# **SCWDS BRIEFS**

A Ouarterly Newsletter from the Southeastern Cooperative Wildlife Disease Study **College of Veterinary Medicine** 

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#### Tom, Beth, and Al

The wildlife management and animal health professions recently lost three highly valued colleagues and cherished friends. Dr. Tom Thorne, a 36-year veteran of the Wyoming Game and Fish Department, and his wife, Dr. Beth Williams, a professor of pathology at the University of Wyoming, were killed in a tragic automobile accident on December 29, 2004. Dr. Al Kocan, recently retired professor of parasitology from Oklahoma State University, died of an apparent heart attack on December 15, 2004.

Tom went to work for the Wyoming Game and Fish Department (WGFD) in 1968, shortly after receiving his D.V.M. at Oklahoma State University. He supervised wildlife research projects and provided veterinary assistance for wildlife translocation projects throughout Wyoming. Tom subsequently became Chief of the Services Division and was named acting director of WGFD in 2002. Although he retired in 2003, Tom returned to WGFD as a wildlife disease consultant and continued this work until his untimely death.

Beth obtained her D.V.M. from Purdue University and her Ph.D. in veterinary pathology from Colorado State University. After completing her graduate studies, Beth joined the faculty of the University of Wyoming in Laramie, where she taught undergraduate and graduate students, conducted research, and worked as a pathologist at the Wyoming State Veterinary Laboratory. Beth and Tom were married in 1979.

The number and significance of the contributions that Tom and Beth made to wildlife conservation and veterinary medicine cannot be overstated, and it is impossible to list all of them here. Although perhaps best known for their work on brucellosis, black-footed ferrets, and chronic wasting disease. Tom and Beth advanced the understanding and management of many diseases in free-ranging wildlife populations. Tom and Beth helped educate current and generations of wildlife health past professionals and they provided much-needed wildlife health expertise to the International Association of Fish and Wildlife Agencies, National Academy of Sciences, U.S. Animal Health Association, The Wildlife Society, and other organizations. They especially promoted the wildlife health profession through their tireless involvement in the Wildlife Disease Association (WDA). Their contributions were recognized in 1996 when they shared the WDA's Distinguished Service Award. Although highly accomplished, Tom and Beth were modest scientists who were more concerned about the success of wildlife research and management projects than who received credit for the work. Tom and Beth enjoyed life and loved to share good meals, wine, and fun with friends.

Al retired in August 2004 after a 30-year career as professor of parasitology in the Department of Pathobiology, College of Veterinary Medicine, Oklahoma State University (OSU). He obtained a B.S. from Hiram College in 1968 and received an M.P.H. in 1969, and a Ph.D. in 1973 from the University of North Carolina at Chapel Hill. From the time of his arrival at OSU in 1974, Al focused most of his research efforts on wildlife health related issues, especially diseases of white-tailed deer and tick-vectored diseases of wildlife, domestic animals. and humans. He was an active member of the Wildlife Disease Association and provided service to the WDA in many

capacities, including serving as Treasurer and on several committees. He was also an active member of several professional societies, especially in the parasitology and vector-borne diseases areas. His career included not only work in the United States, but also in Africa and the Caribbean, which led to significant recognition by the international wildlife, domestic animal, and human health communities. For example, was visiting scientist at the he Onderstepoort Veterinary Institute, South Africa, visiting lecturer at Friday Harbor Marine Laboratories at the University of Washington, and also served as a senior research fellow at the Windward Islands Research and Education Foundation. Granada, West Indies. His distinguished career included many honors and awards, such as the Family Alumni Achievement Award from Hiram College, the Beecham Award for Research Excellence from OSU, and the MSD AgVet Award for Creativity. Al was an avid outdoorsman and enjoyed both recreation and science with his many friends.

Tom. Beth. and Al were major forces in the recent publication of revised editions of two books. Infectious Diseases of Wild Mammals and Parasitic Diseases of Wild Mammals, both of which are indispensable references for the wildlife health profession. These books and numerous other works by Tom. Beth. and Al will ensure that their knowledge and efforts will continue to be available for future scientists, although these are hollow substitutes compared to the opportunity for personal interactions with these three individuals. Early in their careers, Tom, Beth, and AI established collaborations with SCWDS that continued for the rest of their lives, and, like their many other colleagues around the world, all of us at SCWDS deeply miss our wonderful friends. (Prepared by John Fischer and Randy Davidson)

## SCWDS Studies on Johne's Disease in Wildlife

Johne's disease is a chronic, progressive disease of the gastrointestinal tract and

lymphoid tissue that is caused by the bacterium Mycobacterium avium subspecies paratuberculosis (Map). Johne's disease is known primarily as a disease of domestic ruminants, especially cattle, but infections also have been found in wildlife. Mycobacterium avium subspecies paratuberculosis has been reported in isolated cases in wild ruminants in the United States, including white-tailed deer, elk, and bighorn sheep. This organism also has been isolated from non-ruminant wildlife in Scotland, including rabbits, foxes, stoats, weasels, badgers, wood mice, Norway rats, European brown hares, jackdaws, rooks, and crows.

For the most effective control of Johne's disease in livestock it is important to determine if wildlife species (both ruminant and non-ruminant) may serve as reservoirs. Surveys to determine if *Map* occurs in free-ranging mammals and birds on livestock premises in the United States recently were completed by SCWDS in collaboration with the Johne's Testing Center, University of Wisconsin. The project was funded by USDA-APHIS-Veterinary Services.

Specimens were collected on both dairy and beef cattle farms in Georgia and Wisconsin with Map prevalence in livestock on the premises that ranged from 0 to 18%. All specimens collected were from free-ranging animals captured on the selected premises and in the case of the dairy farms, mostly in the immediate vicinity of the dairy barns. Wildlife species sampled represented those species with the highest potential for exposure to contaminated materials and those species that posed the highest risk for contamination of livestock feed or forage. Specimens were collected from almost 800 animals representing 25 mammalian and 22 avian species. Samples of liver, intestine, intestinal lymph nodes, and feces were harvested and cultured for Map.

*Mycobacterium avium* subspecies *paratuberculosis* was found in 36 samples collected from 26 free-ranging animals (genetic analysis is still underway for a few mycobacterial isolates). *Map* was found in wild animals on 6/7 (85.7%) farms where infected livestock were present but not on either of the 2 farms with test-negative livestock. Culture-positive specimens came from 11 wild and feral species: nine-banded armadillo, common snipe, Eastern cottontail, European starling, feral cat, hispid cotton rat, house sparrow, northern short-tail shrew, opossum, raccoon, and striped skunk. *Map* was detected in fecal specimens from 5 animals (3 species); tissue isolates were also obtained from 3 of these animals.

The detection of *Map* in a wide range of wildlife species in Wisconsin and Georgia is the first report of *Map* in non-ruminant wildlife in North America but is consistent with recent survey results in Scotland. Individuals of some species of wildlife may live for several years and can have home ranges that cover areas large enough to include more than one farm. Wildlife could shed *Map* over a period of time and potentially on the farm where the organism was acquired, as well as on nearby farms, depending on movement patterns of the individual animals.

Wildlife may not be a significant contributing factor in the maintenance of *Map* on farms with infected livestock herds. Given the enormous premise contamination that occurs through the large volume of contaminated fecal material produced by infected farm animals, the contributions of local wildlife are not likely to be epidemiologically significant. However. wildlife might be an epidemiologically significant factor for Map control on farms that have eliminated all Map from livestock on the premises and on Map-free farms in the same geographic area as farms with infected livestock. Transmission among wildlife species, independent of continual contact with infected domestic livestock, might result in the establishment of carriers or local wildlife reservoirs. If wildlife contamination were to occur in a sufficient volume and on a site shared by susceptible livestock (calves), transmission of Map from wildlife back to livestock could interfere with Johne's disease control programs. Studies

on the pathogenesis and shedding of *Map* in wildlife are planned to assess the risk that *Map*-infected wildlife pose to livestock. (Prepared by Joe Corn and Becky Manning)

#### WNV Antibody Persistence in Pigeons

While much of the West Nile virus (WNV) surveillance in North America has concentrated on dead bird testing, serologic testing of live wild birds is also a useful tool for investigating WNV epidemiology. However, the duration of the antibody response, laboratory test performance, and the persistence of maternal antibodies can complicate the interpretation of serologic results. Because little information is available regarding the antibody dynamics during infection with the North American strain of WNV. SCWDS conducted a study to determine the long-term persistence of WNV antibodies in naturally infected rock pigeons (Columba livia), to compare the long-term utility of commonly used WNV serologic techniques, and to determine the persistence of maternal antibodies to WNV in squabs derived from these naturally infected birds. This study was funded by the Georgia Department of Human Resources through the Centers for Disease Control and Prevention.

Thirty pigeons, 20 seropositive for WNV and 10 seronegative controls, were captured in April 2003 in Atlanta, Georgia, and housed in a mosquito-free environment for 60 weeks. Blood samples were taken every 3 weeks and tested serologically with plaque reduction neutralization tests (PRNT), epitope-blocking ELISA, and the hemagglutination inhibition (HAI) test. Five squabs hatched from seropositive birds also were tested by PRNT for 6 weeks.

The 20 birds that had antibodies to WNV at the time of capture remained antibody-positive during the 60-week study period; the 10 control birds that had no detectable WNV antibody remained antibody-negative. PRNT titers for 16 of these birds did not vary by more than a two-fold dilution throughout the 60-week testing period, and the titers of the four remaining pigeons varied only four-fold (two dilutions). HAI results were inconsistent with

PRNT results, but good agreement was observed between ELISA and PRNT results. Neutralizing maternal antibodies to WNV in the squabs lasted for an average of 27 days.

The pigeons used in this study were naturally infected field-collected birds. The dates of WNV infection therefore were unknown, and an absolute estimate of antibody persistence could not be determined. This study has shown, however, that the minimum duration of antibody persistence to WNV in rock pigeons is 15 months, there is little longterm variation in antibody titers, and there is no serological evidence of viral recrudescence. Based on these findings, the population immunity to WNV can be expected to increase as WNV establishes itself in North America.

The persistence of antibodies to WNV in an avian species for over a year complicates interpretation of multi-year studies involving serologic surveillance of wild bird populations. Because the antibody titers in this study remained at high levels, it suggests that pigeons maintain neutralizing antibody titers to WNV for several years. Seroprevalence of WNV antibodies in longlived avian species therefore may increase while transmission of the virus in an area remains stable over time. Because variation in the persistence of antibodies to WNV may exist among species, antibody persistence in other avian species should be evaluated.

The results of this study proved to be highly test-dependent, and serologic results should be interpreted with this in mind. The HAI test was not as effective as the PRNT or ELISA in our study. However, neutralizing antibodies detected by PRNT and ELISA generally are considered to persist longer than HAI antibodies, so the results of this study may reflect differences in the timing of infection in individual birds. Those pigeons positive by HAI in this study potentially represent more recent infections. To our knowledge, this is the first report detailing the duration of avian maternal antibodies to the North American strain of WNV. Columbiformes are unique because in addition to the maternal antibodies transferred through the egg yolk, they receive both maternal and paternal antibodies through crop milk after hatching. The role of nestlings in WNV amplification cycles may be reduced by maternal antibody persistence. In the case of pigeons, the additional opportunity for transfer of passive immunity from not only the hen but also the cock increases the proportion of squabs with resistance to WNV infection. How maternal antibody persistence in pigeons compares to indigenous North American avian species is unknown. When determined, this information will help elucidate variations in disease resistance among avian WNV populations. (Prepared by S.E.J. Gibbs)

#### **Feral Swine Distribution Maps**

Feral swine can serve as reservoirs for pseudorabies virus (PRV) and *Brucella suis*, and these pathogens have been detected in feral swine populations throughout much of their range in the United States. When the current USDA-APHIS PRV and *Brucella* eradication programs among domestic swine are successfully completed, feral swine will persist as a potential source for disease reintroduction to domestic herds.

SCWDS has been working with USDA-APHIS–Veterinary Services to develop a rapid and cost-efficient strategy to evaluate this risk. Specifically we are developing site-specific information as to the status of PRV and *B. suis* in feral swine in areas of significant domestic swine production. As part of this effort, SCWDS has developed geographic information system (GIS)-based maps that detail the distribution of feral swine populations in the United States.

Two maps of the nationwide distribution of feral swine have been completed. One map depicts the actual distribution of feral swine and the second identifies feral swine distribution at the county level. These maps were produced using data provided primarily by the respective state and territorial natural resources agencies but also with some data from agriculture agencies and universities. Providing current density information was not an objective because reliable population density estimates do not exist on this scale, and densities can vary dramatically from year to year, as well as between seasons and specific habitat types.

Feral swine distribution has increased dramatically in the past 16 years. This is easily seen when comparing the 2004 map to the feral swine distribution map developed by SCWDS in 1988 (see figures on next page). More detailed versions of can be these maps viewed at www.scwds.org. The number of counties reporting established populations increased from 462 in 1988 to 1,042 in 2004, a 225% increase. This increase in feral swine distribution can be attributed to humanassisted and natural movement of the animals. In addition to the disease risks posed by feral swine, the expanding range of these animals has a significant impact on natural resources and agricultural crops.

The 2004 distribution maps currently are being used to prioritize surveillance for PRV and *B. suis* in feral swine. Together, these maps and accompanying surveillance data will enhance understanding of the epidemiology of these diseases in feral swine and the significance of infected feral swine to the domestic swine industry in the United States. In addition, these maps will be useful in conducting risk assessments regarding the potential role of feral swine in the epidemiology of emerging or foreign animal diseases and in developing response plans for emergency animal disease outbreaks that involve feral and domestic swine. (Prepared by Jay Cumbee and Brian Chandler)

#### Wildlife Piroplasms

A new study funded by the University of Georgia Research Foundation has been initiated at SCWDS to investigate piroplasm infections in southeastern wildlife. Piroplasm infections are caused by intraerythrocytic protozoans in the genera Babesia, Theileria, and Cytauxzoon. They are among the most common infections of free-living animals worldwide, and many are important pathogens of domestic animals and humans. In recent years, an increasing number of piroplasms have been documented in hosts other than their presumed primary host, often with fatal results. Despite their importance, the natural history of piroplasms in the United States is not completely known. Characterizing and delineating the host-range and distribution of these piroplasms is the first step to understanding the natural history and diversity of these parasites, developing better diagnostic assays, assessing the risk of piroplasmosis in domestic and wild animals and humans, and targeting control efforts, such as precautions against translocation of vertebrate hosts or tick vectors.

Historically, babesiosis in humans was caused by either *B. microti* in the United States or *B. divergens* in Europe. The manifestations of babesiosis in humans range from subclinical infection to severe disease resulting in death, with a case fatality rate of 5% for *B. microti* and 35% for *B. divergens*. In recent years, at least three additional piroplasms in the United States and one in Europe have emerged as new zoonotic agents.

Babesia microti, the primary cause of human babesiosis in the United States, is maintained in nature by several species of rodents as reservoir hosts and the tick *Ixodes scapularis* as the vector. Most human cases are reported in the Midwest and Northeast, where *I. scapularis* is most common. Babesia microti also has important considerations for veterinary health because fatal infections have been diagnosed in domestic dogs and an otter in a zoological park.

In addition to *B. microti*, three undescribed species of piroplasms have been recognized as human pathogens in the United States. In the western United States, two piroplasm species (WA1- and CA1-types) emerged, causing dozens of human cases. Based on molecular data, mule deer and bighorn sheep are suspected reservoirs of the WA1-type, but to date no tick vector has been identified. The CA1-type has only been reported from humans and a domestic dog.





Two cases of babesiosis in Missouri and Kentucky were caused by a Babesia species (MO1-type) determined to be closely related or identical to Babesia divergens, a bovine parasite in Europe. Recently, a case of babesiosis in Washington was caused by this species (MO1-type) instead of the more common WA1- or CA1-types found in the western United States. This B. divergens-like species (MO1-type) is suspected to be maintained in rabbits, and Ixodes species are suspected vectors. Additional human babesiosis cases have been reported from Georgia, Maryland, Texas, and Virginia, but the Babesia species associated with these infections were not identified.

Multiple Babesia species have been detected in medium-sized mammals, including B. lotori in raccoons, B. lotori in skunks, and a B. divergens-like species (MO1-type) in cottontail rabbits. Recently, other researchers molecularly characterized Babesia species from raccoons, skunks, and a fox from Massachusetts using partial 18S rRNA and beta-tubulin gene sequences. These three species were shown to be related to, but distinct from. *B. microti*. Another aim of our new study is to determine the prevalence and identity of Babesia in medium-sized mammals in the Southeast. To date, we have detected Babesia in striped skunks, raccoons, a gray fox, and a black bear.

Two piroplasms of wildlife that have important animal health implications are B. odocoilei and Cvtauxzoon felis. Although usually subclinical in white-tailed deer, B. odocoilei causes acute, often fatal, babesiosis in captive elk, reindeer, and caribou. Infected wild white-tailed deer have been reported from Massachusetts, Oklahoma, Texas, and Virginia. Although the vector, *I. scapularis*, is established throughout the Southeast, B. odocoilei has only been reported in Texas and Virginia. Previously we have found a high prevalence of piroplasms in blood smears from white-tailed deer throughout the Southeast, but because of morphologic similarities it is unknown if these represent infections with B. odocoilei or Theileria cervi, another common piroplasm of deer. Because the presence of B. odocoilei may pose a health threat to cervids in zoological parks or to efforts to restore elk populations in Arkansas, Kentucky, and Tennessee, the presence and distribution of this pathogen need to be investigated. SCWDS is developing a polymerase chain reaction (PCR) assay that differentiates *B. odocoilei* and *T. cervi* to test white-tailed deer to determine the distribution of *B. odocoilei* in the Southeast.

a parasite commonly Cytauxzoon felis, detected in bobcats and panthers, often causes severe disease in domestic cats, but the natural history of this pathogen is poorly Dermacentor variabilis is the understood. suspected vector, and bobcats and panthers serve as wildlife reservoirs. SCWDS has developed a PCR assay to detect C. felis and is using this assay to determine the prevalence of this parasite in wild felids (bobcats, In addition, we are amplifying panthers). multiple C. felis gene targets from wildlife samples to compare these parasites with those detected in domestic cats. Thus far, the C. felis from wild felids is indistinguishable from C. felis detected in domestic cats (fatal and nonfatal infections). As this study was recently initiated, testing of samples from mammals will continue to delineate the host relationship and distribution of piroplasms in the southeastern United States. (Prepared by Michael Yabsley)

### Deer Hunter Contracts Bovine TB

The appearance of bovine tuberculosis (TB) in Michigan's white-tailed deer herd in 1994 had obvious implications for the cattle industry and resulted in revocation of Michigan's TB-free status in 2000. In addition to the concerns of many for the welfare of the state's animal industry and deer herds, public health-care workers warned of the potential for people to contract the infection from diseased wildlife or livestock. During the past deer hunting season these concerns were realized when a hunter acquired *Mycobacterium bovis* from an infected deer.

The deer involved in the incident was killed in Alcona County, inside the tuberculosis endemic region of northeastern Michigan. The hunter cut his hand while field-dressing the deer and was not wearing gloves as recommended. At the time, he noticed lesions consistent with TB in the deer's chest. He later sought medical attention and subsequently was diagnosed with cutaneous tuberculosis. Health officials cultured *M. bovis* from the patient and confirmed that the isolate is the same strain as the one found in cattle, deer, and other wildlife species in the state. Physicians expect him to recover fully with the appropriate antibiotic therapy.

The current case in Michigan is only the second human case of tuberculosis attributed to the *M. bovis* strain found in deer and cattle in northeastern Michigan. The first case was found incidentally in an elderly man who died of unrelated causes, and the source of his exposure to *M. bovis* was not definitely identified. A small number of other Michigan residents have been diagnosed with bovine TB, but none of the isolates cultured were related to the strain currently found in Michigan wildlife and cattle. Most of those patients were born outside the United States and likely were infected before immigrating. At least two patients with bovine-TB were elderly Michigan natives who could have contracted the infection at a time when milk was not commonly pasteurized and *M. bovis* was present throughout the state's cattle herds.

Humans may contract M. bovis by aerosol inhalation, ingestion of contaminated food, or through wounds. Historically, people often acquired the infection by consuming unpasteurized milk or other dairy products contaminated with the bacterium. Cutaneous transmission, in contrast, always has been an uncommon route of exposure. Nonetheless, hunters are cautioned to wear gloves when dressing deer in areas with bovine TB, and if a carcass appears diseased in any way they should contact the state wildlife agency and arrange for diagnostic testing of the animal. Complete recommendations for the safe handling of wild game can be found on the website of the Michigan Department of Natural Resources at www.michigan.gov/dnr. (Prepared by Kevin Keel)

#### Human Recovers from Rabies

Rabies usually is fatal once clinical disease appears. Only five humans are known to have survived infection and all of them received post-exposure treatment. In October 2004, a previously healthy 15-year-old girl from Fond du Lac County, Wisconsin, became the first human documented to recover from clinical rabies who had not had pre- or post-exposure prophylaxis for rabies. The girl reportedly was bitten by a bat she was handling approximately 1 month prior to developing clinical disease. Medical attention was not sought and postexposure prophylaxis (PEP) was not administered after the bite. When clinical symptoms of fatigue, nausea, vomiting, and incoordination began, she was examined by a pediatrician and referred to a neurologist, and she was admitted to the local hospital on the following day. After learning of the bat bite, medical personnel assayed and detected rabies-specific antibody in the patient's serum and cerebral spinal fluid (CSF). Rabies RNA was not detected by polymerase chain reaction (PCR), and efforts to isolate the virus were unsuccessful, thus genetic typing of the virus was not possible. However, the history of a bat bite in this girl suggests that the bat variant of rabies virus was the cause of the morbidity. For more information about rabies and bats see SCWDS BRIEFS Vol. 20, No. 3.

Supportive care and neuroprotective measures, including a drug-induced coma and ventilator support, were administered to the patient. She remained comatose for 7 days, at which time a significant increase in CSF antirabies antibodies was detected. The coma medications gradually were attenuated, and after 33 days she was admitted to a rehabilitation unit. By mid-December the patient was able to talk, walk with assistance, and feed herself soft foods. Currently, her chances for a full recovery are not known.

Although the best prevention against rabies is avoidance of potentially infected animals, rabies in humans is preventable with the proper medical attention following exposure to an infected animal. According to the Centers for Disease Control and Prevention (CDC), persons bitten or scratched by a potentially rabid animal should immediately wash the wound thoroughly with soap and water, capture the animal (if this can be done safely by avoiding direct contact), contact local or state public health officials, and visit a physician for treatment and evaluation regarding the need for PEP.

There is no proven treatment for clinical rabies, and although this patient recovered from rabies, the reasons for recovery in this unusual case are unknown. Clinicians and the public should recognize the risk of contracting rabies by direct contact with bats or wild carnivores and should not regard it as a treatable disease based on the outcome of this case. A complete report of this case can be found in the December 24, 2004, issue of CDC's Weekly Morbidity and Mortality Report at www.cdc.gov. (Prepared by Rick Gerhold)

Information presented in this Newsletter is not intended for citation as scientific literature. Please contact the Southeastern Cooperative Wildlife Disease Study if citable information is needed.

Information on SCWDS and recent back issues of SCWDS BRIEFS can be accessed on the internet at WWW.SCWDS.org. The BRIEFS are posted on the web cite at least 10 days before copies are available via snail mail. If you prefer to read the BRIEFS on line, just send an email to gdoster@vet.uga.edu, and you will be informed each quarter when the latest issue is available.