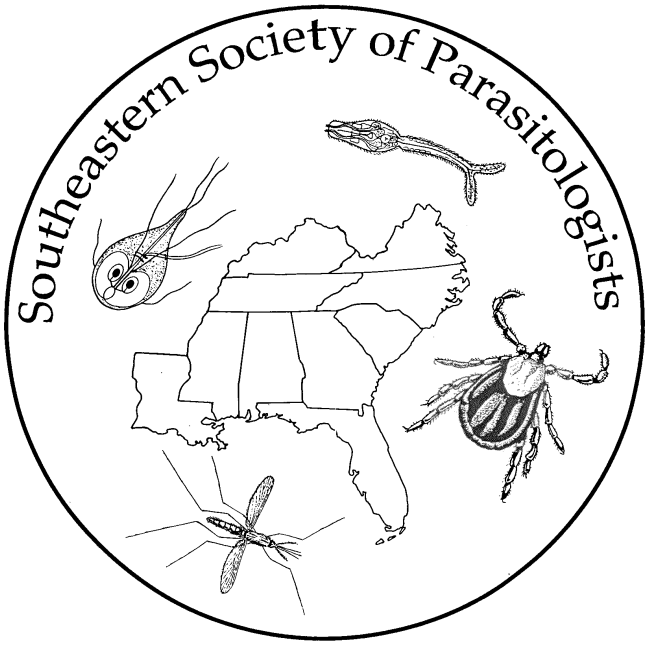
SOUTHEASTERN SOCIETY OF PARASITOLOGISTS

(Affiliate of The American Society of Parasitologists)

PROGRAM AND ABSTRACTS



March 29-31, 2006

Hosted by: University of Tennessee College of Veterinary Medicine,
Glenstone Inn and Gatlinburg Convention Center
Gatlinburg, Tennessee

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Southeastern Society of Parasitologists
2006 Program Summary

SSP Presidential Symposium
Wednesday, 29 March 2006, 5:30 – 7:15 p.m.
Location –Gatlinburg A

Vector Borne Diseases

Presiding: Sharon Patton (for Jennifer A. Spencer), Department of Comparative Medicine, University of Tennessee College of Veterinary Medicine, Knoxville Tennessee.

1. **5:30 p.m. Dr Jennifer A. Spencer**. College of Veterinary Medicine, Auburn University, AL

   Vector-borne diseases: Ticks, bugs, and pathogens of importance to human and animal health.

2. **5:45 p.m. Dr Mario J. Grijalva,** Tropical Disease Institute, Biomedical Sciences Department, College of Osteopathic, Ohio University and Infectious Disease Research Laboratory, School of Biological Sciences, Catholic University of Ecuador.

   Chagas disease in Ecuador: Moving from research to control program implementation.

3. **6:15 p.m. Dr Ed Breitschwerdt**, College of Veterinary Medicine, North Carolina State University, Raleigh, NC

   Concurrent infection with Anaplasma, Bartonella and Ehrlichia species.

4. **6:45 p.m. Dr Byron L. Blagburn,** College of Veterinary Medicine, Auburn University, AL

   Lean, Mean, Transmission Machines: The Biology of Ixodid Ticks.

Thursday, 30 March 2006
Southeastern Society of Parasitologists I
Location –Gatlinburg A

Presiding: Charles Faulkner, University of Tennessee
† Byrd-Dunn Student Presentation

8:15 **Presentation Loading**

8:30 † 5. **YOUNG, JOY AND GEORGE W. BENZ**. Middle Tennessee State University. Rapid colonization of neonate lemon sharks by monogeneans.

8:45 † 6. **MCELWAIN, ANDREW AND GEORGE W. BENZ.** Middle Tennessee State University. Reconsidering phylogeny within Sphyriidae (Siphonostomatoida, Copepoda).

9:00 † 7. **COOK, JOSHUA O[1], ROBIN M. OVERSTREET[1] AND R. RAMA KRISHNA[2].** The University of Southern Mississippi, Gulf Coast Research Laboratory, University of Southern Mississippi, Ocean Springs, MS[1] and ANGR Agricultural University, Andhra Pradesh, India[2]. Pathology associated with fatal myxozoan infections in farmed carp, *Catla catla*, from India.

9:15 † 8. **BAKER, TIFFANY G.[1] AND ISAURE DE BURON[2]**. Grice Marine Laboratory[1] and Biology Department[2], College of Charleston, SC. Stock identification of the Atlantic croaker, *Micropogonias undulatus* Linnaeus, using macroparasites as biological tags

9:30 † 9. **LUCAS, A[1]., W. S. SWECKER[1], G. SCAGLIA[2], D. S. LINDSAY[1], F. C. ELVINGER[1], J. P. FONTENOT[2], AND A. M. ZAJAC[1]**. VA/MD Regional College Of Veterinary Medicine[1] and Dept. Of Animal And Poultry Sciences[2], Virginia Tech. Population dynamics of *Eimeria* spp in grazing beef calves in Virginia.

9:45 † 10. **GOODWIN, DAVID G., SOLANGE M. GENNARI[2], DANIEL K. HOWE[3], J.P. DUBEY[4], ANNE M. ZAJAC[1], AND DAVID S. LINDSAY[1]**. Virginia Tech[1], Universidade de São Paulo[2], University of Kentucky[3], USDA Animal Parasitic Diseases Laboratory[4]. Prevalence of antibodies to *Encephalitozoon cuniculi* in Brazilian horses

10:00 **Break and Presentation Loading**

10:15 † 11. **LASCANO, MAURICIO S.[1,3], ANITA VILLACIS[2], AND MARIO J. GRIJALVA**[3]. Department of Biological Sciences, Ohio University, Athens, OH[1]. School of Biological Sciences, Catholic University, Quito, Ecuador[2]. Tropical Disease Institute, Biomedical Sciences Department, College of Osteopathic Medicine, Ohio University, Athens, OH[3]. Molecular characterization of *Trypanosoma rangeli* isolates from Ecuador.

10:30 † 12. **PINTO, C. MIGUEL[1,2], SOFÍA OCAÑA[1], MAURICIO LASCANO[3], AND MARIO J. GRIJALVA[3]**. Laboratorio de Investigación en Enfermedades infecciosas, Escuela de Ciencias Biológicas, Pontificia Universidad Católica del Ecuador, Quito, Ecuador[1]; Present address: Department of Biological Sciences, Texas Tech University, Lubbock, TX, USA[2]; Tropical Disease Institute, College of Osteopathic Medicine, Department of Biomedical Sciences, Ohio University, Athens OH, USA[3]. Infection by trypanosomes in marsupials and rodents associated to human dwellings in Ecuador

10:45 † 13. **WIMSETT, ASHLEY, AMY ANDERSON, AND CHRIS HALL**. Department of Biology, Berry College, Mount Berry Ga. Humoral recognition of *Trypanosoma cruz*i antigens in raccoons from the Berry College campus.

11:00 † 14. **PIERCE, EMILY, BRAD MEERS, AND CHRIS HALL.** Department of Biology, Berry College, Mount Berry Ga. Vertical Transmission of North American Type II and a South American Type I isolates of *Trypanosoma cruzi* in BALB/c mice.

11:15 † 15. **VINCENT, AMANDA G. AND JEFFREY M. LOTZ.** Gulf Coast Research Laboratory, The University of Southern Mississippi. Evolution of virulence: transmission from dead hosts.

11:30 † 16. **HARRIS, SHAWNA AND CLAIRE A. FULLER.** Murray State University. Immunity in dragonfly naiads (Odonata: Anisoptera): indicators of water quality.

11:45 † 17. **SAVAGE, MASON Y. AND MICHAEL J. YABSLEY.** Southeastern Cooperative Wildlife Disease Study, Department of Population Health, College of Veterinary Medicine, University of Georgia. Sequence polymorphisms in the mismatch-repair (TcMSH2) and glutathione-S-transferase (Tc52) genes of *Trypanosoma cruzi* isolates from United States wildlife.

12:00 – 1:30 p.m. Lunch Break and Presentation Loading

Thursday, 30 March 2006
Southeastern Society of Parasitologists II
Location –Gatlinburg A

Presiding: Michael Yabsley, University of Georgia
† Byrd-Dunn Student Presentation

1:15 **Presentation Loading**

1:30 † 18. MURPHY, STACI M.[1], MICHAEL J. YABSLEY[1], M. PAGE LUTTRELL[1], DAVID E. STALLKNECHT[1], AND SUSAN E. LITTLE[2]. Southeastern Cooperative Wildlife Disease Study, Department of Population Health, College of Veterinary Medicine, University of Georgia[1] and Department of Pathobiology, College of Veterinary Medicine, Center for Veterinary Health Sciences, Oklahoma State University[2.] Experimental inoculation of raccoons (*Procyon lotor*) with *Anaplasma phagocytophilum*, *Ehrlichia chaffeensis*, *Ehrlichia canis*, *Ehrlichia ewingii*, and *Borrelia lonestari*.

1:45 † 19. ESLICK, RENÉ M. and VINCENT A. CONNORS. University of South Carolina Upstate. Production of the reactive oxygen species, superoxide, by cells from the *Biomphalaria glabrata* (Pulmonata) embryonic cell line.

2:00 † 20. MINTER, JENNIFER AND EDWIN ROWLAND. Ohio University. Fate of intracellular *Trypanosoma cruzi* inhibited from egressing the host cell.

2:15 † 21. HARTMAN, ANGELA[1], ROBERT C. WILLIAMS[1], ALEXA C. ROSYPAL[2] AND DAVID S. LINDSAY[1]. Virginia Tech[1] and University of North Carolina Chapel Hill[2]. Efficacy of the FTA-filter based method to detect DNA of *Cryptosporidium parvum* and *Toxoplasma gondii* from oocysts.

2:30 † 22. MITCHELL, SHEILA M., ANNE M. ZAJAC, AND DAVID S. LINDSAY. Virginia Tech. Development of *Cystoisospora canis* in cell culture: Evidence for unizoic cyst formation in vitro.

2 :45 † 23 CASSELL, MEREDITH[1], JEANNINE S. STROBL[2], CHRIS REILLY[2], AND DAVID S. LINDSAY[1]. Virginia Tech[1] and Virginia College of Osteopathic Medicine[2]. Efficacy of histone deacetylase inhibitors against *Toxoplasma gondii*

3 :00 † 24 GAJI, RAJSHEKHAR Y. AND DANIEL K. HOWE. Department of Veterinary Science, University OF Kentucky, Lexington, KY. GAGACGC is a critical cis-acting element required for SnSAG1 gene expression in *Sarcocystis neurona*.

3:15 <u>Break and Presentation Loading</u>

3:30 † 25. STOCKDALE, HEATHER D.[1], G. SHANE WEST[2], TED HANKES[3], KENNETH L. MCMILLAN[4], MARK WHITLEY[5], CHRISTINE C. DYKSTRA[1], JENNIFER A. SPENCER[1] AND BYRON L. BLAGBURN[1] Auburn University College of Veterinary Medicine[1], Vestavia Animal Clinic, Birmingham, AL[2], Alford Animal Veterinary Hospital, Birmingham, AL[3], Pell City Animal Hospital, Cropwell, AL[4], and Cobb Animal Clinic, Greensboro, NC[5]. *Tritrichomonas foetus* induced large-bowel diarrhea is an emerging disease in domestic cats.

3:45 † 26. CARREÑO, ABIGAIL D.[1], A. RICK ALLEMAN[2], ANTHONY F. BARBET[2], GUY H. PALMER[3], SUSAN M. NOH[3] AND CALVIN M. JOHNSON[1]. Department of Pathobiology, Auburn University[1]; Department of Pathobiology, University of Florida[2]; and Department of Veterinary Microbiology and Pathology, Washington State University[3]. *In vivo* endothelial cell infection by *Anaplasma marginale.*

4:00 27. ROSYPAL, ALEXA C.[1,2], DWIGHT D. BOWMAN[3], DANIEL HOLLIMAN[1], GEORGE J. FLICK[1], AND DAVID S. LINDSAY[1]. Virginia Tech[1], University of North Carolina at Chapel Hill[2], Cornell University[3]. Effects of high hydrostatic pressure processing on *Ascaris suum* eggs.

Friday, 31 March 2006
Southeastern Society of Parasitologists III
Location – Gatlinburg A

Presiding: Heather Stockdale and Abigail Carreno, Auburn University

8:15 Presentation Loading

8:30 28. DOFFITT, CYNTHIA M.[1], LINDA M. POTE[1], AND D. TOMMY KING[2]. Mississippi State University[1] and USDA/APHIS/WS[2]. Morphological comparison and identification of cercariae released by the rams-horn snail *Planorbella trivolvis.*

8:45 29. GRINSTEAD, C. BRAD[1], OSCAR J. PUNG[1] AND KRAIG KERSTEN[2]. Georgia Southern University[1] and Armstrong Atlantic State University[2]. Distribution of hydrobiid snails and their parasites in salt marsh along the Skidaway River in coastal Georgia.

9:00 30. BURON, ISAURE DE[1] AND WILLIAM A. ROUMILLAT[2]. Biology Department, College of Charleston, SC[1] and Inshore Fisheries Section, Marine Resources Research Institute, South Carolina Department of Natural Resources[2]. Histopathology of philometrid nematodes in the southern flounder *Paralichthys lethostigma.*

9:15 31. FULLER, CLAIRE A. Murray State University. The relationship between the abiotic environment and the immune function of the Caribbean termite, *Nasutitermes acajutlae.*

9:30 32. STIEVE, ERICA[1], KIMBERLEE BECKMEN[2], AND SHARON PATTON.[1] University of Tennessee College of Veterinary Medicine[1] and Alaska Department of Fish and Game[2]. Seroprevalence of *Neospora spp.* and *Toxoplasma gondii* in caribou, moose, wolf, and fox populations in Alaska.

9:45. **33.** **AHN, JUN, JOSHUA ELLIS, DAAIYAH COOPER, HALEY JACKSON, DANA DARMOHRAY, AND ALAN F. SMITH**. Department of Biology, Mercer University, Macon, GA. Detection of the causative agents of Lyme disease and ehrlichiosis in individual southern black-legged ticks collected from white-tailed deer of the Piedmont National Wildlife Refuge.

10:00 **Break and Presentation Loading**

10:15. **34.** **YABSLEY, MICHAEL J.[1], THIERRY M. WORK[2], AND ROBERT A. RAMEYER[2].** Southeastern Cooperative Wildlife Disease Study, Department of Population Health, College of Veterinary Medicine, University of Georgia, Athens, Georgia[1] and U.S. Geological Survey, National Wildlife Health Center, Honolulu Field Station, Honolulu Hawaii[2]. Molecular phylogeny of *Babesia poelea* from brown boobies (*Sula leucogaster*) from Johnston Atoll, Central Pacific.

10:30. **35.** **PALMIERI, JAMES R., Ph.D[1], KATHERINE BARTER, BS, MT[2], MELISSA HRICKO, BS[3] AND MARIE PALLOTT[4]**. Department of Microbiology, Division of Biomedical Sciences, Virginia College of Osteopathic Medicine[1], Virginia Polytechnic Institute Department of Biochemistry[2,4] and Center for Molecular Medicine and Infectious Diseases Virginia Maryland Regional College of Veterinary Medicine[1,2,4] and Virginia Commonwealth University, Department of Clinical Laboratory Science, Richmond VA[3]. Low temperature induction of *Acanthamoebae* trophozoites from dormant cysts influences their ability to phagocytize bacteria.

10:45. **36.** **STONE, SHARON[1], VINA DIDERRICH-FAULKNER[2], PAUL E. SUPER[3] AND, CHARLES T. FAULKNER[1].** University of Tennessee College of Veterinary Medicine, Knoxville TN[1], Lincoln Memorial University, Harrogate TN[2], and Great Smoky Mountains National Park, Lake Junaluska, NC[3]. Endoparasitic Infections in Migratory Birds from the 2003-2005 Banding Seasons at the Purchase Knob, Great Smoky Mountains National Park, Lake Junaluska, NC.

11:00. **37.** **DIDERRICH-FAULKNER, VINA[1], LEAH ALLEN[1], CHARLES T. FAULKNER[2], AND PAUL E. SUPER[3].** Lincoln Memorial University, Harrogate TN[1], and University of Tennessee College of Veterinary Medicine, Knoxville TN[2], and Great Smoky Mountains National Park, Lake Junaluska, NC[3] Patterns of positivity: Endoparasitic infection and host behaviors and in migratory birds from the 2005 banding seasons at the Purchase Knob, Great Smoky Mountains National Park, Lake Junaluska, NC.

2. SPENCER, JENNIFER A. College of Veterinary Medicine, Auburn University, AL.--Vector-borne diseases: Ticks, bugs, and pathogens of importance to human and animal health.

A disease that is transmitted to humans or other animals by an insect or other arthropod is called a vector-borne disease. Insects are a major cause of human mortality and morbidity, largely as a result of infectious pathogens being transmitted by blood-feeding species. Transmission of these vector-borne diseases is governed by complex interactions between the parasites, the vectors and the hosts. Domestic and non-domestic animals often serve as a reservoir for these pathogens until susceptible human populations are exposed. Nearly half of the world's population is infected by vector-borne disease that result in high morbidity and mortality. Rational control of these diseases requires an understanding of the links between various biological, environmental, agricultural and socio-economic factors.

3. GRIJALVA, MARIO J. Tropical Disease Institute, Biomedical Sciences Department, College of Osteopathic Medicine, Ohio University and Infectious Disease Research Laboratory, School of Biological Sciences, Catholic University of Ecuador.--Chagas disease in Ecuador: Moving from research to control program implementation.

Chagas disease affects an estimated 300,000 people in Ecuador. An additional 3 million people are at risk of infection with *Trypanosoma cruzi*. Research efforts to document the status of disease in the country have been ongoing since 1992. The results have shown anti-*T. cruzi* seroprevalence, infestation with the triatomine insect intermediate host, and other biological and epidemiological aspects that characterize Chagas disease in the country. These results have prompted the country to go from a state of denial in the early 90s to the formation of a National Chagas Disease Control Program. The challenge for researchers now is on how to guide the national control efforts towards a long-term elimination of the disease.

4. BREITSCHWERDT, EDWARD B. College of Veterinary Medicine, North Carolina State University, Raleigh, NC.--Concurrent infection with *Anaplasma, Bartonella* and *Ehrlichia* species.

Current research supports the hypothesis that the common evolutionary history of *Anaplasma, Bartonella* and *Ehrlichia* species has resulted in a modern day complex of pathophysiological interactions among these organisms. Following tick transmission, polymicrobial infections contribute to highly variable disease expression and increased severity of illness in both animals and human patients. Confirming infection caused by a single tick borne pathogen can be very challenging, particularly when evaluating

chronic as compared to acute illness. The microbiological confirmation of polymicrobial tick borne infections, using current diagnostic modalities, can be much more difficult than confirming a solo infection. It has been recognized for some time that *A. phagocytophilum* can induce disease in cats, dogs, horses and human beings, and can also infect numerous other wild animal species that serve as reservoir hosts. *B. vinsonii (berkhoffii)*, initially isolated from a dog with endocarditis in our laboratory, was subsequently isolated from coyotes, foxes and a human with endocarditis in Europe. Infection with *B. henselae*, identified in the early 1990's as the predominant cause of cat scratch disease (CSD) and bacillary angiomatosis and peliosis hepatis in immunocompromised individuals, is now known to be a much more prevalent infection in dogs than previously recognized. *E. canis, E. chaffeensis, E. ewingii* and seemingly *E. ruminantium* can infect both dogs and people. Importantly, organisms in these genera are transmitted world wide to dogs and people by a diverse spectrum of tick species. Widespread variation in the number and types of tick borne pathogens found in different geographic locations further complicates the diagnosis of polymicrobial infections. For the future, researchers should continue to define the frequency and the medical implications of polymicrobial tick-transmitted infections in companion animals.

5. BLAGBURN, BYRON L. College of Veterinary Medicine, Auburn University, AL.-- Lean, mean, transmission machines: The biology of ixodid ticks.

Ticks are second only to mosquitoes in their ability to transmit diseases. They are also important as primary disease agents and as such can cause irritation, anemia, hypersensitivity reactions, and toxicosis. The ixodid or hard ticks are important to human and animal health. They differ from argasid (soft ticks) in their structure, their unique physiologic mechanisms, and their life cycles, including the environmental locations of their developing stages. Important ixodid tick species include *Rhipicephalus sanguineus*, *Dermacentor variabilis, D. andersoni*, *Ixodes scapularis, I. pacificus*, *Amblyomma americanum* and *A. maculatum.* Ixodid ticks are well adapted for transmission of disease agents. Factors that affect disease transmission are both intrinsic and extrinsic. Intrinsic factors include host-seeking behavior, host preference, seasonality of feeding and degree of tick/host contact, and duration of attachment. Extrinsic factors include host abundance, range, seasonality (climate), diurnal patterns, and susceptibility to agents. Ticks are known to harbor multiple pathogens. It is likely that competition between pathogens within the developmental environment in the tick also affects pathogen survival. Successful transmission of disease agents requires that ticks overcome attempts of the host to eliminate them (i.e. coagulation, platelet aggregation, pain/itch responses). They combat host responses by inhibiting calcium-binding proteins, immunoglobulins, interleukins, histamine, complement, and T-cell activity. Recent research has resulted in an increased understanding of tick transmitted protozoal diseases. Agents for which we now have improved understanding of their biology, improved diagnostic procedures, or effective treatments include babesiosis, cytauxzoonosis, and hepatozoonosis. We also now have effective tick control products for use on both humans and animals. Research has demonstrated that proper use of these products combined with vaccines (when available), knowledge of the behavior of ticks, and use of tick avoidance strategies can greatly reduce the likelihood of infection with tick-transmitted agents.

6. YOUNG, JOY AND GEORGE W. BENZ. Middle Tennessee State University.--Rapid colonization of neonate lemon sharks by monogeneans.

Sharks are commonly infected by monogeneans (Monogenea); however, few data exist regarding how soon after birth neonates become infected. Fifty-one lemon sharks, *Negaprion brevirostris*, captured about Bimini, Bahamas were examined for the presence or absence of monogeneans. Thirty-one percent of the sharks were infected by *Dermophthirius nigrellii*. Twenty-five of these sharks were neonates less than 8-10 weeks old and some of these were as young as 3-4 weeks old. Twenty-four percent of the neonates were infected by *D. nigrellii*, with the youngest sharks estimated to be 3-4 weeks old. These results indicate that lemon sharks can be infected by *D. nigrellii* soon after birth. This phenomenon may have important husbandry implications when neonate sharks are selectively acquired for aquarium operations with the assumption that they will not yet be infected by monogeneans. This study was partially supported by an Undergraduate Research, Scholarship and Creative Activity Scholar award to J. Young from Middle Tennessee State University.

7. MCELWAIN, ANDREW AND GEORGE W. BENZ. Middle Tennessee State University.--Reconsidering phylogeny within Sphyriidae (Siphonostomatoida, Copepoda).

Sphyriidae (Siphonostomatoida, Copepoda) contains 9 genera and 16 species whose transformed adult females are highly modified mesoparasites of fish. The only published phylogeny for these copepods appeared well supported by morphological data, implied a pattern of coevolution between these parasites and their hosts, and has been widely accepted. Reconsideration of that analysis prompted us to reject those results in favor of more conservative and ambiguous set of results that do not support any definitive conclusions regarding sphyriid (Sphyriidae) coevolution or historical ecology. Based on our analysis we conclude that because the highly transformed habitus of sphyriids befuddles some considerations of homology and results in a paucity of morphological data that can support a phylogenetic analysis, we must await the results of molecular studies to provide a robust phylogeny for these parasites.

8. COOK, JOSHUA O[1], ROBIN M. OVERSTREET[1] AND R. RAMA KRISHNA[2]. The University of Southern Mississippi, Gulf Coast Research Laboratory, University of Southern Mississippi, Ocean Springs, MS[1] and ANGR Agricultural University, Andhra Pradesh, India[2]. --Pathology associated with fatal myxozoan infections in farmed carp, *Catla catla*, from India.

Farmed catla carp (*Catla catla*) fingerlings from a mass mortality in aquaculture ponds in Veerammagunta, India, exhibited heavy infections of *Myxobolus* sp. in the gill filaments. Macroscopic cysts surrounded the cartilaginous rods of multiple filaments and in some cases even extended around the gill rakers. Chronic inflammation involved the infected gill filaments with hyperplasia and fusion of the lamellae. Host capillaries

persisted within involved filaments, allowing for the transport of blood within the cyst. Myxozoan spores were not found among any host tissues except the gills. The heavy infection of *Myxobolus* sp. can adversely affect host gill filaments, reducing the host's ability to respire. Some bacteria, perhaps occurring post-mortem, infiltrated the body musculature, but the myxozoan infection was the apparent cause of mortality of this commercially valuable fish.

9. BAKER, TIFFANY G.[1] AND ISAURE DE BURON[2]. Grice Marine Laboratory[1] and Biology Department[2], College of Charleston, SC.--Stock identification of the Atlantic croaker, *Micropogonias undulatus* Linnaeus, using macroparasites as biological tags

The Atlantic croaker, *Micropogonias undulatus*, is a commercially and recreationally important fish present from the Chesapeake Bay to the Gulf of Mexico. Though croaker in the Gulf of Mexico are known to be genetically distinct from those in the Atlantic Ocean, it is uncertain whether those within the Atlantic Ocean comprise one or multiple unit stocks. In this study macroparasites were used as biological tags to identify stocks present along the Atlantic coast of the United States. Croaker specimens were collected from fall 2002 – spring 2005 via trawling efforts ranging from New Jersey to Florida. Results concerning the chosen tags and croaker stock structure based on those tags will be presented. Funded by a MARFIN grant NA17FF2885.

10. LUCAS, A[1]., W. S. SWECKER[1], G. SCAGLIA[2], D. S. LINDSAY[1], F. C. ELVINGER[1], J. P. FONTENOT[2], AND A. M. ZAJAC[1]. VA/MD Regional College Of Veterinary Medicine[1] and Dept. Of Animal And Poultry Sciences[2], Virginia Tech.--Population dynamics of *Eimeria* spp in grazing beef calves in Virginia.

Species of the coccidian genus *Eimeria* infect cattle worldwide causing economically significant production losses and clinical disease in some animals. The objective of this study is to characterize coccidia populations in naturally infected beef calves in Virginia. Rectal fecal samples were collected from 68 calves monthly for 5 consecutive months beginning in May, 2005. Calves were 26-76 d of age at the start of the study and grazed with their dams in groups of 6 cow/calf pairs with a stocking density of 0.67- 0.73 hectares/animal unit. No incidence of clinical coccidiosis was seen during the study. Numbers of *Eimeria* spp. oocysts were determined by the modified McMaster's test and coccidia species were identified by examination of oocysts recovered with the Wisconsin sugar flotation test. Oocyst counts were log transformed and geometric means were determined. The mean oocyst count in May was 140 oocysts per gram (OPG) and oocysts were detected in 85% of samples. The percentage of positive samples increased in subsequent months, reaching 100% in September. Monthly geometric mean oocyst counts from June to September were 377, 905, 621 and 848 OPG. *Eimeria bovis* was the most common species seen every month (present in 84% to 100% of positive samples between May and September). Other common species (present in more than 50% of the samples from at least 3 collections) were *E. zuernii, E. alabamensis*, *E. auburnensis*, *E. pelita, E. canadensis E. illinoisensis* and *E. cylindrica/ellipsoidallis*. Less common species (*E. bukidonensis, E. subspherica, E. brasiliensis* and *E. wyomingensis*) were present in 50% or fewer samples from at least 3 collections. All 12 *Eimeria* spp were seen every month with the exception of *E.*

wyomingensis in May and *E. bukidonensis* in July. Results indicate that grazing Virginia beef calves are infected by the second month of life with a diverse population of *Eimeria* spp.

11. GOODWIN, DAVID G., SOLANGE M. GENNARI[2], DANIEL K. HOWE[3], J.P. DUBEY[4], ANNE M. ZAJAC[1], AND DAVID S. LINDSAY[1]. Virginia Tech[1], Universidade de São Paulo[2], University of Kentucky[3], USDA Animal Parasitic Diseases Laboratory[4].--Prevalence of antibodies to *Encephalitozoon cuniculi* in Brazilian horses

Encephalitozoon cuniculi is a zoonotic intracellular parasite found in rabbits and other domestic animals. It is regarded as an emerging human pathogen. Little is know about this parasite in horses. Recently 72 horses from 3 Israeli farms were tested, 60% (43 horses) were seropositive for *E. cuniculi* by indirect fluorescent antibody assay (IFA). *Encephalitozoon cuniculi* was implicated as causing abortion in a Quarter horse mare from Kentucky. Stages were detected using transmission electron microscopy and PCR detected *E. cuniculi* in the placenta. *Encephalitozoon cuniculi* was also detected in the kidney and brain of a stillborn Clydesdale foal from South Africa using light microscopy and Gram staining (*E. cuniculi* spores are Gram positive). We have recently developed a direct agglutination test to detect IgG antibodies to *E. cuniculi* in serum. The serum from 554 Brazilian horses was tested for the prevalence of *E. cuniculi* antibodies using the agglutination test. A majority of the samples were from older horses (>10 years) that were culled and sent to a slaughterhouse. Sixty-nine (12%) of the horses were positive for antibodies to *E. cuniculi* at a 1:50 dilution. Positive samples are currently being examined for IgG antibodies using IFA and Western blot tests. Supported in part by a Clinical Research grant from the Virginia- Maryland Regional College of Veterinary Medicine to DSL.

12. LASCANO, MAURICIO S.[1,3], ANITA VILLACIS[2], AND MARIO J. GRIJALVA[3]. Department of Biological Sciences, Ohio University, Athens, OH[1]. School of Biological Sciences, Catholic University, Quito, Ecuador[2]. Tropical Disease Institute, Biomedical Sciences Department, College of Osteopathic Medicine, Ohio University, Athens, OH[3]. --Molecular characterization of *Trypanosoma rangeli* isolates from Ecuador.

Entomological surveys in Manabi and Loja provinces of Ecuador revealed high incidence of Triatominae insects (Hemiptera: Reduviidae) vectors of Chagas disease. This disease affects approximately 16 million people in South America. The epidemiology of Chagas disease has been hampered by *Trypanosoma rangeli,* very similar to *T. cruzi* in terms of morphology, life cycle, and biochemical features. Both parasites share the same mammal reservoirs and insect vectors. The serological diagnostic methods that are currently available for Chagas disease usually fail to discriminate efficiently between infections caused by *T. cruzi* or *T. rangeli*, causing false-positive results for Chagas. This study attempts to elucidate the origin of the strains of *T. rangeli* found in Ecuador, and to understand the interactions of this parasite with *T. cruzi* and its vectors. *Rhodnius ecuadoriensis*, *Triatoma carrioni, Panstrongylus chinai, P. rufotuberculatus*, and *Erasmus mucronatus* were collected on 23 rural

communities of Loja. *R. ecuadoriensis* and *P. howardi* were collected in five communities of Manabi. Salivary glands, haemolymph, intestinal contents, and feces were obtained from the insects. PCR tests were run on those samples. PCR amplification products were excised from the gels, cloned, and the nucleotide sequences are being determined by automated sequencing. The isolation of *T. rangeli* strains and the study of their molecular characteristics are essential achievements since this is the first report of *T. rangeli* in Ecuador. Moreover, the final results of this study will provide important insights about the influence of *T. rangeli* in the epidemiology and seroprevalence of Chagas disease in the country. Support for this study was provided by the Vice President for Research of Ohio University through the Student Enhancement Award Program.

13. PINTO, C. MIGUEL[1,2], SOFÍA OCAÑA[1], MAURICIO LASCANO[3], AND MARIO J. GRIJALVA[3]. Laboratorio de Investigación en Enfermedades infecciosas, Escuela de Ciencias Biológicas, Pontificia Universidad Católica del Ecuador, Quito, Ecuador[1]; Present address: Department of Biological Sciences, Texas Tech University, Lubbock, TX, USA[2]; Tropical Disease Institute, College of Osteopathic Medicine, Department of Biomedical Sciences, Ohio University, Athens OH, USA[3].-- Infection by trypanosomes in marsupials and rodents associated to human dwellings in Ecuador.

The genus *Trypanosoma* comprises species that parasitize mammals, including humans; among them, *T. cruzi* causes Chagas disease affecting more than 18 million people in the Americas. This report shows the results of surveys of mammal reservoir hosts of trypanosomes in the provinces of Manabí, Loja and Guayas. We conducted trapping sessions focused in nonvolant mammals in and around human residences (< 50m from the houses). We collected all the trapped animals and obtained blood samples to perform trypanosome searches using direct microscopy and hemoculture. Animals were considered positive if flagellates were observed by any of the two tests performed. Parasite identifications were performed by morphological characteristics and by polymerase chain reaction. We captured 222 animals belonging to 9 species, 2 marsupials and 7 rodents. Twenty two animals (9.9%) were positive for trypanosomes. Fifteen (6.8%) were infected with *T. cruzi* (4 of 36 *Didelphis marsupialis*; 1 of 8 *Philander opossum*; 1 of 85 *Mus domesticus*; 9 of 64 *Rattus rattus*). Eleven *R. rattus* (17.2%) harbored *T. lewisi*, five of them presented mixed infections with *T. cruzi*. Additionally, 1 of 3 *Oryzomys xantheolus* were infected with *T. rangeli*. None of the individuals of *Proechimys decumanus* (n = 11), *Akodon orophilus* (n = 4), *Sigmodon peruanus* (n = 3), and *R. norvegicus* (n = 8) showed trypanosomes. Most of the isolates belong to *T. cruzi* suggesting that *R. rattus* are important reservoir hosts of Chagas disease due to the high numbers of infected animals and their close relationship with human dwellings.

14. WIMSETT, ASHLEY, AMY ANDERSON, AND CHRIS HALL. Department of Biology, Berry College, Mount Berry Ga. --Humoral recognition of *Trypanosoma cruz*i antigens in raccoons from the Berry College campus.

Trypanosoma cruzi is a protozoan parasite endemic to the Southeastern United States. Sera from raccoons trapped on the Berry College campus were used to assess parasite prevalence and the nature of the humoral responses to *T. cruzi* infection in this important reservoir host species. Twenty three raccoons were trapped and sero-conversion tested by ELISA. Of those sera tested, 74% tested positive for *T.cruzi* exposure. Western blot analysis was performed by using the positive sera to identify the nature of recognition. Antigen preparations were run on PAGE gels under denaturing conditions and transferred onto nitrocellulose membranes. Membranes were divided into strips with each incubated with a different positive raccoon sera. Membranes were developed with DAB and the molecular weights of positive bands determined by comparison with molecular weight ladders. We found that all sero-positive raccoons recognized an antigen of approximately 47-50 kD. Some sero-positive animals also recognized an additional antigen at approximately 75 kD. The difference in recognition profiles appears correlated to the titer of parasite specific antibody. Further characterization of these antigens and their ability to induce protection in experimentally infected mice continues to be a priority.

15. PIERCE, EMILY, BRAD MEERS, AND CHRIS HALL. Department of Biology, Berry College, Mount Berry Ga. --Vertical Transmission of North American Type II and a South American Type I isolates of *Trypanosoma cruzi* in BALB/c mice.

Vertical transmission of *Trypanosoma cruzi* is well documented. What is less certain is whether all strains exhibit similar transmission patterns. This is especially of interest in North American versus South American strains of the parasite since in North America *T. cruzi* has evolved with placental mammals as the principal sylvatic reservoir hosts, while in South America marsupial species have dominated. Female BALB/c mice were infected with either the Brazil strain (BS), a Type I South American isolate, or the Lemur Isolate (LI), a Type II strain of *T. cruzi* from North America. Breeding pairs were established and monitored for reproductive success. Pups generated from these breedings were weighed and sacrificed at two weeks after birth. Tissues were harvested from pups for PCR analysis using the TCZ primer set. BALB/c mice infected with the BS of *T. cruzi* failed to generate any off-spring. Those infected with the LI produced off-spring in comparable numbers to those of uninfected control females. Of those pups born to LI infected female mice, approximately 50-60% were found to be infected. This data suggests that the LI has a greater propensity toward vertical transmission than the BS. This is consistent with the co-evolution of this strain in placental mammals and possibly represents an increased reliance on this mechanism for transmission.

16. VINCENT, AMANDA G. AND JEFFREY M. LOTZ. Gulf Coast Research Laboratory, The University of Southern Mississippi. --Evolution of virulence: transmission from dead hosts.

Evolution of virulence suggests a pathogen may evolve toward lesser virulence to ensure host survival and consequently pathogen survival. A measure of pathogen lifetime reproductive success is the basic reproduction number R_0, which is the average number of new infections produced by a single infection. R_0 is maximized when virulence is 0 thus, evolving lesser virulence would seem the best evolutionary strategy. However, other pathogen life history traits may be dependent upon virulence, resulting in constraints and tradeoffs. In particular, an inverse dependence of transmission on virulence can maximize R_0 at virulence $\neq 0$ resulting in a balance between transmission and virulence at an optimal level. Models of evolution of virulence typically assume that host death ends infectivity. But what about systems where pathogens are transmitted from dead hosts? We present data obtained through experimental infections for two pathogens, necrotizing hepatopancreatitis-bacterium (NHPB) and Taura syndrome virus (TSV), affecting penaeid shrimp aquaculture where transmission occurs primarily through cannibalism of dead infected hosts. In these systems, transmission is unrelated to the host being alive. After host death, virulence is not a component of R_0 but it does affect the probability of transmission from dead hosts. Virulent strains may not only kill hosts more rapidly than less virulent, but they may also reach a higher lethal load thus having a greater probability of transmission after host death. When dead hosts are the source of pathogen transmission, R_0 is not maximized. Instead R_0 increases as virulence increases.

17. HARRIS, SHAWNA AND CLAIRE A. FULLER. Murray State University.--Immunity in dragonfly naiads (Odonata: Anisoptera): indicators of water quality.

It is important that scientists have an early-warning system capable of identifying environmentally stressed organisms before the stressors cause irreparable population or regional harm. Terrestrial insects with aquatic stages are good biological indicators of both short and long-term environmental change because their life cycles expose them to multiple stressors in both aquatic and terrestrial environments. Currently, however, there are no studies examining immunity as an indicator of environmental health in insects. We collected larvae of the dragonfly *Libellula lydia* in seventeen ponds in western Kentucky. Hemolymph was collected from the larvae and assayed for prophenoloxidase, phenoloxidase, and protein content of hemolymph. Individuals were measured for head width and body length. Water and sediment was analyzed from each pond for metals, organophosphates, pcb's, temperature, and pH. Increased levels of prophenoloxidase, phenoloxidase, protein content of hemolymph, and increased body size have been linked to an increased ability to resist infection in insects. Our hypothesis is that as water quality decreases, immune parameters will decrease. This study was funded by a research grant from USGS.

18. SAVAGE, MASON Y. AND MICHAEL J. YABSLEY. Southeastern Cooperative Wildlife Disease Study, Department of Population Health, College of Veterinary Medicine, University of Georgia.--Sequence polymorphisms in the mismatch-repair (TcMSH2) and glutathione-S-transferase (Tc52) genes of *Trypanosoma cruzi* isolates from United States wildlife.

Trypanosoma cruzi, causative agent of American trypanosomiasis or Chagas disease, can cause fatal myocarditis in many species including humans and dogs. Molecular characterization of *T. cruzi* isolates from the United States is limited. Recently a multi-locus sequencing typing approach has been used to characterize *T. cruzi* isolates from South America. Two of these genes, the mismatch-repair (TcMSH2) and glutathione-S-transferase (Tc52) have shown multiple nucleotide and amino acid substitutions between *T. cruzi* I and *T. cruzi* II, the two major genetic groups of *T. cruzi*. In this study, we amplified and sequenced the variable regions of these two genes from 15 raccoon and 5 opossum isolates of *T. cruzi* from Georgia and Florida. Based on studies conducted in South American reservoirs, we hypothesized that raccoons and opossums will be infected with genetically distinct *T. cruzi* and because of the wide geographic separation we hypothesize that these US isolates of *T. cruzi* will differ from those from S. America. In this study, sequences of the TcMSH2 and Tc52 genes from *T. cruzi* from opossums and raccoons were separated into two main groups corresponding to *T. cruzi* I and *T. cruzi* II, respectively. Only limited nucleotide and amino acid substitutions were noted between the US and South American samples. Future work will include obtaining sequence data from additional isolates from host species over a wide geographic range in order to 1) better understand the genetic variability within this highly diverse species and 2) test for any *T. cruzi* genotype and host species associations.

19. MURPHY, STACI M.[1], MICHAEL J. YABSLEY[1], M. PAGE LUTTRELL[1], DAVID E. STALLKNECHT[1], AND SUSAN E. LITTLE[2]. Southeastern Cooperative Wildlife Disease Study, Department of Population Health, College of Veterinary Medicine, University of Georgia[1] and Department of Pathobiology, College of Veterinary Medicine, Center for Veterinary Health Sciences, Oklahoma State University[2]. -- Experimental inoculation of raccoons (*Procyon lotor*) with *Anaplasma phagocytophilum*, *Ehrlichia chaffeensis*, *Ehrlichia canis*, *Ehrlichia ewingii*, and *Borrelia lonestari*.

Previous studies found serologic and/or molecular evidence for infection of raccoons with several tick-borne pathogens of human and veterinary importance. In the current study, we investigated the experimental susceptibility of raccoons for five of these emerging tick-borne pathogens. Each inoculation trial consisted of three raccoons; two were each inoculated with either *E. chaffeensis*-infected DH82 cells, *E. canis*-infected DH82 cells, *E. ewingii*-positive dog blood, *Anaplasma phagocytophilum*-infected tick cells, or *B. lonestari* spirochetes, and one negative control raccoon was inoculated with either uninfected dog blood or cell cultures. Infections were assessed by a combination of polymerase chain reaction (PCR), indirect fluorescent antibody testing, and/or cell culture isolation methods every 3-6 days for at least 30 days post-inoculation (DPI). Both *A. phagocytophilum*-inoculated raccoons were PCR positive by 3 and 12 DPI and seroconverted (≥1:64) on 24 and 30 DPI, respectively. Both raccoons remained PCR and IFA positive until 74 DPI. One *E. chaffeensis*-inoculated raccoon was PCR-positive

from 9 to 27 DPI, culture positive on 9 DPI, and seropositive from 12 to 48 DPI. None of the *E. ewingii*- or *E. canis*-inoculated raccoons seroconverted and only one raccoon was PCR positive for *E. canis* on 3 DPI. The *B. lonestari*-inoculated raccoons were PCR negative throughout the study, but seroconversion was evident in one raccoon from 3 until 30 DPI. These data suggest that raccoons are not important reservoirs of *E. canis*, *E. ewingii*, or *B. lonestari*; however, they may be involved in the natural maintenance of two important zoonoses, *A. phagocytophilum* and *E. chaffeensis.*

20. ESLICK, RENÉ M. and VINCENT A. CONNORS. University of South Carolina Upstate. --Production of the reactive oxygen species, superoxide, by cells from the *Biomphalaria glabrata* (Pulmonata) embryonic cell line.

Previous work in our laboratory has shown that cells from the immortalized *Biomphalaria glabrata* embryonic (Bge) cell line exhibit increased motility and phagocytosis of target particles following stimulation with the cytokine, interluekin-1 (IL-1). These responses were similar to those reported for hemocytes from normally schistosome-susceptible *B. glabrata* following injection of IL-1, which has been shown to result in increased hemocyte phagocytic activity, hemocyte superoxide production, and parasite killing in the snail. The purpose the work reported herein was to determine if phagocytic stimulation of Bge cells also resulted in the production of superoxide in these cells. To determine if superoxide was produced in Bge cells a modified in vitro phagocytosis assay was developed. In short, cells cultured in 8-well chamber slides were exposed to treatments containing phaogocytosis targets (Zymosan) in the presence or absence of both the superoxide indicator, nitro-blue-tetrazolium (NBT), and the superoxide scavenger, superoxide dismutase (SOD). Results show that SOD drastically inhibits the reduction of NBT to formazan, confirming that Bge cells are capable of producing superoxide. These results further suggest that Bge cells may be useful as a potential *in vitro* model for the analysis of hemocyte mediated killing mechanisms in the snail. This work was supported by a Teaching and Productive Scholarship grant from the Teaching Enhancement Committee of USC Upstate.

21. MINTER, JENNIFER AND EDWIN ROWLAND. Ohio University.--Fate of intracellular *Trypanosoma cruzi* inhibited from egressing the host cell.

The protozoan parasite *Trypanosoma cruzi* is the causative agent of Chagas disease. The chronic stage of infection is characterized by a production of neutralizing antibodies in the vertebrate host. A polyclonal antibody, anti-egressin, has been found in our lab to inhibit egress of parasites from the host cell late in the intracellular cycle, after the parasites have transformed from the replicative amastigote into the trypomastigote. It was also found in our lab that cells in the late stages of parasite infection become permeable to molecules as large as antibodies, leading to the possibility that anti-egressin affects the intracellular parasites. This project addresses the fate of the intracellular trypomastigotes that have been inhibited from egressing the host cell. Extracellular *T. cruzi* trypomastigotes have been found to agglutinate in the presence of anti-egressin containing chronic mouse serum. The possible agglutination effect on intracellular parasites will have been determined by artificially releasing the parasites from the host cell. Also, anti-egressin treated infected cells were transferred to an uninfected culture, which resulted in a decrease of extracellular trypomastigotes relative

to the control after the secondary infection period. Further evidence of the infectivity of these parasites will have been obtained by artificially releasing the trypomastigotes from the anti-egressin-treated host cells and measuring their infectivity on fresh host cells in vitro. These measures will help to elucidate the fate of the parasite in the anti-egressin treated cultures and will provide insight to the effectiveness of the antibody to hinder progression of the infection.

22. HARTMAN, ANGELA[1], ROBERT C. WILLIAMS[1], ALEXA C. ROSYPAL[2] AND DAVID S. LINDSAY[1]. Virginia Tech[1] and University of North Carolina Chapel Hill[2].-- Efficacy of the FTA-filter based method to detect DNA of *Cryptosporidium parvum* and *Toxoplasma gondii* from oocysts

Researchers have developed a method to detect protozoa such as *Cyclospora cayetanensis*, *Cryptosporidium parvum*, *Enterocytozoon bieneusi*, and *Encephalitozoon intestinalis* using a filter-based protocol. FTA filters have been proposed as a useful method to replace traditional time-consuming DNA extraction methods for detecting parasites from food, environmental, water, and stool samples. This study was performed to determine if FTA filters could be used to extract DNA from oocysts of *C. parvum* and *T. gondii*. Extractions were performed on *C. parvum* oocysts, *T. gondii* oocysts, *T. gondii* tachyzoites, and on *T. gondii* oocysts treated with bleach to break down oocyst walls. Each sample of 1 x 10^5 oocysts or tachyzoites/ml was applied to FTA filters and prepared as DNA templates using modified methods of Orlandi and Lampel, 2000. The parasite sample spotted test filters were each used directly as template DNA for PCR. An 18SrRNA primer set was used to detect *C. parvum* and an ITS1 primer set was used to detect *T. gondii*. *Cryptosporidium parvum* oocysts were readily detected. For *T. gondii*, DNA was detected from 1 x 10^5 *T. gondii* tachyzoites, while no DNA was detected with 1 x 10^5 sporulated *T. gondii* oocysts or 1 x 10^5 sporulated *T. gondii* oocysts treated with bleach. An additional amplification was conducted using PCR product from oocyst samples as template DNA and *T. gondii* was still not detected. This study suggests that FTA filter-based extraction may be useful in detection of *C. parvum* while other methods need to be developed for *T. gondii*. Supported in part by a BRIDGE grant to DSL.

23. MITCHELL, SHEILA M., ANNE M. ZAJAC, AND DAVID S. LINDSAY. Virginia Tech. -- Development of *Cystoisospora canis* in cell culture: Evidence for unizoic cyst formation in vitro.

Endogenous stages of *Cystoisospora* species develop in the small intestine of their definitive host but extraintestinal stages can also occur in the mesenteric lymph nodes (MLN) and other tissues of both definitive and paratenic hosts. Little is know about the biology of these stages. Few attempts have been made to grow *Cystoisospora canis* in cell culture. Cover slips with bovine turbinate (BT) cell or African green monkey kidney cell (CV-1) monolayers were infected with 1 X 10^5 excysted *C. canis* sporozoites. Cover slips were removed on various days, fixed, and stained for light microscopy. Sporozoites invaded both cell types. A clear parasitophorous vacuole was present around most sporozoites viewed on days 2 and 10 post infection for both cell types. On

days 15 and 16 post infection, cyst wall developed surrounding the sporozoites in BT and CV-1 cells, respectively, appearing to fill in the parasitophorous vacuoles. These stages structurally resemble the unizoic cysts seen in the MLN of paratenic hosts. No division of sporozoites was observed at any observation period. There was no difference in sporozoite lengths and widths in BT and CV-1 cells from 2 – 17day PI, respectively. In both cell types there were slight differences in cyst wall dimensions from days 2 –17. Viability of the zoites within cysts is currently being assessed. Transmission electron microscopy is also currently being done to further characterize these stages. Supported in part by a BRIDGE grant to DSL.

24. CASSELL, MEREDITH[1], JEANNINE S. STROBL[2], CHRIS REILLY[2], AND DAVID S. LINDSAY[1]. Virginia Tech[1] and Virginia College of Osteopathic Medicine[2]. --Efficacy of histone deacetylase inhibitors against *Toxoplasma gondii*

Toxoplasma gondii is a well-recognized cause of disease in congenitally infected infants and immunocompromised individuals. Histone deacetylase (HdAC) is a regulatory enzyme that is necessary for chromatin structure and gene expression. One study indicated that the histone deacetylase inhibitor (HDI), apicidin, had activity against *T. gondii* in vitro. The present study was done to determine if the HDI suberoylanilide hydroxamic acid (SAHA) and trichostatin A (TSA) had activity against the RH strain of *T. gondii*. TSA and SAHA inhibit class I and class II HdACs defined by their homology to yeast HdACs, Rpd3 and Hda1, respectively. Activity of these 2 agents was evaluated using tachyzoite counting and monolayer protection assays. Briefly, 48-well plates containing HS68 cells were infected with *T. gondii* tachyzoites and allowed to incubate for 1 hour. The media containing all non-infective parasites were then removed. Medium containing the various concentrations of SAHA or TSA or vehicle only was the placed in the infected wells. On day 5 of the incubation period the numbers of tachyzoites present in the media were determined by counting in a hemocytometer. The EC50 of SAHA was 30 nM and that of TSA was 108 nM. The cell monolayers were stained with crystal violet and examined for lesions (plaques). SAHA and TSA showed complete protection of monolayers at 100 nM and 200 nM concentrations, respectively. Nicotinamide, an inhibitor of NAD^+ -dependent class III HdACs, had no activity against *T. gondii* in our assays. HDI have potential as new treatments for toxoplasmosis. Supported by a grant from the Carilion Biomedical Institute.

25. GAJI, RAJSHEKHAR Y. AND DANIEL K. HOWE. Department of Veterinary Science, University OF Kentucky, Lexington, KY.--GAGACGC is a critical cis-acting element required for SnSAG1 gene expression in *Sarcocystis neurona*.

Sarcocystis neurona is an obligatory intracellular parasite in the phylum Apicomplexa, and is the major cause of equine protozoal myeloencephalitis (EPM) in horses. Intracellular development of the parasite is accomplished by a complex process known as endopolygeny during which there is temporal regulation of gene expression. With the objective of characterizing regulatory elements, gene loci were isolated from a *S. neurona* genomic library and the 5' and 3' flanking regions were sequenced. Examination of 5' flanking regions of *SnSAG1, SnSAG2, SnSAG3, SnNTP* and *Sarco-21* genes did not reveal standard eukaryotic promoter elements. However, a 7-base

conserved motif, GCGTCTC, was identified by comparative analysis. This motif is present in either orientation, and there may be single or multiple copies upstream of the transcription start sites. This motif is similar to the WGAGACG/CGTCTCW sequence that has been shown to play a role in gene transcription in *Toxoplasma gondii*. To test the activity of potential regulatory sequences, we have implemented a dual luciferase assay for *S. neurona* gene expression using firefly and renilla luciferase reporter genes. The results showed that inclusion of both 5' and 3' flanking regions is required for efficient transcription of the reporter molecule. Furthermore, functional analysis of 5' flanking region of *SnSAG1* using a series of deletion mutants indicated that the GAGACGC motif, located 129 bases upstream of the major transcription start site, is critical for transcription. These studies aimed at characterizing *S. neurona* promoters may provide insight into the mechanisms that control gene expression during parasite development.

26. STOCKDALE, HEATHER D.[1], G. SHANE WEST[2], TED HANKES[3], KENNETH L. MCMILLAN[4], MARK WHITLEY[5], CHRISTINE C. DYKSTRA[1], JENNIFER A. SPENCER[1] AND BYRON L. BLAGBURN[1] Auburn University College of Veterinary Medicine[1], Vestavia Animal Clinic, Birmingham, AL[2], Alford Animal Veterinary Hospital, Birmingham, AL[3], Pell City Animal Hospital, Cropwell, AL[4], and Cobb Animal Clinic, Greensboro, NC[5].--*Tritrichomonas foetus* induced large-bowel diarrhea is an emerging disease in domestic cats.

The first reported clinical case of intestinal trichomoniasis in cats was in 1956. Since 1996, there have been numerous reported cases of feline trichomoniasis. Our laboratory has received four separate feline fecal specimens from cats throughout Alabama and North Carolina in which trichomonads were observed by direct fecal examination (direct smear). The cats demonstrated chronic diarrhea and bacteria overgrowth. These cats were treated with a combination of metronidazole, fenbendazole and enrofloxacin. Prior to treatment, trichomonads from each of the four naturally infected cats were recovered using TF InPouch assay (Biomed Diagnostics, White City, Oregon) and successfully cultured in TYM Diamond's Media. Of the four clinical cases presented to our laboratory, all cats have successfully resolved the trichomonad infection. However, one has had a relapse of diarrhea with no trichomonads present and a second was euthanized and diagnosed with FIP. Scanning electron micrographs and transmission electron micrographs were taken of the organisms isolated from infected cats and demonstrate the unique characteristics of *T. foetus.* Our laboratory at Auburn University plans to identify the regional prevalences of *T.foetus* in symptomatic and asymptomatic cats as well as host specificities of bovine and feline isolates of *T. foetus*.

27. CARREÑO, ABIGAIL D.[1], A. RICK ALLEMAN[2], ANTHONY F. BARBET[2], GUY H. PALMER[3], SUSAN M. NOH[3] AND CALVIN M. JOHNSON[1]**.** Department of Pathobiology, Auburn University[1]; Department of Pathobiology, University of Florida[2]; and Department of Veterinary Microbiology and Pathology, Washington State University[3].--*In vivo* endothelial cell infection by *Anaplasma marginale.*

Anaplasmosis is an arthropod-borne hemoparasitic disease of cattle and other ruminants. The causative agent is the gram negative bacterium, *Anaplasma marginale.* Infection of bovine erythrocytes by *A. marginale* has been well established *in vivo*, as well as *in vitro*. Recently, *A. marginale* has been propagated *in vitro* in bovine and primate vascular endothelial cell cultures. This finding provides evidence that infected endothelial cells may initiate MHC-Class-I restricted CTL responses in infected cattle. To determine the extent to which endothelial cells are susceptible to *A. marginale* infection *in vivo*, a dual staining technique was applied to tissues from a splenectomized calf experimentally inoculated with 10^9 organisms from the St. Maries strain of *A. marginale.* Sections of lung, kidney, and hemal lymph node were collected, embedded in freezing compound, frozen in isopentane/liquid nitrogen, and cryosectioned at 5 microns. Sections were co-labeled with monoclonal antibody ANAF16C1, recognizing *A. marginale* major surface protein 5 (MSP5) conjugated to Alexa Fluor 488 and a polyclonal rabbit antibody against human von Willebrand Factor (an endothelial cell marker) conjugated to Alexa Fluor 568. Nuclei were stained with 284nM 4', 6-diamidino-2-phenylindole, dihydrochloride (DAPI). Sections were evaluated by conventional wide field and confocal fluorescence microscopy using a BioRad MRC 1024 Scanning Laser Confocal Microscope. As expected, non-endothelial cells within vascular lumens were the major reservoir for *A. marginale*. In addition, *A. marginale* fluorescence co-localized to capillary endothelial cells of the lung, hemal lymph node, and kidney. These results suggest that endothelial cells may serve as a cellular reservoir for *A. marginale in vivo*. (This work was funded by a grant from the USDA-CREES).

REGULAR PAPERS

28. ROSYPAL, ALEXA C.[1,2], DWIGHT D. BOWMAN[3], DANIEL HOLLIMAN[1], GEORGE J. FLICK[1], AND DAVID S. LINDSAY[1]. Virginia Tech[1], University of North Carolina at Chapel Hill[2], Cornell University[3]. --Effects of high hydrostatic pressure processing on *Ascaris suum* eggs.

High hydrostatic pressure processing (HPP) has been shown to be an effective non-thermal means of inactivating non-spore forming bacteria and some protozoa from various food products. Little information is available regarding the effects of HPP on metazoan parasites. Outbreaks of food-borne disease have been associated with importation of food contaminated with fecal material. *Ascaris suum* is used as a surrogate model metazoan parasite for the human roundworm, *Ascaris lumbricoides*, to study the effects of treatments on the inactivation of eggs in sludge. The present study was conducted to determine the effects of HPP on *A. suum* eggs. Unembryonated *A. suum* eggs were collected from sewage and examined prior to HPP treatment. Eggs were subjected to 138 to 552 Mpa for 10 to 60 seconds in a commercial HPP unit.

Embryonation was induced after HPP treatments by incubating eggs in 0.01 N sulfuric acid at room temperature. After 21 days, 100 eggs were examined per treatment using a light microscope and the percent of embryonated eggs was determined. Morphological alterations were not observed in the HPP treated eggs. Embryonation was induced in 38-76% eggs that were subjected to 138 and 270 Mpa. No embryonation was observed in eggs exposed to pressures of 241 Mpa or more for 60 seconds or in eggs exposed to 276 Mpa for 10 to 30 seconds. These results indicate that HPP treatment could be used to protect contaminated food items by inactivating *A. suum* eggs and may also have potential in reducing food-borne illness resulting from fecal contamination. This study was financially supported in part by an Animal Health and Disease grant to DSL and GJF.

28. DOFFITT, CYNTHIA M.[1], LINDA M. POTE[1], AND D. TOMMY KING[2]. Mississippi State University[1] and USDA/APHIS/WS[2]. -- Morphological comparison and identification of cercariae released by the rams-horn snail *Planorbella trivolvis.*

The rams-horn snail, *Planorbella trivolvis*, is commonly found in commercial channel catfish (*Ictalurus punctatus*) ponds in northwest Mississppi. This snail has been found to be an adequate first intermediate host in several digenetic trematode life cycles, including *Bolbophorus damnificus* and *Clinostomum complatum*. These trematodes can cause mortality and the formation of unsightly cysts in the musculature of channel catfish. Infected snails were collected and four major types of cercariae were isolated from these snails. These cercariae are classified as *Bolbophorus* sp., *Clinostomum* sp., amphistome-type, and armatae-type. Morphological comparisons of these cercariae were made using scanning electron microscopy and light microscopy. The cercariae were further identifed using molecular analysis. This
information will be used to complete unknown digenetic trematode life cycles.

29. GRINSTEAD, C. BRAD[1], OSCAR J. PUNG[1] AND KRAIG KERSTEN[2]. Georgia Southern University[1] and Armstrong Atlantic State University[2].--Distribution of hydrobiid snails and their parasites in salt marsh along the Skidaway River in coastal Georgia.

Hydrobiid snails are the first intermediate host of the trematode *Microphallus turgidus*. As part of a study concerned with the temporal and geographic distribution of *M. turgidus* in southeast Georgia salt marshes, hydrobiid snail populations along the Skidaway River were examined to determine their distribution and the prevalence of trematode infection. To do so, sediment was collected from three to seven 0.5 m^2 quadrats along 20 vertical transects beginning in the high marsh and ending in the low marsh creek bed. Sediment samples were sieved to concentrate snails and then examined in the laboratory to determine hydrobiid number and species and the prevalence of trematode infection. Two species of hydrobiid snail, *Spurwinkia salsa* (99% of specimens) and *Onobops jacksoni* (1% of specimens) were collected. Hydrobiid snail density was greatest in the higher cordgrass (*Spartina alternifora*) zones. These zones were also characterized by short- and medium-form cordgrass, the greatest cordgrass density, and the highest richness of other invertebrate species. Hydrobiid snails may prefer the higher cordgrass zones because they are regularly

inundated by tidal flooding but, unlike the low cordgrass zone or creek bed, are subjected to the effects of water currents or the presence of swimming predators for shorter periods of time. Approximately 2.5% of *Spurwinkia* snails were infected with 3 different trematode cercariae; 1 monostome (*Phagicola diminuta*) and 2 types of xiphidiocercariae (possibly *Maritrema* sp. and *Microphallus turgidus* or *Microphallus basodactylophallus*).

30. BURON, ISAURE DE[1] AND WILLIAM A. ROUMILLAT[2]. Biology Department, College of Charleston, SC[1] and Inshore Fisheries Section, Marine Resources Research Institute, South Carolina Department of Natural Resources[2] --Histopathology of philometrid nematodes in the southern flounder *Paralichthys lethostigma.*

In South Carolina estuarine systems, the southern flounder is infected by two species of parasitic philometrid nematodes, one of which is of particular interest because individuals are found lined up along the base of consecutive dorsal and anal fin rays. Most worms are found on the pigmented side of the fish and in smaller fish induce a visible bulging of the body. Histological studies showed that worms lay between the depressor and erector muscles of each fin element. Their presence at the base of the dorsal and anal fin rays displaced or thinned the fin inclinator muscles to the extent of eventually causing complete atrophy of these muscles. Some large worms were found to cross the host's median septum, compress the erector muscles and displace uninfected adjacent fin element muscle groups. Because of the importance of these muscles in controlling fin movements, severe infection of the southern flounder by these worms undoubtedly restricts the ability of the fish to capture food and escape predators, consequently affecting their fitness and overall fate in their habitats. Funded by SC Sea Grant Consortium.

31. FULLER, CLAIRE A., Murray State University. -- The relationship between the abiotic environment and the immune function of the Caribbean termite, *Nasutitermes acajutlae.*

Termites are responsible for up to 20% of recycling of woody debris and their relative importance as degraders increases from the temperate zones to the tropics. Changing environmental conditions such as global warming could influence susceptibility of termites to disease either by increasing encounters with pathogens or altering their ability to produce an immune response. I examined the relationship between the abiotic environment (air, soil and internal nest temperature, air and internal nest relative humidity, soil pH and light levels at nests) and two measures of immunity, phenoloxidase (PO) activity and hemolymph protein concentration, using backwards stepwise regressions. I found that PO activity increased significantly with increasing external air temperature ($P < 0.02$) and nest volume (a proxy measurement of termite size; $P < 0.001$). However, protein concentration in the hemolymph decreased with extermal air temperature ($P < 0.001$) and with the relative humidity of the nest interior ($P < 0.02$). No other environmental variable was significantly related to either immune parameter. Although termites came from a number of discrete habitats (rain forest, dry forest, wetlands, mangrove lagoon and beaches) with different abiotic environments, habitat per se did not appear to influence immunity. This study suggests that the abiotic environment can exert a strong influence on PO activity and hemolymph protein concentrations. Previous studies by other authors have shown that each of these

immune parameters is related to susceptibility of insects to bacterial and fungal pathogens. Thus, changing temperature and humidity may influence prevalence of termite diseases.

32. STIEVE, ERICA[1], KIMBERLEE BECKMEN[2] , AND SHARON PATTON.[1] University of Tennessee College of Veterinary Medicine[1] and Alaska Department of Fish and Game[2]. --Seroprevalence of *Neospora spp.* and *Toxoplasma gondii* in caribou, moose, wolf, and fox populations in Alaska.

Neospora caninum is a protozoan parasite recently distinguished from *Toxoplasma gondii* (Bjerkås et al., 1984) and named by Dubey et al. (1988). It causes abortions in cattle and neurological problems in dogs. McAllister et al. (1998) determined the dog as the definitive host and additional published data have since included the coyote as a definitive host in a sylvan life cycle of the protozoan. This project measured the seroprevalence of *Neospora caninum* in wildlife populations in Alaska including animals that may serve as definite or intermediate hosts. We analyzed serum samples from caribou (n=205), moose (n=201), and wolf (n=200) collected during the 1998-2004 field seasons. We evaluated each sample for antibodies to both *Neospora* (IFA) and *Toxoplasma* (MAT) because of the antigenic similarities between the two parasites and the potential for cross reactivity. The seroprevalence follows: caribou (10.7% seropositive for *Neospora*; 0.5% seropositive for *T. gondii*), moose (0.5% / 0%), wolf (6.0%/ 7.5%). There may be species or environmental differences that alter the seroprevalence in the caribou, moose, and wolf populations that share the same geographical area.

33. AHN, JUN, JOSHUA ELLIS, DAAIYAH COOPER, HALEY JACKSON, DANA DARMOHRAY, AND ALAN F. SMITH. Department of Biology, Mercer University, Macon, GA. -- Detection of the causative agents of Lyme disease and ehrlichiosis in individual southern black-legged ticks collected from white-tailed deer of the Piedmont National Wildlife Refuge.

Adult and nymphal ticks were collected from the carcasses of freshly harvested, white-tailed deer (*Odocoileus virginianus*) at the check station during two prescribed hunts in 2004 (Oct. 21-23 and Nov. 4-6) at the Piedmont National Wildlife Refuge (Jones Co. and Jasper Co., GA). The two major tick species were Southern black-legged ticks (*Ixodes scapularis*) and lone star ticks (*Amblyomma americanum*). Collected specimens were stored individually at -200C, total genomic DNA was extracted from individuals, and aliquots provided templates for the PCR-generation of agarose-gel-electrophoretic identifiable amplicons. Primers were designed from specific gene sequences as follows: a 353-bp *Borrelia* spp. FLA-I gene fragments; a 459-bp *Borrlia burgdorferi* (Lyme disease) rOmpA gene fragment; a 164-bp *Ehrlichia* spp. 16s rRNA gene fragment; and a 900-bp *Ixodes* spp. nuclear ribosomal gene cluster fragment (ITS2). The presence of the *Ixodes*-specific amplicon served as an added control and the confirmed by sequencing the agarose gel-purified amplicons. Of the deer ticks examined, 6.32% and 1.26 % tested positive for *Ehrlichia* and *B. burgdorferi*, respectively. Faculty Research and Development Grants from the College of Liberal Arts, Mercer University provided funding for the project.

34. YABSLEY, MICHAEL J.[1], THIERRY M. WORK[2], AND ROBERT A. RAMEYER[2]. Southeastern Cooperative Wildlife Disease Study, Department of Population Health, College of Veterinary Medicine, University of Georgia, Athens, Georgia[1] and U.S. Geological Survey, National Wildlife Health Center, Honolulu Field Station, Honolulu Hawaii[2]. -- Molecular phylogeny of *Babesia poelea* from brown boobies (*Sula leucogaster*) from Johnston Atoll, Central Pacific.

The phylogenetic relationship of avian *Babesia* with other piroplasms remains unclear, mainly because of a lack of objective criteria such as molecular phylogenetics. In this study, our objective was to sequence the entire 18S, ITS-1, 5.8S, and ITS-2 regions of the rRNA gene and partial β-tubulin gene of *B. poelea*, first described from brown boobies (*Sula leucogaster*) from the central Pacific, and compare them to those of other piroplasms. Phylogenetic analyses of entire 18S rRNA gene sequence revealed that *B. poelea* belonged to the clade of piroplasms previously detected in humans, domestic dogs, and wild ungulates in the western United States. The entire ITS-1, 5.8S, ITS-2, and partial β-tubulin gene sequence shared conserved regions with previously described *Babesia* and *Theileria* species. The intron of the β-tubulin gene was 45-bp. This is the first molecular characterization of an avian piroplasm.

35. PALMIERI, JAMES R., Ph.D[1], KATHERINE BARTER, BS, MT[2], MELISSA HRICKO, BS[3] AND MARIE PALLOTT[4]. Department of Microbiology, Division of Biomedical Sciences, Virginia College of Osteopathic Medicine[1], Virginia Polytechnic Institute Department of Biochemistry[2,4] and Center for Molecular Medicine and Infectious Diseases Virginia Maryland Regional College of Veterinary Medicine[1,2,4] and Virginia Commonwealth University, Department of Clinical Laboratory Science, Richmond VA[3].--
Low temperature induction of *Acanthamoebae* trophozoites from dormant cysts influences their ability to phagocytize bacteria.

Free-living *Acanthamoeba* are commonly found in aquatic biofilms. Acanthamoebae exist in both the trophozoite feeding and the highly resistant and dormant cyst stage. Acanthamoebae and pathogenic bacteria are closely involved in complex symbiotic relationships. In a study by Abd (2003), *F. tularensis* (tularemia) survived and developed within *A. castellanii*. How *F. tularensis* exists in its silent stage in nature and its relationship to naturally occurring acanthamoebae in soil and aquatic environments remains unknown. When acanthamoebae are placed in adverse conditions, (pH, nutrient depletion, increased salinity), trophozoites change into highly protective cysts. Presently, we are studying the mechanisms of cold temperature on *F. tularensis* survival and colonization in trophozoite and cyst forms of *Acanthamoeba*. In nature, *F. tularensis* may survive for extended periods in cold environments while acanthamoebae may remain in the cyst stage for up to 24 years. At lower temperatures, we found a unique and unreported variation in the behavior or three species of *Acanthamoeba* [*A. astronyxis* (nonpathogenic), *A. castellanii* (semi pathogenic) and *A. culbertsoni (*highly pathogenic)]. When dormant cyst stages are lowered to 18C-3C, cysts revert back to trophozoites. As temperatures are lowered to -2C, all newly formed trophozoites eventually encyst. Experiments are presently underway to determine if these cold-induced trophozoites are capable of feeding on *F. tularensis* which may explain how

Francisella exists in its silent-cycle and may elucidate its relationship with *Acanthamoeba* in nature. This relationship represents one of the most scientifically intriguing questions to be answered concerning the epidemiology and natural transmission of tularemia.

36. STONE, SHARON[1], VINA DIDERRICH-FAULKNER[2], PAUL E. SUPER[3] AND, CHARLES T. FAULKNER[1]**.** University of Tennessee College of Veterinary Medicine, Knoxville TN[1], Lincoln Memorial University, Harrogate TN[2], and Great Smoky Mountains National Park, Lake Junaluska, NC[3].--Endoparasitic Infections in Migratory Birds from the 2003-2005 Banding Seasons at the Purchase Knob, Great Smoky Mountains National Park, Lake Junaluska, NC.

During the summers of 2003, 2004, and 2005, fecal samples (n=265) representing 40 species of passerine birds were examined for parasitic infection in conjunction with the All Taxa Biodiversity Inventory (ATBI) and the Monitoring Avian Productivity and Survivorship (MAPS) projects conducted in the Great Smoky Mountain National Park. Evidence of parasitic infection was detected in 47 samples examined. Coccidia oocysts possibly *Isospora* sp. or *Atoxoplasma* sp. were present in 25 samples, Trichostrongyle-type eggs probably from *Trichostrongylus tenuis* were found in 5 samples, *Syngamus* eggs were found in 13 samples, *Capillaria* sp. eggs were found in 13 samples, Spiruid-type eggs were found in 1 sample, and tapeworm eggs were found in 6 samples. Five birds had mixed infections. The occurrence of the coccida oocysts in 12 different passerine species is of interest because this group of parasites has only been previously reported in the wild from the Eastern blue bird, *Sialia sialis,* and a song sparrow, *Melospiza melodia.* Continuing research in this population of migratory passerine birds is directed at the understanding the relationship of parasitic infection with host variables (foraging habitat, diet, and migratory behavior) and geographic variables (vegetation cover and elevation) in the Great Smoky Mountain National Park.

37. DIDERRICH-FAULKNER, VINA[1], LEAH ALLEN[1], CHARLES T. FAULKNER[2], AND PAUL E. SUPER[3]. Lincoln Memorial University, Harrogate TN[1], and University of Tennessee College of Veterinary Medicine, Knoxville TN[2], and Great Smoky Mountains National Park, Lake Junaluska, NC[3]--Patterns of positivity: Endoparasitic infection and host behaviors and in migratory birds from the 2005 banding seasons at the Purchase Knob, Great Smoky Mountains National Park, Lake Junaluska, NC.

The high elevation (>5000 feet) of the Great Smoky Mountains National Park is a unique environment for studying ecologic relationships between avian hosts and factors influential for parasite transmission. The effects of foraging habitat, diet, and migratory behavior on patterns of parasitic infection are poorly understood. Birds identified as ground or canopy foragers were analyzed according to their foraging habitat and its potential impact on parasite infection status. Subsequently birds were classified as insect-eaters or seed/fruit/berry eaters, and neotropical migrants or resident/short distance migrants for analyses of diet and migratory behavior on parasitic infection. In each of these comparisons there was no statistical association (chi-square, $p > 0.05$) between foraging habitat, diet and migratory behavior and parasitic infection. Despite these results, insect-eating birds exhibit a tendency to harbor parasitic infections ($p=0.07$). However, our results may be confounded by the observation that many

species of birds inhabiting the high elevation zone of the Great Smoky Mountains are dietary generalists during the juvenile stages in their life cycle and traditionally defined seed-eaters also consume insects prior to adulthood. We also recognize that the breeding season for birds in 2005 was delayed by a late snowfall May, and the age structure of captured birds included fewer juveniles samples compared to the previous 2 banding seasons.

POSTERS

29. SCHAUMBURG, COLLIN AND PAUL SIKKEL. Murray State University.--Do parasite loads vary with host and environmental parameters in bluegill sunfish (*Lepomis macrochirus*)?

Parasites have been found to affect the health, behavior, and life history of fishes (Bartoli et al 2000), including reproduction (e.g., mate choice and parental investment), and habitat choice. Centrarchid sunfishes (*Lepomis*) are among the best-studied North American fishes. Considerable data exist on their reproductive habits, diet, and habitat choice. While descriptive studies have characterized parasite communities of sunfishes elsewhere, little is known about parasite communities on fishes in western Kentucky, or the relationship between parasites and the behavior and ecology of sunfishes generally. As a first step in addressing this lack of knowledge, I conducted a field study on parasite loads associated with the gill lamellae of bluegill sunfish (*Lepomis macrochirus*), I hypothesized that: 1) males will have a higher rate of parasitism than females due to decreased mobility associated with defending the nest; 2) parasite loads will be higher in vegetated habitats; 3) gill parasites will be most abundant on the first; and 4) parasite loads will change throughout the spawning season. I collected fish by rod and reel and cast netting at different sites in Western Kentucky. Fish were sexed and processed in the laboratory to determine loads of parasitic monogeneans. Sampling of fish was conducted throughout the spring and summer spawning season (water temperature >65^{o}F). Parasites were found to be significantly more abundant on the second gill arch, and parasite abundance differed significantly over time. Although parasite loads tended to be higher for males, the difference was not significant.

30. O'BRIEN, MIKE AND JESSICA SHAW. Murray State University. Murray KY.--Blood and fecal parasites of the tiger salamander (*Ambystoma tigrinum nebulosum*) according to life stage.

This study provides a unique look at differences in parasite prevalence within two life stages of tiger salamanders, *Ambystoma tigrinum nebulosum*. Individuals in the paedomorphic life stage reach maturity while retaining larval characteristics such as gills and a membranous tail. It is also these characteristics which bind them to an aquatic lifestyle. Metamorphic individuals commonly transform before becoming adults and lose their larval characteristics, allowing them to enter the terrestrial environment. These life stages separate the individuals into two ecosystems (aquatic and terrestrial) which suggest that differences between parasites in the same host species might occur

according to life phase. Juveniles were also sampled for parasites, providing a third life stage for comparison. Within juvenile stages a cannibalistic morph, characterized by a wider mouth and enlarged "teeth", also occurs. We hypothesized that metamorphic adults should have higher prevalence of blood parasites (e.g., trypanosomes) due to increased exposure to vectors and that cannibals should have a higher prevalence of intestinal parasites because of increased exposure after eating infected larvae. Blood and fecal samples of tiger salamanders were collected from ponds in south-central Colorado in 2004-05. Blood smears (N =128) were prepared in the field while parasite eggs (N > 100) were separated from fecal samples with a sucrose gradient floatation and prepared for observation under a microscope. Results of both analyses will be presented. Support for this research was received from a grant of the National Science Foundation.

31. BARTER, KATHERINE[1] AND JAMES R. PALMIERI[2]. Virginia Polytechnic Institute Department of Biochemistry[1] and Center for Molecular Medicine and Infectious Diseases Virginia Maryland Regional College of Veterinary Medicine[1], and Department of Microbiology Division of Biomedical Sciences Virginia College of Osteopathic Medicine[2].-- Gentamicin and the symbiotic relationship between *Francisella* and *Acanthamoeba*: The threat of an epidemiological crisis.

Acanthamoeba are protozoa that inhabit a variety of environments. Most species of *Acanthamoeba* are nonpathogenic, however, several species are pathogenic and known to cause granulomatous amoebic encephalitis or amoebic keratitis in humans. *Acanthamoeba* exist in both the feeding trophozoite and dormant cyst stages. The cyst stage is highly resistant to physical, chemical and radiological conditions. Previous research indicates many bacteria species survive within both stages of *Acanthamoeba.* This symbiosis may have contributed to the ability of bacteria to survive and adapt their existence within macrophages. Previous research indicates many pathogenic bacteria utilize the same genes to grow in macrophages as inside *Acanthamoeba*. It is hypothesized that interactions and adaptations undergone by microorganisms living in *Acanthamoeba* may have contributed to the evolution of intracellular bacteria. In 2003, Abd et al reported that *Acanthamoeba castellanii* growth dropped 25% when co-cultured with *F. tularensis* compared to when grown alone. In their study, 250µg/mL of Gentamicin was added to a centrifuged *Acanthamoeba-Francisella* pellet to minimize extracellular *F. tularensis* contamination. The resultant decrease in *Acanthamoeba* population was attributed to unidentified toxic affects of *F. tularensis*. Our laboratory recognized the significance of *Francisella* colonization of *Acanthamoeba*, but attributed the decrease in amoeba growth to the addition of Gentamicin, not the presence of *Francisella*. We investigated the effects of increasing concentrations of gentamicin on the growth of three species of *Acanthamoeba*: We found that *Acanthamoeba* grown in increasing concentrations of Gentamicin resulted in a drop in population of the three tested species of *Acanthamoeba*.

NOTES

Southeastern Society of Parasitologists

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1990 Grover C. Miller
1991 Burton J. Bogitsh
1996 Sharon Patton
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