

REPORT OF THE COMMITTEE ON BRUCELLOSIS

Chair: Dr. Sam D. Holland, Pierre, SD
Vice Chair: Dr. Claude E. Barton, Nashville, TN

Mr. John B. Adams, VA; Dr. L. Garry Adams, TX; Dr. J. Lee Alley, AL; Mr. Keith E. Aune, MT; Dr. Terry L. Beals, OK; Dr. C. Carter Black, GA; Dr. Carole A. Bolin, MI; Dr. Richard E. Breitmeyer, CA; Mr. Wayne Brewster, WY; Mr. Marc Bridges, MT; Mr. John S. Cargile, TX; Dr. Max E. Coats, Jr., TX; Dr. Thomas F. Conner, OH; Dr. Miguel M. Cordoba, MEX; Mr. Ed Corrigan, WI; Dr. Donald S. Davis, TX ; Dr. Debbi A. Donch, MD; Dr. John C. Doyle, OK; Dr. Mark L. Drew, ID; Dr. Anita J. Edmondson, CA; Dr. Philip H. Elzer, LA; Dr. Steven R. England, NM; Dr. Brian H. Espe, AR; Dr. Donald E. Evans; KS; Ms. Darla R. Ewalt, IA; Dr. Adele Faul, South Africa ; Dr. James M. Foppoli, HI; Dr. Tony G. Frazier, AL; Mr. Bob Frost, CA; Dr. Arnold A. Gertonson, CO; Dr. Michael J. Gilsdorf, MD; Mr. L. Wayne Godwin, FL; Dr. William L. Hartmann, MN; Dr. Robert A. Heckert, MD; Dr. Bob R. Hillman, TX; Dr. E. Ray Hinshaw, AZ; Mr. Majon Huff, CO; Dr. David L. Hunter, MT; Mr. Jon G. Johnson, TX; Dr. Terry Kreeger, WY; Dr. Maxwell A. Lea, Jr., LA; Dr. Thomas F. Linfield, MT; Dr. Jim Logan, WY; Dr. Phillip M. Mamer, ID; Dr. Bret D. Marsh, IN; Ms. Barbara M. Martin, IA; Dr. Charles E. Massengill, MO; Mrs. Phyllis Menden, WI; Dr. Andrea Mikolon, CA; Mr. Rick S. Nabors, TX; Mr. Richard E. Nelson, VT; Dr. Don L. Notter, KY; Dr. Steven C. Olsen, IA ; Dr. Angela Pelzel, TX; Dr. Glenn Plumb, WY; Dr. Valerie E. Ragan, MD; Dr. Jack C. Rhyan, CO; Dr. Thomas J. Roffe, MT; Dr. John J. Schiltz, IA; Dr. David D. Schmitt, IA; Dr. Roy A. Schultz, IA; Dr. Gerhardt Schurig, VA; Dr. Clarence J. Siroky, ID; Dr. William C. Stoffregen, IA; Dr. Robert Stout, KY; Dr. David A. Stringfellow, AL; Dr. Paul L. Sundberg, IA ; Mr. George Teagarden, KS; Dr. Tom Thorne, WY; Dr. Kenneth J. Throlson, ND; Mr. Rick Wallen, WY; Dr. James A. Watson, MS; Dr. Gary M. Weber, DC; Ms. Diana L. Whipple, IA; Dr. Margaret A. Wild, CO; Dr. Richard D. Willer, AZ; Dr. Larry L. Williams, NE; Mr. Steve Wolcott, CO; Dr. Taylor Woods, MO; Dr. Glen L. Zebarth, MN; Dr. Ernest W. Zirkle, NJ.

The Committee on Brucellosis met on Tuesday, October 26, 2004, from 12:30 pm to 5:30 pm. There were 28 Committee members and 35 visitors in attendance. A total of 10 presentations were given at the meeting. There were 14 reports, resolutions, and proposals submitted to the Committee for action. The summary of presentations and actions of the Committee follows.

Drs. Debra Donch and Arnold Gertonson, United States Department of Agriculture (USDA), Animal and Plant Health Inspection Ser-

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vice (APHIS), Veterinary Services (VS), presented the cooperative brucellosis program status report for FY04. There were seven (7) cattle herds affected with brucellosis during FY04 with one (1) being disclosed in Missouri, two (2) in Texas, and four (4) in Wyoming. The State of Missouri was granted brucellosis Class Free status early in the year, prior to disclosure of the one (1) affected herd. Current regulations and program standards allow a state to retain class free status when one (1) affected herd is disclosed in the state within a two (2) year period, provided certain requirements are met to contain the outbreak and assure that spread to additional herds has not occurred. Thus, Missouri did not lose its Class Free status because of this single herd. The two (2) herds in Texas were separate and unrelated outbreaks and their disclosure did not affect the brucellosis status of the state. Three (3) of the four (4) herds disclosed in Wyoming were located in the western part of the state in close proximity to the elk winter feed-grounds. The fourth herd is located in the northeast corner of the state and is not associated with known affected elk populations. The investigation of this herd is ongoing at the time of this report. Because of multiple herd outbreaks during the year, the state was reduced from brucellosis Class Free to Class A status. With the exception of the herd in northeast Wyoming, the remaining six (6) herds have been depopulated. At the end of FY04, 48 states held brucellosis Class Free status with Texas and Wyoming continuing in Class A status. The complete text of this status report is included in these proceedings.

Dr. Pamela Ibarra, Director of the Brucellosis Campaign in Mexico, Secretariat of Agriculture, Livestock, Rural Development, Fisheries and Food, presented a status report on the program to eradicate brucellosis from livestock in Mexico. The herd infection rate reported for the year was 2.95% with a cattle infection rate 0.62%. There were 1,893 cases of human brucellosis reported with most being caused by *Brucella melitensis*.

Dr. Steve Olsen, Research Scientist, USDA, Agriculture Research Service (ARS), National Animal Disease Center (NADC), Ames, IA, presented a time specific presentation entitled, "Update on Bison RB51 Efficacy Experiments at NADC". The text of his presentation is included in these proceedings.

Dr. Steve Olsen gave a second presentation on "Genotyping *B. abortus* Isolates." Based on analysis of the *Brucella abortus* genomic sequence, variable regions that may be useful for epidemiologic investigation were identified. Although the *B. abortus* genome is very stable, these intergenic, non-coding regions containing repeated strings of nucleotides, are more apt to mutate than coding regions. An assay has been developed, the "HOOF-Prints" assay, which evaluates multiple loci containing these tandem repeats. The array of alleles identified by this technique creates a genotype for each isolate. For optimal

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performance of this assay, multiple colonies from each animal must be obtained for analysis. Data collected from *B.abortus* strains passed in vitro suggests that patterns of an isolate are stable. Analysis of isolates from culture collections indicates that genotypes differ between outbreaks. Multiple isolates from an infected bovine herd suggest that multiple, closely-related genotypes may be present within individuals; however, a single genotype will predominate. Ongoing work is attempting to characterize rate of change in individual loci and develop a statistical model that will estimate the degree of relationship between isolates from different herds.

Dr. Max Coats, Texas Animal Health Commission (TAHC), gave a presentation on the status of the fluorescent polarization assay (FPA) test and standards for cut-off values for its use in livestock markets. Basically, this was a sharing of data and experience, since Texas is the only state presently using the FPA test in this way. First point testing for brucellosis at livestock markets is still a vital part of the Texas brucellosis program. The unavailability of the CITE test created a major problem in the evaluation of sero-positive cattle at Texas livestock markets. The TAHC selected the FPA test to replace the CITE test. Dr. Coats described the process through which the FPA test was adapted for use in market testing. He reported that at a cut-off point of 50 millipolarization (mP) units the FPA test virtually duplicates the CITE test in providing serologic data for accurately evaluating titered animals in market channels.

Mr. Rick Wallen, Wildlife Biologist, National Park Service, Yellowstone National Park (YNP), presented an update on the bison management plan for Yellowstone. This was a follow up to the bison management plan presented by Mr. Wallen at San Diego, CA, in October 2003. The complete text of this presentation is included in these proceedings.

Dr. Frank Galey, Dean, College of Agriculture, University of Wyoming and Chair, Wyoming Brucellosis Coordination Team, gave a report on the creation and activities of the Wyoming governor's Brucellosis Coordination Team during the past year. As a result of several Wyoming cattle herds being disclosed to be affected with brucellosis during the past year, the Wyoming Governor and Legislature formed the Brucellosis Coordination Team comprised of 19 members and 10 technical advisors. The Team was charged with developing a list of issues, best management practices, and recommendations for four areas of concern. The areas of concern include managing brucellosis in cattle and minimizing transmission between species, how the state's agencies should best respond to subsequent cases, human health implications, and how to reduce and eventually eliminate brucellosis from the state's wildlife, paying special attention to the elk feed-grounds. The final report is expected to be presented to the Governor by January 2005. The complete text of this presentation is included in these pro-

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ceedings.

Mr. Keith Aune, Montana Department of Fish, Wildlife, and Parks, gave an update on the progress of feasibility and environmental assessment studies being done for a bison quarantine facility just north of Yellowstone National Park. This report was a follow-up to the proposed feasibility study given in October 2003, in San Diego. The complete text of this presentation is included in these proceedings.

Dr. Thomas F.T. Linfield, Montana State Veterinarian for the Montana Department of Livestock, and current chair of the Greater Yellowstone Interagency Brucellosis Committee (GYIBC), gave a report on the activities of that organization during the past year relative to the brucellosis situation in wildlife in the Greater Yellowstone Area (GYA). The complete text of this presentation is included in these proceedings.

Dr. Max Coates, TAHC, presented a report on the two Texas herds of cattle disclosed to be affected with brucellosis during FY04. The two herds were unrelated and were proven to be affected with *B. abortus* by microbiological culture. The herds were depopulated.

Dr. Jim Logan, former Wyoming State Veterinarian, presented individual reports on the four Wyoming cattle herds disclosed to be affected with brucellosis during FY04. Three of the four outbreaks were bacteriologically confirmed and were depopulated. The fourth is still being evaluated. Dr. Logan shared some of the problems and frustrations that resulted when the state of Wyoming was declassified from brucellosis Class Free status to Class A. As a result of this flurry of outbreaks, the Wyoming Livestock Board has established statewide surveillance testing rules requiring a negative brucellosis test within 30 days prior to change of ownership or interstate movement of test eligible cattle.

Committee Chair Sam Holland, South Dakota State Veterinarian, gave an update on the status of brucellosis in the bison herd on Triple U Ranch at Pierre, South Dakota. This bison herd is thought to have been affected with brucellosis since the early 1960's. This herd has been used as a study herd since early 2002 and is scheduled for a post-release assurance test in February 2005.

There were no charges referred to the Brucellosis Scientific Advisory Subcommittee during the year, therefore, no meeting was held and there is no Subcommittee report for this year. However, three (3) issues were raised by the Committee during this meeting and referred to the Subcommittee for consideration during the coming year. The three (3) charges for FY05 are:

1. Review the state of the science and determine the level of confidence of recently developed techniques for DNA fingerprinting (genotyping) *B. abortus*;
2. Review the feasibility and capabilities for establishing a bulk

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- milk brucellosis surveillance test for *B. militensis* in goats; and
3. Review the feasibility and capability of matching DNA from sero-positive blood to DNA from hair on corresponding back-tags of MCI reactors.

Vice Chair Dr. Claude Barton gave the report of the Brucellosis Subcommittee on Education in the absence of Dr. Brian Espe, Subcommittee chair. The Subcommittee report was approved by the Committee and is included in these proceedings.

Dr. Carter Black, Georgia Assistant State Veterinarian, gave the report from the Feral Swine Subcommittee on Brucellosis and Pseudorabies. He also gave the report from the joint working group to review brucellosis eradication and recommend policies for harmonization of the swine brucellosis uniform methods and rules and the pseudorabies program standards. The text of this report is included in these proceedings.

Seven (7) resolutions were presented for consideration by the Committee. Three (3) resolutions were approved and forwarded to the Committee on Nominations and Resolutions for approval by the general membership. They addressed:

1. Providing long-range funding for research, program support and field studies on feral swine.
2. Reduction and elimination of brucellosis in wildlife in the GYA.
3. Development of protocols to allow conduct of critical research related to *Brucella* species.

Two (2) recommendations were approved by the Committee. The first recommendation was that USDA-APHIS-VS should make changes to the Swine Brucellosis UM&R as soon as possible to harmonize definitions and testing schedules with the Pseudorabies Program Standards. There are differences in the definitions between the PRV Program Standards and the Swine Brucellosis UM&R and also differences in the testing schedules for PRV Qualified Herds and Swine Brucellosis Validated Free Herds. Advancement of state status in the Swine Brucellosis Program should be based on the commercial production operations and not be affected by feral and/or transitional herds. The definitions of feral, transitional and commercial swine herds, as used in the Pseudorabies program standards needs to be included in the Swine Brucellosis UM&R. The recommended changes for the Swine Brucellosis UM&R were:

Part I Definitions

Feral or wild swine

~~Swine that have lived all (wild) or any part (feral) of their lives as free-roaming animals. Those swine that are free-roaming.~~

Commercial production swine - Those swine that are continuously

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managed and have adequate facilities and practices to prevent exposure to either transitional or feral swine.

Transitional production swine - Those feral swine that are captive or swine that have reasonable opportunities to be exposed to feral swine.

Part V Validated Swine Brucellosis – Free Herds

A. Initial Validation or Revalidation

4. Swine growout premises on which no adult breeding swine are maintained may be validated or revalidated as Swine Brucellosis free if all samples are tested Swine Brucellosis negative when establishing a Qualified Negative growout premises on which no adult breeding swine are maintained.

Part VII Program Stages

State II

2. b. During the 2-year period prior to the request for Stage II status, the State's commercial breeding swine population.....

3. States must develop and adopt a management plan that adequately separates and addresses control of the interface of feral and transitional production swine and commercial swine. The plan is to be reviewed by the National Center for Animal Health Programs staff.

Stage III (Free)

A. Establishment of status

2. Herd Infection Rate

(Change) During the 2-year qualification period, no more than one SB-infected commercial breeding swine herd was identified;.....(no change)

4. States must develop and adopt a management plan that adequately separates and addresses control of the interface of feral and transitional production swine with commercial swine. The plan is to be reviewed by the National Center for Animal Health Programs staff.

C. Termination of status

4. Infection is disclosed in a commercial swine herd with evidence of spread to other commercial swine herds.

The second recommendation approved by the Committee included 3 parts:

1. USDA-APHIS-VS, should encourage states in the Greater Yellowstone Area (GYA) to distribute information on the technology and diseases status of brucellosis in the GYA.
2. USDA-APHIS-VS should continue to educate the livestock industry and legislators at both state and national levels on the importance of surveillance for brucellosis and continued support to complete the eradication effort.
3. USDA-APHIS-VS should appoint a team to review brucellosis

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handout materials used in various states to determine general availability and need for additional current information.

The following statement was suggested for use as a guide in following up on recommendation 2:

Brucellosis in the Greater Yellowstone Area (GYA)

Brucellosis in Cattle

The knowledge of brucellosis in cattle is extensive, based on over 65 years of a cooperative state-federal program to eliminate the disease from the domestic cattle and bison from the United States. Currently, the disease has been eliminated from all states except for Texas and more recently Wyoming.

We understand how the disease is spread within herds, how the prudent use of vaccine can reduce the risk of exposed animals becoming infected, and how other management tools to minimize the risk of a herd becoming affected.

What we know about the disease and the tools needed must continue to be practiced in GYA where there is a risk of exposure to the disease.

Brucellosis in Bison

Brucellosis in captive domestic herds of bison has been eliminated, but the disease is still prevalent in the Yellowstone National Park bison. It has been shown that up to 50 percent of the Park bison may be affected with the disease. These bison represent an apparent threat to domestic cattle operations when they migrate from the park and on rare occasions could mix with cattle on traditional cattle grazing lands near the park.

The knowledge of the disease in bison is much more limited. Effective vaccines are being developed, but even if developed, administration will be a major hurdle to overcome.

Experimentally it has been shown that infected bison can spread the disease to susceptible cattle, but natural occurrences of this are difficult to document.

Brucellosis in Elk

Infected elk in the GYA are probably the most serious threat to the domestic cattle population, as they range over a larger geographic area and are able to commingle with cattle throughout the year.

The amount of infection varies widely in these elk, with the highest infection rates in those elk which are routinely fed in the winter one or more of the elk feed grounds. The concentration of animals on these feed grounds is conducive to the spread of brucellosis, particularly if there are infected animals aborting.

Over the years vaccine has been used in a few feed grounds and it has been shown to lower the spread of the disease in the vaccinated groups. Development to better vaccines and delivery method, while an

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admirable goal, the chance of a significant lowering the incidence of the disease will require vaccination of very large numbers of elk, which will be difficult at best.

Elk have been implicated in recent findings of brucellosis in cattle herds in Wyoming. The initial cattle herd found in 2004 was adjacent to an elk feed ground and there was apparently contact between cattle and elk.

There are no easy solutions to the problems of brucellosis in the GYA, but as long as the disease persists in the area, domestic cattle herds remain at risk and must utilize all known management tools to minimize this risk.

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**REPORT OF THE EDUCATION SUBCOMMITTEE
ON BRUCELLOSIS**

Chair: Dr. Brian Espe
Oklahoma City, OK

The Education Subcommittee on Brucellosis met on October 24, 2004. Five members and visitors were present. Dr. Espe was unable to attend. The meeting was conducted by Dr. Barton, Vice Chair of the Committee on Brucellosis.

Dr. Espe had drafted and forwarded an informational release on brucellosis in the Greater Yellowstone Area (GYA) for consideration by the Subcommittee. The document, with minor additions, was adopted for use in informational outreach activities and is included in this report.

It was noted that Dr. Arnold Gertonson, Animal and Plant Health Inspection Service (APHIS), Veterinary Services (VS), has been assigned to be the liaison official between USDA and the three states in the GYA. In this position Dr. Gertonson will be a key link for the distribution of current information on brucellosis in the area.

Additionally, the following recommendations were selected for submission to the Committee on Brucellosis:

1. Continue to encourage states in the GYA to distribute information on the technology and disease status of brucellosis in the Yellowstone area.
2. Continue to educate the livestock industry and legislators at both state and national levels on the importance of surveillance for brucellosis and continued support to complete the eradication effort.
3. Appoint a team to review brucellosis handout materials used in the various states to determine general availability and need for additional current information.

Brucellosis in the GYA

Brucellosis in Cattle

A massive amount of knowledge about brucellosis in cattle has been gained in more than 65 years of cooperative state-federal efforts to eliminate this serious disease from domestic cattle and bison in the United States. Currently, the disease – which causes abortions and lowered milk production – has been eliminated from all states except for Texas and, more recently, Wyoming.

We have learned how the disease is spread within herds, how the prudent use of vaccine reduces the risk of exposed animals becoming infected, and how other management tools can be used to minimize the risk of a herd becoming affected.

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All of the knowledge gained must be employed in the GYA where there is a high risk of exposure to the disease from wildlife.

Brucellosis in Bison

Brucellosis in captive domestic herds of bison has been eliminated in the United States, but the disease is still prevalent in the bison in YNP and Grand Teton National Park. It has been shown that up to 50 percent of the Park bison may be infected with the disease. These bison represent a clear threat to domestic cattle operations when they migrate from the Park and occasionally mix with cattle on nearby traditional cattle grazing lands.

There is less knowledge of the disease in free-ranging bison than in domestic cattle. Effective vaccines for bison are being developed; but even if developed, administration of such vaccines will be a major hurdle to overcome.

Experimentally it has been shown that infected bison can spread the disease to susceptible cattle, but natural occurrences of this are difficult to document.

Brucellosis in Elk

Infected elk in the GYA are probably the most serious threat to the domestic U.S. cattle population. Elk range over a larger geographic area and are more likely to commingle with cattle throughout the year.

The amount of infection varies widely in these free-ranging elk herds. The highest infection rates are usually found in those elk that are routinely fed in the winter on one of the elk feed grounds. The concentration of animals on these feed grounds makes the spread of brucellosis more likely, particularly when infected animals abort.

Over the years, vaccine has been used in elk on most feed grounds. Spread of the disease has been lower in vaccinated groups. Development of better vaccines and methods of delivery is an admirable goal. However, the likelihood of significantly lowering the incidence of brucellosis will require continued and long-term vaccination of very large numbers of elk. Doing this thoroughly over time would be difficult at best and expensive.

Elk have been implicated in recent findings of brucellosis infection of cattle herds in Wyoming. The initial infected cattle herd found in November of 2003 was adjacent to an elk feed ground and the disease spread appears to be by contact between cattle and elk.

There are no easy solutions to the problems of brucellosis in the GYA. But as long as the disease persists in the area, domestic cattle herds remain at risk and herd owners must utilize all known management tools to minimize this risk.

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REPORT OF THE FERAL SWINE SUBCOMMITTEE ON BRUCELLOSIS AND PSEUDORABIES

Co-Chairs: Dr. Carter Black, Atlanta, GA
Dr. Max Coates, Austin, TX

The subcommittee was called to order at 1:00 pm on October 24, 2004 with 36 attendees.

Dr. Phil Elzer, Research Scientist, Louisiana State University, reported on his work with *B. abortus* RB51 and *B. suis* VTRS1 vaccines. The VTRS1 vaccine is a rough strain of *B. suis* and like RB51 there is no O chain polysaccharides. VTRS1 adequately colonizes pigs and protects sows better than RB51 when challenged. VTRS1 vaccine appears to be superior to RB51 vaccine in swine.

Dr. Lowell Miller presented information on his work, which is sponsored by United States Department of Agriculture (USDA) that deals with immuno-contraception in domestic and feral swine. The Wildlife Research Center is working on a vaccine to stimulate antibodies to GnRH. GnRH is a small peptide hormone which, when injected into females, will stop estrus. The center is investigating an oral vaccine application.

Ned Hahn provided an update on his ongoing effort to fingerprint feral pig pseudorabies (PRV) isolates. The goal is to fingerprint pseudorabies virus DNA from recent out breaks, to improve the data base and to develop a method to determine the source of infection. The main work is with glycoprotein C. There appears to be several strains of PRV virus circulating between and among feral and domestic populations.

Dr. Joe Corn, Southeastern Cooperative Wildlife Disease Study (SCWDS), presented an outstanding paper on their work in describing the distribution of feral swine in the United States and the distribution of PRV and brucellosis in feral swine. The SCWDS has developed a map of feral and domestic swine populations in the United States. By overlaying the two maps, the area of risk for feral and domestic swine interface may be assessed and may facilitate the development of strategies for preventing commingling. These areas may be considered as rational targets for disease surveillance.

Seth Swafford spoke on the mission of USDA Animal and Plant Health Inspection Service (APHIS), Wildlife Services (WS) and related their feral swine activities. Feral swine damage includes the negative impact on endangered species, property damage, crop damage and damage to livestock, as well as negative effects on domestic swine. From the agency's contacts, most public concerns relate to disease transmission. A new focus for USDA-APHIS-WS is cooperative regulatory disease management.

Dr. John Korsland, National Swine Programs Liaison, spoke on 'The

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View from USDA, VS. Dr. Korsland reported that all states are at Stage III for swine brucellosis except Texas. There were two infected transitional herds last year, one in California and one in Hawaii. Dr. Korsland suggested that it is time to update the Swine Brucellosis Control/Eradication Uniform Methods and Rules. Further, only three states had not yet achieved stage 5 in the pseudorabies eradication program and that only Texas had not been recognized as a free state in accordance with the swine brucellosis Uniform Methods and Rules (UM&R).

Dr. Carter Black spoke to the issue of changes to the swine brucellosis UM&R necessary to harmonize the Swine Brucellosis Control and Eradication UM&R with the PRV Eradication Program Standards. After some discussion the changes were evaluated and were unanimously recommended to the Brucellosis Committee as urgently needed changes to the Swine Brucellosis Control and Eradication UM&R and further, there was consensus that the appointed Harmonization Working Group should continue their assessment of the need to make additional changes to the UM&R if necessary to complete the harmonization of the two swine program documents and provide their recommendations at the next meeting of the committee.

There was a unanimous desire of the Subcommittee to forward the changes in the form of resolutions to the Committee on Brucellosis and the Committee on Pseudorabies and to recommend their favorable consideration.

A PROPOSED FEASIBILITY STUDY OF BISON QUARANTINE: UPDATE

Keith Aune, Montana Department of Fish, Wildlife and Parks,
Bozeman, MT

Dr. Jack Rhyan, Veterinary Service, Fort Collins, CO

Introduction

There has been a long history in North America of restoring wildlife populations by capturing animals from robust populations and transplanting them to new habitats or augmenting existing populations near extinction. In the Greater Yellowstone Ecosystem, there is an extensive history of capturing, holding, transporting and relocating wildlife as a species conservation strategy. Yellowstone elk were routinely captured and widely distributed in the mid 1900's to restore wild elk throughout North America. Bison and antelope have been captured and moved from Yellowstone to augment populations elsewhere. Yellowstone has even been a recipient of such transplanted wildlife for restoration including rocky mountain wolves from Canada and bison from Texas and northern Montana.

As it applies to the bison management dilemma surrounding Yellowstone National Park (YNP), there have been many discussions

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about quarantine procedures and using this growing population to establish other free-ranging bison herds. Several quarantine options have been considered, and USDA-APHIS has established a protocol that would apply to this situation (Interagency Bison Management Plan, Appendix B). Federal funding was appropriated for this activity but has not been expended. Despite frequent discussions of quarantine proposals and the acquisition of federal funding for this activity a specific plan has not yet been developed and approved.

Concurrent with the discussion of quarantine in the GYA, there have been frequent discussions and meetings regarding bison conservation strategies in North America and the potential for restoring the species to grassland ecosystems. The World Conservation Union (IUCN)-Bison Specialist Group of North America supported a project to examine the status of bison, which presents several conservation recommendations (Boyd, 2003). This project outlines the current status of bison and identifies the few free-ranging and genetically pure bison herds in North America. There are about 8,300 plains bison classified as such in only 13 conservation herds and they present the best source stocks for restoration efforts. Nearly 2/3 of these bison are from diseased herds while the remainder is found in small fragmented populations with limited potential as a reliable source for restoration efforts.

YNP could become a source of genetically pure bison to be reintroduced into historical habitats thereby contributing to the continued conservation of this species. Currently, the bison population in YNP is above the management trigger level for aggressive removals and there have been annual habitat and weather dependent movements of bison out of YNP causing conflict and concern in the states of Montana, Idaho and Wyoming. The major elements of this conflict include the presence of brucellosis, a nationally regulated disease in YNP bison, and managing the population size and distribution of Yellowstone bison. As we attempt to manage brucellosis, many bison are routinely hazed or captured, tested and slaughtered to minimize the risk of transmission to cattle. Despite the successful management of the risk for transmission of brucellosis and the spatial-temporal separation of bison and cattle accomplished under the current management plan, there are no strategies in place to restrain the base population of bison.

We propose that some bison migrating from YNP could be placed through a quarantine program to restrain population growth and ultimately be used for the restoration of this species in other portions of North America. This selected removal program along with other population regulating tools such as a limited hunting program, as well as natural mortality, could operate in consort to remove an annual increment of bison from the herd to help maintain a relatively stable population and curb range expansions in a confined ecosystem. In addition, such a program could be constructed and implemented to conserve

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the genetics of YNP bison in these newly established populations or even enhance the genetic diversity of existing managed bison herds in North America.

This approach to bison conservation will require many government and private sector partnerships including cooperating participants from the Montana sporting public, various conservation groups, Native Americans, and the affected state/federal agencies. The overall mission of using animals from this robust Yellowstone bison population to restore other populations in North America has benefits as well as challenges.

Project Goal

There are three main project goals described below in this proposed feasibility study for bison quarantine.

1. Develop quarantine procedures, using the best available science and adaptive management strategies, that will allow bison from YNP to be accepted for translocation and utilized for the establishment of new public and Native American bison herds or to augment existing populations in North America.
2. To conserve the genetics of free-ranging Yellowstone bison through the creation of additional bison herds in other habitats in North America without transmitting brucellosis onto these landscapes.
3. To examine the feasibility of quarantine protocols and the reintroduction of bison to large grassland systems as a conservation strategy that may benefit the management of bison in the GYA where populations are expanding beyond social tolerance limits.

The overall project goals are consistent with historical conservation strategies applied to wildlife restoration efforts in North America and previously validated for several species of ungulates (elk, bison and antelope) found within the Yellowstone Ecosystem. The proposed project will also contribute to the conservation of a genetically diverse bison population in which, to date, no cattle genes have been detected (Halbert 2003). In so doing it will lead to the establishment of new herds of similar genetic composition to reinforce the long-term conservation of wild bison genes at locations beyond the borders of the Yellowstone Ecosystem. Recent work by Halbert (2003) has confirmed the diverse genetics of bison from YNP and present significant genetic concerns for many other Department of Interior bison herds. The bison processed through the quarantine program could also be utilized for periodic introduction into existing public bison herds to remove animals with domestic cattle genes and improve genetic diversity to further ensuring conservation of the species.

The proposed study will encourage bison conservation without risk of disease transmission to the landscapes upon which bison will be

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introduced. This research will test the feasibility of implementing quarantine procedures that meet and exceed the existing approved quarantine standards established by USDA-APHIS. The project will develop and implement additional adaptive procedures to improve these quarantine standards and carefully quantify the risks associated with utilizing improved test protocols. Finally, this project will explore the feasibility of using quarantine and translocation as a population regulation tool supplemental to traditional sport hunting and natural processes. It is increasingly clear that bison populations in the GYA are growing and expanding despite the limitations of the landscape to support these populations. A suite of tools including limited hunting, quarantine and translocation, and occasional agency removal will be necessary to regulate population growth and influence bison distribution.

This research project is designed to remain consistent with the existing Interagency Bison Management Plan (IBMP) during all phases. Population triggers established in the plan determine the availability of negative calves for quarantine procedures-Phase I. The program maintains the availability of habitats west of the Yellowstone River for wild free-ranging bison by concentrating quarantine activities on the east side of the Yellowstone River. This geographic compartmentalization of various management activities minimizes management conflicts for implementing the IBMP.

Project Status

The Quarantine Feasibility Study proposal has been in a review process for the past year and several elements of the study design have been modified based upon input from various scientists and stakeholder groups. The final draft has been put out for one last scientific review and the Environmental process for Phase I is completed and Phase II will be started very soon.

As currently proposed, this research project will detain up to 200 sero-negative bison calves (100 in each test group brought into Phase I each year for two years) captured during management actions in the GYA in accordance with the IBMP and EIS for up to 3 years to determine if latent infection occurs and if the current USDA-APHIS protocol for quarantine would efficiently screen for brucellosis. The sero-negative bison calves selected for this research will be contained at an existing bison research facility leased by USDA-APHIS near Corwin Springs, MT for the first year (Phase I). Approximately half of these bison kept during Phase I will be euthanized and specific target tissues will be collected and submitted for culture. The remaining live bison will be available to advance through additional phases of the protocol. During each progressive phase of the study additional facilities (Phase II-Dome Mountain and Phase III- at an undetermined location) are to be developed. Bison will be bred in Phase II and then

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moved to Phase III for calving once a pregnancy is established. If a suitable Phase III site cannot be acquired then these test groups will be rotated back into the Phase I facility. Bison will be maintained in quarantine through the completion of one successful calving and after repeated negative serologic tests.

A detailed analysis and review of the quarantine procedures and testing protocols will be performed at the end of Phase I for use in further environmental and decision processes relative to advancing the study to Phase II and III. The *a priori* hypothesis for Phase I research is rejected if there is evidence of sero-conversion in 5% of each quarantine test group or *Brucella abortus* is cultured from more than 5% of the test animals euthanized during Phase I. Rejecting the Phase I hypothesis could terminate the project or would result in modifications of the subsequent research steps.

The Phase I environmental impact analysis has been released and is under public review. The comment period for the Montana Environmental Policy Act Assessment closes on November 12, 2004 after which comments will be evaluated and a final record of decision will be posted by the end of November. National Environmental Policy Act compliance was satisfied through the publication of a final IBMP/EIS in 2000 and a determination for categorical exclusion produced by USDA-APHIS. A decision to proceed with phase I could result in bison placed under quarantine this winter as they become available. A second Environmental Analysis will begin soon to evaluate the impacts associated with Phase II of this study. A decision to move forward to Phase II will be made next summer after serologic and culture results are available from the first test group and a impact study has been completed for the phase II site at Dome Mountain.

COOPERATIVE STATE-FEDERAL BRUCELLOSIS ERADICATION PROGRAM STATUS REPORT - FISCAL YEAR 2004 "Prove the Negative"

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Fiscal Year (FY) 2004 for the Cooperative State-Federal Brucellosis Eradication Program proved to be a year reflective of adages of final eradication activities for a program nearing its goal. Accomplishment of the goal of total eradication was challenged by findings of last remaining vestiges of disease, recrudescence of disease, and spill-over of disease from wildlife reservoirs. After having met all requirements

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and being designated as a Class Free state for brucellosis, Missouri found a singleton brucellosis affected herd. Testing in Texas disclosed brucellosis in a herd that had been clean for several years. And, Wyoming discovered brucellosis affected herds whose most likely source of exposure is the infected wildlife in the same geographic area. The finding of the brucellosis-affected herds in all these situations exemplifies both the effectiveness of the surveillance program and the commitment to the goal of total eradication. The overall status of the Cooperative State-Federal Brucellosis Eradication Program in the United States for FY 2004 stands at 48 states designated as Class Free for brucellosis and two states, Texas and Wyoming, designated as Class A for brucellosis. Puerto Rico and the Virgin Islands also maintain their Class Free status for brucellosis.

Seven (7) new brucellosis affected cattle herds were disclosed in FY 2004. This compares to only two (2) new brucellosis affected cattle herds disclosed in FY 2003, nine (9) new affected herds in FY 2002, six (6) in FY 2001 and fourteen (14) in FY 2000. Two (2) of the seven (7) FY 2004 brucellosis affected herds were found in Texas, a Class A status state. One (1) brucellosis affected herd was found in Missouri three months after receiving Class Free state status. The other four (4) brucellosis affected cattle herds were found in Wyoming which was a Class Free state at the time of the finding of their first affected herd, but was reclassified to Class A status subsequent to the finding of additional brucellosis affected herds.

The first of the two (2) brucellosis affected herds disclosed in Texas in FY 2004 was identified through first-point testing at a livestock auction market. Subsequent culture confirmation on December 17, 2004 identified *Brucella abortus* biovar 4 in the market reactor beef cow. This animal originated from a herd with premises in Comal and Hays counties in central Texas. The herd of origin premises were depopulated as well as a fence line contact adjacent herd. No additional brucellosis affected herds were identified in the epidemiologic investigation, traceback and area herd testing conducted in this investigation. The second brucellosis-affected herd in Texas in FY 2004 was found in Leon County located in eastern Texas. This finding was also subsequent to a market reactor. The herd owner has opted to remain under quarantine and complete the necessary testing. The epidemiological investigation continues as well as the testing of the index herd and area herds. At this time it is thought that the most likely sources of infection for the two brucellosis-affected herds in Texas in FY 2004 are the purchase of infected replacement heifers or a potentially long-standing low-level infection in the herd.

Missouri attained Brucellosis Class Free status on February 26, 2004. On May 5, 2004, a new brucellosis affected cattle herd was disclosed in Bates County, Missouri. This herd was discovered through

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market cattle inspection (MCI) slaughter surveillance testing. All animals on this index premises were depopulated. A Task Force of state and federal veterinary staff was immediately convened to conduct adjacent herd and area herd testing in a timely manner. All herds located within two miles of the affected premises tested negative. Herds located within one mile of the affected premises will be retested within one year. Whole herd (adult) vaccination was utilized in the high-risk area (one mile). Animals having been moved out of the affected herd since January 2002 were traced. No additional brucellosis affected animals or herds were identified. Per 9 CFR, Part 78, a Class Free State may retain its status if only one brucellosis affected herd is found within a 24 month period, provided the affected herd is immediately depopulated and an appropriate epidemiologic investigation is conducted within the prescribed 60-day timeframe. The epidemiological investigation must confirm that brucellosis has not spread from the affected herd. A review of the epidemiological investigation and herd testing for this case was conducted in July 2004. The review concluded that all requirements to maintain Class Free state status had been fulfilled.

In Wyoming, the traceback of four MCI reactor animals that went to slaughter in late November 2003, led to the finding of a brucellosis affected beef herd in Sublette County (western Wyoming). Bacteriologic culture results reported out in December 2003 confirmed *Brucella abortus* biovar 1 infection in this herd. The most likely source of infection for this herd was determined to be infected elk on adjacent feed grounds. Wyoming took immediate actions and depopulated the herd (in accordance with the 9 CFR, Part 78) to maintain its Class Free status. However, on January 21, 2004, a second brucellosis-affected herd was confirmed in Washakie County. This herd, a terminal feedlot, was disclosed pursuant to epidemiological trace-outs of cattle from the index herd in Sublette County and was depopulated as well. With the discovery of the second herd, Wyoming no longer met the standards for Class Free status. Docket No. 04-009-1, "Brucellosis in Cattle; State and Area Classifications; Wyoming" was published in the Federal Register on February 20, 2004. This Docket amended the brucellosis regulations concerning interstate movement of cattle by changing the classification of Wyoming from Class Free (attained October 1, 1983) to Class A, effective February 13, 2004. Since that time, two additional brucellosis-affected herds have been found in Wyoming. In June of 2004 a reactor cow was identified during a herd test for interstate movement. *Brucella abortus* biovar 4 was confirmed by bacteriologic culture. This herd, located in Teton County, was depopulated. Although the source of infection has not definitively been determined, the epidemiology investigation reveals no contact with infected cattle, however the index herd did have contact with elk and bison, thus wildlife is

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thought to be the most likely source of infection. The fourth brucellosis affected herd is currently still under investigation. This herd is located in Campbell County in northeast Wyoming. In June of 2004 this herd sent approximately 50 head of cattle to a livestock auction market in South Dakota. Two cows tested suspect for brucellosis. Subsequent bacteriologic culture results were reported as *Brucella abortus* biovar 1 infection in these two animals. All remaining animals on the index premises tested negative as did all adjacent and area herds. No discernable source of infection has been identified in the epidemiologic investigation in this case. Additional activities, including testing of elk in this area of the state are underway. The index herd remains under quarantine and additional herd testing is scheduled. The final classification and status of the herd quarantine is pending.

USDA-APHIS is amending the regulations for the Brucellosis program (9 CFR Part 78) "by adding the Fluorescent Polarization Assay (FPA) to the lists of confirmatory and official tests for determining the brucellosis disease status of test-eligible cattle, bison, and swine." The FPA has proved to "provide an efficient, accurate, automated, and cost-effective means of determining the brucellosis status of test-eligible cattle, bison, and swine." The proposal to amend the regulations by adding the FPA to the list of official tests for determining the brucellosis disease status of test-eligible cattle, bison, and swine was published in the Federal Register on May 6, 2004. Comments were solicited until July 21, 2004. Comments received have been reviewed. The posting of the final rule is imminent.

USDA-APHIS is also amending the regulations for the brucellosis program (9 CFR Part 51) to allow the payment of "indemnity for sheep, goats and horses destroyed because of brucellosis. This action makes it easier to eliminate affected herds/flocks and infected animals as sources of infection by encouraging herd and flock owners to cooperate with our brucellosis eradication program. This action is intended to help reduce the incidence of brucellosis and the likelihood of it spreading within the United States." The final rule was published in the Federal Register on July 13, 2004 with an effective date of August 12, 2004.

Brucellosis in the Greater Yellowstone Area (GYA):

USDA-APHIS continues to recognize the importance of cooperating with the federal and state agencies in management of the wild bison and elk in the GYA. USDA-APHIS will respond to the Governor of Idaho's request for assistance in addressing issues regarding elimination of brucellosis from the GYA.

A new Greater Yellowstone Interagency Brucellosis Committee Memorandum of Understanding (MOU) has been drafted and is currently under review by the Secretaries of USDA and Department of Interior, and the Governors of Montana, Wyoming and Idaho. The pre-

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vious MOU has expired.

The agencies are continuing to evaluate research regarding the safety and efficacy of *Brucella abortus* strain RB51 vaccine in bison, elk and other species. As per the Interagency Bison Management Plan (IBMP), when the agencies determine that RB51 vaccine is safe for use in bison, the vaccine will be subcutaneously injected, by hand, to calves and non-pregnant yearlings that are captured outside of Yellowstone National Park (YNP). YNP vaccinated over 100 head of bison captured at the Stephens Creek capture facility during the spring of 2004. This research may require the use of outdoor research facilities to accommodate statistically significant numbers of animals in the experimental and control groups.

YNP vaccinated over 100 head of bison captured at the Stephens Creek capture facility (northern boundary area) during the spring of 2004. It is expected that the Montana Environmental Protection Act (MEPA) process regarding brucellosis of bison in the western boundary area will be completed late 2004 or early 2005. It is expected that vaccination of bison will begin in the western boundary area in early 2005.

YNP is evaluating methods for remote delivery of vaccine to wild bison. The National Environmental Protection Act (NEPA) process to evaluate remote delivery has been started by YNP.

USDA-APHIS and the State of Montana are currently evaluating sites and protocols for a bison quarantine feasibility study. The purpose of this feasibility study is to determine if bison that are captured outside of YNP can be released onto Native American and public lands after they have completed an extensive and conservative quarantine protocol. Two sites have been selected for the first and second phases of the study. Up to 100 head of bison, if captured, may be placed in the first phase of the study. It is expected that the first phase of the study will begin early in 2005. NEPA and MEPA processes are expected to be completed by the end of 2004. Before APHIS and the State of Montana release bison from quarantine, the agencies will be confident, using the best science and tests available at the time, that the bison are not infected with brucellosis.

The Record of Decision (ROD) for the management of bison that nomadically move from YNP into Montana continues to be utilized by the agencies that signed the ROD. The management actions prescribed in the IBMP are meant to minimize the risk of brucellosis transmission from bison to cattle in the GYA. The IBMP is not a plan to eliminate brucellosis from bison in YNP or bison and/or elk in the GYA.

During the 2003 – 2004 bison management season in the western boundary area, 59 hazing operations resulted in approximately 1,434 bison hazed back into YNP and 82 were unsuccessfully (in the first attempt) hazed. Twenty bison were captured in the western boundary

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area in 4 capture operations. Eight bison were tested brucellosis seronegative and release into YNP. Twelve bison were tested brucellosis seropositive and were slaughtered. Two bison were lethally removed.

During the 2003 – 2004 bison management season in the northern boundary area, YNP captured 464 bison. One hundred eleven bison were vaccinated, 264 bison tested brucellosis seropositive and were sent to slaughter, and 198 bison tested brucellosis seropositive and were released into YNP. One bison died in the capture facility. Numerous hazing operations were also performed with YNP and one hazing operation outside of YNP resulted in 14 bison successfully hazed back into YNP.

The hazing, capture and lethal removal operations were cooperative joint agency operations as per the IBMP. The agencies are still in Step 1 of the IBMP because a remote vaccine delivery system is not yet available and cattle are still present on the Royal Teton Ranch.

Four brucellosis affected cattle herds in Wyoming were identified during FY 2004. Three herds were depopulated and one herd owner elected to test out of quarantine. Contact and surrounding cattle herds were brucellosis tested and found negative. Testing of contact and surrounding cattle and elk herds will continue this fall. The most likely source of the brucellosis infection in these cattle herds is believed to be brucellosis infected elk.

Surveillance Activities:

The surveillance statistics for the cattle brucellosis eradication program are based on data available as of October 1, 2004. Normal reporting time allowances for states to gather and submit monthly data and priority emergency disease response activities precluded the ascertainment of all data for FY 2004. Therefore, the following FY 2004 annual statistics regarding the cattle brucellosis eradication program contain estimated data.

As of September 30, 2004, 48 States, Puerto Rico, and the Virgin Islands continue to maintain Brucellosis certified Class Free status. Two states, Texas and Wyoming, are Brucellosis designated as Class A status. Approximately eighty-four percent of all beef and dairy cattle in the United States are located in Class Free States and approximately sixteen percent are located in the two Class A States.

There were seven new brucellosis affected herds found in FY 2004. As previously detailed, the first two new affected herds in FY 2004 were found in Texas and Wyoming in December 2003. Texas found another brucellosis affected herd in August of 2004. Wyoming subsequently found additional brucellosis affected herds in January and July of 2004. Missouri found one brucellosis affected herd in May of 2004.

Brucellosis milk surveillance detected no brucellosis affected dairy herds in FY 2004. Based on available data, 200 suspicious brucellosis ring test (BRT) laboratory reports resulted in 65 herds

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being blood tested for a herd blood test rate (HTR) of approximately 32.5 percent. Repetitive brucellosis ring testing was conducted in the majority of the herds not blood tested. Negative repetitive brucellosis ring tests and epidemiological investigations revealing no evidence suggestive of infection in the herd lead to successful case closures on these cases. Other herd suspicious BRT's were the result of Strain 19 vaccination titers.

There were approximately 8.3 million MCI blood tests conducted in FY 2004. Of these, approximately 5.5 million samples (66.3 percent) were collected at slaughter plants and approximately 2.8 million (33.7 percent) were collected during first point testing at livestock markets. First point testing at markets is primarily conducted in the Central and Southern regions, where the majority of the states that have recently attained Class Free status and the two Class A states are located. Market testing has been the primary surveillance method which has identified newly affected herds.

The total number of cattle tested for brucellosis in FY 2004 was approximately 9.1 million. Of these, approximately 770,500 (8.5 percent) were sampled on farms or ranches and approximately 8.3 million (91.5 percent) were tested under the MCI program. The MCI surveillance continues to be effective in finding reactor animals.

There were approximately 4.04 million calves vaccinated for brucellosis in FY 2004. Approximately 10,100 head of adult cattle were vaccinated in FY 2004 pursuant to the finding of affected herds in the area.

Five brucellosis affected herds were depopulated in the U.S. in FY 2004. Additional adjacent fence-line contact herds were depopulated as well. MCI reactor cattle were also purchased for further diagnostic testing to resolve reactor classified titers. Approximately \$400,000.00 was spent in indemnity monies in FY 2004. Depopulation continues to be the preferred method of handling affected herds as recommended in the Brucellosis Emergency Action Plan.

The United States continues to face challenges in the final phases of eradication of brucellosis. As demonstrated by the finding of brucellosis affected cattle herds in Wyoming, the brucellosis situation in the GYA poses a significant threat to cattle herds in the area. The development of the National Animal Identification System and the National Surveillance Unit will greatly increase our capabilities to find the last remaining cases of brucellosis in our nation's cattle herd and to remain diligent in our quest to find infection early on. Now is the time to prove the negative.

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UPDATE ON GOVERNOR OF WYOMING'S BRUCELLOSIS COORDINATION TEAM

Dr. Frank Galey, Dean, University of Wyoming College of Agriculture, and Chair, Wyoming Brucellosis Coordination Team, Laramie, WY

Wyoming has experienced several new cases of brucellosis (due to *Brucella abortus*) in cattle in the past year. The cases of most interest are in the Greater Yellowstone Area. One case was directly traced to an elk origin whereas the other is very likely due to elk or bison due to reported commingling of animals.

As a result of these cases, the Governor and Legislature of the State of Wyoming formed a Wyoming Brucellosis Coordination Team, which I was asked to chair. This team consists of 29 individuals including 19 members and 10 technical advisors. We were charged with developing a list of issues, best management practices, and recommendations for four topics. Those topics include managing brucellosis in cattle and minimizing transmission between species, how the state's agencies should best respond to subsequent cases, human health implications, and lastly, how to reduce and eventually eliminate brucellosis from the state's wildlife paying special attention to the elk feeding grounds.

The team was given one year to complete this task. We have covered the first three topics in detail and are currently working on the last topic (wildlife brucellosis). General recommendations developed by the team and current progress on the recommendations related to wildlife will be reported.

THE GREATER YELLOWSTONE INTERAGENCY BRUCELLOSIS COMMITTEE - 2004 ANNUAL REPORT

Dr. Thomas F. T. Linfield, Montana State Veterinarian, Helena, MT

The Greater Yellowstone Interagency Brucellosis Committee (GYIBC) was formally established in 1995, when a Memorandum of Understanding (MOU) was signed by the Secretaries of Interior and Agriculture and the Governors of Montana, Wyoming, and Idaho, in an effort to collectively address the problems caused by brucellosis in elk and bison in the Greater Yellowstone Area (GYA). Member agencies represented in GYIBC include the State and Federal agencies responsible for management of wildlife, livestock, and lands in the GYA. The GYIBC has an Executive Committee, a Technical Subcommittee, and an Information and Education Subcommittee. The Goal of the GYIBC is to protect and sustain the existing free-ranging elk and bison populations in the GYA and protect the public interests and economic viability

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ity of the livestock industries of the States of Idaho, Wyoming, and Montana. A major focal point of the GYIBC is to facilitate the development and implementation of brucellosis management plans to control and eventually eliminate brucellosis from the wildlife in the GYA.

In 2003, the Executive Committee recognized an annual report would be a valuable means to inform numerous and diverse stakeholders of GYIBC activities. This annual report is intended to provide the reader the highlights of GYIBC activities for 2003 calendar year and includes the Goal, Mission, and Objectives of the GYIBC, as well as discussion on MOU review and revisions, research activities, management activities and plans, necessary environmental analysis, and information and education efforts.

The members of the GYIBC Executive Committee recognized the need to revise and update the original MOU. The most significant changes proposed were to more aggressively address elimination of brucellosis from the GYA and to include Tribal representation on the GYIBC. Tribal representation is addressed by including the Chairman of the Board of Directors of the Inter-Tribal Bison Cooperative (ITBC) as a representative of Native American Tribes.

The final year of a three-year study was conducted to determine the environmental persistence of *Brucella abortus* (strain RB51) in infected fetal tissues. It was found that *Brucella abortus* bacterium remained viable on fetuses placed out during February for 80-90 days versus 20-30 days for fetuses placed out in mid-May. Preliminary results indicate that UV-B and temperature work in a complex fashion to kill the bacterium present on fetal tissues. Similarly, the final year of a three-year study was conducted to monitor the disappearance of bison fetuses placed within and adjacent to Yellowstone National Park (YNP). Those fetuses placed in YNP were scavenged more rapidly than those placed in adjacent environs. On average, fetuses were scavenged within 18 days, although disappearance ranged from one to 78 days. Approximately half of the fetuses moved more than 100 feet, with one fetus moving two miles, across a frozen lake.

As part of the Interagency Bison Management Plan (IBMP), a study was proposed to determine the feasibility of a quarantine process for seronegative bison calves from YNP. If successful, "disease-free" bison may be considered for YNP bison conservation efforts and potential restoration projects on suitable State, Federal, and Tribal lands. The proposed study is a 3-phased project, with the initial phase potentially beginning, with up to 100 seronegative bison calves, in January 2005.

The first report of brucellosis caused by *Brucella abortus* in Rocky Mountain bighorn sheep was discovered at the Wyoming Game and Fish Sybille Wildlife Research facility near Laramie, WY. Nine (4 female, 5 male) captive, adult Rocky Mountain bighorn sheep were in-

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ected with *Brucella abortus* biovar 4 as a result of natural exposure to an elk fetus aborted by a research elk.

In November 2003, four Market Cattle Inspection (MCI) suspects were traced to a Wyoming premise, which, following herd testing, was determined to be an infected herd. Ultimately, the source of infection was determined to be from infected elk, which were fed on a feedground adjacent to the cattle herd. An additional cattle herd in Wyoming was subsequently determined to be infected in early 2004, ultimately causing Wyoming to lose its Class Free status.

As part of the IBMP, ninety-eight hazing operations were conducted during the year ending in June of 2003; 12 at the North boundary and 86 at the West boundary. The IBMP is a cooperative State-Federal effort aimed at minimizing the risk of brucellosis transmission from infected bison to cattle while maintaining a wild, free-ranging bison population. Most efforts are directed at maintaining spatial and temporal separation of bison and cattle. Adjustments to the IBMP, such as initiation of vaccination programs, are considered annually and the plan may be modified based on the concept of adaptive management.

The 2003 Montana Legislature authorized the Montana Fish, Wildlife and Parks (MT FWP) Commission to consider initiating a bison hunt. MT FWP, in cooperation with the Montana Department of Livestock, began scoping for an environmental review of a proposed bison hunt. The environmental review is scheduled for completion in the fall of 2004. If the State decides to move forward with a bison hunt, it could begin as early as Fall 2004.

Under the Idaho Brucellosis Management Plan, the Idaho Fish & Game Department hired a veterinarian in October 2003. This veterinarian has completed a work plan addressing the issues and goals of the Governor's Brucellosis Task Force report. The objective is to plan and implement management practices to maintain separation between elk and cattle, decrease and eventually eliminate elk dependence on supplemental winter feed and conduct brucellosis surveillance in elk.

As part of the Wyoming Game and Fish Department's integrated Brucellosis-Feedground-Habitat (BFH) program, a total of 570 elk were trapped and tagged at six feedgrounds this past winter. A total of 227 test-eligible female elk were bled for brucellosis evaluation. A total of 2,569 elk calves were vaccinated at 19 state feedgrounds. The strain 19 vaccination program was initiated for the first time during 2003 since 1989-1991 on the National Elk Refuge. Implementation of habitat improvements projects was greatly impeded last fiscal year due to incomplete environmental assessment of the projects and severe drought conditions, which prevented the necessary prescribed burns.

As part of Montana's Elk-Brucellosis Management Plan, active surveillance was conducted through hunter-harvested elk from elk management units of the GYA. Based upon these surveys, Montana elk

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are relatively free of brucellosis with seropositive rates of 1-4%.

Work continued on the Bison and Elk Management Plan for the National Elk Refuge (NER) and Grand Teton National Park. Three problems have emerged as key resource issues needing attention: (1) nonendemic infectious diseases, (2) degradation of native habitats, and (3) brucellosis. The year 2003 was spent primarily analyzing seven different proposed alternatives. Several options for controlling brucellosis are being addressed that would not require reductions in winter feeding or numbers of elk on the NER.

There were many Information and Education Subcommittee activities in the GYIBC in 2003. As part of their regular April meeting in Jackson, WY, the GYIBC held a public open house and panel discussion featuring the Governors from the three states and Undersecretaries from the Departments of Agriculture and Interior. Among other activities, a commitment was made to revitalize the GYIBC website as a vehicle for disseminating brucellosis-related information to the public. As a result, the GYIBC website was resigned and brought up to date on many facets. The revised website can be viewed at: <http://gyibc.com>.

IMPLEMENTATION OF THE INTERAGENCY BISON MANAGEMENT PLAN BY YELLOWSTONE NATIONAL PARK

Rick Wallen and Glenn Plumb
Yellowstone National Park, WY

Introduction:

Much of the controversy surrounding bison management at Yellowstone revolves around the fact that some (approximately 50%) bison are known to have been exposed to brucellosis. While brucellosis has been known from this population since early in the last century (USDI and USDA 2000), the proportion of bison that are infectious at any time of the year is unknown.

The use of spatial and temporal separation of bison from cattle on private and public lands surrounding Yellowstone National Park (YNP) provides a significant assurance to prevent the transmission of brucellosis from wild bison to domestic livestock. To further minimize the risk of transmission, cattle that occupy Special Management Areas (SMA) are being vaccinated for brucellosis. Implementation of the Interagency Bison Management Plan (IBMP) demonstrates a commitment to eventual eradication of brucellosis from the Yellowstone bison population. The interagency partners have agreed to work within their respective authorities and areas of jurisdiction to implement deliberate, stepwise measures that manage the risk of transmission while building a foundation for the eventual elimination of brucellosis in the bison population.

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Nearly all Yellowstone bison select habitats within YNP during the summer months. However, the winter landscape makes forage less available to bison because of snow depth and snow structure characteristics. Thus, the area available to bison during the most difficult months of winter are extremely reduced relative to year around distribution. Special management areas along the north and west boundaries of YNP have been designated to direct our management program which will in turn protect the brucellosis class free status for the state of Montana. Three separate zones are defined within each special management area.

Zone 1 = An area within YNP where bison are managed more intensively to assure that bison do not commingle with cattle on lands immediately outside the park.

Zone 2 = An area immediately outside YNP where bison will eventually be provided winter habitat, for use from 1 November through either early or mid May.

Zone 3 = An area immediately outside zone 2 whereby bison will be intercepted and hazed back in to acceptable tolerance areas, or removed if necessary.

Interagency Bison Management Plan (IBMP):

YNP is collaborating with two other federal and two state agencies to implement the IBMP. The management plan has two main objectives, to protect a free ranging wild population of bison and manage the population in a way that will avoid the risk of brucellosis transmission from bison to cattle (USDI and USDA 2000).

The key principles of the management strategy include the spatial and temporal separation of bison from cattle, a core area of suitable bison winter range outside the Park boundary which will be phased in to use by bison being tied to an increasing increment of bison and cattle vaccinated against brucellosis over time, and finally a minimum population size to protect the conservation value of this unique and valuable genome. Bison that enter the SMA and challenge the area of tolerance are subject to a moderately complex management decision process. This decision process is what generates the vast majority of conflict between constituencies and the interagency partnership. Hazing is considered as a management tool for implementing the spatial and temporal separation of bison and cattle. Should hazing become ineffective at managing bison distribution, bison will be captured. The decision regarding how to handle captured bison is an agency specific decision depending on which SMA bison are captured. At present, the options are only two fold. In early winter, disease management is the primary focus. In late winter, if the population is greater than 3000 bison, agencies have the option to initiate population control measures by cropping bison, only if they are captured in the SMA, or continue

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testing bison captured to further pursue disease management goals.

The IBMP has been implemented for four years now. The status of the accomplishments is currently being reviewed by an interagency review team and will be incorporated into the IBMP administrative record. Hazing of bison to manage distribution on the winter range has been initiated in both SMA's during each of the four winters of operation. Patterns that have evolved in the west SMA show that groups of adult male bison are generally 10 or less and hazing occurs from late September until early June. While generally, movements into the special management zones by groups of adult females begin in late winter and occurs well in to the parturition period. Movement of bison into the northern SMA, by groups of adult females, occurs earlier than at the west SMA and ceases prior to parturition. Movements by adult males into SMA's constitute a lower proportion of the hazing events at the northern SMA. Over the last few years population abundance has leveled off around 4000 animals. In three of those four years more than 200 bison per year have been removed from the population by management actions.

The results of the status review will provide the interagency managers information regarding whether to move to the second step in our adaptive management procedures. While some challenges still exist, the plan is moving forward in accomplishing both of the established goals. Spatial and temporal separation of bison and cattle has been successful.

The IBMP also directs the National Park Service (NPS) to initiate a program to vaccinate bison. The contingency was that vaccinating bison at the SMA's would be initiated once a safe vaccine has been identified. A review of the literature describing the bio-safety parameters of RB51 was completed and signed in to the administrative record by the YNP superintendent. In the spring of 2004, 113 calf and yearling bison were vaccinated at the north SMA (Table 1).

Table 1. Demographics of bison vaccinated at the Stephens Creek capture pen in Feb and March of 2004.

	Males	Females	% of estimated subgroup in the population
Calves	32	46	~ 23 %
Yearlings	18	17	~ 10 %

In addition to in-chute parenteral vaccination of bison at the north SMA, the NPS also has a responsibility to develop a strategy for deliv-

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ering vaccine to wild free-ranging bison that never go to the SMA's. In order to move forward with remote vaccination, the NPS must complete an environmental planning process to evaluate the alternatives. The purpose and need for this planning process are five fold:

- Meet the NPS mission to preserve native wildlife species as a component of a naturally operating ecosystem and protect them from exotic organisms;
- Address the NPS responsibility to implement the IBMP;
- Decrease the probability of individual bison shedding *Brucella* organisms;
- Demonstrate systematic progress in further reducing the risk of disease transmission from bison to livestock; and
- Decrease the percentage of Yellowstone bison infected with Brucellosis

We anticipate this planning process to take 18 months with an expected decision document being issued in January of 2006.

Remote delivery of a brucellosis vaccine presents many challenges. Delivery tools are currently very limited with ballistic delivery of vaccine in bio-absorbable bullet packages showing the most promise. Olsen et al (2002) suggested that ballistic delivery of RB51 vaccine may require a greater dose than would be recommended through syringe injection delivery. Likewise, Roffe et al. (2001) identified the short distances required for the BTI pneumatic delivery system to be successful. YNP has studied those challenges to evaluate the feasibility of success in developing a remote delivery vaccination program. A partnership with Colorado State University has resulted in new ideas for encapsulating the RB51 vaccine. Photo encapsulation of vaccine has been shown to be successful in the laboratory. A relatively high percentage of the live bacteria in the vaccine dose survive the photo polymerization process. In addition, the ballistics of the hydrogel delivery package are very comparable to the traditional bio-bullet system. Field trials are currently in progress to compare the efficacy of this encapsulation methodology with the traditional lyophilization and compaction method (S. Olsen, pers. comm.).

Field evaluations of bison behavior have led to greater confidence in approaching bison to close distances on a consistent basis. A park based program is in place for gaining new knowledge about movement patterns using a system of randomly placed radio transmitting devices to monitor individual animal movements. In addition, aerial surveys by park biologists combined with ground based monitoring aid in documenting abundance of the population and seasonal distribution.

An interagency surveillance program to monitor brucellosis prevalence is also in place led by Montana/APHIS at the west SMA and by NPS at the North SMA. Blood samples are collected from bison cap-

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tured at the SMA's and serology tests are conducted to determine exposure to *Brucella abortus*. A small sample of bison are randomly captured by NPS field staff throughout the park and tested for brucellosis exposure as well.

Conclusion

The IBMP protects the state of Montana interests by maintaining the Brucellosis class -free status designated by APHIS and when fully implemented should systematically reduce the incidence rate of brucellosis infected animals. The IBMP also concurrently achieves the NPS Mission by conserving Yellowstone Bison population and providing for suitable core winter range areas outside of YNP. The interagency partnership continues to implement the IBMP in a very deliberate manner utilizing transparent decision trees and a documented administrative record.

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NADC STUDIES ON BISON BRUCELLOSIS VACCINES AND MOLECULAR TECHNIQUES FOR *BRUCELLA* EPIDEMIOLOGIC TRACEBACKS

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The regulatory programs for elimination of brucellosis within the U.S. began in 1934 as a State-Federal cooperative program to reduce the cattle population during severe drought conditions. As the Brucellosis Eradication Program for cattle nears completion in the United States after 70 years of regulatory efforts, the persistence of *Brucella abortus* in wildlife reservoirs remains a concern for reintroduction of brucellosis to cattle. In the Greater Yellowstone area, sero-prevalence in bison is approximately 50% , whereas sero-prevalence in female adult elk over-wintering on feedgrounds is estimated at 35%. Within the last year, *Brucella*-infected cattle herds in Wyoming were identified which lead to the loss of that state's Brucellosis-Free status. Molecular

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tools for epidemiologic tracebacks on infected herds have previously not been available for brucellosis.

Based on analysis of the *Brucella abortus* genomic sequence, scientists at the United States Department of Agriculture, Agriculture Research Service, National Animal Disease Center (NADC) identified variable regions that may be useful for epidemiologic investigations (Bricker et al 2003). Although the *B. abortus* genome is very stable, these intergenic, noncoding regions containing repeated strings of nucleotides, are more apt to mutate than coding regions. An assay has been developed, the "HOOF-Prints" assay, which evaluates multiple loci containing these tandem repeats. The array of alleles identified by this technique creates a genotype for each isolate. For optimal performance of this assay, multiple colonies from each animal must be obtained for analysis. Data collected from *B. abortus* strains passed *in vitro* suggests that patterns of an isolate are stable. Analysis of isolates from culture collections indicates that genotypes differ between outbreaks. Multiple isolates from an infected bovine herd suggest that multiple, closely-related genotypes may be present within an individual, however, a single genotype will predominate. When combined with epidemiologic work, data from the HOOF-Prints assay suggest that a wildlife reservoir, most likely elk, were responsible for transmitting *B. abortus* to the Wyoming cattle. The HOOF-Prints assay looks very promising for use in future epidemiologic traceback efforts. However, additional data is needed, including characterization of the rate of change in individual loci and comparisons of isolates from sequential herds in a single outbreak. It is anticipated that a statistical model will be developed that will numerically estimate the degree of relationship between isolates from different herds.

A major component of the Brucellosis Eradication Program for cattle has been calfhood vaccination. In 1996, the *Brucella abortus* strain RB51 vaccine (SRB51) was approved by the Animal and Plant Health Inspection Service as a brucellosis vaccine for bovine calves between the ages of 4 and 12 months of age at dosages between 10 and 34 billion colony-forming units (CFU). Since that time, SRB51 has essentially replaced the *B. abortus* strain 19 vaccine which had been the official brucellosis vaccine for cattle in the United States since the 1940's.

Ongoing studies at NADC continue with the objective of developing a safe and efficacious brucellosis vaccine for free-ranging wildlife. The remainder of this report will summarize our work in bison (*Bison bison*).

We have previously reported that parenteral vaccination of bison with 1×10^{10} CFU of SRB51 is clinically safe, induces immune responses that are similar to responses of cattle, and is efficacious in preventing abortion or fetal infection following experimental challenge with a virulent strain of *B. abortus* (Olsen et al. 1997, Olsen et al. 1998,

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Olsen et al. 1999, Olsen et al. 2003). Other studies suggested that alternate methods of delivery of brucellosis vaccines to bison, such as bio-bullets, may influence immunologic responses (Olsen et al. 2002).

At this time, we have completed 7 efficacy studies evaluating vaccination of bison with RB51. These studies included 47 hand vaccinated, 21 ballistic vaccinated, and 25 non-vaccinated bison that were experimentally challenged in mid-gestation with *Brucella abortus* strain 2308 in accordance with the standard bovine challenge model. Bison were sacrificed at the time of abortion, or approximately 24 hours after parturition when observations on calf viability had been completed. Twenty-nine maternal or fetal tissue or fluid samples were collected for bacteriologic evaluation with additional samples obtained for histologic evaluation.

Abortion was defined as the birth of a *Brucella*-infected, nonviable fetus. Infection was defined as recovery of a single CFU from any tissue. Control and vaccinated bison were statistically compared to determine if differences in abortion or infection occurred. In addition, as brucellosis transmission is associated with infection in reproductive or mammary gland tissues, statistics were used to compare treatment groups for differences in infection within these tissues.

Data from our studies indicates that hand or ballistic vaccination with RB51 statistically ($P<0.05$) reduced abortions, uterine/mammary infections, and maternal infections. The reduction in abortions or uterine/mammary infections was a trend that was consistent across all studies. Our data suggests that irregardless of parental or ballistic delivery, RB51 protects bison against *Brucella* infections or abortions. Although it doesn't provide absolute protection in bison, our data suggests that RB51 is a viable vaccine candidate for use in bison.

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