mattly Ranches. We have reviewed the draft Conway Ranch Strategic Facility Plan (Plan) dated April 2016 and would like to take this opportunity to share our general comments on the Plan.

The Plan identifies the master vision for grazing Mattly and Conway Ranches to include domestic sheep. We request the Plan be changed to simply identify livestock grazing as the master vision for the property. As we have communicated in previous letters and discussions, we remain concerned with the potential effects of domestic sheep grazing on Mattly and Conway Ranches to the state and federally endangered Sierra Nevada bighorn sheep (*Ovis canadensis sierrae;* Sierra bighorn).

Our main concern is the potential for disease transmission, specifically pneumonia, between domestic sheep and Sierra bighorn due to the proximity of Mattly and Conway Ranches to the Mt. Warren herd unit. The Mt. Warren herd unit was identified in the 2007 Sierra Nevada Bighorn Sheep Recovery Plan (Service 2007) as a herd unit essential for the recovery of the subspecies and it is also designated critical habitat. The California Department of Fish and Wildlife (CDFW), our partner in recovering the Sierra bighorn, has been successful in increasing the abundance and distribution of Sierra bighorn through augmentations and translocations. These efforts have been necessary in order to ensure the persistence of the subspecies throughout its historic range. A disease outbreak in the Mt. Warren herd unit would not only affect animals

Board of Supervisors

occupying this area but it could also spread to animals in the surrounding area known as the Northern Recovery Unit. Currently, there are two other herds located in the Northern Recovery Unit, one of which is a newly introduced herd located in Yosemite National Park. Presently, we do not think that the current management of Mattly and Conway Ranches provides for the effective separation of domestic sheep and Sierra bighorn. This lack of effective separation limits our ability to recover Sierra bighorn.

The Plan also states that the current grazing lease expires in November 15, 2017, and that domestic sheep grazing may continue "unless it is prohibited through federal action, on North Conway Ranch, South Conway Ranch, and Mattly Ranch, with appropriate seasonal restrictions and BMPs applied." We request that Mono County remove this language from the Plan because it is inconsistent with the U.S. Fish and Wildlife Service's authority under the Endangered Species Act of 1973, as amended (ESA; 50 CFR §402.13).

As a result, we request an opportunity to meet in person to provide more detailed information on our concerns and authorities under the ESA. We are also hoping to arrange a meeting with F.I.M. Corporation to provide an opportunity for information sharing as well. If you have any questions regarding our comments or to arrange a meeting, please contact me or Lee Ann Carranza at (775) 861-6300.

Sincerely,

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Edward D. Koch State Supervisor

ccs:

Tony Dublino, Mono County, South County Office, Mammoth Lakes, California F.I.M. Corporation, Smith, Nevada

Dr. Tom Stephenson, Senior Environmental Scientist, California Department of Fish and Wildlife, Inland Deserts Region, Bishop, California

Literature Cited

U.S. Fish and Wildlife Service. 2007. Recovery Plan for the Sierra Nevada Bighorn Sheep. Sacramento, California. xiv + 199 pages.



United States Department of the Interior Pacific Southwest Region FISH AND WILDLIFE SERVICE Reno Fish and Wildlife Office 1340 Financial Blvd., Suite 234 Reno, Nevada 89502 Ph: (775) 861-6300 ~ Fax: (775) 861-6301



1.

December 12, 2016 File No. 2016-CPA-0077E

Board of Supervisors Mono County P. O. Box 715 Bridgeport, California 93517

Subject: Conway/Mattly Ranch Item for December 13, 2016 Board of Supervisors Meeting

Dear Honorable Mono County Board of Supervisors:

Thank you for notifying our office of the Conway/Mattly Ranch agenda item for tomorrow's Board of Supervisors Meeting. The agenda item is identified as Item 13/Regular Agenda: Presentation Regarding Conway/Mattly Ranch Public Outreach and Authorization for Request for Proposals for Grazing Lease. Although our office has great interest in this topic due to the potential effects of domestic sheep grazing on Mattly and Conway Ranches to the state and federally endangered Sierra Nevada bighorn sheep (*Ovis canadensis sierrae;* Sierra bighorn), we are not able to attend the meeting in person.

As stated in our June 17, 2016 comment letter on the April 2016 draft Conway Ranch Strategic Facility Plan, we request the County simply identify livestock grazing as the master vision for Conway/Mattly Ranches as opposed to limiting consideration to only domestic sheep operations.

Since the current grazing lease expires on November 15, 2017, we appreciate the interest and need for the County to pursue this matter expeditiously; however, we respectfully request the County reschedule this discussion in order for us and California Department of Fish and Wildlife (CDFW) to identify other options of consideration by the Board and to be able to attend the meeting in person. Please note, we would also like to work together in helping craft language for any future Request for Proposals for livestock grazing on Mattly and/or Conway Ranches.

Our main concern is the potential for disease transmission, specifically pneumonia, between domestic sheep and Sierra bighorn due to the proximity of Mattly and Conway Ranches to the Mt. Warren herd unit. The Mt. Warren herd unit was identified in the 2007 Sierra Nevada Bighorn Sheep Recovery Plan (Service 2007) as a herd unit essential for the recovery of the subspecies and it is also designated critical habitat. Our partner in recovering the Sierra bighorn, CDFW, has been successful in increasing the abundance and distribution of Sierra bighorn

Board of Supervisors

through augmentations and translocations. These efforts have been necessary in order to ensure the persistence of the subspecies throughout its historic range. A disease outbreak in the Mt. Warren herd unit would not only affect animals occupying this area but it could also spread to animals in the surrounding area known as the Northern Recovery Unit. Currently, there are two other herds located in the Northern Recovery Unit, one of which is a newly introduced herd located in Yosemite National Park.

I believe that recovery of Sierra bighorn is a shared goal among the County, CDFW and the Service. The Service believes that recovery of this species is within reach. However, presently the current management of Mattly and Conway Ranches does not provide for the effective separation of domestic sheep and Sierra bighorn. This lack of effective separation limits our ability to recover Sierra bighorn and puts the recovery effort within the Northern Recovery Unit at risk.

Although Lee Ann Carranza, Assistant Field Supervisor, reached out to the County and F.I.M. Corporation to offer a meeting in which to facilitate information sharing on the subject, we have not scheduled any meetings. However, we remain interested and our offer to meet remains. Ideally such a meeting would include CDFW. If you have any questions regarding our comments or would like to arrange a meeting, please contact me or Lee Ann Carranza at (775) 861-6300.

Sincerely,

Carolyn Swed, Acting Field Supervisor

ccs:

Tony Dublino, Mono County, South County Office, Mammoth Lakes, California F.I.M. Corporation, Smith, Nevada

Dr. Tom Stephenson, Senior Environmental Scientist, California Department of Fish and Wildlife, Inland Deserts Region, Bishop, California

Literature Cited

U.S. Fish and Wildlife Service. 2007. Recovery Plan for the Sierra Nevada Bighorn Sheep. Sacramento, California. xiv + 199 pages.

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OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

💻 Print

MEETING DATE February 21, 2017

Departments: Public Works

TIME REQUIRED	2.5 hours (15 minute presentation;2.25 hour discussion)	PERSONS APPEARING
SUBJECT	Direction to Staff re Conway Ranch Request for Grazing Proposals	BEFORE THE BOARD

Tony Dublino

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

Presentation by Tony Dublino regarding potential issuance by County of a Request for Proposals for Grazing at Conway Ranch.

RECOMMENDED ACTION:

Receive presentation and provide direction to staff regarding the issuance of an RFP for grazing on Conway Ranch, including, but not limited to, one of the following options: 1. Direct staff to prepare RFP for sheep grazing at Conway and Mattly Ranch. Any such proposal will require indemnification as well as applicant funding of any necessary CEQA. Once prepared, present to Board for approval, posting and publishing. 2. Direct staff to prepare RFP for cattle grazing at Conway and Mattly Ranch. Any such proposal will require indemnification as well as applicant funding of any necessary CEQA. Once prepared, present to Board for approval, posting and publishing. 3. Do not direct staff to prepare an RFP allow current grazing lease to expire without subsequent lease in place.

FISCAL IMPACT:

None at this time.

CONTACT NAME: Tony Dublino

PHONE/EMAIL: 760.932.5453 / tdublino@mono.ca.gov

SEND COPIES TO:

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR PRIOR TO 5:00 P.M. ON THE FRIDAY 32 DAYS PRECEDING THE BOARD MEETING

MINUTE ORDER REQUESTED:

🔽 YES 🗖 NO

ATTACHMENTS:

Click to download

D <u>Staff Report</u>

Public Correspondence Received

History

Time	Who	Approval
2/16/2017 4:01 PM	County Administrative Office	Yes
2/16/2017 2:32 PM	County Counsel	Yes
2/16/2017 2:27 PM	Finance	Yes



MONO COUNTY DEPARTMENT OF PUBLIC WORKS

Post Office Box 457 • 74 North School Street • Bridgeport, California 93517 (760) 932-5440 • Fax (760) 932-5441 • monopw@mono.ca.gov

Jeff Walters, Public Works Director

Garrett Higerd, PE County Engineer

Date: February 21st, 2017

To: Honorable Board of Supervisors

From: Tony Dublino, Environmental Services Manager

Subject: Conway Ranch Grazing Request for Proposals (RFP)

Recommended Action: Receive presentation and provide direction to staff.

- 1. Direct staff to prepare RFP for sheep grazing at Conway and Mattly Ranch. Any such proposal will require indemnification as well as applicant funding of any necessary CEQA. Once prepared, present to Board for approval, posting and publishing.
- 2. Direct staff to prepare RFP for cattle grazing at Conway and Mattly Ranch. Any such proposal will require indemnification as well as applicant funding of any necessary CEQA. Once prepared, present to Board for approval, posting and publishing.
- 3. Do not direct staff to prepare an RFP allow current grazing lease to expire without subsequent lease in place.

Fiscal Impact: None at this time.

Discussion: The County's Conway and Mattly Ranches (CR) are grazed by domestic sheep under a lease agreement that expires in November of 2017.

The lease generates approximately \$19,000 per year in revenue from the lessee FIM Corporation, and the County pays FIM Corporation \$6,000 per year for "Irrigation Specialist" Services for meadow irrigation, resulting in a net revenue of approximately \$13,000 per year for grazing-related activities at CR.

The current lessee FIM has consistently expressed interest in continuing to graze sheep at CR, and would like to extend the lease. However, as a government agency, the County must engage in a Request for Proposals (RFP) or bid process under its own rules (Mono County Code section 3.050.020 and 030) and under State law (Government Code section 25537.)

And grazing at CR is not without controversy. Specifically, interested parties and wildlife agencies have raised potential environmental impacts to special status species such as Bi-State Sage Grouse and Sierra Nevada Bighorn Sheep.

Under state law, the County must review the impacts of any proposed grazing activities under the California Environmental Quality Act before committing itself to a final course of action, and adopt any necessary mitigation measures. Recommended Actions 1 or 2 would provide direction for staff to develop a thoroughly mitigated RFP for a given type of livestock, and bring the final RFP back to the Board for approval. Under these recommended actions, any RFP would require respondents to fund the requisite CEQA effort, as well as indemnify the County against any associated litigation.

<u>History</u>

The County acquired Conway Ranch through a variety of grant funds in 1998. Shortly thereafter, the County resolved (R00-29) to lease parts of the Conway Ranch for the purpose of sheep grazing. A request for proposal process initiated, and was won by FIM Corporation.

During the ensuing years, sheep grazing on Conway become a regular activity during the summer and fall. Grazing activities have been controlled by a series of five-year leases, the most recent of which will expire in November 2017. The leases include terms and conditions, and those terms and conditions have generally been upheld and the overall impact of the sheep on Conway Ranch has been minimal. The meadows are in good to excellent condition, the livestock is light on the land, and each spring there is little sign of their presence from the preceding grazing season.

Despite the absence of significant physical environmental impacts, the presence of domestic sheep on Conway Ranch has become a critical concern for wildlife agencies involved with the recovery of Sierra Nevada Bighorn Sheep (SNBS) due to the issue of disease transmission between domestic sheep and SNBS.

This is a highly controversial matter that has been debated in front of the Mono County Board numerous times. In 2007, during a debate on the designation of critical habitat the Board (R07-81) took issue with the taxonomy of SNBS, and requested that federal grazing allotments be removed from the critical habitat designation, in an effort to preserve the County's agriculture and grazing economy (all those allotments have since been closed to grazing). Conway Ranch was not mentioned in the Board's 2007 Resolution.

In the last five years, the Board has heard comments on the subject of disease transmission from many agencies and individuals as CR planning efforts and the creation of the Conservation Easement have moved through the public process.

Through the process creating the CR Conservation Easement, references to 'sheep grazing' were changed to a more generic 'livestock grazing,' largely in deference to the concerns over disease transmission.

Throughout 2016, public outreach relating to Conway generated the concept that grazing activities at Conway should continue, thus giving rise to the item today.

Today's Recommended Actions

Leading up to today's item, the County distributed a Request for Letters of Intent from grazing operators that would generally describe their approach to grazing and proposed mitigations at CR. The County received seven responses. Two were from sheep operators, and five were from cattle operators.

County staff met with USFWS and CADFW to discuss the various mitigations proposed in the letters, to obtain a sense of whether those mitigations would result in less than significant

impacts to Bi-State Sage Grouse and/or SNBS. Agency representatives did not offer conclusive input regarding proposed cattle mitigations and whether they would effectively eliminate potential impact to Bi-State Sage Grouse, but did state that the mitigations proposed for sheep grazing would not mitigate risk of disease transmission to SNBS, and suggested that no mitigations whatsoever could mitigate that potential impact.

The Recommended Actions reflect what staff believes are the most commonly held public opinions about what should happen at CR in the future – that is, sheep grazing, cattle grazing, or no grazing.

The Conservation Easement for Conway Ranch allows all three of these recommended actions. It does not require the County to graze livestock, but permits livestock grazing. The Easement primarily requires the County maintain conservation values, which will greatly influence grazing activities as well as non-grazing activities. Any activities conducted on CR must comply with the Conservation Easement, which can be viewed in its entirety at the following web address:

http://www.monocounty.ca.gov/facilities/page/conway-ranch-conservation-easement

If you have any questions regarding this item, please contact me at (760) 932-5453.

Respectfully submitted,

Dullino

Tony Dublino Environmental Services Manager



Range of Light Group Toiyabe Chapter, Sierra Club Counties of Inyo and Mono, California P.O. Box 1973, Mammoth Lakes, CA, 93546 Rangeoflight.sc@gmail.com RELEIVED

February 2, 2017

3 - 6 2017

OFFICE OF THE CLERK

Mono County Board of Supervisors PO Box 715 Bridgeport, CA 93517

RE: Conway Ranch Sheep Grazing Renewal

To: The Honorable Mono County Board of Supervisors:

The Sierra Club Range of Light Group and its 394 members have a deep and active interest in the health of the Eastern Sierra's lands, waters and wildlife. Our Executive Committee met recently and discussed the issues related to the management and protection of the Conway and Mattly Ranch properties.

Our group strongly encourages the Mono County Board of Supervisors to support the purpose and intent of both the Endangered Species Act and the Sierra Nevada Bighorn Sheep Recovery Plan by not renewing the lease for domestic sheep grazing on Conway and Mattly Ranch properties. There is a large body of evidence published in the peerreviewed literature that documents the fatal health risk that domestic sheep pose to the Sierra Nevada Bighorn Sheep. We urge the Board to acknowledge the science and support the state and federal agency experts who ask for sheep grazing on these properties to end. We believe the best use of this property is to manage it to emphasize the protection and conservation of wildlife and wildlife habitat while retaining the pastoral viewshed that is highly valued by the community and the public. If the County continues grazing, we encourage the County to consider livestock other than domestic sheep or goats.

We hope you give the CDFW and USFW staff a chance to give a presentation on the current science and research data to the Board and the public before determining whether an RFP for grazing is warranted.

Sincerely,

"g- Lauton

Lynn Boulton, Chair Range of Light Group

Conway/Matley Ranch

Mono County, California

Legal description of sustainable agriculture in the United States, [U.S. code Title 7, Section 3103]: states

Sustainable agriculture means an integrated system of plant and animal production practices having a site specific application that will over the long term:

Satisfy human food and fiber needs

Enhance environmental quality and natural resources based on which the agriculture economy depends

Makes the most efficient use of nonrenewable resources and on-farm resources and integrate, where appropriate natural biological cycles and controls

Enhance the quality of life for farmers and society as a whole

The history of the Conway/Matley ranch is tied to sheep and as was presented in a proclamation that USED to hang in the courthouse in Bridgeport, California. I feel it is necessary to highlight a few issues to get a full understanding of the issues.

History of sheep in California

The first sheep came to the United States in 1496, on the second voyage of Christopher Columbus. The first to the Sierras came at about 1800. The start however, was in 1769 when Franciscan Fathers of the early Spanish Colony commenced raising sheep at their missions. By 1819, the total on 19 mission ledgers showed a record population of 186,233 sheep. When California became a state in 1850, there were less than 20,000 sheep in the entire state. This changed again during the gold rush and the need to feed the hungry miners. They, at one was paying from \$7 to \$16 per sheep. Large numbers of sheep were brought in to furnish mutton, the peak in 1856, when more than 200,000 sheep were driven in to California and Nevada from New Mexico, the Midwest, and from Mexico. According to "historical" documents, the sheep were easy to handle, they had an abundance of pasture, extremely hardy and disease was "uncommon". In 1865, a writer pointed out that there was vast pastures on both sides of the Sierra Nevada where herds could be grazed. He suggested that the "sheepmen" follow the old Spanish trail. The use of the Sierra was the main factor in developing the sheep industry. They found that taking if they took lean and weak sheep taken into the hills and high pastures to graze they returned fat and strong. The herders followed the range across the range to the south end of INYO county, then along the eastern edge of the Sierra, re-crossing the crest to the west side in the vicinity of Yosemite Valley and sometimes as far north as Lake Tahoe, then back along the foothills to the central valley and winter lambing grounds. It was during these days that a penniless young man by the name of John Muir, took, on June 3, 1869, 2050 sheep into the Southern Inyo. Working as a sheepherder for \$30.00 a month, with an assistant, a few dogs and a rifle, he started across the Sierra. He began to gradually move the sheep higher and higher and thru successive belts of forests as the snow melted, stopping for weeks at a time at the best meadows. He mapped and documented the Sierra on his summer as a shepherd. He was so taken by the Sierra that he became an advocate for their preservation. In 1903, after much correspondence, President Theodore Roosevelt, agreed to a visit and camping trip to view for himself, John Muir's mountains. He wrote in his journals about John Muir and their 4 day camping trip. The President wrote this: I was interested and a little surprised to find that, unlike John Burroughs, John Muir did not like birds, their songs of even most animals. What he like was the geology, the rocks, the canyon and the ruggedness of the Sierra Mountains. He climbed a number of mountains including Cathedral Peak, Dana Peak and hiked the old Indian trail down Bloody Canyon to Mono Lake. In John Muirs mind, the animals were a detriment to the natural world. He however wanted to save "HIS" home, The Sierra, from man.

Disease Transmission

Dr. John Boyntons letter told a lot about the cattle industry and the destruction that they can cause to land and riparian areas, and how they can affect the water

quality. The USFWS says that disease transmission between domestic and wild bighorn sheep is a possibility. What they do not tell you, however, is that the pneumonia between the two species can only be done by the exchange of fluids. They have to touch noses. In the 248 years that domestic sheep have been in Nevada and California, there have NEVER been any documented cases of disease transmission in the wild. There have been die-offs where there were NO domestic sheep in the area. The facts about disease transmission is one that will NEVER be won. Scientist and Doctors from all walks of life and from all over the country, have been studying the prospect of transmission for decades. The USFWS and CDFW, will never change their story on the issue, regardless of how many other reports are done. They have an agenda. There have been studies done in Idaho, Wyoming and Colorado, where the bighorn are captured, transported, tagged and placed in tiny 10x10 corrals with domestic sheep. It is true the in the case of the bighorn, they died. Yes, they had pneumonia. WALA, the "best available science" proves that disease transmission is possible. The USFWS have the "proof" they need to make things happen, and at this point it cannot be dis-proven. What you do not see however is the actual necropsy report of the death of the bighorn, in total. The necropsy will find that although the bighorn died from pneumonia, it was the DNA of it's own system that killed it, not the domestic. When a bighorn is stressed its natural immune system fails and the bighorn can get pneumonia from the pathogens it carries in its lungs. The whole of the issue is the "he said, she said" situation. No side will win. Both sided have their own proof and evidence. Especially the bighorn. Their own documents in the recovery plan, clearly shows in the maps that the Northern Unit has very little suitable habitat. [maps included] This does not mean that we all have to buy into what they say is fact. I find it ironic that one of the studies that CDFW highlights in their research { California Department of Fish and Game, 2009], was co-written by sheep men and women. Joe Echinique, Nancy East, Amy Baumer, Marcy Haworth, Marianne F. Leinassar, Chris Popouchis, Tom Stephenson, DianeWeaver and Genny Wilson.

Marianne F. Leinassar is the President of FIM corporation, and the daughter if Fred Fulstone, the very people whom you are trying to kick off the Conway/Matley. Funny huh. To find a leasee, who, if cattle are preferred, would be able to absorb the cost to do all the necessary changes to the property, would be rare. To install the fences needed to do proper rotation, to fence off the water suppy to insure water quality, to fence off the watering facilities that would be needed to be added, could be a challenge. The sage grouse issue of NO fences would also cause a problem as well as the mule deer corridor. To get all the necessary changes through the ESLT easement would be another. The property is not big enough to have the necessary AUMs to make it profitable for anyone.

There are ways to keep sheep on the property IF it necessary, which FIM has done in the past. They have changed season of use to earlier in the season, so as not to be there when the bighorn are in RUT. They have made all changes that the county have imposed. The Conway/Matley ranches are the only losers in this situation. The easement is forever, not until next year or 10 years down the road. There are No easy fixes here, and no MONEY to fix them either.

Thank you for your time on this matter, Marilyn "MAX" Symonds, 1-15-2017 330 W. Nye Ln. #1 Carson City, Nevada 89706 1-775-430-2380

Conway/Matley Ranch

Mono County, California

The problems on the Conway/Matley Ranches, unfortunately cannot and will not ever be solved. In reality the property cannot be returned to its previous glory, because simply there is not enough MONEY. The meadows will not look the way they did in the 50s and 70s. Time and circumstances have changed too much and there are too many restrictions now. If it were only one problem for the ranches maybe there would be a chance. Unfortunately that is not the case. The largest issue, if there were only one, would be as I said above, if financial. The County does not have the resources to fix and maintain all the issues. The water situation is maybe the biggest problem, because there is no way to deliver the 6cfs of water that is due to them for the ranch. The ditches would require not only re-aligning the ditch, but repairing many places on the ditch on the Chichester portion and on the Webb portion. The BLM/FS portion not only needs to be cleared but also repaired just to get the water down to the ranch, not to mention all the work on the existing gates on the Virginia Creek side. There are many gates that need repaired on the ranch and more that need to be added in order to get the water distributed to the places on the ranch where it will be beneficial. The existing ditches need to be cleaned and maintained each year. FIM Corporation and NRCS in 2015 did a day trip to Conway Summit to research what could be done and what it would cost to repair only that part of the ditch. NRCS said that the things that needed to be done are considered as maintenance and they could not do them at this time. Mr Webb would not allow FIM on

his property to maintain or repair work to the ditch from the moles and ground squirrels that had burrowed into the sides of the ditch banks causing the ditch to flood onto the Virginia Lakes road and onto Conway Summit road at 395. The ranch itself, {CONWAY}, is in desperate need of plowing and re-seeding the grasses in the irrigated meadow that are on the property. The grasses are so old they are RANK, which means nothing wants to eat the grass because it is very tough. Like sawgrass. The historical buildings need to be at least stabilized to secure their future. When you take into consideration the sage grouse and bighorn sheep issues, you have a huge mess. The restrictions which have been introduced for these wildlife, will have to be taken into consideration before anything can be done, IF the money could be found. The next issue for the ranches would be the conservation easement itself, and the restrictions from it. The Federal Government wants their way, the State of California wants their way, and the County is tied up in litigation. Where is the answer for Conway/Matley? FIM Corporation has spent over \$25,000. of their own money repairing and maintaining the ditches and waterways since they started leasing the ranches. They pay a hefty lease each year and it is going UP each year. You asked for EIS money, ditch work, and maintenance be burdened on the lease. That is not right. The BOS is seeking bids for the lease of the property and is considering cattle to graze instead of sheep because they have been told that there is a huge risk to the bighorn sheep that have been located in the Lundy Canyon area. The disease transmission issue with bighorn has been the source of litigation for over 30 years and will not be resolved anytime soon. The issues could be argued from both sides because there are as many doctors and scientist that side with the governments as there are that

say the claims they have are false. I have personally done research on both sides of the issue and know from my own research that there is not a clear answer. They only answer at this time, is that the State and Federal governments have an agenda, they have been told what they have to do, and they want it done, their way. The money that FIM pays to the County has to be used to pay the maintenance fees to ESLT for the easement, so there is not monetary value for the County. FIM does what it can with what they have to work with, but for the good of the property it may not be enough. They are only able to get about 2cfs of water to the property instead of the 6 it is supposed to use. This is in violation of the Walker River Water Authority, and the 6cfs is not going to beneficial use as intended, because it cannot make it to the ranch. Under the present condition and restrictions there will never be able to achieve the "desired conditions". Even if the County were able to find the money to make ALL the repairs and modifications needed to get the proper water, and grasses in, who would do the maintenance and upkeep. The county is looking to maybe put cattle on the property, and that is an option, but when you add in the fences needed, the watering areas needed and the water quality issues involved, it may not be feasible at all. The amount of AUM's would not offset the cost to do the EIS and the modifications. The benefit just is not there financially. The loser in this whole ball game, is of course is the Conway/Matley ranches. They are the ones that are in the middle of this mess, and there is no clear way out. With the easement in place, if would even be almost impossible to sell the property, because the easement is FOREVER. The restrictions are FOREVER and the wildlife issues are basically FOREVER. Not a lot of solutions here. I personally have spent, hundreds of hours and days researching the die-off of bighorn sheep,

disease transmission issues between domestic and wild sheep, the issues surrounding sage grouse, bighorn and mule deer. I have analyzed the situation from all the angles and there is really no answer for MONO COUNTY. You can't even use the historical value, because of All the restrictions that have been imposed. Mono County has virtually built themselves into a corner and the walls are too thick to get out.







file:///C:/Users/owner/Desktop/bighorn_CA.gif

1/20/2015









Map 3



TO: Mono County Board of Supervisors FROM: Dr. Dale Matson REGARDING: Sheep Grazing On Conway Ranch DATE: 02-11-17

Dear Supervisors,

While I am a resident of Fresno County, Bighorn Sheep are of great interest and concern for me. Bighorn Sheep also live on the western slopes of eastern Fresno County in the Sierra Nevada Mountains. What puts bighorn sheep at risk in the Eastern Sierra could put the Fresno County sheep at risk also. Many of these sheep reside in Kings Canyon National Park, a large part of KCNP being in Fresno County. I have spent weeks backpacking in the higher elevations solely for the purpose of photographing these magnificent, scarce and elusive sheep.

These endangered sheep live on the razor's edge between being reestablished and becoming extinct. The two biggest impediments to their flourishing are predation by mountain lions and the spread of disease from domestic sheep and goats. The Sierra Nevada Bighorn Sheep have no immunity and deadly pathogens carried by domestic sheep and goats quickly spreads through a SNBS herd unit. The Conway/Mattley Ranch near Lee Vining is close to both the Mt. Warren and Mt. Gibbs herd units.

These sheep need advocates especially because they are endangered. The mountain lion, which is an apex predator and has no natural enemies, has "Protected" status in California even though it is neither endangered nor a threatened species.

I have included a link to a blog article I wrote after visiting the Mt. Gibbs herd on a Mono Lake Committee Field Seminar last September. We had to climb to 11,000' just to get close enough for photographs of the sheep with telephoto lenses. We also visited the Lundy Canyon area the following day.

http://midsierramusing.blogspot.com/2016/09/the-mono-lakecommittee-field-seminar_13.html

I also had the good fortune of getting close enough to a group of SNBS from the Mt. Langley herd to take close up photographs of these magnificent animals. It was one of the best experiences I have had in my 70 plus years of life. I wrote about this rare opportunity last year also on my blog including some of the closest photographs ever of these sheep. I think the photographs do a lot in explaining the difficult life they lead. They are both hardy and fragile. In the last photograph you can see evidence of a mountain lion attack on the young ram. http://midsierramusing.blogspot.com/2016/04/the-endangered-sierranevada-bighorn.html I hope each of you has had the opportunity to see and become familiar with these animals that are residents of your county. For me it was a privilege to see and photograph the Sierra Nevada Bighorn Sheep. I cannot attend your meeting but hope that you also will decide to advocate for the endangered bighorn sheep.

Respectfully, Dr. Dale Matson 9223 N. Ashford Ave. Fresno, CA 93720 (559) 434-9549

Helen Nunn

From: Sent: To: Subject: Shannon Kendall Monday, February 13, 2017 7:51 AM Helen Nunn FW: Sierra Nevada Bighorn Sheep

-----Original Message-----

From: Roxanne Romo [mailto:roxanneromo1@gmail.com] Sent: Saturday, February 11, 2017 2:22 PM To: Shannon Kendall <<u>skendall@mono.ca.gov</u>> Cc: Stacy Corless <<u>scorless@mono.ca.gov</u>>; Larry Johnston <<u>ljohnston@mono.ca.gov</u>>; Fred Stump <<u>fstump@mono.ca.gov</u>>; Bob Gardner <<u>bgardner@mono.ca.gov</u>>; John Peters <<u>jpeters@mono.ca.gov</u>> Subject: Sierra Nevada Bighorn Sheep

February 11, 2017

Dear Mono County Supervisors,

I urge you to protect the local bighorn sheep populations of the Lundy Canyon area. The county has a responsibility through it's conservation easement to protect wildlife, not domestic sheep. If the domestic sheep pose a threat then they don't belong there plain and simple.

Our public lands and the environment are currently being threatened on so many fronts. Please do your part to preserve our little corner of this planet.

Respectfully,

Roxanne Romo Crowley Lake

Helen Nunn

From:	Helen Nunn
Sent:	Friday, February 10, 2017 3:50 PM
То:	Helen Nunn
Subject:	Sierra Nevada Bighorn Sheep

Subject: Sierra Nevada Bighorn Sheep

To: The Mono County Board of Supervisors,

In the interest of protecting the Sierra Nevada Bighorn Sheep, I implore you to please find an alternative to domestic sheep grazing on the Conway/Mattley Ranch near Lee Vining, in close proximity to the Mt. Warren and Mt. Gibbs herd units, close enough to cause them to die of pneumonia. Domestic sheep carry pathogens that kill bighorn, the reason for their rapid demise, and **it is imperative to keep domestic and bighorn sheep apart**. Researchers and volunteers have spent countless hours and effort to keep the Sierra Nevada Bighorn Sheep alive and the herds growing. DO NOT impede this progress. DO NOT allow the animals to suffer because of fiscal concerns.

Find an alternative to domestic sheep grazing on the Conway/Mattley Ranch, please!

--Jeanette Smith jeanette@wildhorsedesign.biz

Helen Nunn

From: Sent: To: Subject: Howard Whitaker <hjameswhitaker@att.net> Friday, February 10, 2017 3:09 PM Helen Nunn Fwd: Terminate Domestic Sheep Grazing

------ Forwarded Message ------Subject: Terminate Domestic Sheep Grazing Date: Fri, 10 Feb 2017 14:56:30 -0800 From: Howard Whitaker ≤hjameswhitaker@att.net> To:skendall@mono.ca.gov

Dear Mono County Board of Supervisors,

For decades, I have been a frequent visitor to Mono County and am deeply appreciative of its many attributes.

I have for many years been concerned about the health and survival of the Lundy Canyon herd of bighorn sheep, particularly the devastating effect of pathogens transmitted by the grazing of domestic sheep on the Conway/Mattley Ranch.

With a simple decision by you supervisors to cease the grazing of domestic sheep on the ranch, an enormous threat to the bighorn herd can be eliminated and for the sake of both the bighorn and all of us who value Mono Country, I ask that you please make that decision.

Sincerely,

Howard Whitaker 2041 Campton Circle Gold River, CA 95670-8301
From: Sent: To: Subject: Shannon Kendall Monday, February 13, 2017 4:12 PM Helen Nunn FW: Conway Ranch and Sierra Nevada bighorn sheep

Dear Mono Supervisors:

I am writing to plead that Conway Ranch no longer be available for domestic sheep or goat grazing.

I recently retired as Chief Scientist of the Pacific West Region of the National Park Service. As a young wildlife biologist, I was first assigned to represent NPS on the Sierra bighorn recovery effort back in 1980, and continued in that capacity until 2014. I am a co-author of the Sierra Nevada Bighorn Sheep Recovery Plan published by U.S. Fish and Wildlife Service and California Department of Fish and Wildlife. I continue to serve on the science advisory team to the FWS bighorn recovery effort.

During those more than three decades, our team strived to bring bighorn back from the brink of extinction. There have been many discouraging, if not frightening moments, but as I'm sure you know the bighorn are now doing very well along a substantial stretch of their original range. This is a great success, but also a very precarious one. As wildlife biologists have learned to their great dismay, a robust population of bighorn can collapse over the span of a year or two when exposed to several diseases frequently carried by domestic sheep and goats. This has happened most recently in the Mojave Desert just in the past few years.

At the moment, Conway Ranch represents the greatest threat to Sierra bighorn along their entire extent. I will not belabor the details because I am confident all that information has been made available to you. Please do the right thing.

Sincerely, David Graber

David Graber, Ph.D. 40854 Oak Ridge Drive Three Rivers, CA 93271 <u>dbear1948@gmail.com</u>

From:Shannon KendallSent:Tuesday, February 14, 2017 7:13 AMTo:Helen NunnSubject:FW: For 2/21/17 Mono Co. Board of Supervisors meeting; re Sierra Nevada Bighorn
Sheep

From: Jeff W. [mailto:jeffwyneken@gmail.com]
Sent: Tuesday, February 14, 2017 12:54 AM
To: Shannon Kendall <<u>skendall@mono.ca.gov</u>>
Subject: For 2/21/17 Mono Co. Board of Supervisors meeting; re Sierra Nevada Bighorn Sheep

Dear Ms. Kendall:

I'm writing to recommend the termination of domestic sheep grazing at Conway Ranch due to the proximity of the grazing allotment to the Lundy Canyon bighorn sheep population and the documented danger of lethal viral infection from domestic sheep to wild sheep. Just one encounter between an infected sheep and a bighorn could result in the swift extirpation of the entire bighorn population.

Bighorn sheep have been seen in the vicinity of the grazing area. As the Sierra Bighorn Sheep is endangered, all prudent steps should be taken to protect it from extinction.

Much work has been done to protect and help recover these populations. Let's be proud and conservative of our magnificent Mono legacy and protect these native animals in their last holdouts.

Thank you.

Jeff Wyneken PO Box 201 Lee Vining, CA 93541

From: Sent: To: Subject: Shannon Kendall Tuesday, February 14, 2017 7:13 AM Helen Nunn FW: Bighorn sheep

-----Original Message-----From: Debbie Boucher [mailto:dboucher2008@gmail.com] Sent: Monday, February 13, 2017 5:18 PM To: Shannon Kendall <<u>skendall@mono.ca.gov</u>> Subject: Bighorn sheep

Please do everything within your power to ensure the health of this species. If that means restricting sheep from grazing at Conway Ranch, than please do that.

Debbie Boucher, Mammoth Lakes resident

Robert R Tyson 211 Mesquite Rd Bishop CA 93514 (650) 475-6293

Sierra Nevada Bighorn Sheep and the Mono County Board of Supervisors -Public Meeting for Feb 21 2017

To the kind attention of the Mono County Board of Supervisors,

Stacy Corless Larry Johnston Fred Stump Bob Gardner John Peters

Shannon Kendall, Clerk of the Board

I write to support you in your deliberations to find an equitable and positive solution to damaging interactions between endangered Sierra Nevada bighorn sheep and domestic sheep in Mono County when the Board meets February 21 2017. I cannot attend this meeting, but I very much wish to contribute my thoughts.

As much as I am concerned that ranchers need to continue sheep grazing in areas common to the bighorn, I am at least as concerned and wish to advocate for the welfare of the bighorn which are fatally affected by the pneumonia they acquire by having contact with domestic sheep. The present state of affairs drives a one-way decline for the bighorn.

We should do all we can to conserve and support native animal populations, especially so magnificent a species as the bighorn. An equitable solution *can* be reached that will conserve and preserve both the bighorn and those who depend on domestic sheep. Let me emphasize this. My passionate interest, at the same time involved and passionate *and* detatched and scientific, may lie first with the bighorn, but I no less believe in the preservation and conservation of the ways of life and livelihoods long practiced around domestic sheep husbandry.

I have fiscal concerns for the County too. Were I a Mono County taxpayer (I live in Bishop) I would worry about fiscal damage if Mono County does not vigorously act to prevent contact between bighorn and domestic sheep. Consequences I see for Mono County and its tax base include paying for impact research and regulatory review, probably lengthy, and litigation inevitable if the County does not provide for bighorn recovery. I ask, in contrast to maintaining status quo, is not supporting the bighorn a prudent policy and wise use of County monies? Does not keeping things as they are not beg other pragmatic aspects of the high side to conserving both populations such as tourism and the revenue support it provides?

I am presently in Italy, where surprisingly I encounter even at this distance awareness of the bighorn, of its majesty, and of its plight. Italians would say that if the County does not protect such an iconic and beautiful creature it would be considered guilty of *mancanza di soccorso*, failure to care for one in need. This is a criminal offense under Italian law.

I heartily commend you to constructing the solution to preserve these magnificent and needed creatures and also to conserve productive domestic grazing. May both thrive, now and for our future.

Respectfully,

Solo Type

Robert R Tyson

From:	Shannon Kendall
Sent:	Wednesday, February 15, 2017 9:14 AM
То:	Helen Nunn
Subject:	FW: Bighorn Sheep, Domestice Sheep and Conway/Mattley Ranch

From: diane dickey [didickeybooks@gmail.com]
Sent: Wednesday, February 15, 2017 7:17 AM
To: Shannon Kendall; Stacy Corless; Larry Johnston; Fred Stump; Bob Gardner; John Peters
Cc: ginniebeth@me.com
Subject: Bighorn Sheep, Domestice Sheep and Conway/Mattley Ranch

good morning shannon kendall, et al,

im writing to ask mono county board of supervisors to please consider a healthy solution for the use of the conway ranch. the proverbial term, "can't we all get along?" be applied to the issue of our endangered sierra neveda bighorn sheep and domestic sheep grazing - can we find a solution to protect all?

i'm confident in the board, the s.n. bighorn sheep foundation, and citizens of california to allow the recovery of the bighorn sheep to continue & flourish.

i'm asking for a good solution, for the beautiful wildlife in the area to be sustained & valued - good news for nature, good news for human beings.

i'm dedicated to the furture of the bighorn sheep, they are a treasure for our wonderful state. i hope you can decide in their favor and help be their good steward, they need your assistance & they need your help.

appreciate your time, many thanks diane dickey supporter of the restoration of the sierra neveda bighorn sheep and conserving, protecting, managing, california's wildplaces. Mono County Board of Supervisors P.O. Box 715 Bridgeport, CA 93517

Hi,

I am writing to ask you to please consider stopping the practice of sheep (or goat) grazing on the Conway Ranch near Lundy Canyon. It can be very dangerous to the resident, endangered big horn sheep in the area due to disease transfer.

There is ample evidence that disease transfer is a real threat and I hope the Fish & Wildlife Services will convince you to stop sheep grazing on the Conway ranch.

Sincerely,

Dennis Switick 1673 Grant Road Mountain View, CA

From: Sent: To: Subject: Shannon Kendall Thursday, February 16, 2017 7:10 AM Helen Nunn FW: Conway Ranch/Sheep Grazing

From: Lynn Boulton [mailto:amazinglynn@yahoo.com]
Sent: Wednesday, February 15, 2017 9:11 PM
To: Shannon Kendall <skendall@mono.ca.gov>
Cc: Fred Stump <fstump@mono.ca.gov>; Larry Johnston ljohnston@mono.ca.gov>; Stacy Corless
<scorless@mono.ca.gov>; Bob Gardner <bgardner@mono.ca.gov>; John Peters <jpeters@mono.ca.gov>
Subject: Conway Ranch/Sheep Grazing

February 15, 2017

Re: Conway/Mattly Ranch - Sheep Grazing

Honorable Mono County Supervisors:

I hope you decide to take domestic sheep grazing off the table at the February 21 board meeting, which will help protect the endangered Sierra Nevada Bighorn Sheep. They are a species unique to the Eastern Sierra and it is a thrill for visitors and locals alike to see them in the wild. I'm glad the CDFW will be presenting the science and explaining the risk that having domestic sheep within 0.5 mile of the bighorns poses to the entire northern unit: the Mt. Gibbs herd, the Yosemite herd, and the Mt. Warren/Lundy herd. The County was listed as a participant in the 2007 SN Bighorn Sheep Recovery Plan. Yet the County has continued sheep grazing on the property contrary to the recommendations in the plan, convinced that domestic sheep could be managed to prevent contact. Now, ten years later, there is much more evidence that nothing short of separation by miles will safeguard the bighorns.

I wish there were mitigations that would allow sheep grazing without putting the bighorns at risk, but there aren't any that I have discovered in my research. Yes, the County could enclose the two properties with double fencing where at least one is a very high fence, but fencing would create new problems for deer, antelope, and sage grouse and all other wildlife. Conway and Mattly ranches are located in a state designated wildlife corridor and essential habitat (see pgs. 75 and 82 of the CEHC Project report). Wildlife needs to be able to freely pass through to/from the Sierras and the Great Basin. It would also be unsightly and mar the viewshed. As a taxpayer I don't want to pay for that solution. I personally would like to see Conway and Mattly Ranches managed for maximum biodiversity and wildlife. This would be much more in line with the terms of the Easement, which was funded by grants that were to offset habitat losses in other parts of the state.

One argument for sheep grazing is to reduce thatch. As it is not really known if or the extent to which thatch truly presents a problem on the property, the Board should consult with experts (e.g. botanical, wildlife, range and ecological restoration specialists) who can make field evaluations and recommendations as to how the property can best be managed for wildlife habitat. Thatch could be burned, mowed, or grazed by other livestock, or adjustments in irrigation could remedy the issue. Sheep grazing is not required to address this question.

From a financial perspective, the County netted \$11-14,000/year on the current lease, which doesn't stimulate the local economy. Compare that to the SNBS recovery project estimated to cost \$22 million over 20 years that all taxpayers finance and which does stimulate the local economy through jobs and tourism. The bighorn sheep are a much better draw to tourists than domestic sheep. For all the talk of preserving the past, the County hasn't invested in preserving the barn, corral, and ranch buildings. They are in disrepair and in need of arrested decay. The ranch buildings clue us in to the ranching past. Without them no one would make that connection. The main draw of the Mono Basin is not the ranches of the Bodie era, but Mono Lake, Yosemite, fishing, fall colors, and the viewshed.

I urge the County to support the Endangered Species Act and to indicate that in Option 3 of Tony Dublino's request to the Board to let the sheep-grazing lease expire due to the risk posed to the SN Bighorn Sheep. The SN Bighorn Sheep recovery has been successful and their numbers have increased over time. The recovery of the entire population, however, will remain precarious as long as the county allows sheep grazing on Conway Ranch. Each species is part of the web of life, the biodiversity of this planet. Each species that becomes extinct creates a hole in that web and a vacuum for other species that consumed it. With each loss, the web of life unravels a little more and a little faster making it harder on the remaining survivors. The Endangered Species Act isn't just saving the listed species; it is saving all of nature.

Regards, Lynn Boulton Lee Vining

From:	Helen Nunn
Sent:	Thursday, February 16, 2017 8:40 AM
To:	Helen Nunn
Subject:	FW: Please protect bighorn sheep

------- Forwarded Message -------Subject: Please protect bighorn sheep Date: Tue, 14 Feb 2017 11:48:04 -0800 From: Jeff Holmquist <<u>jholmquist@ucla.edu</u>> To: Fred Stump <<u>stumpsuper@gmail.com</u>>, scorless@mono.ca.gov, ljohnston@mono.ca.gov, bgardner@mono.ca.gov, jpeters@mono.ca.gov

Dear Supervisors:

The Sierra Nevada Bighorn Sheep is an icon of our beloved mountains, and I think that you would agree that we should keep this magnificent animal here in our mountains for current and future residents to enjoy. Indeed, I think that your record of environmental stewardship will be a key part of your legacy as County Supervisors.

Our local bighorns have increased in abundance over recent years, but the continued survival of this species is by no means assured. Because of the serious disease risk that domestic sheep pose to bighorns, I urge you to end grazing by domestic sheep at Conway Ranch. This disease risk to mountain bighorns is well-established in the peer-reviewed scientific literature, has been identified as a key area of concern for Sierra Nevada Bighorn Sheep, and has been recognized by courts in other states.

Ending domestic sheep grazing at Conway Ranch makes good fiscal sense. If domestic sheep grazing continues, expensive environmental analyses will be required, and the matter may end up in court, which is good for no one.

Continued sheep grazing at Conway Ranch just doesn't pencil out, given the environmental and fiscal risks. Cattle grazing, though not completely benign from an ecological standpoint, would be far preferable to continued sheep grazing at Conway ranch.

I believe that Mono County should join with our neighbors in Yosemite National Park in doing everything possible to ensure the continued survival of our native bighorn sheep. In case you haven't seen it yet, here is a beautifully-filmed, nine-minute video that explains the uniqueness of our bighorn, outlines the disease threat posed by domestic sheep, and includes a remarkable reintroduction scene.

https://www.youtube.com/watch?v=qCf47SrgDss

Yosemite National Park and other federal and state agencies have taken heroic measures to save our bighorns-- shouldn't Mono County do everything that we can do to help out?

During the 1990s, my wife and I were deciding whether or not to relocate to Mono County. While visiting, we were lucky enough to see a herd of bighorn on the slopes of the Wheeler Crest. There was something primordial about that, seeing these animals gliding effortlessly across impossibly steep terrain. The bighorns also awed the First Peoples who lived here-- we know this from their rock carvings. Seeing the bighorns was an important experience for us, and, well,

here we are. We've been fortunate to see them a few times since, both in the Sierra and in the White Mountains. I can't imagine our mountains without them.

Thanks for your attention and for your service as Supervisors.

Sincerely,

Jeff Holmquist

Paradise/Swall Meadows

760-937-6317



California Program Office 980 Ninth Street, Suite 1730 | Sacramento, California 95814 | tel 916.313.5800 www.defenders.org

VIA U.S. MAIL AND EMAIL

February 16, 2017

Mono County Board of Supervisors c/o Shannon Kendall, Clerk of the Board P.O. Box 715 Bridgeport, CA 93517 <u>skendall@mono.ca.gov</u>

Re: Conway Ranch Grazing Lease Renewal

Dear Supervisors Corless, Johnston, Stump, Gardner and Peters;

We are writing on behalf of Defenders of Wildlife (Defenders) regarding management of activities, and specifically domestic sheep grazing, on the Conway Ranch property in Mono County. Defenders is a non-profit national environmental organization with 1.2 million members and supporters nationally, including 170,000 in California. Defenders is dedicated to protecting all wild animals and plants in their natural communities. To this end, Defenders employ science, public education and participation, media, legislative advocacy, litigation, and proactive on-the-ground solutions to slow accelerating species' extinction rates, halt the associated loss of biological diversity, and minimize wildlife habitat loss.

The Conway Ranch was acquired by Mono County (2000) using State of California and federal grants. This ranch is managed under the terms/conditions of a conservation easement designed to protect various wildlife species and their supporting habitat. Authorized livestock grazing is a discretionary land use allowed per this easement. The current grazing lease for this property authorizes domestic sheep grazing and expires in November, 2017.

Defenders is very concerned over continued domestic sheep grazing on the Conway Ranch property because of its proximity to habitat occupied by the endangered Sierra Nevada bighorn sheep (*Ovis canadensis sierrae*). There is undisputed professional documentation, as well as scientific research, which demonstrates that the bacterium *Mycoplasma ovipneumoniae* is transmitted from infected domestic sheep to bighorn sheep. This transmission occurs through direct contact between the two species, and almost always results in bighorn sheep death as the result of pneumonia.

Once transmitted, *Mycoplasma ovipneumoniae* spreads rapidly within a bighorn sheep population due to their gregarious behavior. This bacterium is known to be persistent for long periods of time, resulting in rapid mortality among bighorn sheep lambs. Over time, this disease-caused mortality results in long-term population decline due to lack of young animal recruitment. Adult bighorn sheep, although less likely to acquire pneumonia, also acquire the disease from domestic sheep. Adult bighorn sheep, while less likely than lambs to succumb quickly following disease exposure, can also die as a result of subsequent environmental stress (i.e., low food supply and extreme temperatures) because their immune system becomes depressed following disease transmission.

The current Conway Ranch Conservation Management Plan indicates that Mono County personnel will seek the advice and recommendations of bighorn sheep management experts relative to potential disease impacts to Sierra Nevada bighorn sheep posed by domestic sheep grazing. The California Department of Fish and Wildlife (CDFW) and United States Fish and Wildlife Service (USFWS) both have numerous seasoned experts which can provide recommendations relative to domestic sheep grazing within proximity to occupied Sierra Nevada bighorn range. We strongly encourage Mono County to take these recommendations into account as this grazing authorization decision is finalized.

Should Mono County elect to renew the domestic sheep grazing lease for Conway Ranch, it is required to comply with the California Environmental Quality Act (CEQA) and prepare an Environmental Impact Report (EIR) that fully discloses the environmental effects of grazing lease renewal. If any authorized domestic sheep grazing would result in an adverse effect upon Sierra Nevada bighorn sheep or its designated critical habitat, Mono County would need to consult with the CDFW under provisions of the California Endangered Species Act (CESA), as well as the USFWS under provisions of the Endangered Species Act (ESA). The EIR which is prepared must also include measures which could be adopted to mitigate any and all adverse Sierra Nevada bighorn sheep effects to a less-than-significant level.

Given that 1) domestic livestock grazing is a discretionary activity that may be authorized on the Conway Ranch by the Mono County Board of Supervisors; 2) the relatively high cost of preparing a legally defensible EIR if continued domestic sheep grazing is proposed through a lease renewal; and 3) the known risk of disease transmission from domestic sheep to bighorn sheep and the potential resulting mortality of a state/federally listed species, we strongly urge the Mono County Board of Supervisors to allow this grazing lease to terminate. Doing so would contribute to the recovery of this endangered species. If discretionary lease renewal is still considered after the above considerations, recommended bighorn-domestic sheep separation guidelines outlined in the Sierra Nevada Bighorn Sheep Risk Assessment (National Park Service 2011¹) and other guidance (i.e., Bureau of Land Management 2016²; Desert Bighorn Council 1990³) should be closely followed.

¹ National Park Service (NPS). 2011. Sierra Nevada bighorn sheep risk assessment: research and recovery actions. Sequoia and Kings Canyon National Parks.

² Bureau of Land Management. 2016. MS 1730 – Management of domestic sheep and goats to sustain wild sheep (public).

³ Desert Bighorn Council. 1990. Guidelines for the management of domestic sheep in the vicinity of desert bighorn habitat.

We appreciate the opportunity to provide the Mono County Board of Supervisors with this important information regarding the endangered Sierra Nevada bighorn sheep.

Sincerely,

Otto Cundahe

Jeff Aardahl California Representative Defenders of Wildlife 980 Ninth Street, Suite 1730 Sacramento, CA 95814 jaardahl@defenders.org

Mass-

Thomas B. Egan California Desert Representative Defenders of Wildlife P.O. Box 388 Helendale, CA 92342 tegan@defenders.org

References

- Bureau of Land Management (BLM). 2016. MS 1730 Management of domestic sheep and goats to sustain wild sheep (public). Headquarters. Washington D.C. <u>https://www.blm.gov/style/medialib/blm/wo/Information_Resources_Manageme_nt/policy/blm_manual.Par.40213.File.dat/MS%201730.pdf</u>.
- Cahn, M.L., M.M. Conner, O.J. Schmitz, T.R. Stephenson, J.D. Wehausen, and H.E. Johnson. 2011. Disease, Population Viability, and Recovery of Endangered Sierra Nevada Bighorn Sheep. The Journal of Wildlife Management 75(8):1753–1766. https://nrm.dfg.ca.gov/FileHandler.ashx?DocumentID=39865&inline=true.
- Desert Bighorn Council. 1990. Guidelines for the management of domestic sheep in the vicinity of desert bighorn habitat. Desert Bighorn Council Technical Staff. Desert Bighorn Council 1990 Transactions. http://www.desertbighorncouncil.com/app/download/7104371504/Guidelines+fo r+Management+of+Domestic+Sheep+in+the+Vicinity+of+Desert+Bighorn+Hab itat 1990+Volume+34.pdf.
- National Park Service (NPS). 2011. Sierra Nevada bighorn sheep risk assessment: research and recovery actions. Sequoia and Kings Canyon National Parks. Three Rivers, California. https://www.bing.com/search?q=sierra+nevada+bighorn+sheep+risk+assessment %3A+research+and+recovery+actions&form=EDGTCT&qs=PF&cvid=2766addb 946548e98884bd1e760bd640&pq=sierra+nevada+bighorn+sheep+risk+assessment %3A+research+and+recovery+actions&cc=US&setlang=en-US.

From: Sent: To: Subject: Shannon Kendall Thursday, February 16, 2017 11:51 AM Helen Nunn FW: remove domestic sheep from Conway Ranch

From: laurie | [mailto:lauriel1@earthlink.net]
Sent: Thursday, February 16, 2017 11:48 AM
To: Shannon Kendall <<u>skendall@mono.ca.gov</u>>
Cc: Stacy Corless <<u>scorless@mono.ca.gov</u>>; Larry Johnston <<u>ljohnston@mono.ca.gov</u>>; Fred Stump
<<u>fstump@mono.ca.gov</u>>; Bob Gardner <<u>bgardner@mono.ca.gov</u>>; John Peters <<u>jpeters@mono.ca.gov</u>>; Subject: remove domestic sheep from Conway Ranch

Dear Mono County Board of Supervisors:

I am writing in support of the removal of domestic sheep from Conway Ranch. Grazing domestic sheep in close proximity to a Bighorn Sheep herd poses a substantial risk of disease to the Bighorn population which could reduce or eliminate the wild herd. Establishing and maintaining the herds of Bighorn as they are today involved a significant outlay of money and effort. This would be lost if the herds were lost. It could also result in significant legal costs to Mono County. I would recommend that uses other than domestic sheep grazing on Conway Ranch be considered and implemented.

My personal experiences with Bighorn Sheep in the High Sierra began in about 1984 when I first hiked to what some call Lamb and Ewe Meadow near Lee Vining Peak. The two scientists who were there monitoring the sheep indicated where I could go to see them. It was an amazing introduction to these wonderful animals. In the intervening years I have revisited that area on a couple of occasions. Mostly, though, I am always on the lookout for them from whatever vantage point I may have in the area between Parker Pass and Lundy Canyon. It is a rare, thrilling moment when I am able to discern them in these most wild places. I love too the excited accounts of others who have encountered them in these areas.

These animals have an extremely tough life. What a tragedy it would be to continue to endanger them unnecessarily when our goal should be to ensure their survival in every possible way. Please discontinue the grazing of domestic sheep on Conway Ranch.

Thank you. L. Lawrence

Bob Gardner Supervisor, District 3 PO Box 564 June Lake, CA 93541

February 16, 2017

Dear Bob,

My husband and I are part-time (summer and fall) residents of Mono City. We write to urge you to support any and all protection for our local population of bighorn sheep. Enormous efforts by various agencies, including our great neighbor Yosemite National Park, and many individuals have gone into reintroducing these beautiful wild creatures and helping them thrive. We have spotted them a number of times on our walks up Lundy Canyon, and the thrill never diminishes. My husband prides himself on his pictures of them, beautifully draped over rocks on a sunny afternoon.

It is heartening to see that a rare species can recover, and yet the bighorn's foothold in our neighborhood is tenuous. As I'm sure you are aware, domestic sheep pose a serious threat to wild bighorn since the former can infect the latter with diseases that decimate the wild animals. We are aware that sheep grazing has a long and cherished history in Mono County. Nonetheless, I think that at this point in Mono County's history it is time to embrace the new - and the wild! The bighorn will thrill the hikers and photographers and others who come to Mono to take in its great beauty. Cattle grazing could replace sheep grazing on Conway Ranch, allowing animal husbandry to continue while protecting the bighorn sheep.

Thank you for all you do for our beautiful section of Mono County. Please add your support to bighorn sheep at the upcoming supervisor meeting when Fish & Wildlife services will make their presentation on the bighorn sheep.

Sincerely,

Jim Merz

Heidi Hopkins

PC Box 409 Lee Vining 93541

CC: Mone County Bound of Supervisors

P.O. Box 128 Lee Vining, CA 93541

P.O. Box 715 Bridgeport, CA 93517 February 16, 2017

Dear Mono County Board of Supervisors,

Last summer as I was walking through high open meadows in Yosemite, I looked up to the crest and saw a Sierra Nevada bighorn sheep silhouetted for a split second against the sky, followed by a second one, before they dropped down the other side, out of sight. I somehow happened to look the moment they were moving up and over.

This serendipitous sighting was a wild moment—an image and experience forever burned in my memory. Having witnessed the initial Yosemite bighorn sheep restoration project in 1986 when 27 animals were relocated to the Yosemite National Park/Inyo National Forest crest-line boundary and having followed the work of partnering agencies and organizations dedicated to protecting and rebuilding the population, I celebrate the return of the bighorn to their native range and the recovery of a species found nowhere else in the world. In celebration, though, I am fully aware their survival remains precarious.

Continued grazing of domestic sheep at Conway Ranch presents a threat to the bighorn. Bighorn rams are known to take off and travel great distances. They have historically been sighted in the Conway Ranch area. Disease transfer from domestic to wild sheep leads to decimation of entire herds. One encounter can undo years of recovery effort. It can undo the very existence of an endangered Sierra Nevada species. Renewing a grazing permit is not worth a loss of this magnitude—a forever loss.

I urge the Mono County Board of Supervisors to terminate domestic sheep grazing at Conway Ranch on behalf of the iconic Sierra Nevada bighorn and in support of the tremendous work of Mono County's neighboring agencies.

Thank you for your consideration.

Sincerely,

Margaret Eissler

David B. Herbst, PhD Sierra Nevada Aquatic Research Laboratory, University of California 1016 Mt Morrison Road, Mammoth Lakes CA 93546 February 15, 2017

Mono County Board of Supervisors P.O. Box 715, Bridgeport, CA 93517 Sent via email for Board distribution to: <u>skendall@mono.ca.gov</u>, County Clerk

Regarding the risk of domestic sheep grazing at Conway Ranch to Sierra Nevada Bighorn Sheep:

Like any large and seasonally migratory mountain animal, year to year variations in movements often depend on weather, temperature, snow cover, availability of food and cover. While there is some indication from population studies that may show what the herds have done in the past, that does not preclude the chance that future conditions may promote movements that result in contact between domestic sheep and the Endangered Sierra Nevada Bighorn Sheep (SNBS). So what are the odds? How does one go about protecting threatened species or sensitive ecosystems for that matter? Answer - evaluate the risks, minimize or ameliorate those risks, and then, include a margin of safety for what limited evidence and data may not anticipate.

What is known is that if contact occurs, there can be dire consequences for transmission of a disease that threatens a federal listed species that has uncertain prospects for sustaining viable populations. The extremes of climate patterns (drought vs deep snowpack seen in just the last 4-5 years) suggest to me that we have not seen all the possible movement patterns of SNBS in future environments. The marginal value of meadow grazing in the Mono Basin for domestic sheep does not, in my opinion, outweigh the removal of a potential risk to the population of a nearby species (Lundy Canyon and environs) for which great efforts have been invested by Yosemite National Park, the California Department of Fish and Wildlife and others to secure long-term survival.

The County and other groups should support these efforts and abide by the standard of including a margin of safety for protecting species and ecosystems at risk. I include the term ecosystem here because SNBS are representative of Sierra Nevada ecosystems. Whatever actions can be taken to guard the structure and function of mountain ecosystems with a margin of safety for protecting the overarching importance of water resources and forest and meadow integrity, are worthwhile, and will benefit not only SNBS, but other wildlife, habitat values, iconic scenery, and rural recreation economies.

I encourage Mono County and federal/state agencies to act to limit the potential for contact between Bighorn and domestic sheep at Conway Ranch, and reconsider how to manage this land for native wildlife values.

Respectfully,

David Herbst

From: Sent: To: Subject: Shannon Kendall Thursday, February 16, 2017 5:01 PM Helen Nunn FW: Sierra Nevada Bighorn Sheep

From: john ljung [mailto:jrljung@gmail.com] Sent: Thursday, February 16, 2017 5:00 PM

To: Shannon Kendall <skendall@mono.ca.gov>

Cc: Lisa Cutting <<u>lisa@monolake.org</u>>; Larry Johnston <<u>ljohnston@mono.ca.gov</u>>; Fred Stump <<u>fstump@mono.ca.gov</u>>; Bob Gardner <<u>bgardner@mono.ca.gov</u>>; John Peters <<u>jpeters@mono.ca.gov</u>>; Stacy Corless <<u>scorless@mono.ca.gov</u>> Subject: Sierra Nevada Bighorn Sheep

Greetings,

We are writing to strongly urge you to cancel the lease which allows domestic sheep to graze on Conway Ranch. Domestic sheep pose an undeniable danger to Sierra Nevada bighorn sheep. Yosemite National Park and wildlife agencies have gone to great effort and expense to return bighorn sheep to the Lundy canyon area. These efforts have been successful so far, although the herd is still in danger from forces beyond our control (avalanche, predators, etc). With this in mind we urge you to remove an acknowledged danger which is in your control: the danger posed by nearby domestic sheep.

Conway Ranch, managed by the county, is designated for open space and wildlife. These uses should be paramount. Any other choices, revenue producing or not, should not compromise the health of the Sierra Nevada bighorn sheep herd which occupies county land bordering Conway Ranch and beyond to the other herds. Allowing domestic sheep to graze there would require expensive monitoring and, worse case scenario, compensation for causing the failure of the Lundy Canyon bighorn herd, and paying for reintroduction of healthy animals. What a nightmare! You should not expose the county to the possibility of expensive litigation around this issue. In addition, the annual lease revenue to the county, minus the management fee, is minimal in view of the entire County budget.

We strongly urge you to vote to terminate the grazing lease allowing domestic sheep on Conway Ranch. Thank you for your attention to this issue.

Sincerely,

John and Mary Ljung



OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

💻 Print

MEETING DATE February 21, 2017

TIME REQUIRED		PERSONS
SUBJECT	Closed SessionHuman Resources	APPEARING BEFORE THE BOARD

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

CONFERENCE WITH LABOR NEGOTIATORS. Government Code Section 54957.6. Agency designated representative(s): Stacey Simon, Leslie Chapman, and Dave Butters. Employee Organization(s): Mono County Sheriff's Officers Association (aka Deputy Sheriff's Association), Local 39--majority representative of Mono County Public Employees (MCPE) and Deputy Probation Officers Unit (DPOU), Mono County Paramedic Rescue Association (PARA), Mono County Public Safety Officers Association (PSO), and Mono County Sheriff Department's Management Association (SO Mgmt). Unrepresented employees: All.

RECOMMENDED ACTION:

FISCAL IMPACT:

CONTACT NAME: PHONE/EMAIL: /

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR **PRIOR TO 5:00 P.M. ON THE FRIDAY** 32 DAYS PRECEDING THE BOARD MEETING SEND COPIES TO:

MINUTE ORDER REQUESTED:

🗌 YES 🔽 NO

ATTACHMENTS:

Click to download

No Attachments Available

History

Time



OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

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MEETING DATE February 21, 2017

Departments: CAO, Sheriff

TIME REQUIRED	10 minutes (5 minute presentation; 5 minute discussion)	PERSONS APPEARING
SUBJECT	Review of Need for Continuation of Local Emergency	BEFORE THE BOARD

Leslie Chapman, Ingrid Braun

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

On January 31, 2017 the Mono County Sheriff declared a state of local emergency as a result of extreme winter weather. The Board of Supervisors ratified this declaration on February 7, 2017, and further declared a continuing state of emergency. Mono County Code Section 2.60.080 requires that the Board of Supervisors review the need for continuing the local emergency every 14 days until it is terminated. This item is provided for that purpose.

RECOMMENDED ACTION:

Review need for continuing the local emergency. If Board determines that need no longer exists, direct staff to prepare a declaration terminating local emergency.

FISCAL IMPACT:

None

CONTACT NAME: Ingrid Braun

PHONE/EMAIL: 760-932-5414 / Ichapman@mono.ca.gov

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR **PRIOR TO 5:00 P.M. ON THE FRIDAY** 32 DAYS PRECEDING THE BOARD MEETING SEND COPIES TO:

MINUTE ORDER REQUESTED:

🗖 YES 🔽 NO

ATTACHMENTS:

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No Attachments Available

History

Time	Who	Approval
2/16/2017 1:51 PM	County Administrative Office	Yes
2/16/2017 11:58 AM	County Counsel	Yes
2/16/2017 2:25 PM	Finance	Yes



OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

Print

MEETING DATE February 21, 2017

Departments: CAO, Finance

TIME REQUIRED	1 hour 20 minutes (20 minutes presentation, 60 minutes discussion)	pe Af
SUBJECT	Mid-Year Budget Review	BE BC

PERSONS APPEARING BEFORE THE BOARD Leslie Chapman, Janet Dutcher

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

Receive analysis of the County's General Fund fiscal performance for the year ended June 30, 2016. Present mid-year budget review and discuss budget updates.

To view documents related to this item which are too large to attach to the agenda, please click on the link below:

http://www.monocounty.ca.gov/auditor/page/2016-17-mono-county-mid-year-budget-review

RECOMMENDED ACTION:

Receive analytical analysis of General Fund fiscal performance for the year ended June 30, 2016. Hear budget updates and approve the mid-year budget adjustments (4/5ths vote required). Provide any desired direction to staff.

FISCAL IMPACT:

Increase in General Fund appropriations of \$147,347 funded with an increase in revenues of \$147,347 and the use of carryover balance of \$0. Increase in Non-General Fund appropriations of \$1,037,409 funded with an increase in revenues of \$626,948 and the use of carryover balance of \$410,461.

CONTACT NAME: Janet Dutcher

PHONE/EMAIL: 760-932-5494 / jdutcher@mono.ca.gov

SEND COPIES TO:

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR **PRIOR TO 5:00 P.M. ON THE FRIDAY** 32 DAYS PRECEDING THE BOARD MEETING

MINUTE ORDER REQUESTED:

🔽 YES 🕅 NO

ATTACHMENTS:

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History		
Time	Who	Approval
2/16/2017 4:23 PM	County Administrative Office	Yes
2/15/2017 9:48 AM	County Counsel	Yes
2/15/2017 4:23 PM	Finance	Yes



DEPARTMENT OF FINANCE AUDITOR-CONTROLLER COUNTY OF MONO

Stephanie M. Butters Assistant Finance Director Auditor-Controller Janet Dutcher, CPA, CGFM Director of Finance P.O. Box 556 Bridgeport, California 93517 (760) 932-5490 Fax (760) 932-5491

Date:	February 21, 2017
To:	Honorable Board of Supervisors
From:	Janet Dutcher, Finance Director
Subject:	Analysis of the County's General Fund fiscal performance for the year ended June 30, 2016. Present mid-year budget review and discuss budget updates

Action Requested:

Receive analysis of the General Fund fiscal performance for the year ended June 30, 2016. Hear budget updates and approve the mid-year budget adjustments (4/5ths vote required). Provide any desired direction to staff.

INTRODUCTION

This analysis prepared by Finance discusses the County's General Fund (GF) fiscal performance for the year ended June 30, 2016. It compares the GF's modified (amended) budget with the actual year-end results, and includes:

- Assessment of the overall health of GF finances.
- Analysis of the preciseness of forecasting last year's revenues and expenditures.
- Fiscal evaluation of revenue and expenditure trends.
- Status of carryover balance, reserves and ability to finance long-term needs.

	Modified	Year-end
GF FISCAL OVERVIEW FOR FY 2016	Budget	Results
Available Financing		
Fund balance	\$ 2,267,606	\$ 3,197,600
Revenues	34,051,277	35,741,532
Total Financing Sources	 36,318,883	38,939,132
Financing Uses		
Transfer to general reserve	38,934	38,934
Expenditures	 36,279,949	31,980,161
Total Financing Uses	 36,318,883	32,019,095
Unassigned Fund Balance	\$ -	\$ 6,920,037

Highlights of last year's fiscal performance include:

- Carry forward of unanticipated fund balance of \$2.3 million not needed to legally balance the FY 2017 adopted budget. We recommend setting aside all or some of this as savings against economic downturns and/or financing of long-term needs.
- Discretionary revenues outperformed the budget by \$3.5 million, or 15.5%.
- Program revenues, much of which are directly impacted by actual expenditures, were \$1.8 million less than projected.
- The GF ended the fiscal year with 18 vacant positions that generated \$1.6 million in budget savings.
- Budget savings of \$1.6 million was realized from services with outside vendors that were not incurred as anticipated.

GF REVENUES

GF activities are financed with two types of revenues: program revenues and discretionary revenues. Program revenues are generated from departmental activities. Discretionary revenues come mostly from taxes including property, sales, and transient occupancy (TOT). The amount of discretionary revenues when compared to the budget and prior years are an indication of fiscal health because these revenues finance the gap between department expenditures and their program revenues. A healthy level of discretionary revenues ensure the GF can sustain current service levels.



GF revenues were \$1.7 million, or 5%, greater than anticipated. Property tax related revenues generated \$2 million more than projected. TOT taxes (excluding amount assigned to tourism fund) reached over \$2.8 million, the highest amount to date.



Specific revenue trends are illustrated below using actual results from the past four years.



GF EXPENDITURES

GF expenditures were \$4.3 million under budget (88.15% of budget). There are primarily two reasons for lower spending. One reason is vacant positions that are budgeted as if filled all twelve months of the fiscal year with the highest level of benefits. This generated budget savings of \$1.6 million. The second results from unneeded services. The GF budget included \$4.9 million to vendors for services of which \$1.6 million remained unspent at the end of the year. Of this amount, \$690,000 is the first time homebuyer program that had little activity until FY 2017. The remainder includes external contracts with subject matter experts that were budgeted in case such services were required but were not needed during the fiscal year.

Salary, Wages, Overtime and Benefits FY 2013 to FY 2016 \$16,000,000 \$14,000,000 \$12,000,000 \$10,000,000 \$8,000,000 \$6,000,000 \$4,000,000 \$2,000,000 \$-FY 2013 FY 2014 FY 2015 FY 2016 Salaries & Wages \$13,418,509 \$13,703,319 \$12,767,691 \$12,652,170 Overtime \$898,482 \$608,691 \$1,060,851 \$888,922 Benefits \$8,017,100 \$8,416,654 \$7,986,642 \$8,307,043

Specific expenditure trends of interest are illustrated below using actual results from the past four years.



FISCAL SUSTAINABILITY

Carryover balance is the aggregate of prior year surpluses and deficits. While the County's GF experienced surpluses these past two years, the fund endured a string of deficits affecting fiscal sustainability of public services. Reserves previously built up over time were used to balance each successive year's budget. Accumulated budget savings eventually disappeared.



There were some years when deficits nearly used up all past surpluses. In those years, tough balancing strategies such as hiring freezes, furloughs and delaying projects were necessary. While economic downturns, revenue shortfalls and deficits may be unavoidable, one strategy for ensuring fiscal sustainability and even fiscal resiliency is diversity of reserves, namely an economic stabilization fund or strategic reserve that serves to sustain pubic services during recessionary times.

STATUS OF GF RESERVES

The general reserve balance illustrated below is available for spending in case of disasters. County budget policy establishes this reserve at 5% to 15% of the average annual general fund expenditures. For FY 2017, the reserve fund returned to the minimum policy amount (5%) after dipping below policy minimum level because of the Round Fire incident.



In FY 2016, the GF unexpectedly received \$770,000 in excess ERAF revenues. With the adoption of the FY 2017 budget, this amount along with some anticipated salary savings from vacant positions was moved into an economic stabilization fund. The balance is currently \$1,117,542. We recommend continuing to invest year-end surpluses into this economic stabilization fund.

GF CARRYOVER BALANCE

Positive operating results for the past two years has replenished the GF carryover. The schedule below shows the status of the carryover balance available for appropriation or further savings.

Available for appropriation or further savings as of June 30, 2016	\$ 6,920,037
Amounts encumbered by the FY 2017 Adopted Budget:	
Balance budget deficit	(3,193,075)
Transfer to general reserve	(256,909)
Transfer to economic stabilization	 (1,117,516)
Unassigned fund balance	\$ 2,352,537

LONG-TERM FINANCING NEEDS FOR PROJECTS AND LIABILITIES

One reason for today's discussion of the County's GF fiscal status is to facilitate a decision about funding certain projects such as the application for SB 844 funding for a new jail or the amount that can be invested in our south county facilities. These are just two projects in need of a long-term financing solution.

When making long-term financing decisions consider the following list of projects and liabilities:

- Jail Construction
- South County Facilities
- CARB Compliance
- Radio Network Replacement
- Long-term solution for Bridgeport Clinic and Inmate Medical Services
- Deferred maintenance on County roads (current GF subsidy at \$850,000)
- Unfunded County Comprehensive Facilities Plan projects
- Impact from future MOU negotiations
- Replacement of aging and outdated elections equipment
- Aging motor pool vehicles in need of replacement beyond available capital reserve balances
- Continued increases to retirement contributions (CalPERS recently lowered the discount rate by ½% over the next three years)
- Continued increases to health insurance premiums
- Accrued vacation and vested sick leave balances payable when employees separate from service

FY 2017 DISCRETIONARY REVENUES – SO FAR

As of January 31, 2017, discretionary revenues to date were \$15.6 million, or 68% of budget. At this time, current unsecured property taxes exceeds the annual budget by \$41,426, or 3%. Current secured property taxes were 60% of budget and \$275,000 more than at this time last year. TOT revenues were 67% of budget and \$361,000 more than at this time last year. Mid-year budget requests conservatively recommend increasing the budget for discretionary revenue by \$185,000.

BUDGET STATUS

The Mono County Budget is on track as of January 31, 2017, and the departments are doing a good job of living within their means. In the General Fund, 55% of revenues were collected and 49% of expenditures were paid during the first seven months of the fiscal year. Non-general fund revenues are tracking at 49% with expenditures at 37%. The 2016-17 Board-Approved budget includes \$383,390 in contingencies and after allocating \$6,000 for replacement plan benefit invoices and recommending using \$14,907 to balance mid-year budget requests, there is a remaining balance of \$362,483 for the remaining five months of this fiscal year.

While still too early to project, it appears there will be savings on the expenditure side. If revenues continue to come in as projected, we will again close the fiscal year with a surplus critical to balancing next year's budget.

	General Fund	Non-General Fund	All Funds
Adopted Budget, as revised		++	
Expense	39,107,471	31,743,612	70,851,083
Revenue	34,485,425	31,338,681	65,824,106
Net Revenue (Cost)	(4,622,046)	(404,931)	(5,026,977)
Midyear Budget Request	1		I
Expense	39,356,346	32,781,021	72,137,367
Revenue	34,734,300	32,020,957	66,755,257
Net Revenue (Cost)	(4,622,046)	(760,064)	(5,382,110)
Actual Revenues & Expenditures (Thru	1	1	
January 31, 2017)			1
Expense	19,304,646	11,641,655	30,946,301
Revenue	18,984,959	15,399,187	34,384,146
Net Revenue (Cost)	(319,687)	3,757,532	3,437,845
Budget Change at Mid-Year			
Expense	248,875	1,037,409	1,286,284
Revenue	248,875	682,276	931,151
Net Revenue (Cost)	_	(355,133)	(355,133)



OFFICE OF THE CLERK OF THE BOARD OF SUPERVISORS

REGULAR AGENDA REQUEST

💻 Print

MEETING DATE February 21, 2017

Departments: Public Works

TIME REQUIRED	1 hour (15 minute presentation, 45 minute discussion) SB 844 Jail Proiect Proposal	PERSONS APPEARING BEFORE THE	Garrett Higerd
SUBJECT	Package	BOARD	

AGENDA DESCRIPTION:

(A brief general description of what the Board will hear, discuss, consider, or act upon)

Update on proposal for jail revenue bond funds to construct a new jail facility on the site of the old County hospital on Twin Lakes Road.

The Mono County General Plan is available at: <u>http://monocounty.ca.gov/planning/page/general-plan-eir</u>

The contract documents (in template form) for the project are located at <u>http://www.bscc.ca.gov/s_cfcformofdocuments.php</u>. These documents would be approved as to form in the proposed resolution and then finalized and executed if the grant were awarded. County Counsel has reviewed the contracts and finds them to be legally adequate and acceptable as to form.

RECOMMENDED ACTION:

Receive update on status of SB 844 Jail Project Proposal Package to construct a new jail facility on the site of the old County hospital on Twin Lakes Road. Approve Resolution R17-_____ authorizing application for adult detention facility construction funds under SB 844 from the Board of State and Community Corrections and adopting a General Plan EIR Addendum. Provide direction to staff.

FISCAL IMPACT:

If Board approves submission of application, if County is awarded funds, and depending on ultimate design of project, Mono County's contribution towards this project is projected to be about \$2,053,000, which includes \$440,000 of in-kind labor for project management, construction management, transition planning and grant administration that will be required over the life of the project (the next four to five years) and a \$1,613,000 cash match of which \$203,000 has already been budgeted. We expect the remaining \$1,410,000 will come from an investment pool loan or bank line of credit having a five-year term, payable semi-annually at as low as 2.50% interest per year. Annual loan payments will be approximately \$302,000, and will cost the County \$98,750 in interest over the term of the loan. Annual loan payments could be funded through a combination of budgetary expenditure savings, expenditure deferrals and if necessary, temporary expenditure reductions.

CONTACT NAME: Garrett Higerd

PHONE/EMAIL: 760-924-1802 / ghigerd@mono.ca.gov

SUBMIT THE ORIGINAL DOCUMENT WITH ATTACHMENTS TO THE OFFICE OF THE COUNTY ADMINISTRATOR **SEND COPIES TO:**

MINUTE ORDER REQUESTED:

VES 🗖 NO

ATTACHMENTS:

Click to download			
D	Staff Report		
D	SB 844 Proposal		
D	EIR Addendum		
D	Resolution		

History

Time	Who	Approval
2/16/2017 7:11 AM	County Administrative Office	Yes
2/16/2017 2:15 PM	County Counsel	Yes
2/16/2017 2:26 PM	Finance	Yes



MONO COUNTY DEPARTMENT OF PUBLIC WORKS

Post Office Box 457 • 74 North School Street • Bridgeport, California 93517 760.932.5440 • Fax 760.932.5441 • monopw@mono.ca.gov • www.monocounty.ca.gov

- Date: February 21, 2017
- To: Honorable Chair and Members of the Board of Supervisors
- From: Garrett Higerd, County Engineer
- Re: SB 844 Jail Project Proposal Package

Recommended Action

Receive update on status of SB 844 Jail Project Proposal Package to construct a new jail facility on the site of the old County hospital on Twin Lakes Road. Approve Resolution R17-

_____ authorizing application for adult detention facility construction funds under SB 844 from the Board of State and Community Corrections and adopting a General Plan EIR Addendum. Provide direction to staff.

Fiscal Impact:

If Board approves submission of application, if County is awarded funds, and depending on ultimate design of project, Mono County's contribution towards this project is projected to be about \$2,053,000, which includes \$440,000 of in-kind labor for project management, construction management, transition planning and grant administration that will be required over the life of the project (the next four to five years) and a \$1,613,000 cash match of which \$203,000 has already been budgeted. We expect the remaining \$1,410,000 will come from an investment pool loan or bank line of credit having a five-year term, payable semi-annually at as low as 2.50% interest per year. Annual loan payments will be approximately \$302,000, and will cost the County \$98,750 in interest over the term of the loan. Annual loan payments could be funded through a combination of budgetary expenditure savings, expenditure deferrals and if necessary, temporary expenditure reductions.

Strategic Plan Alignment: Public Safety, Infrastructure

Background:

On January 3, 2017, the following project alternatives were discussed for completion of a Senate Bill 844 funding application:

- (1) Construct new facility at site of old Mono County Hospital on Twin Lakes Road; or
- (2) Construct jail annex at site of Frontier warehouse adjacent to existing jail.

The Board of Supervisors provided direction to, "pursue option (2) Construction of jail annex at site of Frontier warehouse adjacent to existing jail as discussed. However, if it is determined that the Frontier site is not available, or not available in a reasonable amount of time which will allow for the grant application to be filed on time, then pursue option (1) Construction of new facility at site of old Mono County Hospital on Twin Lakes Road". On January 17, 2017, the Board of Supervisors was informed that the jail annex alternative is not feasible because Frontier is not willing to work with the County to transfer ownership of the site.

Accordingly, staff has prepared a complete SB 844 Project Proposal for a new jail facility at the old hospital site on Twin Lakes Road. Please see the proposal package attached. Our consultants and I have refined the proposal and worked to reduce the costs where possible within the SB 844 cost estimating requirements. For example, the buildings have been shifted about 100 feet to the north (closer to the existing clinic) to realize the following benefits:

- Better utilization of the existing outpatient clinic lobby and office space in the existing Bridgeport clinic to reduce costs. This refinement reduces the new jail footprint by approximately 1,300 square feet.
- The shared parking lot that serves the clinic and paramedic station can be paved with this project and there will be less asphalt to maintain long term.
- The new jail would be constructed more squarely on the existing hospital footprint, potentially reducing the risk that we get into geotechnical engineering issues for the new foundations.
- A larger portion of useable land would be left to the south that could potentially be used for job training, a garden, etc. in the future.

To summarize the proposal, this project would:

- Provide medical and dental exam rooms and mental health service support space.
- Provide appropriate space to conduct evidence based programs that are necessary to reduce recidivism.
- Provide a respiratory isolation cell capable of accommodating an inmate with a contagious disease and a safety cell needed for inmates in mental health crisis.
- Correct operational safety, fire and life safety, access compliance, and energy efficiency deficiencies that exist at the current jail.
- Be staffed with approximately the same staffing levels as today, and therefore not increase long term operational costs.
- Replace a mandatory facility that was not designed to adequately meet current needs and will be nearing the end of its useful life in the coming years (generally estimated at 50 years for jails). Maintenance costs will increase as the jail continues to age and a funding source like this may not be available when the County will be forced to plan for a replacement jail.
- Be co-located with the existing Bridgeport medical clinic and the addition of state-ofthe-art dental and mental health space in the jail clinic, that can be secured and made accessible to the public at regularly scheduled times, may help the County to attract service providers who would otherwise be unwilling to serve Bridgeport for a few inmates, but may be willing to serve the community if they could expand their client base.
- Demolish the decommissioned hospital building and redevelop the site.
• Be an appropriate use of the hospital site and a benefit to the community, based on public input at Planning Commission and Bridgeport RPAC meetings.

Cost Estimate and Budget Narrative:

Please see the proposed Budget Summary Table on page 4 of Section 2 of the Proposal. The Conceptual Budget Estimate prepared by VANIR estimates the total construction costs, including a 20% contingency and 3% adjustment for our remote location, at approximately \$17,182,000. The BSCC requires a 5.04% annually-compounding escalation factor for the project duration and an additional 5.5% contingency, which adds \$4,132,000. We have reduced these costs as much as possible by accelerating the proposed schedule by six months. We would accomplish this by demolishing the hospital building immediately after learning our project has been selected. The escalation factors and additional contingencies explain why Line Item 1 "Construction" of the Budget Summary Table is \$21,314,000. The values in the cash match and in-kind match columns were determined after the \$25,000,000 maximum state reimbursed amount was reached.

The proposed cash match is \$1,613,000 and, because \$203,000 has already been identified and budgeted for this project, the amount of the loan needed is approximately \$1,410,000. At approximately 2.5% interest, annual loan payments will be approximately \$302,000, and will cost the County \$98,750 in interest over the term of the loan. Annual loan payments would need to be budgeted starting in the 2017-2018 Fiscal Year through the term of the loan. Payments could be funded through a combination of budgetary expenditure savings, expenditure deferrals and if necessary, temporary expenditure reductions.

In accordance with CEQA, a General Plan EIR Addendum has been prepared for this project. Please see it attached.

I believe this project is a responsible use of state and local funding and provides a good long term value to the County and therefore recommend that the attached resolution be approved.

Please contact me at 924-1802 if you have any questions regarding this item.

Respectfully submitted,

Janet Higerd

Garrett Higerd County Engineer

Attachments: (SB 844 Jail Proposal, Mono County General Plan EIR Addendum #17-01, Draft Resolution)



SENATE BILL 844, ADULT LOCAL CRIMINAL JUSTICE FACILITIES CONSTRUCTION FINANCING PROGRAM PROPOSAL FORM

This document is not to be reformatted.

SECTION 1: PROJECT INFORMATION

A. APPLICANT INFORMATION AND PROPOSAL TYPE								
COUNTY NAME				STATE	STATE FINANCING REQUESTED			
Mono County				\$ 25,	,000,000			
SMA	ALL COUNTY	-	MEDIUM	COUNT			LA	
(Below 200,00	DO GENERAL COUNT	ΓY				(7	· 00,001 م	
		FROFC	PLEASE CHEC	K ONE (ONLY):			
IN		FACILIT	۲X	REGIO				
B: BRIEF PROJE	CT DESCRIPTION							
FACILITY NAME								
Mono County	/ Jail							
PROJECT DESCR	IPTION							
Replacement	t jail facility tha	t will p	provide adequate	e hous	ing and a	opropria	te pro	ogram and treatment
STREET ADDRES	S							
221 Twin Lak	es Road							
				STATE				
Bridgeport							025	17
Bridgeport				UA			935	17
C. SCOPE OF W	ORK – INDICATE F	ACILITY	TYPE ANDCHECK A	LL BOX	ES THAT APF	PLY.		
FACILITY TYPE	(II, III or IV)	NEV	V STAND-ALONE FACILITY	RENOVATION/ REMODELING		C	CONSTRUCTING BEDS OR OTHER SPACE AT EXISTING FACILITY	
D. BEDS CONST construction as a	RUCTED – Provide a result of the project	the nun ct, <u>whet</u>	nber of BSCC-rated b her remodel/renovati	eds and on or ne	I non-rated sp ew construction	oecial use on.	beds th	at will be subject to
	A. MINIMUI SECURITY E	M BEDS	B. MEDIUM SECI BEDS	URITY	C. MAXII	NUM SECU BEDS	RITY	D. SPECIAL USE BEDS
Number of beds constructed, remodeled	4		20			22		2
			E. BEDS DECO	6 REMO MMISSIC	/ED/ DNED	F.	NET B	EDS AFTER COMPLETED PROJECT
TOTAL BEDS (A+B+C+D)	48			0				0

E. APPLICANT'S AGREEMENT						
By signing this application, the authorized person assures that: a) the County will abide by the laws, regulations, policies, and procedures governing this financing program; and, b) certifies that the information contained in this proposal form, budget, narrative, and attachments is true and correct to the best of his/her knowledge.						
PERSON AUTHORIZED TO SIGN AGREEMENT						
Leslie Chapman		County Administrati	ve Officer			
AUTHORIZED PERSON'S SIGNATURE			DATE			
F. DESIGNATED COUNTY CONSTRUCTION A	DMINISTRATOR					
This person shall be responsible to oversee contractor, and must be id	onstruction and ad entified in the Boar	minister the state/county a d of Supervisors' resolution	greements. (Must be county staff, n.)			
COUNTY CONSTRUCTION ADMINISTRATOR						
Garrett Higerd		County Engineer				
DEPARTMENT Public Works			TELEPHONE NUMBER			
STREET ADDRESS			100-324-1002			
P.O. Box 457						
CITY	STATE	ZIP CODE	E-MAIL ADDRESS			
Bridgeport	CA	93517	ghigerd@mono.ca.gov			
G. DESIGNATED PROJECT FINANCIAL OFFIC This person is responsible for all financial and contractor, and must be identified in the Board	ER I accounting projec d of Supervisors' re	t related activities. (Must solution.)	be county staff, not a consultant or			
PROJECT FINANCIAL OFFICER						
Janet Dutcher		Finance Director				
DEPARTMENT			TELEPHONE NUMBER			
Finance			760-932-5494			
STREET ADDRESS P.O. Box 457						
CITY	STATE	ZIP CODE	E-MAIL ADDRESS			
Bridgeport	CA	93517	jdutcher@mono.ca.gov			
H. DESIGNATED PROJECT CONTACT PERSO	N					
This person is responsible for project coordin consultant or contractor, and must be identified	ation and day-to-da ed in the Board of S	ay liaison work with the BS Supervisors' resolution.)	CC. (Must be county staff, not a			
PROJECT CONTACT PERSON						
Garrett Higerd		County Engineer				
DEPARTMENT			TELEPHONE NUMBER			
			/60-924-1802			
P.O. Box 457						
CITY	STATE	ZIP CODE	E-MAIL ADDRESS			
Bridgeport	CA	93517	ghigerd@mono.ca.gov			

SECTION 2: BUDGET SUMMARY

Budget Summary Instructions

(This must be part of the 35 page narrative) Definitions of total project costs for purposes of this program (state reimbursed, county cash contribution, and county in-kind contribution) can be found in the "Budget Considerations" page 22 of the Senate Bill (SB) 844, Construction of Adult Local Criminal Justice Facilities (ALCJF's) Request for Proposals (RFP). The county cash and in-kind contributions are collectively the county contribution. Those defined costs in the RFP shall be the guide for accurately completing this budget summary section.

In the Budget Summary Table that follows in part D of this section, indicate the amount of state financing requested and the amount of cash and/or in-kind contributions allotted to each budget line-item, in total defining the total project costs. It is necessary to fully include <u>each</u> eligible project cost for state-reimbursed, county cash, <u>and</u> county in-kind contribution amounts.

The in-kind contribution line items represent <u>only</u> county staff salaries and benefits, needs assessment costs, transition planning costs and/or current fair market value of land. An appraisal of land value will only be required after conditional award and only if land value is included as part of the county's contribution.

The total amount of state financing requested cannot exceed 90 percent of the total project costs. The county contribution must be a minimum of 10 percent of the total project costs (unless the applicant is a small county petitioning for a reduction in the county contribution amount). County contributions can be any combination of cash or in-kind project costs. Small counties requesting a reduction in county contribution must state so in part A of this section. The County contribution must include all costs directly related to the project necessary to complete the design and construction of the proposed project, except for those eligible costs for which state reimbursement is being requested.

State financing limits (maximums) for all county proposals are as follows. For proposed regional ALCJF's, the size of the lead county determines the maximum amount of funds to be requested for the entire project. In the small county category the lead county may request up to an additional 10% of the maximum amount as an incentive to build a regional facility:

- **\$70,000,000** for large counties;
- **\$30,000,000** for medium counties; and,
- **\$25,000,000** for small counties.

A. <u>Under 200,000 Population County Petition for Reduction in Contribution</u>

Counties with a population below 200,000 may petition the Board of State and Community Corrections (BSCC) for a reduction in its county contribution. This proposal document will serve as the petition and the BSCC Board's acceptance of the county's contribution reduction, provided the county abides by all terms and conditions of this SB 844 RFP and Proposal process and receives a conditional award. The county (below 200,000 population) may request to reduce the required match to an amount not less than the total non-state reimbursable projects cost as defined in Title 15, Division 1, Chapter 1, Subchapter 6, Construction Financing Program section 1712.3. If requesting a reduction in match contribution, check the box below to indicate the county's petition.

By checking this box the county hereby petitions for a contribution reduction request as reflected in the proposal budget.

B. <u>Readiness to Proceed Preference</u>

In order to attest that the county is seeking the readiness to proceed with the proposed project, the county included a Board of Supervisors' resolution doing the following: 1) identifying and authorizing an adequate amount of available matching funds to satisfy the counties' contribution, 2) approving the forms of the project documents deemed necessary, as identified by the board to the BSCC, to effectuate the financing authorized in SB 844 3) and authorizing the appropriate signatory or signatories to execute those documents at the appropriate times. The identified matching funds in the resolution shall be compatible with the state's lease revenue bond financing. Additionally see Section 6 "Board of Supervisors' Resolution" for further instructions.

This proposal includes a Board of Supervisors' Resolution that is attached and includes language that assures funding is available and compatible with state's lease revenue bond financing. <u>See below for the description of</u> <u>compatible funds.</u>

<u>County Cash Contribution Funds Are Legal and Authorized</u>. The payment of the county cash contribution funds for the proposed adult local criminal justice facility project (i) is within the power, legal right, and authority of the County; (ii) is legal and will not conflict with or constitute on the part of the County a material violation of, a material breach of, a material default under, or result in the creation or imposition of any lien, charge, restriction, or encumbrance upon any property of the County under the provisions of any charter instrument, bylaw, indenture, mortgage, deed of trust, pledge, note, lease, loan, installment sale agreement, contract, or other material agreement or instrument to which the County is a party or by which the County or its properties or funds are otherwise subject or bound, decree, or demand of any court or governmental agency or body having jurisdiction over the County or any of its activities, properties or funds; and (iii) have been duly authorized by all necessary and appropriate action on the part of the governing body of the County.

<u>No Prior Pledge</u>. The county cash contribution funds and the Project are not and will not be mortgaged, pledged, or hypothecated by the County in any manner or for any purpose and have not been and will not be the subject of a grant of a security interest by the County. In addition, the county cash contribution funds and the

Project are not and will not be mortgaged, pledged, or hypothecated for the benefit of the County or its creditors in any manner or for any purpose and have not been and will not be the subject of a grant of a security interest in favor of the County or its creditors. The County shall not in any manner impair, impede or challenge the security, rights and benefits of the owners of any lease-revenue bonds sold by the State Public Works Board for the Project (the "Bonds") or the trustee for the Bonds.

<u>Authorization to Proceed with the Project</u>. The Project proposed in the County's SB 844 Financing Program proposal is authorized to proceed in its entirety when and if state financing is awarded for the Project within the SB 844 Financing Program.

C. <u>California Environmental Quality Act (CEQA) compliance</u>

Has the county completed the CEQA compliance for the project site?

Yes. If so, include documentation evidencing the completion (preference points).

No. If no, describe the status of the CEQA certification.

D. Agreement Not to Lease Beds if Increasing County Capacity

If the proposed project results in a net increase in rated beds in the county, will the county certify and covenant in writing that the county will not lease housing capacity from the proposed SB 844 project to any other public or private entities for a period of 10 years beyond the completion date of the adult local criminal justice facility?

Yes. The County agrees that if the project results in a net increase in rated beds, the County will not lease capacity in the SB 844 financed adult local criminal justice facility to any other public or private entity for a period of 10 years beyond the completion date of the adult local criminal justice facility. The County further agrees to certify and covenant in writing to this requirement in future agreements as provided by the BSCC and/or SPWB.

No. (Project not eligible for SB844 funding)

E. Budget Summary Table (Report to Nearest \$1,000)

LINE ITEM	STATE REIMBURSED	CASH CONTRIBUTION	IN-KIND CONTRIBUTION	TOTAL
1. Construction	\$ 21,114,000	\$ 200,000		\$ 21,314,000
2. Additional Eligible Costs*	\$ 1,045,000	\$ 95,000		\$ 1,140,000
3. Architectural	\$ 2,321,000	\$ 525,000		\$ 2,846,000
4. Project/Construction Management	\$ 520,000	\$ 545,000		\$ 1,065,000
5. CEQA	\$ O	\$ O		\$ 0
6. State Agency Fees**	\$ O	\$ 116,000		\$ 116,000
7. Audit		\$ 13,000	\$ O	\$ 13,000
8. Needs Assessment		\$ 119,000	\$ 40,000	\$ 159,000
9. Transition Planning		\$ O	\$ 150,000	\$ 150,000
10. County Administration			\$ 250,000	\$ 250,000
11. Land Value			\$ O	\$ O
TOTAL PROJECT COSTS	\$ 25,000,000	\$ 1,613,000	\$ 440,000	\$ 27,053,000
PERCENT OF TOTAL	92.41%	5.96%	1.63%	100.00 %

* Additional Eligible Costs: This line item is limited to specified fees and moveable equipment and moveable furnishings (eligible for state reimbursement or cash contribution), and public art (eligible for cash contribution only)

** For State Agency Fees: State reimbursable costs include Real Estate Due Diligence only. State Fire Marshal fees may only be claimed as cash match.

Provide an explanation below of how the dollar figures were determined for <u>each</u> of the budget categories above that contain dollar amounts. Every cash contribution (match) line item shall be included with a reporting of the full amount budgeted unless a line item is not an actual cash contribution project cost for the county. (In that case, indicate so below.) For each budget category explanation below, include how state financing and the county contribution dollar amounts have been determined and calculated (be specific).

1. Construction (includes equipment and furnishings) fixed (state reimbursement/cash match): The cost of construction is estimated to be \$21,314,000 including cost escalation to the anticipated mid-point of construction and a construction contingency. Total construction cost estimate was prepared by a professional cost estimator with knowledge of jail construction and costs. The estimate is based on conceptual building and site plans that were prepared by Nacht & Lewis Architects in conjunction with the County and Sheriff's Office for the new facilities. The conceptual planning identifies all site work, infrastructure and utility improvements within the project boundary and offsite improvements that required for the new construction. The County is estimating State reimbursement to be \$21,114 for construction within the

project boundary. The County is contributing a cash match of \$200,000 for demolition of existing structures on the site.

- 2. Additional Eligible Costs (specified allowable fees, moveable equipment and furnishings, and public art) A total need of \$ 1,140,000 is estimated for additional costs, of which the county is requesting state reimbursement of \$1,045,000 and a match of \$ 95,000.
 - a) Define each allowable fee types and the cost of each: State Reimbursement: \$345,000 for Construction Testing and Inspection and \$60,000 for Consultant Plan Checking Services. County Cash Match: \$70,000 for Geotechnical Study and Topographic Surveying and \$25,000 for Permits and Fees,
 - b) Moveable equipment and moveable furnishings total amount: State Reimbursement: \$640,000 for Furniture Fixtures & Equipment, County Cash Match: \$0
 - c) Public art total amount: 0
- 3. Architectural(state reimbursement/cash match):
 - a) Describe the county's current stage in the architectural process: The County has completed conceptual planning and programming for the new facilities. The County is prepared begin schematic design.
 - b) Given the approval requirements of the State Public Works Board (SPWB) and associated state reimbursement parameters (see "State Lease Revenue Bond Financing" section in the RFP), define which portions/phases of the architectural services the county intends to seek state dollar reimbursement: The County is seeking reimbursement for the cost of design services through Preliminary Plans and Working Drawings, Bidding and Construction Support for construction within the Project Boundary.
 - c) Define the budgeted amount for what is described in b) above: \$2,846,000
 - d) Define which portion/phases of the architectural services the county intends to cover with county contribution dollars: The County intends to cover the cost design services through Preliminary Plans and Working Drawings and Bidding and Construction Support for the construction that is outside of the Project Boundary.

Define the budgeted amount for what is described in d) above: \$525,000

- 4. Project/Construction Management Describe which portions/phases of the construction management services the county intends to claim as: The County will hire an experienced Construction Management Firm to assist the County with development of construction contracts Division 0 and 1 specifications, marketing and bidding the project, review of bids, construction phase support including but not limited to construction inspection, change order review and negotiation, pay application review and processing, project closeout and commissioning. The County will seek state reimbursement of \$ 520,000 for these services.
 - **a) Cash** \$545,000
 - **b) In-Kind** \$0.00
- 5. CEQA may be state reimbursement (consultant or contractor) or cash match The County has completed CEQA requirements for this project. The County is not

including the cost of CEQA as part of its match.

- 6. State Agency Fees Counties should consider approximate costs for the SFM review which may be county cash contribution (match). \$16,000 for the due diligence costs which may be county cash contribution (match) or state reimbursement. The County is budgeting a cash match \$116,000 for State Fire Marshal review. The County will seek State reimbursement of \$0 for due diligence costs.
- 7. Audit of Grant Define whether the county is intending to use independent county auditor (in-kind) or services of contracted auditor (cash) and amount budgeted: A cash match of \$13,000 is budgeted for the independent County Auditor to audit the grant.
- 8. Needs Assessment Define work performed by county staff (in-kind), define hired contracted staff services specifically for the development of the needs assessment (cash match) : The County hired an experienced Jail Architecture team to prepare the needs assessment. The cost of \$119,000 has been identified as cash match. The County utilized in house staff for an additional \$40,000 in-kind match.
- 9. Transition Planning Define work performed by county staff (in-kind), define the staff hired specifically for the proposed project (cash match): The County plans to utilize county staff for transition planning with costs totaling \$150,000 over the course of the project.
- 10. County Administration Define the county staff salaries/benefits directly associated with the proposed project. The Project Management Team's efforts to manage and administer the project is expected to cost \$250,000.
- 11. Site Acquisition Describe the cost or current fair market value (in-kind): N/A

SECTION 3: PROJECT TIMETABLE

Prior to completing this timetable, the county must consult with all appropriate county staff (e.g., county counsel, general services, public works, county administrator) to ensure that dates are achievable. Please consult the "State Public Works Board (State Capital Outlay Process)/Board of State and Community Corrections Processes and Requirements" section, page 30 of the RFP for further information. Complete the table below indicating start and completion dates for each key event, including comments if desired. Note the <u>required time frames</u> for specific milestone activities in this process. The BSCC Board intends to make conditional awards at its June 2017 board meeting.

KEY EVENTS	START DATES	COMPLETION DATES	COMMENTS
Site assurance/comparable long-term possession <u>within 90 days</u> <u>of award</u>	6/8/2017	8/7/2017	
Real estate due diligence package submitted within 120 days of award	6/8/2017	8/7/2017	
SPWB meeting – Project established <u>within 18 months of</u> <u>award</u>	8/7/2017	10/11/2017	
Schematic Design with Operational Program Statement <u>within 24</u> <u>months of award</u> (design-bid-build projects)	8/30/2017	2/1/2018	Includes 1-month for BSCC and CSFM Review
Design Development (preliminary drawings) with Staffing Plan	2/1/2018	10/29/2018	Includes 4-months for BSCC, CSFM reviews and DOF approval
Staffing/Operating Cost Analysis approved by the Board of Supervisors	1/30/2018	10/9/2018	
Construction Documents (working drawings)	10/31/2018	9/26/2019	Includes 5-months for BSCC, CSFM reviews and DOF approval
Construction Bids or Design-Build Solicitation	9/30/2019	11/19/2019	30 days for bidding and 2-months for review of bids, and BOS approval and award of contract.
Notice to Proceed <u>within 42 months</u> of award	11/19/2019	12/23/2019	Allows 16-days for DOF approval prior to NTP.
Construction (maximum three years to complete)	1/6/2020	4/30/2021	16-months for construction and project closeout.
Staffing/Occupancy <u>within 90 days</u> of completion	4/30/2021	7/29/2021	

SECTION 4: FACT SHEET

To capture key information from Section 5: Narrative, applicants must complete this Fact Sheet. Minimal information is requested. Narrative information or explanations are not to be included on this Fact Sheet nor as part of the tables in this section. Explanations of what is provided in these tables may be included in the Narrative section of the Proposal Form. Proposal narratives may include reference back to one or more of these specific tables (e.g., refer to Table 3 in Section 4 Fact Sheet).

Tab	Table 1: Provide the following information				
1.	County general population	14,202			
2.	Number of detention facilities	1			
3.	BSCC-rated capacity of jail system (multiple facilities)	48			
4.	ADP (Secure Detention) of system	27			
5.	ADP (Alternatives to Detention) of system	47%			
6.	Percentage felony inmates of system	0.66%			
7.	Percentage non-sentenced inmates of system (this is not for the pretrial information that is requested in the rating criteria Section 5)	56%			
8.	Arrests per month	55			
9.	Bookings per month of system	40			
10.	"Lack of Space" releases per month	0			

Table 2: Provide the name, BSCC-rated capacity (RC) and ADP of the adult detention facilities (type II, III, and IV) in your jurisdiction (county)

	Facility Name	RC	ADP
1.	Mono County Jail	48	28
2.			
3.			
4.			
5.			
6.			
7.			
8.			

Table 3: List of the offender assessments used for determining programming

	Assessment tools	Assessments per Month
1.	Offender Registration Survey	25 to 35
2.		
3.		
4.		
5.		
6.		

SECTION 5: NARRATIVE

Section 5 is limited to 35 pages (the budget narrative from Section 2 must be included in the 35 page limit) and must be <u>double-spaced</u> with <u>one-inch margins</u>. All narrative (Section 5) must use no smaller than <u>12-point Arial font</u> and be ordered in the 6 subject areas listed below. If the narrative can be written in less than the maximum 35 pages, please do so (avoid "filler"). Pictures, charts, illustrations, or diagrams are encouraged in the narrative. Data sources must be identified.

If the project is for a regional ALCJF (must meet the requirements outlined in the "Eligible Projects" section, "Limit on Number of Projects/Set Asides" (pages 9 and 10) section of the RFP), clearly indicate so. Include the names of the partnering counties and their individual data that support the project and respond to the requested narrative points.

In order for a proposal to be able to compete and be rated by the ESC, it must include the following:

- Any locked facility constructed or renovated with state funding awarded under this program shall include space to provide onsite, in-person visitation capable of meeting or surpassing the minimum number of weekly visits required by state regulations for persons detained in the facility. (Gov. Code § 15820.946 subd. (f))
- Any county applying for financing authority under this program shall include a description of efforts to address sexual abuse in its adult local criminal justice facility constructed or renovated. (Gov. Code§15820.946 subd. (g))

The Proposal structure is designed to allow county applicants to demonstrate how their proposed project meets the need for ALCJFs as stated in SB 844, and how proposed expenditures of public funds meet the identified need and are justified. The presentation of information about the proposed project should allow both applicants and raters to make a step-by-step connection between the need addressed by the project and its associated budget request.

The raters will ask many questions about the proposed project as they evaluate, including but not limited to:

- What need is the project designed to meet?
- What construction work does the county propose is necessary to meet this need?
- How will offender programming and/or treatment be served in the proposed new or renovated facility?
- What is the county plan of action to accomplish the legal, design, and build steps required for this project?
- What is the total project cost, what are the funding sources, and how will the county allocate expenditures of these funds?

• Will the county be prepared to proceed with the project in a timely manner if financing is approved?

SB 844 describes the purpose for which ALCFJ construction financing is to be awarded. Additionally, the legislation states specific factors to be considered in assessing how well a proposal suits those purposes. In each section of the proposal, the rater (1) assesses how well the narrative addresses the general merit factors that apply to this section, and (2) assesses special factors mentioned in the SB 844 legislation as criteria for funding.

- a. General merit is assessed on a 13-point scale:
 - 0 Fails to meet minimum standards for financing
 - 1-3 Reaches minimum standards despite deficiencies
 - 4-6 Generally adequate
 - 7-9 Good
 - 10-12 Excellent
- b. Special merit factors are scored from 0 to 4; depending on the factor, it may be scored on a 0-4 range, or as yes/no (0/4).

For an ALCJF construction project, county applicants must answer the following questions:

1. Statement of Need: What are the safety, efficiency, and offender programming and/or treatment needs addressed by this construction proposal? Please cite findings from the needs assessment (through 2019) submitted with this proposal.

General Merit Factors

- A. To what extent does the need described in the proposal match the legislative intent of SB844?
- B. Does the applicant provide a compelling case for the use of state financing to meet this need?
- C. How well is the description of need supported by evidence provided by the applicant?

2. Scope of Work: Describe the areas, if any, of the current facility to be replaced or renovated, and the nature of the renovation, including the number of cells, offices, classrooms or other programming/treatment spaces to be replaced or added and the basic design of the new or renovated units.

General Merit Factors:

- A. How will the planned replacement, renovation, or new construction meet the needs described in Question 1 (Statement of Need)?
- B. How well does the proposed project plan suit general operational requirements for the type of facility in the proposal, including factors such as safety, security and efficiency?
- C. Where applicable, how well does the proposed project meet specific needs for programming and treatment space?

Special factors (Gov. Code, §15820.946, subd. (c)) Funding consideration:

- A. Does the county plan for seeking to replace compacted, outdated, or unsafe housing capacity that will also add treatment space; or,
- B. Does the county plan for seeking to renovate existing or build new facilities that provide adequate space for the provision of treatment and rehabilitation services, including mental health treatment.
- 3. Programming and Services: Describe the programming and/or treatment services currently provided in your facility. Provide the requested data on pretrial inmates and risk-based pretrial release services. Describe the facilities or services to be added as a result of the proposed construction; the objectives of the facilities and services; and the staffing and changes in staffing required to provide the services.

General Merit Factors:

- A. How clearly described are the facility's current programming and/or treatment services?
- B. If improvements to programming and/or treatment services are expected as a result of the planned construction project:
 - Are the improvements to programming and/or treatment services clearly described?
 - How strong is the evidence provided by the applicant that the programming and/or treatment services planned for inmates upon project completion will help reduce recidivism or meet inmates' health and treatment needs while incarcerated?
 - What are the applicant's current efforts and the plan to provide a post-release continuum of care? Describe the applicant's efforts to collaborate with community based partners in this effort.
- C. If improvements are designed to replace compacted, outdated, or unsafe housing capacity:
 - How are the program and treatment service needs of the facility population expected or planned to be met?
 - Are the improvements to housing deficiencies clearly described?
 - To what extent will the deficiencies be remedied by the proposed construction?
- D. How thoroughly does the staffing plan and lines of authority (including interagency partnerships, if relevant) in program and treatment management meet operational objectives?

Special Factors (Gov. Code, § 15820.946, subd. (b)) Mandatory Criterion:

- A. The county provided documentation that states the percentage of its inmates on pretrial status between January 1, 2015 and December 31, 2015 in the narrative of question 3.
- B. Provided a description of the county risk-assessment-based pretrial release program is <u>provided</u> in the narrative of question 3.
- 4. Administrative Work Plan: Describe the steps required to accomplish this project. Include a project schedule, list the division/offices including personnel that will be responsible for each phase of the project, and how it will be coordinated among responsible officials both internally and externally.

12

General Merit Factors:

- A. How clearly described are the elements of the work plan: timeline, assigned responsibilities, and coordination?
- B. Can the scope of work described in Question 2 (Scope of Work) feasibly be accomplished within the time allotted?
- 5. Budget Narrative: Describe the amounts and types of funding proposed and why each element is required to carry out the proposed project. Describe how the county will meet its funding contribution (match)requirements for all project costs in excess if the amount of state financing requested and how operational costs (including programming costs) for the facility will be sustained.

General Merit Factors:

- A. Is the allocation of effort in the budget appropriately matched to the objectives described under project need, scope of work, offender treatment and programming, and administrative work plan?
- B. Are the budgeted costs an efficient use of state resources?
- C. Rate the applicant's plan for sustaining operational costs, including programming over the long term.

6. Readiness to Proceed (Gov. Code, § 15820.946, subd. (b)(2)) Funding Preference:

- A. Did the county provide a board resolution: 1) authorizing an adequate amount of available matching funds to satisfy the counties' contribution 2) approving the forms of the project documents deemed necessary, as identified by the board (SPWB) to the BSCC, to effectuate the financing authorized by the legislation, 3) authorizing the appropriate signatory or signatories to execute those documents at the appropriate times. The matching funds mentioned in the resolution shall be compatible with the state's lease revenue bond financing. See page 2 of the Proposal Form and Instructions for more information regarding "compatible funds".
- Note: Finance and the SPWB will ultimately make the final determination of any fund sources compatibility with the SPWB's lease revenue bond financing. For multicounty proposals or regional proposals, applicants must attach their MOUs. All partners in the regional application must provide a board resolution.
- B. Did the county provide documentation evidencing CEQA compliance has been completed? Documentation of CEQA compliance shall be either a final Notice of Determination or a final Notice of Exemption, as appropriate, and a letter from county counsel certifying the associated statute of limitations has expired and either no challenges were filed or identifying any challenges filed and explaining how they have been resolved in a manner that allows the project to proceed as proposed.

The evaluation factors to be used and the maximum points that will be allocated to each factor are shown in the table below.

EVAI	UATION FACTOR	Scoring Method	Percentage	Weighted Score
1.	Statement of Need	0-12	15%	18
2.	Scope of Work	0-12	15%	18
	SF A/B: Feasible plan to replace compacted housing/expand program/treatment space	0-4		4
3.	Programming and Services	0-12	25%	30
	SF A: Documents pretrial inmate percentage	0/4		4
	SF B: Describes risk assessment-based pretrial release process	0/4		4
4.	Administrative Work Plan	0-12	10%	12
5.	Budget Narrative	0-12	10%	12
6.	A. Readiness: Board Resolution	0/12	10%	12
	B. Readiness: CEQA Compliance	0/12	15%	18
тот	AL POINTS	96	100%	132

Notes:

Special Factor
Scored on a 0 to 12 pt. range
Scored on a 0 to 4 pt. range
Scored 4 if pass, 0 if fail

0/12 Scored 12 if pass, 0 if fail

1. STATEMENT OF NEED

What are the safety, efficiency, and offender programming and/or treatment needs addressed by this construction proposal? Please cite findings from the needs assessment (through 2019) submitted with this proposal.

The Mono County Sheriff's Office operates the County's only jail facility which is located at 100 Emigrant Street in downtown Bridgeport, California. It is located on a downtown block bound by Emigrant, Sinclair, Bryant, and School Streets. The jail shares the block with the Mono County Sheriff's Office, the Mono County Office of Education, the Mono County Library, the historic jail facility, and a Frontier Communications substation and warehouse.

The jail is rated as a Type II detention facility by the Board of State and Community Corrections (BSCC) with a current rated capacity of 48 beds. When the jail was constructed in 1988 it had a design capacity of 24:

The original design also featured non-rated beds in isolation cells and a small female holding area. The kitchen and laundry were only designed to accommodate 24 inmates. The board rated capacity has since been increased. The maximum security pod has been double bunked to double the capacity and the dormitory capacities have been increased to the minimum limits allowed by code (10). Additionally, the female holding was expanded to incorporate the required dayrooms space for a dormitory, and an interview room was converted to an inmate worker dormitory. Finally, the isolation cells have become rated beds. The existing board rated capacity is illustrated in the following table:

Inmate Housing Classification and Unit Bed Capacities						
Housing Unit	Classification	BSCC	Total Number			
		Rated Capacity	of Fixed Beds			
A	Max Male/Female	16	16			
В	Min/Med Male Dorm	10	10			
С	Min/Med Male Dorm	10	10			
D	Min/Med Female Dorm	4	4			
Isolation	All Male/Female	4	4			
E	Inmate Workers	4	4			
Total		48	48			

At first glance, the jail may seem adequate because it is not overcrowded and the facility has been very well maintained. A closer examination, however, reveals that the jail lacks inmate programs and services that are required of a modern jail, post realignment. Furthermore, because of changes in the building code, the existing building has significant code deficiencies which would need to be improved if the facility was encumbered as part of an SB844 renovation project. The cost of these improvements would require the majority of any available state funding and therefore would not be a cost-effective approach. If there were site area available, a detached building addition (which is the norm for past state-funded projects) would be possible. However, since the existing jail site has no space for expansion, a replacement jail on a new site is the only realistic option. We believe this application will show that this is the most responsible use of state funding and which best allows for the incorporation of inmate programs and services which are the focus of the SB844 funding.

Currently there is no dedicated space for inmate programs. Inmate programs therefore occur in a portion of the corridor where intake and classification occurs at the entry to the jail facility. This is not a safe for effective practice, but is a result of the outdated facility design. While a holding cell has been converted into a medical examination room, it is too small to provide anything other than triage processes. There is no dental suite. Because Bridgeport a small community, remote from metropolitan areas, they currently lack all local services to provide medical, mental health, or dental services. Inmates are transported approximately an hour one-way to receive any of these services, and when snow is present (as it often is in winter), transportation to and from services may take an entire day. The most appropriate solution would be to provide proper medical, dental, and mental health spaces at the jail facility; but there is no room on the site to expand the existing facility.

In 2016 the County retained a consultant team to prepare a 2017 Jail Needs Assessment Update. The effort for the Jail Needs Assessment Update focused on compiling and analyzing historical jail inmate population trends which have been used to prepare an updated jail inmate profile with average daily population (ADP) inmate projections through 2034 as well as identifying the detention facility's physical plant and primary building system's strengths and weaknesses. This information was used to determine the continued operability and overall economic viability of the structure. The Assessment focused on factors that included (a) configuration and intended security levels, (b) defined uses of the facility, (c) physical condition of the building elements, (d) quality of the space, and (e) ability to achieve intended security and level of safety for staff and inmates as well as compliance with Title 15 and 24 standards.

Unlike many county jails throughout the State, overcrowding has not been an issue for the Mono County Jail. The County does not foresee a need to add any beds beyond the current rated capacity of 48. It is important to note that most of the crime in Mono County is committed by tourists who live outside the area. Because of this, the bed capacity is not impacted by population trends in Mono County itself. Spikes in ADP are driven by events which draw tourists to Mono County; Mammoth Lakes in particular.

However, the findings of the Needs Assessment Update indicates that the AB 109 sentenced (N3) inmates have and will continue to affect the jail system since their sentences are generally longer than traditional county sentenced inmates. According to the findings, the Mono County jail has booked an average of one AB 109 newly sentenced felon a month into the jail facility. This is significant for the small County Jail. The Jail does not have the appropriate space to adequately satisfy the programming and treatment services of the AB 109 inmates.

In-Person Visitation

• The proposed SB 844 Jail Replacement project includes ample in-person visitation for inmates. **Refer to Question 2 – Scope of Work** response on page 16.

County's Sexual Abuse Prevention Efforts

• **See Attachment 3:** Mono County's Sexual Abuse Prevention Plan relative to strategies for preventing sexual abuse in the jail.

As part of the Needs Assessment Update study, the existing jail was assessed for building code and operational deficiencies. The following is a summary of the major safety, efficiency, and program/treatment needs identified in the Assessment which our construction project addresses.

Mono County Jail 2017 Jail Needs Assessment Update Findings and Conclusions

Background

The jail facility affronts Emigrant Street to the north and is surrounded on all other sides by existing buildings. East of the jail, on the same block and separated by

approximately 10 feet is a metal warehouse owned and operated by Frontier Communications. Directly to the south, physically connected to the jail (and mostly separated by a fire wall) is the Sheriff's Office. Directly to the west, on the same block and separated by approximately 20 feet is Mono County Office of Education. It is important to note that the attached Sheriff's Office, constructed in 1963, is required to be an essential services building. While that building is mostly constructed of concrete, it does have some wood framing making it a non-compliant, archaic structure by current standards. This means that the jail is bound on all sides by Type V buildings of combustible construction, with no room to expand.

BSCC Operational Safety Requirements

- Ligature points are present in cells, dormitories and showers which could be used by inmates to hang themselves.
- Power outlets in cells and dormitories, which are not secured, may lead to electrocution.
- Porcelain plumbing fixtures (non-security grade) in the maximum security cells can be broken or dismantled, exposing porcelain shards and exposed metal parts which can be fabricated into weapons.
- Light fixtures (non-security grade) on the wall above lavatories in the maximum security cells can be broken and fashioned into weapons and/or expose lethal opportunities for electrocution.

Fire and Life Safety Requirements

Corridors are not rated. Doors along corridors use sliding doors which are not smoke tight. Transfer ducts allow for air passage from the corridor to cells and other rooms including the kitchen. Some corridor walls are non-rated steel panel construction.

- The kitchen is not properly separated from the exit corridor; the corridor door to the walk-in-refrigerator area of the kitchen has been removed.
- The lobby, designed as a corridor, is not properly separated from the administration area due to the open pass-through.
- Central control, which also acts as the Sheriff's 911 dispatch and therefore is the location that fire alarms alert to, is not properly separated from the remaining jail by 2-hour construction.
- Some portions of the jail are not adequately separated from the non-compliant Sheriff's Office to the south.
- The covered recreation yard and circulation are not fire sprinklered. The recreation yard is not properly separated from the non-compliant Sheriff's Office.
- The vehicle sallyport was constructed south of the fire wall and therefore is technically part of the non-compliant Sheriff's Office. It framed with wood construction. It is not properly separated from the interior portions of the Sheriff's Office- separated by glass block.
- The required jail exit to the east, which also leads to the recreation area, travels through a room that also serves as a detox cell. Because exits are not allowed to travel through occupied areas and because this exit also occurs under a nonsprinklered canopy, it is not a compliant exit.

Access Compliance Requirements

- There is no accessible parking for staff or visitors.
- Public toilets are not accessible; they do not provide the proper toilet clearances.
- Visitation booths do not provide adequate maneuvering clearances.
- Staff and service provider toilets are not compliant;
- Toilets in dormitories are not accessible.
- No accessible maximum security cells are provided.
- No showers are compliant; they include a non-compliant curb at their entry.
- No accessible holding or detox cells are provided.
- Some portions of the corridor along the exit route are not wide enough to accommodate door clearances.
- Essentially, the jail facility fails to provide any accessibility requirements of the current code cycle.

Efficiency

- Inmate program functions are held in the corridor where inmates circulate between housing, intake, the kitchen, and laundry.
- Inmates need to be transported at least 55 miles for any medical and dental exams.
- The kitchen was only designed for 24 inmates.

Programming and Treatment

 There are no program rooms to be used as classrooms and a wide variety of programming and evidence-based counseling. • There is insufficient treatment space and support staff areas which can be used for inmates with medical, dental and mental health treatment needs.

Physical Plant and Functional Needs

In order to better understand and identify space deficiencies in the Mono County Jail, the Needs Assessment Study compared a summary breakdown of the amount of square feet per rated inmate bed in each functional use area from a database. The database contains functional use area space allocations by rated bed capacity for 78 new generation adult jail/detention facilities constructed with State bonds beginning in the early 90s. The following table provides a comparison of the Mono County Jail to the 27 jails in the small county jail database.

Co	Comparison of Mono County Jail Functional Use Allocations with Other California Small County Jails						
	California Small County Jail Database Mono County Jail						
	Functional Use Area (FUA)	Number of Facilities	Average Square Footage (GSF)	Percent (%) of Total Square Footage	Gross Square Footage (GSF)	Percent (%) of Total Square Footage	
1	Administration	22	2,896	7.05%	1,142	14.16%	
2	Visiting/Lobby/Public	21	2,993	7.29%	377	4.67%	
3	Food Service	18	4,401	10.72%	691	8.57%	
4	Central Control	19	729	1.78%	256	3.17%	
5	Maintenance/Utility/Storage	23	1,879	4.58%	336	4.17%	
6	Laundry	16	661	1.61%	194	2.41%	
7	Intake/Release	23	3,727	9.08%	275	3.41%	
8	Medical/Mental Health	19	1,453	3.54%	84	1.04%	
9	Inmate Programs	21	1,285	3.13%	0	0.00%	
10	Housing/Dayroom	27	17,992	43.83%	3,943	48.89%	
11	Interior Circulation	25	3,034	7.39%	767	9.51%	
	Total Interior Jail Space 41,050 100.00% 8,065 100.00%						

Note: Areas shown for the California Small County Jail Database are averaged. Small county jails are those with an inmate population of less than 200.

The information shows that the Mono County Jail contains 8,065 square feet of interior space while the state-wide average of 27 facilities is 41,050 square feet. Even though the Mono County Jail facility is highly efficient, there is no room for growth in any of the defined functional use spaces, particularly in inmate programs and services. Based on this comparative data, the 29 year old jail facility has nearly three times less operational space for inmate functions compared to other modern, new generation jail facilities in California. The Comparison also shows that the average floor area per bed among small counties is 365.2 square feet while the Mono County Jail only contains 188.7 square feet per bed/bunk.

Medical/ Mental Health and Dental Care Needs



The jails only medical exam room is located in the jails Intake area Two critically deficient areas in the jail are: (1) medical / mental health / dental care; and (2) inmate program space. The Mono County Jail Medical Unit consists of a single 80 sf exam room with zero dedicated medical or mental health treatment space and housing. When an inmate requires segregation for potential communicable diseases, an isolation cell is utilized, but this does not meet the requirements of a respiratory isolation room. Inmates requiring constant

monitoring, including mental health cases, also are

temporarily housed isolation cells. The facility offers no x-ray or other advanced medical service to inmates. There is no telemedicine currently being offered within the facility. There is no office space for treatment staff. Inmates needing even the most basic medical

and dental care are transported 55 miles away for these services, which can be an allday trip during the winter. The Mono County Jail could contract with regional medical and dental providers to service inmates in the jail if they had space within the facility for the necessary procedure rooms and support areas.

Inmate Programming Space Needs

Another example of the space deficiencies in the jail occurs relative to inmate program space. There is no dedicated space for programming and the existing practice of holding programs in the circulation corridor presents operational, security, and fire safety challenges. This area contains no sanitary facilities for



Jail programs are run in the jails booking area corridor.

inmates or staff. The jail needs a dedicated program space that can accommodate small and larger group counseling sessions and treatment programming for incarcerated inmate populations. With the implementation of AB 109, which is shifting inmate populations to longer term offenders, the lack of program space severely limits the jail's ability to provide offenders the variety of programming options that criminal justice research has shown will aid in reducing offender recidivism. The average building area dedicated to program space in similarly sized detention facilities throughout California is 1,285 square feet. The Mono County Jail essentially has no useable programming space and instead uses a 12' length of corridor that is only 10' wide (and reduced to 6' wide once you consider the shelving and door swings in this area.) In an era where evidence-based programming is seen as the key to rehabilitation, this is a huge disparity.

Summary Observations

The jail lacks medical and dental exam rooms, mental health service support space, and appropriate space to conduct evidence based programs that are necessary to reduce recidivism. Community service providers have expressed full support for expanding inmate services but the current lack of program and treatment space does not support the services offered at the jail by these agencies.

The jail also lacks a respiratory isolation cell capable of accommodating an inmate with a contagious disease. Additionally, there is no safety cell needed for inmates in mental health crisis. The nearest hospital facility offering medical and mental health services is 55 miles away.

The jail has no toilet or shower facilities which meet current disabled accessible code requirements. There are no accessible parking spots, visitation booths, or other accessible features along the accessible path of travel and therefore the jail is noncompliant relative to ADA and California Title 24 requirements.

Relative to fire and life safety, the facility has no rated corridors, no smoke barriers, and relies on a vehicle sallyport which is part of an archaic, non-compliant building which is not entirely separated. Many walls which are required to prevent the passage of smoke have openings in them. Exiting is compromised by programs and detox functions.

Cells and dormitories have a variety of ligature points creating suicide risks. Cells and dormitories are not constructed per maximum security requirements. There is no space on the existing site to expand the facility in any direction, and too many code deficiencies in the existing jail to allow for a renovation.

Therefore a replacement jail facility where evidence-based programs and medical and mental health services can be successfully delivered efficiently and cost effectively is necessary. The construction of this new facility fulfills a critical need of the Mono County jail and it provides a safe, highly structured and service focused custody environment where inmates of all security classifications can receive services and participate in evidence based programs.

2. SCOPE OF WORK

Describe the areas, if any, of the current facility to be replaced or renovated, and the nature of the renovation, including the number of cells, offices, classrooms or other programming/treatment spaces to be replaced or added and the basic design of the new or renovated units.

The existing Mono County jail facility is non-compliant relative to fire/life safety and ADA. The remote location of the jail requires that clinical support areas be located onsite. Furthermore, the existing jail building and site cannot be expanded. For these reasons, we believe that a replacement jail facility is the only viable solution. The proposed project will provide a secure, efficient, and compliant facility which includes the following program elements:

- (a) 48 replacement beds (2 administrative segregation beds, 22 maximum security beds, 24 dormitory beds)
- (b) New program rooms directly adjacent to housing.
- (c) New visitation space including a contact family visitation room.
- (d) A respiratory isolation cell with an ante room.
- (e) A safety cell.

- (f) New weather protected/enclosed recreation yards directly accessible from the dayrooms of the four large housing units plus an additional large exterior yard.
- (g) A new Central Control with visual control over new housing and program areas.
- (h) A new clinic area supporting medical, dental, and mental health services. The clinic will serve both jail inmates, probationers and potentially the public.
- (i) A new intake area with a covered vehicle sallyport, property storage, and laundry.
- (j) A new kitchen properly sized for 48 inmates.
- (k) New parking including accessible spaces for sheriff staff, the public, and clinical staff.

The SB 844 jail project will remedy the deficiencies that impact operations at the Mono County jail. The new facility will provide not only adequate housing but also appropriate program and treatment space. These spaces can provide inmates with a wide range of education, prevention and intervention services and opportunities to develop job skills. When inmates with this type of preparation return to the communities, they typically meet with much greater success and have much lower rates of recidivism. The foregoing offers a more detailed description of the SB 844 Project Scope.

Project Overview

The project consists of two new buildings on a new 3-acre site. The main building contains approximately 19,800 sf of Type I-B construction. This locked facility includes housing, control, administration, the public lobby and visitation, program space, a medical/dental/mental health clinic, and intake area. A stand-alone 2,100 sf kitchen building attaches to the main building with a secure covered walkway. The kitchen has free egress into a secure yard, which allows for more economical Type V construction.

Separate public and staff parking lots are located on the north side of the building which is the public face. The housing and yards all face south which is the secure side of the facility. The vehicle sallyport is attached to intake and is located on the west side of the facility with dedicated access from Twin Lakes Road. *(See Attachments 1 & 2: Mono*)

County Jail Proposed SB 844 Project Concept Plans)

Central Control

Central control is located in the center of the main building and has a 270 degree view of all inmate spaces and circulation. Central Control has CCTV coverage of remote areas of the jail which will decrease incident response time and improve staff and inmate safety. The Central Control room will also house the County's 911 answering point. This will allow the Sheriff to continue to use the current correctional deployment plan. This is an essential service and as such, requires the appropriate construction standards.

<u>Housing</u>

The new facility is planned for 48 board-rated beds. These beds can be classified as follows:

- Two 11-bed maximum security pods; each includes five standard double cells and one single ADA compliant cell. The cells are equipped with rear chases. The dayrooms include dining/game tables, a sink, an ADA shower, a janitor closet, and space for a video-visitation kiosk. Dedicated covered yards are directly accessible from the dayrooms. These pod design allows inmates to remain outside their cells as much as possible. Direct access to yards maximizes their recreation time.
- Each of the two 10-bed medium security dormitories includes five double bunks. The dayrooms are open to the sleeping area. The dayrooms include dining/game tables,

a sink, an ADA toilet, an ADA shower, a janitor closet, and space for a video-visitation kiosk. Dedicated covered yards are directly accessible from the dayrooms. These pods are designed to maximize recreation time.

- Each of the 2-bed administrative segregation pods includes one single standard cell and one single ADA compliant cell. The cells have rear chases. The dayroom provides a built-in counter with a sink and seating area, an ADA shower, and a janitor closet. The security level of the pod indicates the likelihood that these inmates spend far less time out of their cells than those inmates in other less secure pods.
- A 4-bed minimum security dormitory with two double bunks has dayrooms which are open to the sleeping area. The dayrooms include a dining/game table, a sink, an ADA toilet, and an ADA shower. Inmates in this dormitory are expected to work in the kitchen or the laundry area much of the day.
- The four large housing units have small dedicated yards. An additional large (over 600 sf) yard is accessible from the main secure corridor. This yard is open to the sky, except for a small portion covering the ADA comby toilet.

Medical, Dental and Mental Health Treatment

The new facility will accommodate medical and dental treatment as well as the mental health treatment needs of jail inmates and probationers. A respiratory isolation room (with ante room) and a safety cell facilitate delivery of these clinical services. Both are located adjacent to central control. The formal clinic area includes an exam room and dental exam/procedure room. A small interview room allows for private interviews, use as a small program room, remote video interviews or tele-psych. There is also space for medical records storage, clean/soiled utility, equipment storage, pharmaceutical storage,

and other support spaces. An inmate toilet, which is located along the main inmate corridor, facilitates urinalysis testing. The clinic design allows separation from other jail functions such as accessibility to probationers who are no longer in custody, but who may require medical, dental, or mental health services.

Programming and Services Rooms

The two program rooms are immediately adjacent to the housing pods. Each is large enough for 8 inmates and has access to a storage closet for materials and equipment. The rooms also have access to an inmate toilet, which is just outside of those rooms and adjacent to central control. The nearby family/contact visit room also may be used as a program room whenever visitation does not occur.

Administration/Staff Services

The Administration/Staff Services area is located between the accessible staff parking and the public lobby. This area includes a reception station, Sergeant's Office, Lieutenant's Office, a print/copy/mail alcove, a storage room, and file alcove. Two ADA compliant uni-sex toilets with showers and changing benches flank a shared locker room. This arrangement accommodates all genders and sexual orientations. An open conference area with seating for 10 and a kitchenette is located in the middle of the space. Central control has views into the conference area. Other spaces in administration include a security electronics room, an MDF room, a staff toilet, janitor/storage room, and a staff entry which includes space for gun lockers.

Public Lobby/Visitation

The building has a connected public lobby that serves the jail and visitation. This public area includes a uni-sex toilet and a janitor closet. The lobby has access to two non-

contact visitation rooms; one is ADA compliant. The lobby also accesses a family/contact visitation room which can be used for family visits, plea bargaining, or other program uses.

<u>Intake</u>

The intake area includes an enclosed, covered, drive-through vehicle sallyport capable of accommodating a full size van. Inside are two report stations, a staff toilet, an inmate toilet, an exam/interview room, a booking counter with live scan, two sober cells, an ADA compliant cell, 2 small cells which can hold between 1 and 4 inmates, and a safety cell. There is a dedicated classification office to facilitate interviews with inmates during booking. Intake is arranged so that access into the secure part of the jail is through a vestibule which includes an accessible shower, access to clothing and fish kit, a clothing/property drop, and a sterile holding cell. The clothing and fish kit area as well as the clothing/property drop back up to a storage room, laundry, and long term storage. The long term storage is located along the release route for inmates from intake and housing.

<u>Kitchen</u>

The kitchen is located in a remote building, connected by a secure covered walkway. The kitchen has two doors located in the direction of the jail to accommodate the flow of food carts to the jail and dirty carts as they return through separate doors. There is a dishwashing area which includes storage racks, a prep area, a cooking area with a hood, and a beverage area. There is a walk-in refrigerator, walk-in freezer, and walk-in dry storage. Kitchen amenities also include a chef office, uni-sex toilet, janitor closet, and dressing area. The kitchen has free exiting into a large yard which is also where deliveries occur.

Expansion

The jail building is designed for expansion, primarily along the east-west secure corridor. There is room to expand upon the clinic or provide additional services to the west. Additional housing wings can be located at the east end of the corridor. The staff area can expand to the east for additional offices and to the north for additional locker space.

Additional Considerations

This facility is designed to serve the County and the community as well as the needs of the jail facility itself. The site is directly adjacent to an existing medical clinic owned by the County that recently closed. The County is actively looking for a permanent health care provider to re-establish local medical service to the public and inmates. With the addition of dental and mental health space in the jail clinic, assuming inmates use the clinic in the morning, this area can be secured and made accessible to the public in the afternoon; not just to probationers, but to any of the County's citizens. This may help the County to attract service providers who would otherwise be unwilling to serve Bridgeport for a few inmates, but may be willing to serve the city if they could expand their client base.

Likewise, the kitchen can be used to cook meals in emergencies, such as for firefighters battling wildfires or other community events. It is located so that it can be secured from the jail and made accessible on the public side. The kitchen could even be used to prepare meals for senior services events held across the parking lot in the conference room of the existing clinic.

3. PROGRAMMING AND SERVICES

Describe the programming and/or treatment services currently provided in your facility. Provide the requested data on pretrial inmates and risk-based pretrial release services. Describe the facilities or services to be added as a result of the proposed construction; the objectives of the facilities and services; and the staffing and changes in staffing required to provide the services.

Current Programs and Services

Mono County Jail currently does not have any space to hold any of the offered programs. Currently, all programs are held in the booking / sobering cell hallway. There is also no space for private mental health counseling and a converted holding cell is used for medical visits. In spite of having no programming and/or appropriate medical space, the jail provides programs to inmates to the maximum degree possible. The jail provides Title 15 programs including (a) law library/legal resources, (b) recreational reading library, (c) in-person visitation, (d) recreation, (e) religious services, (f) telephone access, and (g) medical and mental health services.

Following is a list of current programming and treatment services provided in the Jail. These programs and services are provided on a limited basis due to the lack of adequate program space.

- High School Diploma GED and remedial education This program is run under the supervision of the Mono County Office of Adult Education. The focus of the Jail Program is to enable inmates in the Mono County Jail the opportunity to continue or enhance their education, obtain a high school diploma, prepare for the General Education Development Test, or complete remedial education in Junior High School/ Elementary subjects.
- *Narcotics Anonymous (NA)* This program is facilitated by volunteers from the community who meet with groups of inmates. The group atmosphere offers an
ongoing support network for addicts who wish to pursue and maintain a drug-free lifestyle.

- Alcoholics Anonymous (AA) This program is facilitated by volunteers from the community who meet with groups of inmates. Its primary purpose is to help alcoholics "to stay sober and help other alcoholics achieve sobriety."
- *Bible Study* This program is facilitated by volunteers from the community, who meet with groups of inmates and study various religious beliefs.
- Attorney Visits The jail provides a room where attorneys can meet with their clients in private. Attorneys arrive at the jail and request to meet with their client on a first come, first served basis.
- Mental Health Crisis Counseling Mono County Health & Human Services meets with inmates who have been placed on suicide watch for harming themselves or making threats to harm themselves, or demonstrate behavior which leads staff to believe behavioral health intervention is necessary. The clinician assesses the inmate and makes recommendations for continued observation or clears them to return to general population.
- Re-Entry and Medi-Cal Counseling Mono County Sheriff's Office contracted with a local non-profit to create our re-entry program. Twice a week, a re-entry coordinator meets with inmates who have no health care coverage and assists them in getting enrolled. The coordinator also ensures that inmates already enrolled in Medi-Cal know how to get their benefits reinstated upon release from custody. In addition, the coordinator works with inmates individually to arrange and ensure continuation of treatment and counseling that the inmate began while in custody, including setting up

medical and mental health counseling services and finding group programs near where the inmate will be residing once released. The coordinator also finds transportation and assists with temporary housing for indigent inmates.

- Drug and Alcohol Treatment The jail provides programs run by Mono County Behavioral Health. Drug and Alcohol counselors facilitate the programs. The program addresses the nature of addiction, underlying causes of addiction, relapse, and finding sober supports.
- Evidence Based Programming Moral Recognition Therapy (MRT) is the only Evidence Based Program currently being offered due to a lack appropriate programming space. MRT is a cognitive-behavioral counseling program that combines education, group and individual counseling, and structured exercises designed to foster moral development in treatment-resistant clients. As long as clients' judgments about right and wrong are made from low levels of moral reasoning, counseling them, training them in job skills, and even punishing them will have little long-lasting impact on their behavior. They must be confronted with the consequences of their behavior and the effect that it has had on their family, friends and community. Poor moral reasoning is common within at-risk populations.

Pretrial ADP

During the period of January 1, 2016 through December 31, 2016 the Mono County Jail had an Average Daily Population (ADP) of 28 inmates. The ADP of inmates on pretrial status over this period was 16. The percentage of inmates on pretrial status for this time period was 57%.

Mono County's Risk-assessment-based Pretrial Release Program

Mono County's methodology for reducing the need for jail custody beds is a product of collective efforts among local agencies including the Sheriff's Department, Superior Court, District Attorney and Probation. The primary alternative to incarceration program has been substantially expanded with the implementation of realignment. Mono County has developed a pre-trial release plan that incorporates a "two tier plan" for releases. A lower tier is implemented when the jail facility is under 90% rated capacity and an upper tier is added when the facility is 90% or over its rated capacity. Lower tier releases account for 47.5% of all Mono County Jail releases during 2016. The basis for these releases include:

- Intoxication Only Release: Under normal conditions, Mono County releases all inmates on their Own Recognizance or, in some instances per Penal Code Section 849(b), who are arrested only for intoxication, including first time driving under the influence misdemeanor offenses, once the subject is sober and deemed no longer a public safety risk.
- PC 4030 Release: In order to protect human dignity, Mono County will release, on their Own Recognizance, any arrestee who does not meet the Penal Code section 4030 criteria allowing for pre-arraignment strip searching and housing within the facility.
- *District Attorney Pre-arraignment Release*: The District Attorney may, after careful review and determination that the inmate/arrestee is not a continuing danger to others or property and has local ties to the community, issue an order to release the subject on their Own Recognizance pre-arraignment.

- Court Ordered Conditional Own Recognizance Release: The Court working with the District Attorney, Probation and Public Defender may order a release requiring a subject to comply with Court-ordered conditions beyond those normally associated with a traditional pre-arraignment Own Recognizance Release (such as a stay away order).
- *Presumptive Bail Schedule Releases*: Any arrestee who is not intoxicated and able to care for him/herself may arrange to be released on bail, before arraignment, at the amount contained in the schedule of bail for the offense, if so allowed by law.
- Bail Release per PC 1270/1275: The Court shall set, raise, reduce or deny bail after taking into consideration the protection of the public, the seriousness of the offense charged, the previous criminal history of the defendant, and the probability of the defendant appearing at trial or hearing. In all cases, public safety shall be the primary concern.
- Once the jail population reaches 90% of the rated capacity of the facility a second tier release plan is implemented in order to manage and control the need for custody beds.
- Pre-Trial/Sentencing Felony Agreement to Appear. The Mono County Sheriff, the Superior Court and the District Attorney have entered into an agreement whereby lower level pre-trial/sentenced felony offenders may be released from the Mono County Jail on an agreement to appear after having met criteria set by the Court in conjunction with the District Attorney. This review for felony release usually applies to offenders who live locally in the area (Mono and adjoining counties) and have no failures to appear in their arrest history. The arrestee would be disqualified if on felony probation or parole or have any other felony arrests on their record. Arrestees will

also be disqualified if they are a serious threat to public safety or arrested on felony violence charges. Also, there cannot be a reasonable likelihood that the arrestee will continue the criminal conduct for which initially arrested. Preference will be given to inmates who currently are employed.

- Alternative Custody Programs: Currently, through the Mono County Probation Department, drug court/diversion is being offered to qualified inmates.
- Alternative Sentencing Program (ASP): The Mono County Court, in conjunction with the District attorney's Office, regularly modifies sentences to allow inmates who are employed to continue their employment. We believe that this effort helps reduce recidivism substantially by allowing the offender to remain employed and thus return to a stable release status.
- *Alternative Work Program*: This is an in-house program that allows inmates who work either within the jail or on County or non-profit projects to receive a reduced sentence on a scale consummate with the time spent working.

Project Objectives for Programs and Services

The new jail facility will provide Mono County its first medical, dental, telepsych/telemedicine, and programming space. The program rooms of different sizes will be used for a wide range of evidence based programs, group therapy, education, prevention and intervention services and opportunities to develop life and vocational skills that will prepare inmates for their return to the community.

Medical, Dental and Mental Health Treatment

Inmate services space is planned to accommodate the medical, dental and mental health treatment needs of the jail population.

- Medical Exam: The medical exam room will improve inmate access to medical services. Unlike the existing medical exam room, which was re-purposed from a holding cell, the new room will be appropriately sized and equipped to support the needs of our medical providers. In addition, this exam room is located such that inmates do not need to cross circulation paths with new arrestees (the current exam room is in the intake area). It will allow for greater access to inmates and to service providers in a more secure, efficient manner.
- Dental Exam: The dental exam room will allow basic dental services to be provided at the jail and thereby eliminate the current practice of transporting inmates out of the jail for basic preventative dental services. This will not only allow for greater access of dental services to inmates but also eliminate the security risk associated with inmates leaving the jail. Providing a dental exam room in the jail will also reduce an unnecessary burden on staffing due to transportation.
- Respiratory Isolation: The respiratory isolation room will allow inmates with contagious diseases to remain at the jail with appropriate treatment and medical isolation. This room will be far superior to our current limited ability to isolate contagious inmates in a sobering cell which is not adequately designed for sick inmates, or keeping them in the general population which allows spread of disease among other inmates.
- Safety Cell: The safety cell will provide a safe location to temporarily hold an inmate in mental health crisis. Without such an amenity, an inmate is at greater risk of harming themselves or others.
- *Telemedicine room:* The telemedicine/telepsychiatry room will allow for one-on-one interviews and/or treatment in a private setting via video conference equipment.

Currently, there is not a secure, private location in the jail for counseling or medical discussions. Telemedicine is vitally important to our inmates due to our remote location and extreme weather, which prevents some providers from reaching us during the winter months.

 Classification / Re-entry Coordinator Office: This space will allow our classification team to interview inmates in a private, secure space and will also house our re-entry team, who provide the inmates with the best transition opportunities for success after incarceration. Currently such interviews are conducted in an open hallway.

Resolving Outdated Housing

The Mono County Jail was built in 1988. It is outdated and a retrofit of the existing building would be cost prohibitive. See Statement of Need. The bed count for the facility would remain the same at 48.

- A new jail will allow the staff greater flexibility to manage higher security inmates by utilizing the double bunked cells for single occupancy when needed. The new jail will also have ADA cells that are compliant with today's expectations and laws. The new jail will also meet our needs to classify and appropriately house inmates with housing space designed for inmate worker housing, administrative segregation, protective custody and medical isolation.
- Finally, the new housing will provide Inmates with direct access to recreation yards and programs spaces from their dayrooms, which maximizes opportunities for inmates to be outside of their cells, with less security risk than currently. These yards will be heated and covered, allowing inmates the opportunity to recreate in our extreme

winter weather conditions. We will also be able to provide a multi-use outdoor recreation area with natural sunlight, meeting Title 15 requirements.

Staffing for Programming, Treatment Services, and Housing

Mono County Sheriff's Office and their justice community partners through the Community Corrections Partnership (CCP) share a common interest in the successful pursuit of SB 844 funding. These partners have collaborated in the planning and development of the conceptual project and will soon begin planning for the staffing needs of new evidence based programs and treatment services at the new facility.

Inmate Programming Services: The justice partners and community service partners, including Mono County Office of Education, Mono County Behavioral Health Department, the County Social Services Department, and Probation will continue to support programming needs at the jail. Funding for these programs will be provided through existing revenue sources such as the Mono County CCP. The CCP has discussed and supports in-custody programming but the lack of programming space at the current jail has stifled the development of new programs. When new programming space is available, the Community Corrections Budget managed by the CCP will be able to fund staffing for unmet jail programming needs.

<u>Medical Services:</u> The County is actively looking for a permanent health care provider to re-establish local medical service in the existing clinic directly adjacent to the proposed project site. The addition of dental and mental health space in the jail clinic, assuming the area can be secured and made accessible to the public at regularly scheduled times, may help the County to attract service providers who would otherwise be unwilling to

serve Bridgeport for a few inmates, but may be willing to serve the community if they could expand their client base.

<u>Mental Health Treatment Services:</u> The Mono County Behavioral Health Department would be able to provide more comprehensive services (including telepsychiatry) once we have appropriate facilities to do so. Psychiatry services are very limited in our remote county and our inmate population would benefit from counseling opportunities unavailable due to lack of resources.

<u>Housing and Custody Staffing:</u> The new facility will not drastically change the day-to-day duties of the correctional officers. Given that the new facility has no increase in beds, we expect to operate at our current staffing levels.

The conceptual design for the new facility includes a fixed post/control room that will become the primary control center. No new space for jail supervision or administration will be created and the new facility will function efficiently with no additional jail staff.

4. ADMINISTRATIVE WORK PLAN

Describe the steps required to accomplish this project. Include a project schedule, and list the division/offices including personnel that will be responsible for each phase of the project, and how it will be coordinated among responsible officials both internally and externally.

Mono County has developed a project management, construction and administrative work plan specifically to address the needs of the proposed SB 844 Mono County Jail Programs, Treatment and Replacement Housing project. This work plan leverages County resources to the fullest while assuring the project scope, project schedule, construction budget, and construction quality are maintained. The work plan methodology described below is consistent with other capital improvement projects undertaken by the County and has been successfully utilized for other adult detention facilities throughout the state.

- County Contracts for Architectural and Engineering Services;
- County contracts with a construction management firm for the construction oversight of the project;
- County submits site assurance and real estate due diligence package;
- County Board of Supervisors contracts with an architectural firm development of plans and specifications, and assistance during the design-bid-build process;
- County submits Project Establishment documents for the State Public Works Board (SPWB) approval and Project Establishment;
- County staff work with the architectural firm to prepare preliminary plans;
- County prepares an Operational Program Statement and Staffing Plan including anticipated operating costs;
- County submits preliminary plans to Board of State and Community Corrections (BSCC) and the State Fire Marshal for review and approval. The submittal will include an Operational Program Statement and Staffing Plan;
- County staff work with the architectural firm to prepare working drawings;
- County submits construction documents to Board of State and Community Corrections (BSCC) and the State Fire Marshal for review and approval;
- Department of Finance approves working drawings and issues a Proceed to Bid;
- Bids are received and evaluated;
- County Board of Supervisors awards a construction contract;

- Department of Finance approves Construction Contract and Issues a Notice to Proceed for construction;
- Construction is initiated;
- Construction, including punch list, is completed;
- Facility is commissioned and construction contract is closed out;
- Occupant move-in proceeds;
- Operation of new facility begins;

Project Management

The Mono County SB 844 Jail Replacement project will be handled from start to finish by a Project Oversight Team and a Project Management Team supplemented by a construction management (CM) consultant and architect/engineer (A/E) consultant.

The County has organized a Jail Construction Committee that has been involved in the programming and conceptual planning of the SB 844 project. The team members consist of representatives of County Administration, Public Works, the Sheriff's Office and County Financial Services.

Jail Construction Committee: John Peters – Mono County Supervisor; Ingrid Braun – Mono County Sheriff; Leslie Chapman – County Administrator; Janet Dutcher – County Finance Director, Stacey Simon – County Counsel, Garrett Higerd – County Engineer.

The Committee is responsible to provide regular updates and review to the Board of Supervisors and provide oversight, direction and guidance to the Project Management Team. Garrett Higerd has also been designated as the County Construction Administrator. The Project Management Team members will consist of representatives of the Sheriff's Office, Administrative Office and representatives of other County agencies as needed.

Project Management Team:

Shawn Minder – Jail Manager; Garrett Higerd – County Engineer; Representatives of other County Agencies as needed including County Counsel, County Administration, Behavioral Health, Information Technology, Community Development, etc.

The Mono County Public Works will provide the overall coordination of the project and provide direct administration and oversight of the Project Management Team. They will also oversee and manage the contracted (CM) firm and (A/E). Garrett Higerd, County Engineer, will serve as construction project management lead.

Sheriff's Office members of the team will represent the interests of the Sheriff and provide technical assistance and operational expertise, and will form the Transition Team. In addition to other responsibilities the Transition Team will be responsible for planning and implementation of new programs and hiring and training new staff.

Finance Director Janet Dutcher will serve as the Financial Officer for the project coordinating with other County offices on all financial aspects of the project, including close coordination with the Auditor-Controller and Treasurer Tax-Collector for cash management of project costs. The Project Management Team will also include the expertise, as needed, of staff from other County Agencies and Departments: Sheriff, Facilities, County Administrative Office, County Counsel, County Probation, Health, and Human Services, and others.

Clerical support for accounting, project record keeping and documentation will be provided by Public Works and the Sheriff's Office. A full-service CM firm will be contracted to provide construction management throughout the complete project. A contracted A/E will be responsible for the development of the design and construction documents and assisting the county in bidding and construction administration.

Public Works will process consultant and contractor payment applications, and in turn, submit them to the State for reimbursement. Designated County staff will respond to the Board of State and Community Corrections (BSCC) on all contract matters, and the Architect will respond to all minimum codes and standards issues. The construction management firm will assist with the monitoring, scheduling, and coordinating all activities on behalf of the County and Sheriff's Office. *(See Attachment 4: Mono County Jail*)

Proposed SB 844 Project Schedule)

5. BUDGET NARRATIVE

Describe the amounts and types of funding proposed and why each element is required to carry out the proposed project. Describe how the county will meet its funding contribution (match) requirements for all project costs in excess of the amount of state financing requested and how operational costs (including programming costs) for the facility will be sustained.

The County of Mono, and planning consultants worked collaboratively to determine the project costs identified in the budget summary table. The following narrative provides an overview of these costs:

Construction Cost (\$21,314,000): With these construction dollars, the County will have the necessary funds to carry out the objectives of this project by constructing the new standalone facility with programming and treatment space and replacement housing. Additionally, the county will construct a new parking area for staff and new accessible parking stalls for visitors. The estimated cost of construction was determined in February 20176 dollars and then escalated to the mid-point of construction based on the anticipated project schedule. Included in the construction (bid) costs are; fixed equipment and

furnishings, change order contingency, and a geographic location factor. Construction cost estimates were provided by a Vanir, a firm experienced with jail construction, and based on a conceptual building plan and site layout prepared by Nacht & Lewis. The County has included \$200,000 to demolish and remediate existing structures on the site as part of its cash contribution.

Additional Eligible Costs (\$1,140,000): Additional eligible costs include: Furniture fixtures and equipment (\$640,000): Includes moveable furnishings for the new facility including furnishings for dayrooms and inmate programs and treatment space. In addition audio visual equipment and office furnishings, shelving will be purchased. Medical equipment such as exam table and tele-med and tele-psych equipment, as well as dental equipment are included; *Materials Testing and Inspection (\$345,000)*: For construction materials and testing and full time inspection services during construction; *Building Department Plan Check Fees (\$60,000)*: The County will contract with a service provider to review the plans for compliance with the Building Code. The County has included (\$95,000) for utility upgrades and site due diligence services occurring prior to project establishment as part of its cash contribution.

Architectural (\$2,846,000): The County will contract with an Architectural firm experienced in Jail design and construction to provide planning, design and construction contract documents as well as construction administration services. All required engineering services will be provided under this contract. The Architect will assist the county with services needed for project establishment. The County has included \$525,000 for these services as part of its cash contribution.

Construction Management (CM) (\$1,065,000): The County intends to contract with an experienced CM firm to support the County's project management efforts and construction phase management of the construction contract. The CM will also provide an independent opinion of construction costs. The CM will assist the county with services needed for project establishment. The County has included \$545,000 for these services as part of its cash contribution.

Cash Match

The Board of Supervisors has unanimously approved submitting this application for SB 844 fund and has acknowledged the funding requirements, including \$1,613,000 in cash match. The County has appropriated, or will appropriate the amount of cash contribution funds after conditional project award but before state/county funding agreements. The Project proposed in the County's SB 844 Financing Program proposal is authorized to proceed in its entirety when and if state financing is awarded for the Project within the SB 844 Financing Program.

Sustaining the Cost of Operations:

The Sheriff's Office and County construction staff are seeking building designs with minimum staffing cost that will translate into the lowest long-term lifecycle cost expense to community taxpayers. The Sheriff's Office has prepared an estimate of staffing needs for the new facility and no new Correctional Officer positions will be required. It is anticipated that the increase in energy costs for the larger new facility will be offset by higher energy efficiency standards.

The County, through its submittal of this SB 844 funding proposal, is committing to staff and operate the new facility. With a conditional funding award the County will commit,

through its budgetary practices and long range financial planning, the funding to operate the new facility. This will include a planning effort of the County CCP to fund the necessary staffing and support for evidence based programs and mental health treatment.

6. READINESS TO PROCEED

Mono County's Board of Supervisors has approved a resolution which is included with this application. The resolution adopts an Addendum to the Environmental Impact Report (EIR) prepared in conjunction with the Regional Transportation Plan and General Plan Update in 2015 evidencing CEQA compliance, appoints the County's representatives for the project, approves the form of the project documents, authorizes the County Administrator to sign and submit the proposal, identifies an adequate amount of matching funds, commits to staff and operate the jail upon project completion, and attests to the County's ownership of the site.





Attachment 3: Mono County Jail Proposed SB 844 Project

Mono County Sexual Abuse Prevention Program

Following are the key elements of Mono County Sheriff's Office Sexual Abuse Prevention Program for the County's current and proposed new Jail Facility.

- All Staff, Service Providers and volunteers who have access to inmates undergo training including mandatory reporting.
- All reported violations are investigated by our multi-agency, sexual assault team.
- All staff and providers, whether employees or volunteers, receive additional training on sexual harassment, and sexual victimization prevention and awareness.
- Classification and medical screening at intake to determine level of risk.
- Separation of those inmates determined through our classification system that are likely to victimize or be victimized.
- Good lighting to inmate areas of the facility.
- Recorded video surveillance of inmate areas, with the exception of privacy areas.
- Regular reviews of phone audio recordings and viewing of all non-legal mail for potential victimization.
- Signage throughout the facility notifying inmates of their rights to be free from sexual abuse and harassment.(English and Spanish).
- Pamphlets and handbooks providing inmate information about their rights and resources regarding victimization. This includes information on how to report and gives options of who to report to. (English and Spanish)
- Phone number to sexual abuse hotline is displayed on all written information, including wall mounted signage.



Attachment 4: Mono County Jail SB 844 Project Schedule

SECTION 6: BOARD OF SUPERVISORS' RESOLUTION

All counties applying for SB 844 financing must include a Board of Supervisors' resolution with the proposal submittal. The resolution must include the requisite components as outlined below. For counties submitting multiple proposals (which requires participation in a regional ALCJF as described in the RFP), separate resolutions for each proposal, with the necessary language contained in each resolution, are required.

The Board of Supervisors' resolution for the project shall be attached to the original proposal and must contain all of the following:

- A. Names, titles, and positions of county construction administrator, project financial officer, and project contact person.
- B. Approving the forms of the project documents deemed necessary, as identified by the board (SPBW) to the BSCC, to effectuate the financing authorized by the legislation.
- C. Authorization of appropriate county official to sign the applicant's Agreement and submit the proposal for funding.
- D. Assurance that the county will adhere to state requirements and terms of the agreements between the county, the BSCC, and the SPWB in the expenditure of state financing and county match funds.
- E. Assurance that authorizes an adequate amount of available matching funds to satisfy the counties' contribution. The identified matching funds in the resolution shall be compatible with the states' lease revenue bond financing. (see page 2 of this form for description of compatible funds)
- F. Assurance that the county will fully and safely staff and operate the facility that is being constructed (consistent with Title 15, California Code of Regulations, Chapter 1, Subchapter 6 section 1756 (j) 5) within 90 days after project completion.
- G. All projects shall provide the following site assurance for the county facility at the time of proposal or not later than 90 days following the BSCC's notice of Intent to Award: 1) assurance that the county has project site control through either fee simple ownership of the site or comparable long-term possession of the site and right of access to the project sufficient to assure undisturbed use and possession of the site; and, 2) will not dispose of, modify the use of, or change the terms of the real property title, or other interest in the site of facility subject to construction, or lease the facility for operation to other entities, without permission and instructions from the BSCC, for so long as the SPWB lease-revenue bonds secured by the financed project remain outstanding.
- H. Attestation to \$_____ as the current fair market land value for the proposed new or expanded facility. This can be claimed for on-site land value for new facility

construction, on-site land value of a closed facility that will be renovated and reopened, or on-site land value used for expansion of an existing facility. It cannot be claimed for land value under an existing operational facility. (If claimed as in-kind match, actual on-site land value documentation from an independent appraisal will be required as a pre-agreement condition.)

I. Regional ALCJF projects only: A Board of Supervisors' resolution from all counties in the regional partnership containing the items identified above, along with a Memorandum of Understanding (MOU) or Joint Powers Agreement (JPA) between each of the partner counties. Please consider the information about regional ALCJFs for the purposes of this funding program as described in the "Eligible Projects" section, "Limit on Number of Projects/Set Asides" sub-section of the RFP, before developing these documents. If preliminary MOUs and JPAs are submitted, final documents must be submitted within 90 days following the notification to the lead county of conditional Intent to Award state financing.

Note: Additionally, refer to "Section 5: Narrative - Readiness to Proceed."

MONO COUNTY GENERAL PLAN EIR ADDENDUM #17-01 New Jail Facility at 221 Twin Lakes Road December 15, 2016

Project Description

Mono County is proposing to demolish an existing building and construct a new jail facility at 221 Twin Lakes Road, Assessor's Parcel Number (APN) 008-080-007-000, in Bridgeport, California. The current jail facility to be replaced is located at 94 N. School Street (along the Emigrant Street side of the parcel) in Bridgeport. (See attached map, Exhibit 1.)

Land Use Designation

The parcel has a Land Use Designation of Public and Quasi-Public Facilities (PF), and is approximately 3.4 acres. The jail facility is a permitted use under "other uses permitted by the public landowner," and this Land Use Designation does not have maximum lot coverage, minimum setbacks, or maximum building height requirements.¹

APN 008-080-007-000 is surrounded by a single, privately-owned parcel with an Agriculture (AG) Land Use Designation. A 0.2 acre parcel to the south is designated PF and owned by the Bridgeport Public Utilities District. To the north, land uses are a mix of multi-family residential – low (MFR-L) and commercial (C), with a few scattered PF parcels. The Eastern Sierra Unified School District offices and elementary school are located approximately 0.1 miles northeast (measured from parcel boundary to parcel boundary). (See attached map, Exhibit 1.)

Existing Uses

The following uses currently exist on the parcel:

Building/Facility	<u>Size</u>	<u>Use</u>		
Old General Hospital	16,796 square feet (sf)	Cold storage		
Medic 7 trailer	1,050 sf	Office space for emergency medic staff		
Medic 7 garage	960 sf	Houses emergency response vehicle		
Twin Lakes Annex	3,222 sf	Conference room, health clinic		
Helipad	N/A	Paved area for emergency airlifts		

The entire parcel is impacted by prior development, with little to no native vegetation remaining. Structures, facilities (such as the helipad), paved surfaces for interior circulation and parking, irrigated turf, and hardened dirt driveways and parking currently cover the parcel. Some native vegetation may be remaining or may have re-established around the edges of the parcel.

Proposed Jail Facility

The proposed jail facility contains the following structures and facilities (see attached site plan, Exhibit 2):

- An approximately 21,796 sf main building that includes housing, control, administration, public lobby and visitation space, program space, medical/dental/mental health clinic, and intake area, as well as a detached kitchen building connected to the main building by a secure covered walkway, with free egress into a secure yard.
- Separate public and staff parking lots on the publicly facing side of the building (north side).
- A secure yard on the secure side of the building (south side).
- Vehicle sallyport attached to the main building intake and located on the west side of the facility with dedicated access from Twin Lakes Road.
- Medical provider offices and a public lobby will be provided in the existing Twin Lakes Annex building.

These facilities will accommodate the following program elements:

- 48 replacement beds (2 administrative segregation beds, 22 maximum security beds, 24 dormitory beds)
- New program rooms directly adjacent to housing.
- New visitation space including a contact family visitation room.
- A respiratory isolation cell with an ante room.

¹ See the 2015 Mono County General Plan Land Use Element, Public and Quasi-Public Facilities (PF) Land Use Designation, available at <u>http://www.monocounty.ca.gov/sites/default/files/fileattachments/planning_division/page/812/2015_land_use_final.08.15_0.pdf</u>.

- A safety cell.
- New weather protected/enclosed recreation yards directly accessible from the dayrooms of the four large housing units plus an additional large exterior yard.
- A new Central Control with visual control over new housing and program areas, and also includes the County's emergency (911) dispatch center.
- A new clinic area supporting medical, dental, and mental health services including staff areas. The clinic could serve both jail inmates, probationers and the public. Offices, the public lobby, and possibly some services may be provided in the existing clinic in the Twin Lakes Annex building located to the north on this same parcel.
- A new intake area with a covered vehicle sallyport, property storage, and laundry.
- A new kitchen properly sized for 48 inmates.
- New parking including accessible spaces for sheriff staff, the public, and clinical staff.

The old general hospital building will be demolished in order to accommodate the new jail structure. Demolition will occur according to applicable standards and regulations, including Mono County building permits and inspections. Any hazardous waste material removal, handling and disposal (e.g., potentially asbestos) shall also comply with applicable standards and regulations, such as permits and inspections by the Great Basin Unified Air Pollution Control District.

The new construction, parking and driveways occur primarily on the footprint of the old general hospital building and/or pavement or hardened dirt used for parking and interior circulation. A small portion of the detached kitchen building (less than 1,900 sf) and some hardened surface for interior vehicle circulation will be constructed on existing irrigated turf. All of these surfaces are previously impacted. Proposed future uses shown on the site plan also occur on previously impacted surfaces, primarily on paved or hardened dirt surfaces currently used for parking, interior circulation, or as the helipad. Construction shall utilize best management practices to prevent impacts from run-off or dust, and stormwater management shall be incorporated into the site design.

The project shall be designed as an essential services facility and will meet applicable standards and regulations of the State Architect, State Fire Marshal, and local regulations of the County, including building codes, fire districts, and any other applicable agencies.

The proposed building is anticipated to have a maximum height of 25', and shall not exceed 35'. This height is similar to the old general hospital building to be demolished, and consistent with the maximum allowable height of a single-family residence. Exterior lighting shall be dark sky compliant (Chapter 23 of the General Plan), and the complex oriented in such a manner that outdoor inmate presence and use is screened by the buildings and covered walkway from public uses (residential, school, public clinic use and parking) to the north and east. Potential Bi-State sage-grouse habitat will be protected by incorporation of applicable conservation measures as indicated in the General Plan Conservation/Open Space Element, particularly Action 2.A.3.e.

Public Outreach

The proposed jail facility has been agendized and discussed at the following public meetings: two Board of Supervisor meetings, one Planning Commission meeting, and one Bridgeport Regional Planning Advisory Committee (RPAC) meeting. Specific outreach to the Bridgeport Indian Colony was completed, including a phone conversation with the Environment Director about the proposed project and invitations to the RPAC meeting. A tribal representative was present at the RPAC meeting, and the County will continue to coordinate with the tribe as needed.

Environmental Review & CEQA Provisions for Preparation of an Addendum to a Final EIR

In 2015, Mono County certified an Environmental Impact Report (EIR) for the Regional Transportation Plan/General Plan Update (SCH #2014061029). The General Plan EIR analysis included the Land Use Designations and associated standards such as permitted uses and development standards, and identified the following significant and unavoidable impacts: biological resources; geology, soils, minerals; public health & safety, hazards, hazardous materials; cultural resources; hydrology, flooding, water quality, water supply; aesthetics, light & glare, scenic resources; and public services and utilities.

As discussed below, an addendum to the 2015 RTP/General Plan Update EIR is the appropriate level of environmental review for the proposed amendments, because none of the conditions set forth in CEQA Guidelines section 15162 exist.

The California Environmental Quality Act (CEQA §15164[a]) states:

"(a) The lead agency or a responsible agency shall prepare an addendum to a previously certified EIR if some changes or additions are necessary but none of the conditions described in Section 15162 calling for preparation of a subsequent EIR have occurred."

In turn, §15162 states that preparation of a subsequent EIR is required where one or more of the following occurs:

"(a) When an EIR has been certified or a negative declaration adopted for a project, no subsequent EIR shall be prepared for that project unless the lead agency determines, on the basis of substantial evidence in the light of the whole record, one or more of the following:

(1) Substantial changes are proposed in the project which will require major revisions of the previous EIR or negative declaration due to the involvement of new significant environmental effects or a substantial increase in the severity of previously identified significant effects;

(2) Substantial changes occur with respect to the circumstances under which the project is undertaken which will require major revisions of the previous EIR or negative declaration due to the involvement of new significant environmental effects or a substantial increase in the severity of previously identified significant effects; or

(3) New information of substantial importance, which was not known and could not have been known with the exercise of reasonable diligence at the time the previous EIR was certified as complete shows any of the following:

(A) the project will have one or more significant effects not discussed in the previous EIR or negative declaration;

(B) significant effects previously examined will be substantially more severe than shown in the previous EIR;

(C) mitigation measures or alternatives previously found not to be feasible would in fact be feasible, and would substantially reduce one or more significant effects of the project, but the project proponents decline to adopt the mitigation measure or alternative; or

(D) mitigation measures or alternatives which are considerably different from those analyzed in the previous EIR would substantially reduce one or more significant effects on the environment, but the project proponents decline to adopt the mitigation measure or alternative."

Discussion of Impacts

The project is on a previously developed and impacted site, and consistent with the Land Use Designation analyzed in the General Plan EIR. Therefore, the proposed project does not require a major revision of the 2015 General Plan EIR due to any new significant environmental effects.

The proposed project also does not substantially increase the severity of the previously identified significant effects, as analyzed below (see Exhibit 3 for a map of these environmental considerations):

Biological Resources

- The site is previously impacted and developed with buildings, pavement, and turf. The parcel was evaluated in the General Plan EIR Biological Assessment and no sensitive plant communities were identified. However, vegetation indicative of wetlands may exist in this area. As noted below in the Hydrology section, this parcel is not mapped in the National Wetlands Inventory as a potential wetland.
- No Bi-State sage-grouse leks are located within five miles of the parcel. (See Environmental Constraints Map, Exhibit 3.)

Geology, Soils, Minerals

- The site is not located in a known Alquist-Priolo earthquake fault as delineated by the State Geologist or based on other evidence. (See Environmental Constraints Map, Exhibit 3.)
- No evidence exists for strong seismic ground shaking, and the topography is not conducive to landslides.
- Seismic-related ground failure, including liquefaction, shall be evaluated via appropriate geotechnical soils reports and studies during project engineering, and the necessary design standards shall be incorporated. The engineering standard

may be higher than for a typical commercial or public structure as this facility is an essential services facility because it houses emergency dispatch.

Public Health & Safety, Hazards, Hazardous Materials

- The site is not located in an area exposed to significant natural hazards, such as avalanche, landslides, rockfall, or volcanic activity. The new construction shall be subject to Mono County Building standards, which includes locally adopted standards for wind and snow loading requirements.
- The parcel is located in a fire hazard area (see Environmental Constraints Map, Exhibit 3); however, defensible space will be maintained in compliance with applicable standards. In addition, this project replaces an existing structure, and the old jail facility is also located within the same fire hazard area. Therefore, the project does not increase the severity of fire hazard impacts.
- The helipad may continue to be used for a period of time after construction; however, the long term plan is to relocate helipad uses to Bryant Field airport, approximately 0.6 miles to the northeast. The distance is insignificant as Bryant Field is located approximately the same distance from the commercial district of Bridgeport, and closer to residential neighborhoods to the northeast and southeast. Therefore, the relocation of helipad uses, such as medical airlifts, to Bryant Field does not impact emergency services. (See land use map, Exhibit 1.)
- Safety hazards related to the transportation or release of hazardous materials are not applicable, except potentially during
 the demolition phase of the project. As noted in the project description, hazardous waste material removal, handling, and
 disposal will be compliant with applicable standards and regulations. If any hazardous wastes are generated by routine
 construction and operations, these materials will be handled and disposed of in compliance with applicable standards and
 regulations.

Cultural Resources

• The site is previously impacted and developed with buildings, pavement, and turf, and no known cultural resources exist on the site. The Bridgeport Indian Colony has been contacted and attended a public meeting about the project, and no concerns have been communicated to the County at this time.

Hydrology, Flooding, Water Quality; Water Supply

- The jail facility will be connected to the Bridgeport Public Utilities District for water and sewer service, and therefore will not violate any water quality standards, or wastewater treatment or discharge requirements.
- The project is not located in a 100-year flood hazard area as mapped by the Federal Emergency Management Agency (FEMA), and does not expose people or structures to flood or related hazards. (See Environmental Constraints Map, Exhibit 3.)
- The parcel is not mapped as a potential wetland in the National Wetlands Inventory database. (See Environmental Constraints Map, Exhibit 3.)
- Impervious surface will increase very slightly on the parcel given less than 1,900 sf the new facility footprint and some hardened surfaces for vehicle circulation will extend into area currently occupied by turf. Stormwater management shall be incorporated into the site design as applicable to prevent impacts of drainage from increased impervious surfaces.

Aesthetics, Light & Glare, Scenic Resources

- The building essentially replaces an existing building, with minimal expansion of the footprint on the eastern section of the parcel and a maximum height not exceeding that of a single family residence. Therefore, the aesthetic impacts and impacts to scenic resources are similar to existing conditions.
- Exterior lighting shall comply with the County's Dark Sky requirements (Chapter 23 of the General Plan) to prevent fugitive light and glare impacts.

Public Services and Utilities

- The proposed project mitigates or reduces impacts identified in the General Plan EIR.
 - The existing Mono County jail facility is non-compliant relative to fire/life safety, Americans with Disabilities Act requirements, and certain programming features. The proposed project will remedy these deficiencies to maintain acceptable public services as required for inmate facilities.
 - The design of the building to meet essential service facility standards for emergency dispatch operations improves the security of the building and therefore the ability of the County to provide emergency communications during disasters.

• The public medical clinic in Bridgeport currently lacks an operator; however, the needed public medical services could be provided through the jail medical facility.

Conclusion

CEQA Sections 15164(c) through 15164(e) states, "An Addendum need not be circulated for public review but can be included in or attached to the final EIR or adopted negative declaration. The decision-making body shall consider the addendum with the final EIR or adopted negative declaration prior to making a decision on the project. A brief explanation of the decision not to prepare a subsequent EIR pursuant to §15162 shall be included in an addendum to an EIR, the lead agency's findings on the project, or elsewhere in the record. The explanation must be supported by substantial evidence."

The information presented above indicates that the proposed General Plan Amendment does not require a major revision of the 2015 General Plan EIR due to substantive changes to the number of significant effects, severity of effects, or the feasibility and or effectiveness of applicable mitigation measures or alternatives previously addressed in the EIR.

Specifically, no substantial changes are proposed to the project which will require major revisions of the 2015 EIR due to the involvement of new significant environmental effects or a substantial increase in the severity of previously identified significant effects;

There are not substantial changes with respect to the circumstances under which the project is undertaken which will require major revisions of the 2015 EIR due to the involvement of new significant environmental effects or a substantial increase in the severity of previously identified significant effects;

There is not new information of substantial importance, which was not known and could not have been known with the exercise of reasonable diligence at the time the 2015 EIR was certified as complete which shows any of the following:

- (A) the project will have one or more significant effects not discussed in the previous EIR or negative declaration;
- (B) significant effects previously examined will be substantially more severe than shown in the previous EIR;

(C) mitigation measures or alternatives previously found not to be feasible would in fact be feasible, and would substantially reduce one or more significant effects of the project, but the project proponents decline to adopt the mitigation measure or alternative; or

(D) mitigation measures or alternatives which are considerably different from those analyzed in the previous EIR would substantially reduce one or more significant effects on the environment, but the project proponents decline to adopt the mitigation measure or alternative."

Therefore, a subsequent EIR is not required because none of the conditions set forth in CEQA Guidelines section 15162 exist for this project.

Exhibit 1: Existing Land Use Map, Bridgeport

Available at http://monomammoth.maps.arcgis.com/apps/Viewer/index.html?appid=8670c63cda0540b39c3ae388cdd7db78





Exhibit 3: Environmental Constraints Map





3. <u>Project Contact Person</u> Garrett Higerd, County Engineer

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SECTION THREE: The form of the project documents, including the Project Proposal, Board of State and Community Corrections (BSCC) Jail Construction Agreement, Project Delivery and Construction Agreement, Right of Entry for Construction and Operation Agreement, Facility Lease, Facility Sublease, Ground Lease, and any other forms of the project documents deemed necessary, as identified by the State Public Works Board (SPWB) to the BSCC, to effectuate the financing authorized by the legislation, are hereby approved.

SECTION FOUR: Leslie Chapman, Mono County Administrative Officer, is hereby authorized to sign the applicant's Agreement and submit the proposal for funding.

SECTION FIVE: The County will adhere to state requirements and terms of the agreements between the County, the BSCC, and the SPWB in the expenditure of state financing and County match funds.

SECTION SIX: An adequate amount of available matching funds to satisfy the County's contribution, as identified in the Proposal budget, will be obtained for the Jail Replacement
 Project in the form of an investment pool loan or bank line of credit having a term of approximately five-years if an award is offered. Annual loan payments will be funded through a combination of budgetary expenditure savings, expenditure deferrals and if necessary, temporary expenditure reductions. This funding is compatible with the states' lease revenue bond financing, which is set forth on page 2 of the BSCC's Proposal Instructions, which are incorporated by this reference.

SECTION SEVEN: The County will fully and safely staff and operate the jail facility that is constructed (consistent with Title 15, California Code of Regulations, Chapter 1, Subchapter 6 section 1756(j)5) within 90 days after project completion.

SECTION EIGHT: The County has project site control through fee simple ownership of the site and right of access to the project sufficient to assure undisturbed use and possession of the site and will not dispose of, modify the use of, or change the terms of the real property title, or lease the facility for operation to other entities, without permission and instructions from the BSCC, for so long as the SPWB lease-revenue bonds secured by the financed project remain outstanding.

SECTION NINE: The County is not claiming land value as in kind match. Therefore, for purposes of this proposal, the current fair market land value for the proposed facility is \$0.

PASSED, APPROVED and **ADOPTED** this 21st day of February, 2017, by the following vote, to wit:

AYES: NOES: ABSENT: ABSTAIN:

> Stacy Corless, Chair Mono County Board of Supervisors

1 2	ATTEST:	APPROVED AS TO FORM:
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