REPORT OF THE COMMITTEE ON TUBERCULOSIS

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The Committee met on October 22, 2007, from 1:00 to 6:00 p.m. at John Ascuaga's Nugget Hotel, Reno, Nevada. There were 148 members and guests in attendance. Kathleen M. Connell and Michael S. VanderKlok presided. In her opening remarks, Connell reviewed the day's agenda and welcomed members and guests. The Chair determined that a quorum was present to conduct business.

Formal presentations began with C. William Hench, National Tuberculosis (TB) Eradication Program, Veterinary Services (VS), Animal and Plant Health Inspection Services (APHIS), United States Department of Agriculture (USDA). Hench provided the current status of the U.S. Bovine TB Eradication Program. The full text of this report is included in these proceedings.

Kathy Orloski, Epidemiologist, National TB Eradication Program, VS-APHIS-USDA, presented an update on the U.S. National Surveillance Program for Bovine TB. The full text of this report is included in these proceedings.

Brian Morrow, Director, Trace First Ltd., provided a presentation entitled Disease Management and Surveillance Using IT Systems Such as Modeling Exercises, an Emergency

Preparedness Toolkit and Surveillance Programs. The full text of this report is included in these proceedings.

Pauline Nol, National Center for Animal Health Programs, Ruminant Health Program-Wildlife, National Wildlife Research Center APHIS-USDA, presented an update entitled The Efficacy of Oral Bacille Calmette-Guerin (BCG) Vaccination in White-tailed Deer. The full text of this report is included in these proceedings.

Phil Durst, Extension Dairy Educator-Northeast Michigan, gave a presentation entitled A Case Study of the Test-and-Remove TB Herds in Michigan: A Description, History and Comparison. The full text of this report is included in these proceedings.

The current status of Mexico's campaign against TB and an update on Mexico's national surveillance program was delivered by MVZ M en C José Alfredo Gutiérrez Reyes, Subdirector de Sanidad en Especies Mayores, Dirección de Campañas Zoosanitarias, Dirección General de Salud Animal, Servicio Nacional de Sanidad, Inocuidady Calidal Agroalimentaria (SENASICA). The full text of this report is included in these proceedings.

REPORT OF THE SCIENTIFIC ADVISORY SUBCOMMITTEE ON TUBERCULOSIS (TB SAS)

Chair: Mitch Palmer

The following presentations were made during the Subcommittee meeting, October 20, 2007.

Cervid Serum Bank Activities and Update in Response to Resolution 21.

Jeffrey T. Nelson National Veterinary Services Laboratories

In response to Resolution 21 passed at the 2006 Annual Meeting, USDA-APHIS-VS began collection of serum samples from various deer species to create a serum bank of well-characterized samples. Skin test data as well as histopathology and culture results, when available accompany samples. Most samples have been submitted from white-tailed deer, fallow deer and reindeer. Lesser numbers of samples have been submitted from other deer species. Serum panels have been provided to various developers of serological assays for test validation. Future work will focus on continued collection of samples and creation of serum panels for test validation. It has been suggested that a similar serum bank be created using samples from cattle. Such a bank could be used to create serum panels for use in bovine TB test evaluation and validation.

An Efficient Cost Effective Serology Assay for Accurate Identification of *M. Bovis* Infected Cattle.

Larry Green PriTest

PriTest's new diagnostic test for cattle tuberculosis infection, SeraLyte-*Mbv*TM, greatly improves the speed and accuracy of TB detection. An accurate test is needed to reach the goal of finally eliminating TB in the United States. A serologic screening test has long been hoped for, but it has not been identified. Many experts in the industry, based on the poor performance of serologic tests previously used to detect *Mycobacterium bovis*, have assumed that no satisfactory serologic test could be identified. It appears by all measures that with the PriTest proprietary imaging method in a well worked out assay protocol, serologic methods for accurately identifying *M. bovis* infected cattle are easily achieved. Most importantly, the test is rapid and economical compared to other methods currently in practice. Since our report to the USAHA in October 2006, two large USDA blinded studies have been completed in cattle with infected animals. PriTest evaluated serum samples from both studies. In addition to the blinded studies, we have tested

extensively to gather data the will be useful in evaluating specificity for the test by testing cattle in presumed *Mycobacterium bovis* free zones. Based on these findings it appears that our serologic test can achieve specificity measurements in the range 95 to 100 percent. We believe the current sensitivity is greater then 90 percent. SeraLyte-*Mycobacterium bovis* enables a paradigm shift from diagnosing only limited surveillance populations and animals with active disease forms of TB, along with testing those in reactor herds, to screening large populations for unsuspected infection with the *Mycobacterium bovis* bacillus. Based on the test results in its current validation work on US and United Kingdom cattle, PriTest is requesting the USAHA TB SAS to assist PriTest in proceeding to a phase 3 field trial; that would include side-by-side comparison testing to current screening and confirmation test methods on the appropriate number of reactor and Accredited Tuberculosis Free herd samples in the US.

Update on Guidelines for the Control of Tuberculosis in Elephants, 2007.

Michelle Miller

Disney Animal Programs, Department of Veterinary Services.

The emergence of TB in elephants in 1996 prompted the formation of an advisory panel to draft guidelines for the control of tuberculosis in elephants. Since that time various modifications of the guidelines have been drafted. The proposed 2007 guidelines incorporate several changes including the addition of serological testing using Chembio's elephant TB Stat-Pak and additional options for culture positive elephants. Proposed guidelines would call for annual testing by the triple culture method (three trunk wash samples) and a single sample of serum collected for analysis by the elephant TB Stat-Pak. Guidelines for treatment and movement restrictions would be based on culture and serological results. A Subcommittee has been formed to review and comment on the proposed guidelines.

Tuberculosis Diagnosis: Analyzing the Parameters of the Interferon-gamma Assay.

Irene Schiller, Roland Hardegger, Annika Kyburz, Alex Raeber, Bruno Oesch Prionics AG

> Ray Waters, Mitchell Palmer, Brian Nonnecke, National Animal Disease Center

Martin Vordermeier, Teklu Egnuni, Veterinary Laboratory Agency, Great Britain

The Bovigam interferon (IFN-g) assay constitutes an ante-mortem, in vitro laboratory based tuberculosis test and is widely used complementary to tuberculin skin testing. The assay is performed in two stages: first, whole blood is cultured with antigens stimulating blood leukocytes to produce IFN-g that is quantified by enzyme-linked immunosorbent assay (ELISA) in a second step. Environmental conditions before and during the culturing of the leukocytes influence the efficacy of in vitro IFN-g production. Optimal conditions are therefore essential. In this study we analyzed the effect of stimulation vessel geometry, temperatures during stimulation, and the stability of antigens stored at different temperatures. Blood from experimentally infected cattle and from tuberculosis negative cattle was stimulated in 24 well tissue culture trays (standard), 48 well and 96 well culture plates with the following antigens: purified protein derivative from Mycobacterium bovis (PPD-B) and from Mycobacterium avium (PPD-A), a fusion protein from early secretory antigenic target 6 (ESAT-6), culture filtrate protein 10 (CFP-10), and pokeweed mitogen. Stimulation was equally efficient in all plate formats. The results with specific antigens correlate with mitogen induced stimulation, CO₂ is not required during incubation, as cultures from an incubator with five percent CO₂ produced similar amounts of IFN-g as without CO₂. However, the temperature used for stimulation was critical. Stimulation at 37°C and 33°C was equally efficient, but a culture temperature of 29° C reduced IFN-g production significantly. At 25°C and 22°C no stimulation was detectable. Antigens are usually stored at 2°-8° C (tuberculins) or at -80° C (recombinant proteins) until usage. We tested in parallel antigen storage of recombinant proteins (ESAT-6:CFP-10 fusion protein, TB10.4, TB27.4, MPB3) at 4°C for 24 hr at 20°C for 8 hrs prior to use in cell culture. Our results show that antigens may be stored at either of these conditions without affecting the efficacy of stimulation. Finally, we compared the activities of tuberculins from five different sources in naturally infected cattle (n=10). Matched PPD-B and PPD-A tuberculins were used at eight dilutions each. Relative potency 30 (RP30) was defined as the tuberculin concentration required to induce 30 percent of the peak response values. RP30 differed by a factor of more than 10 between the PPD-B with the highest and lowest potency. Therefore, tuberculins of different sources may give different results and the overall assay performance may be improved by optimizing tuberculin concentrations.

Update on Chembio Tuberculosis Assays

Konstantin Lyaschenko Chembio Diagnostics Systems

Multiple animal species are susceptible to tuberculosis (TB) that has serious zoonotic and regulatory concerns. The current testing methodologies are inadequate for most of the non-domestic species. To improve TB control programs, new diagnostic tools that would be simple, rapid, accurate, and inexpensive are urgently needed. Chembio developed two serological assays, PrimaTB STAT-PAK and ElephantTB STAT-PAK, using lateral-flow technology to detect specific antibody in animals infected with *Mycobacterium tuberculosis* or *M. bovis*. These two rapid tests were approved by USDA, Center for Veterinary Biologics in 2007. In addition, the Multi Antigen Print ImmunoAssay (MAPIA) was proposed for elephants, particularly, as confirmatory test and treatment monitoring tool. The results of clinical evaluation of PrimaTB STAT-PAK assay showed 87 percent sensitivity and 99 percent specificity in macaques. Extended studies with ElephantTB STAT-PAK (100 percent sensitivity and 97 percent specificity in elephants) demonstrated its potential to be a valuable animal-side diagnostic tool in multiple zoo animals as well as in a number of free-ranging wildlife species involved in maintaining *M. bovis* reservoirs worldwide.

An Evaluation of New Technologies for Diagnosis of *Mycobacterium bovis* Infection in Cattle.

Robert M. Meyer Kathy A. Orloski Veterinary Services

Drs. Bob Meyer and Kathy Orloski discussed the identification of tuberculosis in two New Mexico dairy herds. Serum samples were collected from a large number of cattle from these herds and submitted blindly to three different companies for analysis by four different assays; PriTest's SeraLyte Mbv assay, Chembio's TB Stat-Pak and MAPIA, and Diachemix FPA. Sensitivity and specificity estimates for each assay were presented. Depending on the assay, sensitivity estimates ranged from 26 to 82 percent and specificity estimates ranged from 76 to 100 percent.

Although no specific assignments were given to the TB SAS nor was any data provided to the TB SAS for evaluation and comment several observations were made. Significant progress has been made in development of serological assays for TB in various species as well as standardization of existing assays such as the Bovigam. Further progress will require continued cooperation between USDA, industry and producer groups. USDA's continued support of the creation of well defined serum banks and use of samples from naturally infected herds, such as that in New Mexico, for test validation will be critical. Continued support from producers as a source of samples with accompanying skin test data will also be needed.

The Committee approved the Subcommittee Report.

State Updates followed, provided by Mike VanderKlok, Michigan, Linda C. Glaser, Minnesota Board of Animal Health, and Tim Hanosh, New Mexico Assistant State Veterinarian. The full texts of these reports are included in these proceedings.

Billy Johnson, Bi-National TB and Brucellosis Committee Coordinator, followed with a report on the Bi-National Committee (BNC) activities. Johnson gave a brief history of this 16-member committee. He discussed TB reviews in Mexico, the waiver conditions document and the current statuses of states. The full text of this report is included in these proceedings.

Formal presentations continued with C. William Hench providing an update on proposed changes to the Code of Federal Regulations (CFR) regarding the bovine tuberculosis program. He summarized the status of proposed changes to the domestic and international rules for the TB program. No written report is available for this presentation.

At the conclusion of the formal presentations, Connell reported on 2006 Resolutions. USDA-APHIS-VS responded promptly in writing to both resolutions from 2006. Connell read the USDA-APHIS-VS responses to the two resolutions to the attendees.

Four Resolutions were approved and forwarded to the Committee on Nominations and Resolutions:

CURRENT STATUS OF THE US BOVINE TUBERCULOSIS ERADICATION PROGRAM FISCAL YEAR 2007

C. William Hench National TB Eradication Program

The cooperative State–Federal–Industry effort to eradicate bovine TB from the United States has made significant progress toward eradication, markedly decreasing the prevalence of the disease. However, the goal of eradication has been elusive despite renewed efforts. Remaining challenges—primarily infected wildlife and infected cattle from Mexico—hinder eradication.

In fiscal year (FY) 2006, there was a rise in the number of cattle herds that were found to be tuberculosis-affected relative to the previous year. These herds were all located in areas where we have discovered affected herds in previous years. In FY 2006, a total of nine affected herds were found. In contrast, seven affected herds, six bovine and one captive cervid, were discovered in FY 2007. Slaughter surveillance for TB continues to exceed our national goals in FY 2007, and three of the newly discovered herds were detected as a result of this surveillance and epidemiological investigations. This shows that slaughter surveillance continues to be an integral part of our eradication program. Nevertheless, TB response plans remain critical in areas where the disease has recently been detected.

At the end of FY 2006, 49 States and Territories were TB Accredited-Free (AF), including Puerto Rico and the U.S. Virgin Islands. Two States, New Mexico and Michigan were regionalized, and Texas was classified as Modified Accredited Advanced (MAA). New Mexico was regionalized in FY 2005 with a small zone in the eastern region of the State classified as MAA and the remainder of the State TB-Free. Michigan was further regionalized during FY 2005. At that time, Michigan was divided into three zones: the Upper Peninsula was classified as AF, 11 counties and portions of two others in the northeastern Lower Peninsula were Modified Accredited (MA), and the remaining counties in the Lower Peninsula were MAA.

In January 2006, as a result of the discovery of three affected herds in the State, Minnesota was downgraded to MAA status. During 2006, the State of Texas once again became eligible and applied for TB-Free Status. Texas' AF status was initially restored with the October 2006 publication of an interim rule in the *Federal Register* and was finalized with the January 2007 publication of a final rule. As a result of these changes during FY 2006, at the end of the year, 49 States and Territories were classified AF, including Puerto Rico and the U.S. Virgin Islands, two States remain regionalized, New Mexico and Michigan, and one state, Minnesota, has MAA status. There has been no change in the status of any State or Territory during FY 2007.

Of the six affected cattle herds discovered in FY 2007, two were beef herds in Minnesota. The two new herds were identified during continued surveillance and epidemiological investigations. The source of this infection has not yet been determined and epidemiological investigation of the subsequent herds is in progress. In addition to the two new beef herds discovered in 2007, surveillance of free ranging white-tailed deer is on-going through hunter-harvested and targeted culling sample collection. As a result of finding these additional herds and infected wildlife, the State of Minnesota and USDA jointly developed a management plan for livestock and wildlife statewide. The goal of this management plan is to determine the extent of the infection in livestock and to determine whether or not the disease has become established in wildlife. All herds affected in Minnesota to date have been depopulated with federal indemnity.

In Michigan, two herds – one bovine and one captive cervid – were detected in FY 2007. Both of these herds are located in northern Lower Michigan in the bovine MA zone. The captive cervid herd was identified through hunter kill surveillance. The bovine herd was a small dairy and was identified through annual surveillance testing. Both of these herds have been depopulated.

The other three herds detected in FY 2007 were all a result of investigations initiated by Food Safety Inspection Service (FSIS) identification of *Mycobacterium bovis* infected animals detected during routine slaughter inspection. Trace testing confirmed infection in a beef herd in western Oklahoma in April 2007. Oklahoma has been classified as AF since 1996. Trace testing

also confirmed infection in a large dairy operation in New Mexico's AF zone. This dairy operation encompasses two premises and totals approximately 12,000 head of cattle. In Colorado, an affected herd was disclosed through epidemiological investigation of a rodeo bull found to be infected at slaughter. Testing of herds in which the affected animal resided confirmed infection in performance cattle (bucking bulls) herd. Colorado has been classified AF since July 1975. The herds in Colorado and Oklahoma have been depopulated with federal indemnity. The New Mexico dairy operation is currently in the depopulation process.

Three affected herds detected prior to FY 2005 remain under quarantine and test and removal herd plans. The first of these herds is a dairy herd in New Mexico which declined to depopulate. Two dairies in Michigan also remain under quarantine and test and removal herd plans. One of these quarantined dairies in Michigan is a reinfected herd. All three herds continue to undergo regular herd testing as part of their herd plans. Michigan herd plans also include requirements for mitigating the risk of infection from wildlife.

FY 2007 herd depopulations were accomplished at a cost of \$1,499,430. Indemnity costs for caudal fold tuberculin test positive animals in affected herds, comparative cervical tuberculin test- or gamma interferon-positive and suspect animals in non affected herds and for certain other situations were \$1,451,926 in FY 2007. Total indemnity costs for all purposes were \$2,951,356.

Due to continuing concern about the level of surveillance for TB in captive cervids, a working group of State-Federal personnel developed a surveillance plan for captive cervids in 2004. That plan, conditionally approved by cervid industry leadership, was presented during the 2004 Annual Meeting of the USAHA Committee on Tuberculosis and discussed and comments and suggestions were made. All of this input was incorporated into a draft Uniform Methods and Rules (UMR) for Captive Cervids. Finalization of this UMR has been delayed while USDA drafts comprehensive revisions of both the bovine and cervid portions of the TB rules in the Code of Federal Regulations (CFR).

Currently there are 15 states and the U.S. Virgin Islands that have achieved and maintained their TB Free status for over 25 years; 22 states that have been TB Free for 15 or more years; seven states that have been TB Free for 10 or more years; three states and Puerto Rico that have been TB Free for five or more years; and two states and two regionalized zones which have had TB Free status for less than five years. Given the six bovine herds discovered this year and the three herds that remain under quarantine from previous years, there were nine affected herds among the estimated 971,400 cattle herds in the United States at the end of FY 2007. Therefore, the national prevalence for FY 2007 is estimated to be 0.0006 percent, or one affected herd per 107,933 U.S. herds. Though TB does exist in the United States, this extremely low level of prevalence should be a significant factor in convincing international trading partners of the very low level of risk with TB in our cattle.

Veterinary Services (VS) has completed its oversight of the operation for the removal of all dairy operations from the El Paso, Texas milk shed. This process was completed during calendar year 2007. There were a total of 10 dairy operations, some with multiple production units, removed to create a buffer zone between the U.S. and the TB affected dairy operations immediately across the border in Juarez, Mexico. All 10 operations have completed close out procedures. During this program, designated VS and Texas Animal Health Commission personnel ensured that every animal leaving the premises was identified and permitted to slaughter or quarantine feedlots. All depopulated cows were inspected at slaughter and had no TB lesions detected. Each depopulated dairy will remain out of operation, in the El Paso area, for at least the next 20 years.

VS continues to work with Mexico on ensuring there is equivalency between the two countries' requirements. To accomplish this, reviews of Mexican State TB programs have been conducted under the umbrella of the U.S. and Mexico BiNational Tuberculosis and Brucellosis Eradication Committee. For this fiscal year there were seven review trips completed. The review teams examined TB program integrity, progress and the level of prevalence. There were two reviewers working under contract, 11 that were VS or International Services (IS) employees, one National Veterinary Services Laboratories employee, and seven that were employed and paid for by State or industry agencies in Arizona, California, Missouri, Texas, and Washington. The financial contributions of those States and industry groups are recognized and appreciated.

In 2007, a five-year plan, Strategic Plan for Reducing the Risk of Importing Tuberculosis Infected Cattle from Mexico 2008-2012, was developed and presented to Mexico. This plan requires that the Mexican TB eradication program achieve equivalency with the U.S. program by the end of 2012.

Extensive efforts have been taken in regards to the U.S. rule making efforts related to tuberculosis. Changes to the CFR will be proposed for the bovine, cervid, international, roping steer and the indemnity regulations. These CFR revisions are currently in various stages of review. Given the complexity of the revisions and the linkage between the bovine, cervid, and international rules, this process is lengthy. The bovine and cervid rules were separated this year to allow the domestic and international bovine rules to proceed at a faster pace. The roping steer regulation has been has been drafted and is currently undergoing final review and economic analysis. Changes to the TB indemnity section of the CFR are also in final stages of review before publication.

Updates on States with Recent Infection

Colorado update: As a result of a rodeo bull identified as infected by FSIS inspection in January 2007 a thorough epidemiological investigation was initiated. One herd was identified as affected and depopulated with federal indemnity. Seventeen contact herds were tested, approximately 950 animals, without any additional findings of infection. Epidemiologic investigations are ongoing and extend to over 30 states but have led to no additional sources of infection at this time.

Michigan update: Two new affected herds were found in FY 2007. One herd was a small dairy and the other was at captive cervid ranch. Both herds were depopulated. The State continues as regionalized in three zones: TB Free, MAA, and MA. Eleven hundred herds are tested in the MA zone annually. Eight hundred randomly-selected cattle herds are tested each year in the TB Free and MA zones. The prevalence of TB in wild deer continues to decrease. The prevalence in wild deer in the core of the Modified Accredited zone (DMU 452) was 2.3 percent in 2006 which is up from 1.2 percent in 2005. Continued monitoring will be necessary to see if this is indeed a trend in prevalence or a one year anomaly.

Two dairy herds in Michigan continue under test-and-removal herd plans and are classed as carry-over herds from FY 2006. One is located in Alpena County, with about 100 head total. This herd was detected through area (annual surveillance, FY 2004) testing and one positive animal was found. The other herd is located in Montmorency County, with about 175 head total. It was detected through area (annual surveillance, FY 2004) testing as well with five reactors found. This is the second time this herd has been found affected. It was originally found positive in 2000 and released in 2002, before being detected again in 2004.

Minnesota Update: There were two positive beef herds detected in FY 2007 through continued epidemiological testing of area and high risk herds. All affected herds have been located in either Roseau or Beltrami Counties. Through FY 2007, all affected herds in Minnesota have been depopulated. Epidemiological investigations for all affected herds continue in Minnesota and additional states. In FY 2006, The Minnesota Department of Natural Resources and the Minnesota Board of Animal Health worked with USDA to develop a surveillance plan in both livestock and wildlife. This surveillance plan calls for risk-based, statewide testing of livestock and wild deer to determine the extent of the TB infections in the State and to also clarify whether the disease has become established in wildlife or not. Additional federal funding has been provided in support of TB surveillance in Minnesota in both cattle and wildlife, funding for fee-basis veterinarians, and federal TB testing teams.

New Mexico update: One dairy herd in the MAA zone of New Mexico continues under a test and remove herd plan. To date, no additional TB has detected in this herd. Five FSIS slaughter inspections identified infected cattle that were traced back to two herds in New Mexico's accredited-free zone. Over 100 caudal fold test (CFT) and gamma interferon suspects were removed from one dairy for laboratory analysis with no further findings of TB. At the other dairy TB infection was confirmed. This dairy is currently under quarantine and depopulation efforts have begun.

As a result of finding this dairy operation affected a Task Force was initiated in August 2007 to assist New Mexico Animal Health Officials with the tremendous load of traces and testing

that needed to be accomplished. At the time of this report the work effort there is winding down with no signs that infection has spread beyond the original herd.

Oklahoma update: A November 2006 FSIS slaughter trace led to a herd which was identified as affected through on farm testing. Herd testing was initially delayed because of inordinately bad winter weather in this part of the country. The herd has been depopulated. Traces from this case extend to several western states and no further findings of TB have been found.

UPDATE ON THE US NATIONAL SURVEILLANCE PROGRAM FOR BOVINE TUBERCULOSIS FISCAL YEAR 2007

Kathy Orloski Veterinary Services

Primary surveillance for bovine TB in the United States (U.S.) consists of slaughter surveillance for granulomas and skin and blood testing in cattle.

The national granuloma submission surveillance program for adult cattle met or exceeded the target rate of five submissions per 10,000 adult cattle killed for the sixth consecutive year as of the third quarter of federal fiscal year (FY) 2007, with 15.2 granuloma submissions per 10,000 adult cattle killed. A total of 10,286 granulomas were submitted from US plants. A total of 36 of the top 40 adult slaughter (90 percent) establishments met the target rate of five submissions per 10,000 adult cattle killed. Four establishments were at 18, 43, 73 and 79 percent of the standard as of the third quarter of federal FY 2007.

A critical component of the granuloma submission program is diagnostic laboratory support. Three diagnostic laboratories provide outstanding support for the national bovine TB surveillance effort. A total of 7,090 (68.9 percent) samples were evaluated by National Veterinary Services Laboratories (NVSL), 1,816 (18.5 percent) by the Food Safety Inspection Service (FSIS) Pathology Laboratory, Athens, Georgia, and 1,298 (12.6 percent) by the California State Diagnostic Laboratory, Tulare, California.

Slaughter surveillance continues to identify new cases of TB in both adult and fed cattle. Twenty-four new cases of TB were found in cattle in U.S. slaughter plants during FY 2007, compared with 28 cases in FY 2006. No cases of TB were detected in bison or captive cervids slaughtered under state or federal inspection during FY 2004 through FY 2007.

Of the 24 new TB cases, six (25 percent) cases occurred in adult cattle. Three cases resulted in the identification of three new affected herds in Colorado (rodeo/beef), Oklahoma (beef) and New Mexico (dairy). The herds in Colorado and Oklahoma have been depopulated with federal indemnity. The New Mexico dairy operation is currently in the depopulation process.

A fourth adult case in a culled dairy cow was traced back to a New Mexico herd of origin; no additional infected animals were found following comprehensive herd testing. This dairy was not depopulated, but has been identified as a high risk herd and will undergo additional testing. A fifth adult case (beef) was from South Dakota; extensive testing did not identify additional infected animals. A sixth adult case (beef) was slaughtered in South Dakota. No additional infected cattle were found during herd testing in South Dakota; however, a vaccination eartag was found to be a duplicate, bringing into guestion whether the herd of origin was correctly identified.

One adult TB case occurred in a rodeo performance bull from Colorado. This animal traveled extensively while performing, resulting in trace back investigations in over 20 states. An additional infected bull was found in a beef and rodeo cattle herd in Colorado where the index case had previously resided. In FY 2006 a TB case occurred in a Kansas Mexican-origin roping steer, resulting in the exposure and depopulation of exposed beef breeding cattle. These cases illustrate the risk that longer-lived, roping and performance cattle may cause to our livestock industries. The Colorado bull was not of Mexican-origin, but may have been exposed to TB-infected Mexican-origin cattle.

The New Mexico affected dairy was identified in April of this year when an adult Holstein cow presented with lesions suggestive of TB during regular slaughter inspection at a federally-inspected plant in Arizona. Tissues from this animal were positive for TB by polymerase chain reaction (PCR) and culture. Epidemiologic investigation found that this cow had originated from a large dairy in eastern New Mexico milking approximately 2,400 adult Holstein cattle. On further investigation, an associated herd milking nearly 3,600 cattle was found to be involved due to movement of cattle from the index herd.

Genotyping is being used to assist in epidemiologic investigations of TB cases. Genotyping confirmed that two isolates of *Mycobacterium bovis* in the Oklahoma affected herd were identical and that this strain was the same as only ten other isolates in the NVSL database.

An evaluation of eight *M. bovis* isolates from roping steers cases detected between 2001-2006, found that the variability in these isolates was similar to that seen in *M. bovis* isolates obtained from feeder cattle detected through slaughter surveillance.

The remaining 18 (75 percent) cases were detected in fed steers or heifers considered to be beef-type cattle. These cattle had been fed in Texas (14 cases), California (three cases) and Kansas (one case). Of these 18 cases, 17 were of Mexican origin. The state of origin for 14 cases with Mexican official eartags include Aguascalientes, Chihuahua, Coahuila, Jalisco, Sonora, one case each; Campeche and Tamaulipas, two cases each; Nuevo Leon, five cases. Three additional cases originated from Mexico, but the state of origin has not yet been identified. One domestic fed cattle case occurred and was traced back to the depopulated beef herd from Oklahoma.

In FY 2006, approximately 1.1 million cattle were imported to the U.S. from Mexico; a majority of these are feeder cattle. A sample of 22 recent TB cases in Mexican origin feeder cattle found these animals resided in the U.S. a median of 10 months (range 4.6 to 16 months). Using FY 2007 TB cases in cattle of Mexican origin and FY 2006 Mexican cattle import records, the overall incidence of TB cases is 1.6 cases per 100,000 imported cattle. This is a substantial decrease from 1995 through 1997, when there were 7.3 to 18.7 infected cattle per 100,000 imports annually. Beginning in 1998 through the present, the annual rate has ranged from 1.0 to 5.4 infected cattle per 100,000 imports. Though this represents a sustained decrease from earlier years, infected cattle continue to be imported from Mexico and present an ongoing risk of TB transmission to U.S. cattle.

National TB surveillance is also accomplished through tuberculin skin testing and gamma interferon testing of livestock. Preliminary data of caudal fold tuberculin tests conducted during FY 2007, show that 961,475 tests were conducted on cattle and bison with 12,488 responders (1.3 percent, 46 states and Puerto Rico reporting). The response fraction in FY 2006 was 1.0 percent. A standard for caudal fold testing was implemented in 2005, based on an expected false positive response fraction of 1 percent (Uniform Methods and Rules, Appendix C, January 2005).

During FY 2007, a total of 231 suspects (2.2 percent) were reported to USDA from the 10,353 captive cervids tested by the single cervical test. The FY 2007 reports are preliminary; however, cervid testing reported in FY 2007 appears to have decreased from the 25,421 cervids reported in FY 2006 (374 responders, 1.5 percent).

The gamma interferon test (GI) has been available as an official test in the national eradication program for bovine TB for three years. Four laboratories throughout the United States are approved to conduct gamma interferon testing (California, Michigan, Texas, National Veterinary Services Laboratory). Collectively, these laboratories reported testing 14,618 blood samples during FY 2007. Ninety-seven percent of tests were for cattle from four states (Colorado 773 tests, Michigan 2,581, New Mexico 9,622, and Texas 1,125).

DISEASE MANAGEMENT AND SURVEILLANCE USING IT SYSTEMS SUCH AS MODELING EXERCISES, AN EMERGENCY PREPAREDNESS TOOLKIT AND SURVEILLANCE PROGRAMS

Brian Morrow
Trace First Ltd

The importance of strong foundations in the creation of efficient systems for disease surveillance is often overlooked. This is, in part, due to a natural tendency to look towards the program-specific aspects of the IT systems rather than the more general requirements. The term "good foundations" encompasses information, processes and systems; for example, you should ask yourself difficult questions.

- Is my data available electronically 24 x 7, 365 days?
- Is it systematically refreshed?
- Is it available in a system my staff can actually use?

Equally important are the processes that are used to capture the data. Do they allow the introduction of errors or do they exclude them? The critical point is, do I generate data or information?

Do not build different systems for each quality or disease program; work to find a common set of requirements and build a system that meets the majority of your requirements for all. You can then generate the information that is required to effectively manage each program. Watch out for common pitfalls such as allowing bad data to accumulate in your systems; remember that it is many, many times more expensive to manually correct bad data after it has gained entry to your systems. Periodic updates and refreshes are essential to not only run your programs effectively, but also because in the event of an emergency or notification requirement your core information must be accurate.

Our team at Trace First has been involved in livestock traceability systems since 1986; our team of world-class experts understands your challenges and can deliver the veterinary information systems you need. Trace First has earned the trust of customers like you and wants to demonstrate what we can do for you.

Our products include:

- Quality Program and Disease Surveillance Management
- Premises Registration and Update Service
- Emergency Preparedness Toolkit.

These systems, individually or combined, can deliver substantial benefits to your department and can be implemented to complement your existing systems.

THE EFFICACY OF ORAL BACILLE CALMETTE-GUERIN (BCG) VACCINATION IN WHITE-TAILED DEER

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National Center for Animal Health Programs, Ruminant Health Program-Wildlife
National Wildlife Research Center

Bovine tuberculosis poses a serious continual threat to the health and economic well-being of both livestock and humans. Free-ranging white-tailed deer (*Odecoileus virginianus*) can be endemically infected with *Mycobacterium bovis*, and serve as a potential reservoir of infection to livestock, other wildlife species, and humans, representing a significant obstacle to disease eradication efforts. An effective vaccination program, involving a mucosal vaccine that can be distributed in the field, would be very useful for disease management on a herd-wide level.

We investigated the efficacy of oral and parenteral BCG in its ability to protect white-tailed deer against disease and shedding caused by *M. bovis* infection. Thirty white-tailed deer were divided into four groups. One group was vaccinated with 10⁶ colony forming units (cfu) BCG (Danish strain 1331) subcutaneously, one group received 10⁹ cfu BCG in culture directly to the oropharynx, one group received 10⁹ cfu BCG via a lipid-formulated oral bait, and the last group received a placebo directly to the oropharynx. Throughout the study, oropharyngeal swab and fecal samples were collected monthly. Three months post-vaccination, all deer were challenged with virulent *M. bovis*. Five months post-challenge, the animals were examined for lesions caused by *M. bovis*. Results indicate that both oral forms of BCG offered significant protection against *M. bovis* challenge as compared to placebo. No differences in shedding among vaccine groups could be detected at the time points examined. These positive results warrant expanded research of oral BCG in white-tailed deer for eventual use in field investigations.

A CASE STUDY OF THE TEST-AND-REMOVE TB HERDS IN MICHIGAN: A DESCRIPTION, HISTORY AND COMPARISON

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> Galen Schalk Michigan dairy producer

Dan Grooms
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It has been almost eight years since the diagnosis of bovine tuberculosis (bTB) in a Michigan dairy herd that was allowed to undergo a test and remove program rather than wholeherd depopulation. Since then, four additional dairy herds have gone, or are going, through a test and remove program. The experience with these herds allows us to evaluate the ability of the test and remove protocol to control bTB while maintaining herds as economic drivers in their communities. In four of five herds, only one animal was ever diagnosed with bTB even though testing including the caudal fold test (CFT), comparative cervical test (CCT) and gamma interferon (ifn-g) repeatedly throughout a two year quarantine period initially and now throughout four and a half years of guarantine. Only one animal was diagnosed in those herds also in spite of the fact that 178 animals, 33 percent of the combined mature herds, have been taken for slaughter, necropsy and histopathology. One herd has become reinfected, although it may have been new infection from the wildlife source. Even so, this compares to a recrudescence of at least 3 of 22 repopulated beef herds following whole herd depopulation. Meanwhile, the test and remove herds are still in business and bringing income into Michigan communities of approximately \$1.4 million per year. This paper presents a case for allowing test and remove as an option for producers where certain conditions are met.

CURRENT STATUS OF MÉXICO'S CAMPAIGN AGAINST TUBERCULOSIS (TB) AND UPDATE ON MÉXICO'S NATIONAL SURVEILLANCE PROGRAM

MVZ M en C José Alfredo Gutiérrez Reyes Subdirector de Sanidad en Especies Mayores Secreteria de Agricultura, Ganaderia, DeSarrollo Rural, Pesca Alimentacion (SAGARPA)

Mr. Gutiérrez provided a report on the current situation of México's Bovine TB Program. He first covered the country's laws and rules, to include:

Regarding laws, the Federal Law of Animal Health was published on July 25, 1995, and the Federal Law on Metrology and Normalization, Chapter III, Articles 52 to 57, was published on June 30, 1992.

Regarding regulations, the Mexican NOM-031-ZOO-1995 is being updated. NOM-046-ZOO-1995, National System of Epidemiological Surveillance, was published on February 19, 1997. NOM-018-ZOO-1994 was published on April 25, 1995, and covered veterinarians approved to conduct verification and other official services in zoosanitary activities. NOM-054-ZOO-1996 deals with qquarantine establishment for animals and their products and was published on June 8, 1998.

Mexico now has 11 states and 14 regions in the eradication phase of the TB campaign. Mr. Gutiérrez reviewed a PowerPoint presentation with some data on the program.

He discussed the modification of the federal regulations regarding campaign phases, mobilization and quarantine procedures and gave the tentative schedule of TB reviews in México for 2008. He also mentioned records for tuberculin tests, quarantined premises, results of epidemiological surveillance from slaughter cattle and laboratory results. Mr. Gutiérrez spent some time reviewing TB cases in Mexican cattle slaughtered in the U.S. and reported to SAGARPA.

Strengths of the Mexican TB program include:

- Federal appropriation of approximately 33 percent of the General Animal Health budget for the TB program through the "Alianza para el Campo";
- Specialized and trained personnel;
- Surveillance systems becoming more efficient;
- Reliable TB program indicators;
- Continuous TB inspections at slaughterhouses and movement training; and
- Equivalent regulations in constant update.

STATE TUBERCULOSIS (TB) UPDATES

Michigan

Michael S. VanderKlok Michigan Department of Agriculture

The Michigan Department of Agriculture, in conjunction with the Michigan Department of Natural Resources, United States Department of Agriculture, Animal and Plant Health Inspection Service, VS and WS, and other state and federal agencies have been working on eradicating bovine TB from all livestock and wildlife species since it was identified in free-ranging white-tailed deer in 1995. Since that time TB has been found in 42 cattle herds and two captive cervid herds, all located in the current modified accredited (MA) zone of the northern lower peninsula. It has also been found in free-ranging white-tailed deer in this same area. As of October, 2007, over 1.3 million TB tests have been completed in Michigan cattle, and over 153,000 free-ranging white-tailed deer have been tested for TB statewide. Evidence to date supports that the disease appears to be confined to the MA area, and the specific strain of TB in Michigan has not spread to any other area of the state, and has not been found in any other state or country. Evaluation of historical testing and epidemiological information demonstrates that the risk of TB infection in cattle herds is related to the location of the herd, and geospacially related to proximity to the northeastern area of lower Michigan. This area contains over 90 percent of all the TB infected wildlife and cattle herds found to date.

Although the current animal identification, movement tracking, annual TB testing of cattle herds, and TB movement testing requirements appear to prevent the spread of disease through cattle movement, the risk of transmission from TB infected wildlife is still present. Future activities will continue to expand the areas of preventing this spillover, and developing more risk-based targeted surveillance strategies in the modified accredited advanced and TB free areas of the state. Future enhancements to the TB program include the following:

- Mandatory Radio Frequency Identification (RFID) cattle movements statewide (implemented March, 2007);
- Increased tracking of cattle movements from the Modified Accredited Advanced (MAA) zone;
- Increased use of technological/passive tracking systems for RFID identified cattle (market flow):
- Increased surveillance in feedlots in MA zone;
- Risk-targeted Surveillance programs in MAA/TB Free;
 - o Increasing emphasis on compliance programs throughout the state including activities at the Mackinac bridge, mobile surveillance patrols, inventory reconciliation, and heightened presence at livestock markets.
 - Wildlife more tools for deer population control/increased focus on enforcement of baiting bans.
- Programs to eliminate the transmission from wildlife into livestock. Implementation of wildlife Risk mitigation plans for herds in the MA zone and evaluation of the use of indemnity. In wildlife populations, more liberal use of deer control permits and continued investigation and research into the potential of vaccines.

Minnesota

Linda C. Glaser Minnesota Board of Animal Health

Since the 2005 discovery of bovine tuberculosis (TB) in northwestern Minnesota, a total of seven infected beef cattle herds have been identified and subsequently depopulated. TB positive white-tailed deer have been found in close association with five of the infected premises and the Minnesota Department of Natural Resources (DNR) has defined a Core Area in order to identify this area of concern. The DNR is taking a multi-pronged approach to reduce deer populations in this area and eliminate TB infected deer, including: implementing a recreational

feed ban, granting landowner hunting permits, developing a special permit area with increased bag limits, assisting producers in construction of deer-proof fencing for their stored feed, and contracting sharp shooters to collect deer in the Core Area. The Minnesota Board of Animal Health adopted the DNR Core Area boundaries; producers in this area are now required to restrict cattle movement and adopt management practices that reduce the interaction between cattle and deer on their premises. Minnesota has TB tested deer statewide and is in the middle of a statewide campaign to TB test cattle herds. With the Core Area measures in place and with completion of statewide surveillance in cattle, the state will apply for reinstatement of TB-Free status in December 2008.

New Mexico Tim Hanosh New Mexico Livestock Board

New Mexico is a Tuberculosis (TB)-free state with a small Modified Accredited Advance (MAA) zone along the eastern edge of the state. The MAA zone has stringent and specific requirements. All herds within the MAA zone are under strict movement control and are TB tested annually. TB testing within the MAA zone revealed no positive cases in 2007. Mitchell dairy, one of the two herds responsible for creation of the MAA zone, underwent a complete herd test in July 2007 with no positive cases. Schapp dairy, the other TB positive dairy responsible for creation of the MAA zone, depopulated shortly after TB was diagnosed in the herd in 2003. Over 20,000 animals were TB tested within the MAA zone during 2007. The majority of testing was performed by state or federal regulatory personnel.

In February 2007, a cow from Cornerstone Dairy in southeastern New Mexico, outside of the MAA zone, was diagnosed with TB via slaughter surveillance. The dairy was quarantined and underwent a complete herd test. Approximately 5,000 animals were TB tested with no positive animals found. The quarantine was lifted with the agreement that the dairy will undergo a complete herd test in December 2007 and again in December 2008.

In April 2007, a cow from the DoRene/Milagro Dairy was diagnosed with TB via slaughter surveillance. The DoRene/Milagro herd consists of two dairies and one heifer-raising facility, all near, but outside of, the MAA zone. The herd was quarantined and a complete herd TB test led to necropsies which confirmed the diagnosis of TB. With the aid of a federal task force, the regulations set forth in the TB Uniform Methods and Rules (UMR) were followed and all ancillary testing was completed with no other positive TB cases found. Approximately 20,000 animals were tested. The owners of the DoRene/Milagro herd accepted a depopulation plan and, as of this writing, 1,950 cows of the original 12,000 head remain to be depopulated. The depopulation will be completed by December 14, 2007. Epidemiology continues with the final epidemiology report due November 30, 2007. The DoRene/Milagro owner plans to repopulate as soon as possible. The DoRene/Milagro herd will be TB tested according to the UMR at six months and again at one year post-repopulation. It should be noted that the DNA fingerprint from the organism isolated from the DoRene/Milagro herd is not the same as the DNA fingerprint from the organism isolated from Mitchell dairy, Schapp dairy or Cornerstone dairy.

There are seven other dairy herds in New Mexico that have been or will be TB tested in the near future. These herds are being labeled as "high gamma" herds. These herds, although testing negative for TB during their last herd tests in 2004, had one or two animals with suspiciously high gamma interferon tests. In an attempt to be as thorough as possible in the search for TB in New Mexico, these herds were added to the list to be TB tested.

REPORT ON BI-NATIONAL BRUCELLOSIS AND TUBERCULOSIS COMMITTEE ACTIVITIES

Billy Johnson, Coordinator

The U.S.-Mexico Bi-National Brucellosis and Tuberculosis Eradication Committee (BNC) was formed in 1993 based on a recommendation by United States Animal Health Association (USAHA) with responsibility to provide oversight on the eradication programs in each country and to provide recommendations for the minimum requirements for the exportation of cattle from Mexico to the United States. The BNC has sixteen members with representation from the livestock industries, research, and State and Federal officials. It should be pointed out that there is no government funding for the Committee members to attend the meetings. These expenses are paid by the members or their sponsoring organizations. The Committee has met three times during the past year, twice in the US in conjunction with the National Cattlemen Beef Association and at the USAHA Annual Meeting and once in Mexico during the Confederation National Ganadera (CNG) meeting. There will be a meeting on Thursday, October 25th during this USAHA Annual Meeting. These organizations as well as other industry groups have worked cooperatively with the BNC since it's beginning by providing space, financial aid and other assistance. By meeting at these locations, cattlemen and other industry and veterinary officials have the opportunity to participate.

The BNC has no authority to pass or implement regulations or procedural changes. However, it has been involved in providing input and recommendations in all phases of the programs since its formation. The BNC worked closely with Animal and Plant Health Inspection Service (APHIS) officials in developing the present requirements and in developing review procedures to be followed in Mexico. The most critical step in forming the BNC was to bring the livestock industries into the process of program development and implementation.

In 1993 when the BNC first was started there was concern by the U.S. cattle producers that their herds were being exposed to large numbers of tuberculosis infected steers and spayed heifers when they were being put on grass before movement to feedlots. At the time over 500 infected animals were being found at U.S. slaughter establishments each year. United States Department of Agriculture (USDA) implemented regulations to refuse entry of Holstein and cross bred Holstein steers into the U.S. and the Border State Veterinarians developed a procedure called the Consensus Document which required each state in Mexico to be enrolled in an eradication program in order to export cattle to the U.S. This program was coordinated by the BNC until USDA-APHIS could publish new regulations controlling the import of Mexican feeder animals. The goal under these regulations was to work towards equivalency between the eradication programs in the two countries.

Status reports are provided at each meeting on the following issues:

- Slaughter reactor traceback efforts;
- Eradication program progress;
- · Research programs in each country;
- State reviews; and
- Interstate and inter zone movement controls.

As the two countries work towards equivalency in the eradication programs, areas of concern continue to arise because of the differences in normal cattle operating procedures and the disease levels in the two countries. These concerns are brought before the Committee for discussion and recommendations to be presented to Secretaria de Agricultura Ganaderia, Desarrollo Rural, Pesca y Alimentacion (SAGARPA) and APHIS.

During the past year the following issues have been presented and discussed.

- 1. Standardization of tuberculins used in Mexico, the United States and Canada. This is progressing and a Committee with representatives from the three countries.
- Approval of designated feedlots for Modified Accredited Advanced states or zones. Designated feedlots are permitted in Modified Accredited and Accreditation Preparatory states. Industry officials in Modified Accredited Advanced States indicate they are not getting sufficient cattle to meet their needs.

- 3. Request for approval of designated pastures in Modified Accredited and Accreditation Preparatory states to operate under conditions similar to designated feedlots.
- 4. Movement of purebred cattle from Accredited Free herds in non-accredited states to all other states.
- 5. Recognition of Mexico's National Identification System for imported cattle to the U.S. and simplification of import documents.
- 6. Proposal to modify outlines for the authorization of designated feedlot so only electronic identification systems are required.
- 7. System for the movement of rodeo bulls between status and non status states.
- 8. A system for sampling slaughter cattle in slaughter plants that are too small to have full time slaughter inspection.
- 9. Request for APHIS to reconsider their requirements that any new zone must contain at least 1,000 herds.
- 10. Request for a cooperative agreement between Mexico and USDA that would allow SAGARPA to funnel user fees back into their program.

The procedures in place allow time for SAGARPA and the Mexico industries and APHIS and the U.S. industries to meet prior to the full BNC meeting to develop their issues and then time after the BNC meeting for SAGARPA and APHIS officials to meet to discuss actions to be taken on the issues.

Although the disease level in imported animals has decreased as programs have been implemented in more states there still are from 15 to 20 infected Mexican steers being found at slaughter in the U.S. and in Mexican rodeo animals used in the U.S. There is still concern by officials in the U.S. that there are a significant number of states and zones in Mexico that have not implemented full surveillance programs for tuberculosis nor eradication programs. These problems will continue to be areas of discussion in the BNC.

Although the BNC was originally established for tuberculosis procedures, brucellosis was later added to the Committee responsibilities. Although the brucellosis programs in most states in Mexico are not progressing at the same rate as their tuberculosis eradication programs, the state of Sonora has progressed well and is requesting Brucellosis Class A status. Also a U.S.-Mexico Tick Committee meets at the same time as the BNC and provides a summary of their meeting to the BNC since most of the BNC members are also involved with tick eradication programs.