PROCEEDINGS

ANNUAL

28th MEETING

THE

AMERICAN

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OF VETERINARY

PARASITOLOGISTS

JULY 17–18 1983

NEW YORK, NEW YORK
AMERICAN ASSOCIATION OF VETERINARY PARASITOLOGISTS
Founded 1956
Affiliated with the American Veterinary Medical Association

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SESSION 1 - EPIDEMIOLOGY
Chairman: EL Roberson and MD Ruff

1. 8:20 A Review of the Fourth International Conference of Institutions of Tropical Veterinary Medicine, INTERNATIONAL CONFERENCE ON IMPACT OF DISEASES ON LIVESTOCK PRODUCTION IN THE TROPICS
   JB Malone
   Baton Rouge, LA

2. 8:30 Prediction of Fascioliasis Risk in Louisiana from Meteorologic Data
   JB Malone, ME Hugh-Jones, and RA Muller
   Baton Rouge, LA

3. 8:45 Trichinella spiralis in Pennsylvania Wildlife
   GA Shad, DA Leidy, KD Murrell, and G Alt
   Philadelphia, PA

4. 9:00 Epidemiologic Approach to the Control of Sheep Nematodes
   RP Herd, CF Parker, and KE McClure
   Columbus, OH

5. 9:15 The Epidemiology of Gastrointestinal Nematodes in Sheep in Pennsylvania
   C Johnstone and S Donoghue
   Philadelphia, PA

6. 9:30 The Horse and Deer Flies Collected from Tethered Cattle in East-Central Mississippi
   RL Combs, Jr.
   Mississippi State, MS

7. 9:45 Echinococcus Scene in Minnesota
   JC Scholotthauer, BE Stromberg, PD Karnes, and NB Ballard
   St. Paul, MN

10:00 Break
SESSION 2 - SYMPOSIUM: TOPICS IN VETERINARY ENTOMOLOGY
Chairmen: JA Hawkins and JW McCall

8. 10:20 Economic Impact and Control of Arthropods Affecting Livestock
   ET Schmidtmann
   Beltsville, MD

9. 10:50 Tabanids as Vectors of Bovine and Equine Diseases
   JA Hawkins
   Mississippi State, MS

10. 11:10 Black Flies: Their Role as Vectors and Pathogens in Animal Health
     EW Cupp
     Ithaca, NY

11. 11:40 The Dung Beetle Connections to Helminth Parasites
     TB Stewart
     Baton Rouge, LA

12. 12:00 Lunch

SESSION 3 - CANINE HEARTWORM
Chairmen: LS Blair and RB Grieve

12. 1:20pm A Review of the 1983 Heartworm Symposium
   LS Blair and RB Grieve

13. 1:30 The Impact of Dirofilaria immitis on the Racing Greyhound Industry in Florida
     CH Courtney, SF Sundlof, and TJ Lane
     Gainesville, FL

14. 1:45 Standardization of Dirofilaria immitis Infections in the Ferret (Mustella patorius furo) by Transplantation of Immature Worms
     PA Tanner, JW McCall, RA Holmes, and JJ Jun
     Athens, GA

15. 2:00 Development of Four Monoclonal Antibodies Against Dirofilaria immitis and Their Potential in the Detection of Circulating Antigen
     CP McArthur, TW Schillhorn van Veen, AM Geary, and JF Williams
     East Lansing, MI

16. 2:15 Diagnosis of Canine Heartworm Infections: Emphasis on Serodiagnosis of Occult Infections
     CA Wagner, SE Pratt, and RM Corwin
     Columbia, MO

17. 2:30 Serologic Patterns of Canine Heartworm Infection: The Louisiana Experience
     LT Glickman, RB Grieve, EB Breitschwerdt, M Mika-Grieve, CJ Patronek, LM Domanski, and CR Root
     Philadelphia, PA
18. 2:45 Effects of Decreased Liver Function on the Thiacetarsamide Efficacy and Arsenic Kinetics in Dirofilaria immitis Therapy
   RA Holmes, KW Prasse, RC Wilson, LWL Cornelius, EL Roberson, MB Bumgardner, and JW McCall
   Athens, GA

3:00 Break

SESSION 4 - CHEMOTHERAPY: GENERAL
Chairmen: WC Campbell and VJ Theodorides

19. 3:30 Metabolic Profile of Albendazole
   RJ Gyurik, RC Parish, and AW Chow
   West Chester, PA

20. 3:45 A Controlled Test of Fenbendazole Against Naturally Acquired Infections of Monezia and Trichuris in Young Cattle
    TJ Kennedy and GH Myers
    Somerville, NJ

21. 4:00 Palatability of Albendazole Feed Premix and Top Dress Formulation for Cattle
    HD Daniels and L Shor
    West Chester, PA

22. 4:15 Effectiveness of the Morantel Sustained Release Bolus in the Prophylaxis of Bovine Parasitic Gastrointestinal Helminthiasis
    BL Presson, TA Yazwenski, TE Greenway, and TJ Newby
    Fayetteville, AR

23. 4:30 Efficacy of Ivermectin Against Oestrus ovis in Sheep
    RA Roncalli
    Rahway, NJ

24. 4:45 Anthelmintic Efficacy of a Febantel-Praziquantel Combination Paste in Dogs and Cats
    RG Arthur and DD Cox
    Shawnee, KS

25. 5:00 The Prophylactic Efficacy of MK-401 on Fasciola hepatica-Infected Ruminants
    RH Fetterer, RS Rew, and LC Casbarre
    Beltsville, MD

26. 5:15 Bovatec® (Lasalocid) – A New Anticoccidial for Beef Cattle
    G Untawale and WE Brandt
    Nutley, NJ

5:30 Break

5:45 Business Meeting

6:30 Society Social
MONDAY, July 18

SESSION 5 - CHEMOTHERAPY: EQUINE AND OVINE
Chairmen: RP Herd and TW Schillhorn van Veen

27. 8:00am An Update on World-Wide Clinical Experience with EQVALAN®
     WDH Leaning
     Rahway, NJ

28. 8:15 Efficacy of Oxfendazole Against Migrating Stages of Strongylus vulgaris
     JOD Slocombe, BM McGraw, PW Pennock, NG Ducharme, and JD Baird
     Guelph, Ontario, CANADA

29. 8:30 Critical and Clinical Studies on VET-220, A Benzimidazole Anthelmintic Against Equine Parasites
     TR Bello
     Southern Pines, NC

30. 8:45 Benzimidazole Resistant Small Strongyles in Horses in Southeastern Pennsylvania: Prevalence and Factors Leading to Development of Resistance
     C Uhlinger and C Johnstone
     Kennett Square, PA

31. 9:00 Antiparasitic Activity of Closantel in Horses
     BP Campbell, J Guerrero, KM Newcomb, and BF Michael
     Washington Crossing, NJ

32. 9:15 Critical Testing of the Activity of Fenbendazole and Febantel Against Parascaris equorum in Ponies
     JA DiPietro, A Paul, TF Lock, and KS Todd
     Urbana, IL

33. 9:30 Efficacy of Albendazole Against Very Immature Fasciola hepatica in Sheep
     VJ Theodorides, J Freeman, and GC Scott
     West Chester, PA

34. 9:45 Efficacy of Albendazole Against Fascioloides magna in Sheep
     BE Stromberg, JC Schlotthauer, and GA Conboy
     St. Paul, MN

10:00 Break

SESSION 6 - QUIZ

35. 10:15 What's Your Diagnosis?
     CH Gardiner
     A.F.I.P.
     Washington, DC
SESSION 7 - COCCIDIA
Chairmen: JP Dubey and KR Kazacos

36. 10:45  Experimental *Isospora suis* Infections in Piglets
           JV Ernst and HR Gamble
           Beltsville, MD

37. 11:00  Enhanced Virulence as the Reason for Apparent Resistance of Avian
           Coccidia to Ionophores
           MD Ruff
           Beltsville, MD

38. 11:15  The Effect of High Fiber Diets on Intestinal *Eimeria* spp. Infections
           in the Domestic Rabbit
           LM Pote and TA Yazwinski
           Fayetteville, AR

39. 11:30  *In Vitro* Cultivation of the Swine Coccidia *Isospora suis*
           R Fayer, HR Gamble, and JV Ernst
           Beltsville, MD

SESSION 8 - PRESIDENT'S ADDRESS

40. 11:45  JP Williams
           East Lansing, MI

12:15pm  Lunch

SESSION 9 - EXPERIMENTAL AND CLINICAL STUDIES
Chairmen: KD Murrell and RM Corwin

41. 1:30  Autoradiographic Tracing of Parasitic Migrations
         JR Georgi
         Ithaca, NY

42. 1:45  Assessment of Pulmonary Migration of Ascarids (*Baylisascaris, Ascaris*)
         by Bronchopulmonary Lavage
         WK Wyand-Ouellette, KR Kazacos, and DB Denicola
         West Lafayette, IN

43. 2:00  Studies on Prenatal and Lactogenic Transmission of *Toxocara canis* and
         *Ancylostoma caninum* I. Experimental Infection of the Bitch Before
         Pregnancy
         EL Roberson and TM Burke
         Athens, GA

44. 2:15  Studies on Prenatal and Lactogenic Transmission of *Toxocara canis* and
         *Ancylostoma caninum* II a. Experimental Infection of the Bitch at
         Mid-Pregnancy and at Parturition b. Longevity of Lactogenic
         Transmission
         TM Burke and EL Roberson
         Athens, GA
SESSION 10 - IMMUNITY
Chairmen: BE Stromberg and JF Williams

45. 2:30 Influences of Selenium Intake on Infections by Nematodes and Coccidia in Rats and Goats
   JC Frandsen
   Auburn, AL

46. 2:45 Pathophysiology of Fasciolioides magna Infection in Sheep
   BE Stromberg, GA Conboy, DW Hayden, and JC Schlotthauer
   St. Paul, MN

47. 3:00 Taenia hydatigena Infections in Old World Monkeys
   S Sulaiman, JF Williams, and D Wu
   East Lansing, MI

48. 3:15 Experimental Visceral Leishmaniasis in Domestic Cats
   CE Kirkpatrick, JP Farrel, and MH Goldschmidt
   Philadelphia, PA

3:30 Break

49. 4:00 Development of Immunity to Ascaris suum in Swine After Natural Infection
    JF Urban
    Beltsville, MD

50. 4:15 Immunodiagnostic Antigens for Swine Trichinosis Isolated Using Monoclonal Antibodies
    HR Gamble and CE Graham
    Beltsville, MD

51. 4:30 Lymphocyte Reactivity to Ostertagia ostertagi L3 Antigens in Type I Ostertagiasis
    PH Klesius, SM Washburn, H. Ciordia, BT Haynes, and TG Snider III
    Auburn, AL

52. 4:45 Antigen-Antibody Complexes from the Sera of Dogs with Filarial Infections
    B Hammerburg
    Raleigh, NC

53. 5:00 Immune Expulsion of Trichinella spiralis from Swine: Density Dependence
    KD Murrell and HR Gamble
    Beltsville, MD

54. 5:15 The Antibody Response to Sarcocystis of Experimentally Infected Cattle and Sheep as Determined in an Enzyme-Linked Immunosorbant Assay
    LC Gasbarre
    Beltsville, MD
Antibody Dependent Cell Mediated Adherence of *Strongyulus vulgaris* Third Stage Larvae, *In Vitro*

TR Klei and MR Chapman

Baton Rouge, LA
1. A REVIEW OF THE FOURTH INTERNATIONAL CONFERENCE OF INSTITUTIONS OF TROPICAL VETERINARY MEDICINE, "INTERNATIONAL CONFERENCE ON IMPACT OF DISEASE ON LIVESTOCK PRODUCTION IN THE TROPICS."
J.B. MALONE, School of Veterinary Medicine, Louisiana State University, Baton Rouge, Louisiana

An Invited Review

2. PREDICATION OF FASCIOLIASIS RISK IN LOUISIANA FROM METEOROLOGIC DATA
J.B. MALONE, M.E. HUGH-JONES, and R.A. MULLER, School of Veterinary Medicine, Louisiana State University, Baton Rouge, Louisiana.

Farms in three geographically diverse areas of Louisiana were studied over a 3 to 5 year period (1978 - 1983) to establish seasonal transmission patterns for bovine fascioliasis. Snail population dynamics (4 to 8 habitats at each location), sentinel calf infections, and herd prevalence rates were recorded and related to local climatologic variables. Previously tested climatologic forecasting systems—the "Mt" system of Ollerenshaw and Rowlands (1959), the "wet-day" system of Ross (1964) and reported mathematical models—will be studied for "fit" to available annual transmission data. The suitability of these methods for use in a climatologic forecasting system for fascioliasis in Louisiana will be reported.

3. TRICHINELLA SPIRALIS IN PENNSYLVANIA WILDLIFE

Tongues from 1170 raccoons, 384 oppossums, 201 muskrats, 168 foxes, 51 skunks, and 17 mink were examined by peptic digestion. The overall prevalence of Trichinella infection was 3.2%, varying in the carnivores from 2.6% in raccoons to 15.1% in red foxes; no muskrats were infected. Prevalence varied regionally, being higher west of the Susquehanna River, where, in the Ridge and Valley Province it reached 9%. Infections were heavy, the mean number of larvae per gram (LPG) was 113.3 in infected animals.

Tongue and diaphragm samples (mainly) from 881 of 1404 hunter-killed, Pennsylvania black bears were also examined by digestion. Of these, 9 of 454 (1.96%) and 10 of 427 (2.34%) were infected in 1981 and 1982, respectively. Intensity of infection varied greatly; 8 with <1 LPG, 8 with between 5 and 112, and 3 heavily infected bears with 348, 465, and 512 LPG, respectively.

4. EPIDEMIOLOGIC APPROACH TO CONTROL OF SHEEP NEMATODES
R.P. HERD, C.F. PARKER, and K.E. McCLURE, The Ohio State University College of Veterinary Medicine, Columbus, and the Ohio Agricultural Research and Development Center, Wooster, Ohio.

The value of an epidemiologic approach to control of sheep nematodes was demonstrated in a study utilizing 6 groups of lambs
4 groups of ewes and lambs. Strategies tested included winter treatment to kill arrested (hypobiotic) larvae, spring treatments to prevent the mid-year rise in pasture infectivity, treat and move at mid-year, and turnout to pasture regrowth after harvesting hay or silage. The various strategies and controls were assessed by comparison of body weights, wool production, fecal egg counts, pasture larval counts, clinical signs and mortality rates.

5. THE EPIDEMIOLOGY OF GASTROINTESTINAL NEMATODES IN SHEEP IN PENNSYLVANIA
C. JOHNSTONE and S. DONOGHUE, Departments of Pathobiology and Clinical Studies, University of Pennsylvania School of Veterinary Medicine, Philadelphia, Pennsylvania.

The pattern of natural infections with gastrointestinal nematodes in crossbred ewes and their lambs was followed for 18 months. A thiabendazole resistant strain of Haemonchus contortus was recognized in the summer of 1981. Breeding ewes showed 2 peaks in their fecal egg counts during 1982 (Jan-Dec). The first, a periparturient rise in egg counts, occurred 9 weeks after the start of lambing and was terminated naturally after weaning. A second, smaller rise occurred in August. Pasture larvae counts showed 2 peaks of pasture contamination: the first in June and July followed the periparturient rise in egg counts 2 months earlier while the second occurred in October and closely followed the second smaller rise in egg counts of the breeding ewes.

Lambs were either weaned and taken to slaughter or weaned, treated with pyrantel pamoate, and moved to "safe" pasture for summer grazing then returned to the permanent ewe pasture for breeding in November. The pattern of fecal egg counts in lambs showed two small peaks. The first occurred in August while the second occurred in November - December following their return to the ewe pasture. Neither peak was high enough to be of concern.

6. THE HORSE AND DEER FLIES COLLECTED FROM TETHERED CATTLE IN EAST-CENTRAL MISSISSIPPI
R.L. COMBS, JR., Department of Entomology, Mississippi Agricultural and Forestry Experiment Station, Mississippi State University, Mississippi State, Mississippi.

Studies to determine the species of Tabanids that consume bovine blood, preferred feeding time (PFT), preferred feeding site (PFS), and peak seasonal activity (PSA) were conducted in Oktibbeha County, Mississippi during 1981-82. Each year, 2 steers were tethered along a timberline. Tabanids that consumed blood were individually collected, killed and above data for each recorded. In 1981, 565 specimens representing 3 genera and 20 species were collected. During 1982, 3335 specimens representing 4 genera and 27 species were collected. Five species, in the genus Tabanus represented 83% of the total collection. These five were T. pallidescens (57.9%), T. sulcifrons (10.0%), T. calens (6.0%), T. lineola (4.9%), and T. gladiator (4.2%). The PFT (morning), PFS (lower legs) and PSA (June 10) was similar for T. pallidescens and T. lineola. T. sulcifrons and T. calens had a PFT of early
morning, a PFS along the animals topline and sides and a PSA of
September 12. T. gladaitor had a PFT of late afternoon, a PFS
along the topline and a PSA of early to mid-August.

7. THE ECHINOCOCCUS SCENE IN MINNESOTA
J.C. SCHLOTTHAUSER and B.E. STROMBERG, Department of Veterinary
Pathobiology, College of Veterinary Medicine, University of
Minnesota, St. Paul, Minnesota, P.D. KARNS, Forest Wildlife and
Populations Groups, Minnesota Department of Natural Resources,
Grand Rapids, Minnesota, and N.B. BALLARD, Department of Biology,
Mankato State University, Mankato, Minnesota.

Endemic Echinococcus granulosus infection in northern
Minnesota has been recognized but poorly documented for many years
in the classic North American timber wolf (Canis lupus) - moose
(Alces alces) cycle. It should not be surprising that the parasite
occurs here since Minnesota supports the largest herd of moose,
estimated to be in excess of 10,000 animals, and the largest pack
of timber wolves, estimated between 1000 and 1200 animals, of any
state in the United States other than Alaska. The incidence of E.
granulosus in timber wolves is thought to be high although
documented recoveries of the parasite are rare. Recent surveys by
personnel from the Minnesota Department of Natural Resources of
viscera of legally hunted and killed moose indicate the annual
prevalence of pulmonary hydatid cysts in these animals is as high
as 39 per cent in the northeast herd. These moose are in the same
area where the timber wolf population is found. Hydatid cysts in
resident human and domestic animal populations are not recorded.

Echinococcus multilocularis infection was first recognized in
Minnesota by Carney and Leiby (J. Paras. 54:714, 1968) in red foxes
(Vulpes vulpes) and deer mice (Peromyscus maniculatus) during 1966
and 1967. The first human case of alveolar hydatid disease in the
contiguous United States was subsequently diagnosed in a
56-year-old female from Lyon County in Minnesota in 1977 (Gamble,
et al., JAMA 241:904-907, 1979). Recent field surveys of red foxes
(Ballard, et al., unpublished data) indicate adult E.
multilocularis infection in 25 to 95 per cent of these animals in
the southwestern prairie biome of Minnesota.

8. ECONOMIC IMPACT AND CONTROL OF ARTHROPODS AFFECTING LIVESTOCK
E.T. SCHMIDTMANN, Livestock Insect Laboratory, U.S.D.A.,
Beltsville, Maryland.

Invited Symposium Speaker

9. TABANIDS AS VECTORS OF BOVINE AND EQUINE DISEASES
J.A. HAWKINS, School of Veterinary Medicine, Mississippi State
University, Mississippi State, Mississippi.

Invited Symposium Speaker
10. BLACK FLIES: THEIR ROLE AS VECTORS AND PATHOGENS IN ANIMAL HEALTH
E.W. CUPP, Department of Entomology, Cornell University, Ithaca, New York.

Invited Symposium Speaker

11. THE DUNG BEETLE CONNECTIONS TO HELMINTH PARASITES
T.B. STEWART, Department of Veterinary Microbiology and Parasitology, School of Veterinary Medicine, Louisiana State University, Baton Rouge, Louisiana.

Invited Symposium Speaker

L.S. BLAIR, Merck & Company, Rahway, New Jersey, and R.B. GRIEVE, Department of Pathobiology, College of Veterinary Medicine, University of Pennsylvania, Philadelphia, Pennsylvania.

An Invited Review

13. THE IMPACT OF FILARIASIS ON THE RACING GREYHOUND INDUSTRY IN FLORIDA
C.H. COURTNEY, S.F. SUNDLORF, and T.J. LANE, Department of Preventive Medicine, College of Veterinary Medicine, University of Florida, Gainesville, Florida.

Florida racing rules prohibit the use of diethylcarbamazine in racing greyhounds because its presence interferes with thin-layer chromatography testing of urine samples for drugs. In response to complaints by veterinarians and greyhound trainers that a high proportion of greyhounds were becoming infected with Dirofilaria immitis as a result of the ban on the use of diethylcarbamazine, the blood of 2004 greyhounds from 4 separate areas (Tampa, n = 163; St. Petersburg, n = 553; Miami, n = 801; and Jacksonville, n = 487) was examined for the presence of microfilariae. Only 11 dogs (0.5%) were found to be infected with D. immitis, 403 dogs (20.1%) were infected with Dipetalonema reconditum, and 2 dogs (0.1%) were infected with a Dirofilaria striata-like microfilaria. To determine the impact of D. reconditum on the canine athlete, the prevalence of this parasite was compared among 483 dogs of different racing grades. Its prevalence in A grade dogs (the fastest) was 45%, B = 35%, C = 34%, D = 33%, and E (the slowest) = 40%. It was concluded that infection by D. immitis in racing greyhounds was a minor problem, the complaints of massive epizootics probably resulted from misidentification of D. reconditum which has no effect on racing performance.
14. STANDARDIZATION OF DIROFILARIA IMMITIS INFECTIONS IN THE FERRET (*MUSTELA PUTORIUS FURO*) BY TRANSPLANTATION OF IMMATURE WORMS

P.A. TANNER, J.W. McCALL, R.A. HOLMES, and J. JUN, Department of Parasitology, College of Veterinary Medicine, University of Georgia, Athens, Georgia.

Immature Dirofilaria immitis larvae collected from heavily infected donor ferrets soon after reaching the heart were highly successful in migrating to and developing in the heart of recipient ferrets after subcutaneous transplantation. Two to three worms of both sexes were used for both transplantation routes. Infections were not established by transfer of young adult worms into the peritoneal cavity. Live worms were recovered from the peritoneal cavity of only 3 of 33 ferrets given the worms by the intraperitoneal route, and worm recoveries in these ferrets were low. In regard to subcutaneous transplantation, worms were recovered from all ferrets receiving worms 93 to 107 days of age. Sixty-seven to 100 percent of the 93 to 107 day old worms reached the heart in recipient ferrets, but migration of older worms (117 to 128 days old) was lower and less predictable. Thus, transplantation of worms at 107 days of age is optimal in view of the relatively high worm burdens in donors and consistent recoveries of near 100% of the transplanted worms in the recipients. Earlier observations indicate that a worm burden of 4 to 6 heartworms is tolerated by ferrets and should produce useful patent infections.

15. DEVELOPMENT OF FOUR MONOCLONAL ANTIBodies AGAINST DIROFILARIA IMMITIS AND THEIR POTENTIAL IN THE DETECTION OF CIRCULATING ANTIGENS


Balb/c mice were immunized with soluble antigen prepared from canine Dirofilaria immitis. Two hundred and sixteen hybridomas produced by somatic cell fusion were screened by a solid-phase RIA and 83 were positive. These were cloned and 4 hybrid lines were selected for further study after growing in tissue culture or as ascites in mice. Hybridoma antibodies were classified using monospecific rabbit antimouse subclass specific reagents. Antibody reactivity was examined against antigens from *D. immitis* and *O. volvulus* by Ochterlony immunodiffusion and immuno-electrophoresis. Immunoprecipitation of radio-labelled antigens by monoclonal antibodies and serum from 40 dogs was examined by SDS-PAGE, followed by autoradiography. Circulating antigens in the serum of *Dirofilaria*-infected dogs, as well as in serum of non-infected dogs spiked with these antigens, could not be detected with this method. Apparently these antigens are blocked by substances present in *Dirofilaria*-infected, as well as non-infected dogs. Results of these and inhibition studies are discussed and their implications for heartworm diagnosis are considered.
16. DIAGNOSIS OF CANINE HEARTWORM INFECTIONS: EMPHASIS ON SERODIAGNOSIS OF OCCULT INFECTIONS
C.A. WAGNER, S.E. PRATT, and R.M. CORWIN, Department of Veterinary Microbiology, University of Missouri, Columbia, Missouri.

Diagnosis of canine heartworm disease is usually dependent upon the demonstration of Dirofilaria immitis microfilariae in blood, especially upon routine examination. Known exposure in endemic areas and clinical signs aid in presumptive diagnosis, and thoracic radiographic findings such as pulmonary arterial hypertrophy may be confirmative. But, because of potential amicrofilaremic infections, a procedure is needed which can accurately detect occult infections, is relatively easy to perform, and can be incorporated into the diagnostic evaluation of dogs for this condition.

An ELISA test is described and its use in the diagnosis of heartworm infection in random-source dogs and clinic patients will be reported for 1982-83. Serologic conversion of experimentally-infected dogs will also be discussed.

17. SEROLOGIC PATTERNS OF CANINE HEARTWORM INFECTION: THE LOUISIANA EXPERIENCE
L.T. GLICKMAN, R.B. GRIEVE, E.B. BREITSCHWERDT, M. MIKA-GRIEVE, G.J. PATRONEK, L.M. DOMANSKI, and C.R. ROOT, Department of Clinical Studies and Pathobiology, University of Pennsylvania School of Veterinary Medicine, Philadelphia, Pennsylvania and the Louisiana State University Veterinary Teaching Hospital, Baton Rouge, Louisiana.

Six hundred and two dogs at the Louisiana State University Veterinary Teaching Hospital were tested for antibodies to Dirofilaria immitis by ELISA using a purified somatic antigen. Each dog was also evaluated for heartworm infection by a complete white blood cell count and a Knott's test for circulating microfilariae. The serologic prevalence of heartworm infection was 34.7%; prevalence increased significantly with age up to 8 years and then decreased. Indoor dogs were significantly less likely to be infected as were dogs receiving diethylcarbamazine therapy. Coat length, sex, breed, and the presence of intestinal parasites were not associated with a positive heartworm ELISA test. ELISA titers showed a positive relationship with both eosinophil and basophil levels. Ninety-nine dogs that were evaluated radiographically were grouped according to results of the Knott's test and radiographic findings. These groups were negative Knott's test and negative radiographs (14 dogs), negative Knott's test and positive radiographs (57 dogs), and positive Knott's test and positive radiographs (28 dogs). The serologic prevalence of D. immitis infection in each of these groups was 35.7%, 56.1%, and 85.7%, respectively. The ELISA when used in conjunction with the Knott's test, exposure history, clinical signs, laboratory results, and radiographic findings, appears to be an accurate diagnostic tool and is useful for studying epidemiologic patterns of heartworm infection.
18. THE EFFECTS OF DECREASED LIVER FUNCTION ON THIACETARSEMIDE EFFICACY AND ARSENIC KINETICS IN DIROFILARIA IMMITIS THERAPY
R.A. HOLMES, K.W. PRASSE, R.C. WILSON, L.W. CORNELIUS, E.L. ROBERSON, M. BUMGARDNER, and J.W. McCALL, College of Veterinary Medicine, University of Georgia, Athens, Georgia.

Since thiacetarsamide is metabolized by the liver and excreted by the liver and kidney, the function of the liver could play a role in the efficacy of the drug. Decreased liver function was obtained by the surgical removal of approximately 60 to 70% of the liver, and confirmed by indocyanine green clearance tests. One month prior to hepatectomy, a known number of adult Dirofilaria immitis were transplanted via jugular vein into Beagles. The dogs were divided into 3 groups: (1) hepatectomy and treatment; (2) sham operated and treatment; and (3) sham operated no treatment. Therapy with thiacetarsamide (1.76 mg/kg bid for 2 days) was started on the third day post hepatectomy. On day 16 post hepatectomy, the dogs were euthanized and the heart and lungs examined and worms counted. There were significant statistical differences between numbers of worms killed in all 3 groups, with Group 1 having the highest kill rate followed by Groups 2 and 3, respectively. The conclusion is that decreased liver function results in a higher efficacy. The mode of action in this finding is not known, but is presumed that with decreased metabolism of the drug, the worms have increased exposure to the drug.

19. METABOLIC PROFILE OF ALBENDAZOLE

The metabolism of the broad-spectrum anthelmintic, albendazole, was examined in cattle, sheep, rats, and mice. After oral administration of radiolabelled drug, nine urinary metabolites were identified. The general metabolic profile was similar in all species studied. Very little albendazole was detected unchanged in urine, but the corresponding sulfoxide was a major urinary metabolite. The corresponding sulfone was not found to be a major component of urine; it is postulated, however, to be an important metabolic intermediate.

20. A CONTROLLED TEST OF FENBENDAZOLE AGAINST NATURALLY ACQUIRED INFECTIONS OF MONEZIA AND TRICHURIS IN YOUNG CATTLE

Fenbendazole has been shown to have excellent efficacy against many of the gastrointestinal nematodes of cattle. The present study was designed to evaluate the efficacy of fenbendazole against naturally acquired burdens of Monezia and Trichuris in young cattle. Working with treatment groups of ten calves, the efficacy of fenbendazole against Monezia was 64 and 94% at 5 and 10 mg/kg, respectively. Eight of the control animals remained infected at slaughter, compared to six of ten in the 5 mg/kg groups and two of ten in the 10 mg/kg group. With Trichuris, efficacy was 61% at 5
mg/kg and 97% at 10 mg/kg. All control animals were infected at necropsy with an average of 131 worms, while seven animals treated with 5 mg/kg fenbendazole harbored an average of 71 worms and four animals receiving 10 mg/kg averaged 10 worms per animal, with 33 worms in one animal.

21. PALATABILITY OF ALBENDAZOLE FEED PREMIX FORMULATION FOR CATTLE

Albendazole is a broad spectrum anthelmintic not yet cleared in the U.S. for the treatment of gastrointestinal nematodes, lungworms, liver flukes, and tapeworms. An albendazole feed premix is being developed for use in situations where treatment of individual animals is not feasible. Palatability of the albendazole premix was examined to determine whether cattle would accept the formulation under typical feeding conditions. Results of the study will be presented.

22. EFFECTIVENESS OF THE MORANTEL SUSTAINED RELEASE BOLUS IN THE PROPHYLAXIS OF BOVINE, PARASITIC GASTROINTESTINAL HELMINTHIASIS
B.L. PRESSON, T.A. YAZWINSKI, T.E. GREENWAY, and T.J. NEWBY, Department of Animal Sciences, University of Arkansas, Fayetteville, Arkansas.

Thirty Holstein calves with previous experience were allocated into two treatment groups (15 animals/group). The morantel sustained-released bolus (13.5 grams of morantel base activity) was administered to each calf in the designated treatment group. Each group was allowed to graze on pastures of equal area and previous contamination for the entire summer grazing season. For the 6-month grazing period the MSRB treated calves passed 93% fewer worm eggs than the control animals, thus substantially reducing pasture contamination by infective larvae. Every two months parasite-free tracer calves were allowed to graze on the two respective pastures. Tracer calves that grazed on the MSRB pasture were found to harbor 92% fewer worms than their control pasture counterparts. At the end of the grazing season, six calves from each group were necropsied and their worm burdens quantified. An 81% reduction in worm burdens (combined populations of Ostertagia ostertagi, O. lyrata, Trichostrongylus axei, Cooperia oncophora, C. punctata, C. mcmasteri, and Oesophagostomum radiatum) was evident in the MSRB treated calves. Finally, the MSRB animals outgained the controls by an average of 62.8 lbs over the entire trial period.

23. THE EFFICACY OF IVERMECTIN AGAINST OESTRUS OVIS IN SHEEP
R.A. RONCALLI, Merck Sharp & Dohme Research Laboratories, Rahway, New Jersey.

The sheep bot fly, Oestrus ovis, is encountered in sheep raised in many parts of the world. The action of ivermectin against this parasite was evaluated in controlled trials conducted
in Brazil. The results of these trials showed that ivermectin given orally to sheep at 200 mcg/kg of body weight was very effective (100%) against the three larvae stages of *Oestrus ovis*.

24. ANTHELMINTIC EFFICACY OF A FEBANTEL-PRAZIQUANTEL COMBINATION PASTE IN DOGS AND CATS
R.G. ARTHUR and D.D. COX, BAYVET Division of Miles Laboratories, Inc., Shawnee, Kansas.

The anthelmintic efficacy of a combination paste formulation containing 3.4% febantel plus 0.34% praziquantel was evaluated in 7 trials utilizing 80 dogs and 56 cats with natural and experimental helminth infections. One-half of the dogs and cats received 10.0 mg febantel + 1.0 mg praziquantel per kg body weight administered per os daily for 3 days. The other one-half of the animals received blank paste vehicle. The following mean critical efficacies were achieved for dogs infected with the following parasites: *Ancylostoma caninum* (28 infected) 97.3%; *Uncinaria stenocephala* (11 infected) 100%; *Toxocara canis* (12 infected) 85.1%; *Toxascaris leonina* (9 infected) 94.3%; *Trichuris vulpis* (20 infected) 100%; *Taenia pisiformis* (16 infected) 100%; *Dipylidium caninum* (10 infected) 100%. Controlled trial efficacy against experimental immature (i.e. 7-day old) *A. caninum* infections in 10 dogs was 100%.

In cats, mean critical efficacy against experimental and natural infections of *Ancylostoma tubaeforme* (14 infected) was 98.5%. Mean critical efficacy against natural *Toxocara cati* (25 infected) and *Toxascaris leonina* (3 infected) infections was 92.5 and 100%. Efficacy against experimental mature (15 infected) and immature (i.e. 7-day old) (10 infected) *Taenia taeniaeformis* infections was 100%.

25. PROPHYLACTIC EFFICACY OF CLORSULON AGAINST FASCIOLA HEPATICA IN CALVES AND SHEEP

A new fasciolicide clorsulon (MK-401) was examined as a potential prophylactic for ovine and bovine fascioliasis. A daily oral dose of clorsulon (5 mg/kg/day) for 21 days after inoculation with *Fasciola hepatica* cysts was highly effective in reducing worm burdens (98%) and preventing liver pathology in treated calves. Clorsulon was much less effective as a prophylactic in sheep, but delayed the onset of liver pathology and reduced the size of flukes recovered from sheep. Although clorsulon prevented development of fascioliasis in treated calves, the host antibody response was qualitatively similar to that of untreated infected calves although the magnitude of the response was reduced. Clorsulon levels in calf blood rose to 2.95 µg/ml within the first week, then fluctuated between 2.75 and 2.9 µg/ml for the next two weeks. Blood clorsulon levels in sheep were 0.5 to 0.6 µg/ml lower than that of calf blood. The difference in bioavailability of clorsulon
between sheep and calves appeared to account for the difference in efficacy of the drug.

26. BOVATEC® (LASALOCID) - A NEW ANTICOCCIDIAL FOR BEEF CATTLE
C.G. UNTAWALE and W.E. BRANDT, Department of Animal Science Research, Hoffmann-La Roche Inc., Nutley, New Jersey.

During the 17 trials conducted in 12 different geographical locations in the United States, the anticoccidial efficacy of lasalocid was evaluated at levels of 10 to 150 gm/ton of feed in feedlot cattle, or at 100 to 300 mg/head/day in pasture cattle. It is concluded from these data that (1) dietary levels of 25 to 30 gm lasalocid/ton of feed in feedlot cattle were significantly efficacious over unmedicated controls in the prevention of natural and induced bovine coccidiosis caused by the predominant species (Eimeria bovis and Eimeria zuernii) of coccidia and (2) in pasture cattle, dietary levels of 100 mg to 300 mg lasalocid per head/day were found significantly effective over unmedicated controls in the prevention of natural coccidial infections.

27. AN UPDATE ON WORLD-WIDE CLINICAL EXPERIENCE WITH EQVALAN®
WDH LEANING, Merck and Company, Rahway, New Jersey.

An Invited Review

28. EFFECTIVENESS OF OXFENDAZOLE AGAINST MIGRATING STRONGYLUS VULGARIS LARVAE IN PONIES
J.O.D. SLOCOMBE, B.M. McGRAW, P.W. PENNOCK, N.G. DUCHARME, and J.D. BAIRD, Departments of Pathology and Clinical Studies, Ontario Veterinary College, University of Guelph, Ontario, Canada.

In a controlled trial, 20 pony foals were reared parasite-free and inoculated with 500 S. vulgaris infective larvae. Two groups, each of four foals, were treated with a placebo, one group on Day 7 and the other on Day 56 following inoculation. Three groups, each of four foals, were treated with an oxfendazole suspension orally at 10 mg/kg of body weight, one group on Day 7, another on Days 7 and 9, and the third on Days 56 and 58.

Three groups of foals, one treated with the placebo on Day 7, another with oxfendazole on Day 7, and the third with oxfendazole on Days 7 and 9 were necropsied on Day 28. Two groups of foals, one treated with the placebo on Day 56 and the other with oxfendazole on Days 56 and 58 were necropsied on Day 91. Efficacy was judged on lesions and numbers of larvae found at necropsy. Oxfendazole was 80.0% effective against early 4th-stage larvae in the early intestinal phase when given only on Day 7 and 94.9% effective when given on Days 7 and 9. Oxfendazole was 96.6% effective against later 4th stage larvae in the mesenteric phase when given on Days 56 and 58.
29. CRITICAL AND CLINICAL STUDIES ON VET 220, A BENZIMIDAZOLE ANTHELMINTIC, AGAINST EQUINE PARASITES

A critical study was done to titrate the anthelmintic dosage of VET 220, a benzimidazole compound, against equine parasites. There were 24 mixed-bred ponies which were naturally-infected with Strongylus vulgaris, S. edentatus, and small strongyle species as determined by fecal EPG, LPG, and larval differential values. Dosages compared were 2.5, 5.0, and 10.0 mg/kg BW and placebo given by nasogastric intubation. All ponies were necropsied at 7 days posttreatment. Strongylus vulgaris and S. edentatus were completely removed by all dosages. Small strongyle removal was variable and dose-dependent. Habronema spp., Gastrophilus spp., and Anoplocephala spp. were not affected.

The appearance of the intestinal lining was dose-dependent: the mucosae of the ponies treated at 5.0 and 10.0 mg/kg BW had a clean, smooth appearance; that of ponies given 2.5 mg/kg BW were clean, but nodular and moderately reactive to embedded immature small strongyles.

A limited clinical titration was done in 12 ponies whose fecal cultures had no S. vulgaris larvae, although other parasites were present. The ponies were treated in pairs at these dosages: 0 (placebo), 0.5, 1.0, 2.5, 5.0 mg/kg BW of VET 220, and at 5.0 mg/kg BW of fenbendazole as a comparative treatment control. Based on pre- and posttreatment EPG and LPG values, VET 220 at dosages of 1.0 to 5.0 mg/kg BW was highly effective and similar to fenbendazole given at 5.0 mg/kg BW. Small strongyles were most responsive to effect of FBZ and of VET 220 at all levels. Strongylus edentatus was removed by FBZ, by VET 220 at 2.5 and 5.0 mg/kg BW, and partially removed by VET 220 at 1.0 mg/kg BW. Trichostrongylus axei was removed by VET 220 at 2.5 and 5.0 mg/kg BW, but not by 1.0 mg/kg BW dosage. Parascaris equorum was removed from the single infected pony by VET 220 at 5.0 mg/kg BW.

30. BENZIMIDAZOLE RESISTANT SMALL STRONGYLES IN HORSES IN SOUTHEASTERN PENNSYLVANIA: PREVALENCE AND FACTORS LEADING TO DEVELOPMENT OF RESISTANCE
C. UHLINGER and C. JOHNSTONE, Departments of Clinical Studies and Pathobiology, University of Pennsylvania School of Veterinary Medicine, Philadelphia, Pennsylvania.

The prevalence of benzimidazole resistance was investigated in 18 stables with a total of 316 horses. Resistance, ranging from 75 to 100% was shown in 15 stables, in horses on rotational drug schedules and in horses treated as infrequently as twice a year. Only 3 herds (on an inadequate strongyle control program, i.e. fewer than two anthelmintic treatments per year) harbored strongyles highly susceptible to benzimidazoles. Strongyle resistance to the following drugs was recorded: cambendazole, fenbendazole, mebendazole, oxfendazole, and thiabendazole. Cross resistance among these drugs was found in herds with benzimidazole-resistant strongyles. Significant fecal egg counts were found in most horses between 6 and 8 weeks after treatment with an effective
anthelmintic. Pyrantel pamoate, benzimidazole-piperazine combinations and piperazine-carbon disulfide combinations were effective against benzimidazole-resistant small strongyles.

31. ANTIPARASITIC ACTIVITY OF CLOSANTEL IN HORSES
B.P. CAMPBELL and J. GUERRERO, Pre-Clinical Research Department, Pitman-Moore, Inc., Washington Crossing, New Jersey.

In a series of controlled experiments, utilizing combinations of natural and experimental parasite infections, the prophylactic and therapeutic anthelmintic activity of Closantel was examined in a total of 33 pony or horse foals and 17 yearling or adult horses. At a dose of 8 mg/kg, Closantel effectively prevented development of Gasterophilus spp. larvae in foals and horses when administered orally at 2 month intervals under natural pasture conditions. Closantel also demonstrated excellent prophylactic activity against migrating larvae of Strongylus vulgaris at doses of 20 and 40 mg/kg. High therapeutic activity against adult S. vulgaris, Strongylus edentatus, Parascaris equorum, Triodontophorus spp., and Anoplocephala perfoliata as well as Gasterophilus intestinalis was found when Closantel was given at doses of 15 or 20 mg/kg.

32. CRITICAL TESTING OF THE ACTIVITY OF FENBENDAZOLE AND FEBANTEL AGAINST PARASCARIS EQUORUM IN PONIES.
J.A. DiPIETRO, A. PAUL, T.F. LOCK, and K.S. TODD, Departments of Veterinary Clinical Medicine and Veterinary Pathobiology, College of Veterinary Medicine, University of Illinois, Urbana, Illinois.

The activity of fenbendazole and febantel was evaluated in 12 pony foals which were inoculated with 2600 P. equorum eggs. Once patent for P. equorum, the foals were randomly assigned to and treated one time intraorally with either 0.5 ml corn syrup/kg (controls; n = 4), 10 mg fenbendazole/kg (n = 4), or 6 mg febantel/kg (n = 4). Foals were necropsied and examined for parasites 10 days after treatment. Fenbendazole and febantel were highly effective against adult and immature P. equorum. Gross lesions attributed to P. equorum were evident in all foals. P. equorum were not found in any of the fenbendazole- or febantel-treated foals. The mean number of adult and immature P. equorum found in the controls was 66.8 (15-166) and 65.0 (21-147), respectively. Strongyle infections were insufficient for efficacy evaluations to be done. Limited activity of fenbendazole and febantel occurred against Draschia megastoma, Habronema majus, and H. muscae adults but not immatures. Neither anthelmintic was effective against Gasterophilus intestinalis or late 4th stage larvae of Strongylus vulgaris. Adverse side effects due to treatment were not observed.
33. EFFICACY OF ALBENDAZOLE AGAINST VERY IMMATURE FASCIOLA HEPATICA IN SHEEP

The objective of this experiment was to determine the age of F. hepatica when effectively killed by albendazole. Forty lambs were allocated in groups of five and kept in an indoor barn and fed hay of good quality and pelleted commercial feed. Three hundred metacercariae were given orally in a gelatin capsule to each lamb (150 metacercariae in the morning and 150 in the afternoon of the same day). Albendazole 5 mg/kg b.w. was administered intraruminally. At the completion of the experiment, 14 weeks post-infection, the efficacy of albendazole was as follows: 100% protection in lambs treated on the day of the infection and for the ensuing 35 days, 90.4% in lambs treated from day 1 to 7 post-infection, 79.7% by lambs treated from day 8 to 14 post-infection, 71.1% in lambs treated from day 15 to 21 post-infection, 63.5% in lambs treated from day 22 to 28 post-infection, and 21.8% in lambs treated from day 29 to 35 post-infection. There was a discernible eosinophilia in all infected animals with the exception of those animals treated with albendazole from day 1 to 35 post-infection.

34. THE EFFICACY OF ALBENDAZOLE AGAINST FASCIOLOIDES MAGNA IN SHEEP
B.E. STROMBERG, J.C. SCHOLTTHAUER, and G.A. CONBOY, Department of Veterinary Pathobiology, College of Veterinary Medicine, University of Minnesota, St. Paul, Minnesota.

Sheep experimentally infected with Fascioloides magna were used in a controlled study to determine the flukicide activity of several doses of albendazole (methyl[5(propylthio)-1H-benzimidazole-2-Yl]carbonate). Sheep, about four months of age, were infected with 100 metacercariae of F. magna and treated with various doses of albendazole 10 weeks after infection. Six weeks after treatment, the sheep were necropsied, flukes were recovered and measured, and the pathology recorded.

The sheep were given different doses of albendazole (5.0, 7.5, 10.0, 15.0, and 2 x 7.5 mg/kg) and were compared to both infected-untreated and uninfected-untreated controls. Comparing treated animals with the untreated controls there was up to a 70% reduction of worms recovered in the 2 x 7.5 mg/kg dosage group. There was also a significant reduction in flukes recovered in the 7.5, 10.0, and 15.0 mg/kg body weight groups. Because a single F. magna fluke has the potential to kill a sheep, it becomes imperative to consider the number of sheep with no flukes after treatment. Using that parameter, 50% of the sheep were protected at 7.5, 10.0, 15.0, and 2 x 7.5 mg/kg body weight.

35. WHAT'S YOUR DIAGNOSIS?
C.H. GARDINER, Armed Forces Institute of Pathology, Washington, D.C.

An Invited Presentation
36. EXPERIMENTAL ISOSPORA SUIS INFECTIONS IN PIGLETS

Four- to 8-day old piglets, taken off sows at 1 to 3 days of age, were inoculated per os with 50,000 to 800,000 sporulated oocysts of Isospora suis. Piglets given more than 200,000 oocysts usually died from the infection. Severity of diarrhea early in the infection was directly related to the number of oocysts given. This relationship was not apparent once piglets developed diarrhea. The prepatent period was 5 to 6 days. The patent period usually lasted 3 to 8 days, but was occasionally longer. Some piglets had diarrhea for several days after oocyst production had ended; some pigs died during this period. One piglet had a rectal prolapse. This animal did not have severe diarrhea during the infection, but did have an extended patent period. A fibrinecrotic membrane was present in the small intestine of piglets necropsied during the infection. Parts of this membrane were passed by some piglets during the course of the infection.

37. ENHANCED VIRULENCE AS THE REASON FOR APPARENT RESISTANCE OF AVIAN COCCIDIA TO IONOPHORES
M.D. RUFF, U.S. Department of Agriculture, Agricultural Research Service, Animal Parasitology Institute, Beltsville, Maryland.

Classic drug resistance, the loss of reduction of efficacy with continuous use of an anticoccidial, has been known for many years with avian coccidia. Cross-resistance to closely related compounds has also been demonstrated. Resistance to monensin, however, has been slow to develop although resistance to other ionophores has developed in the field. Studies at our laboratory have shown that strains resistant to monensin can be developed. Furthermore, in some cases, decreased control has been shown to result from increased virulence rather than because of classic drug resistance. This means that even changing to an unrelated class of compounds may not affect control.

38. THE EFFECT OF HIGH FIBER DIETS ON INTESTINAL EIMERIA SPP. INFECTIONS IN THE DOMESTIC RABBIT
L.M. POTE and T.A. YAZWINSKI, Department of Animal Science, University of Arkansas, Fayetteville, Arkansas.

The purpose of the study was to establish the type and prevalence of parasites in the domestic rabbit. Additional objectives were to determine the interaction of low or high fiber diets on intestinal Eimeria spp. infections with respect to oocyst output and speciation, morbidity, fecal and urine output, histological observations, digestibility of the feedstuffs, weight gain, and long term growth. Fecal samples collected from commercial rabbitries showed Eimeria spp. and Passalurus ambiguus to be the primary parasites present. Correlations were established as to management practices of the rabbitries, the sex and age of rabbits, and the type and severity of parasitic infections found.
It was observed that rabbits had the highest coccidia infections while does had the highest *P. ambiguus* infections. A digestibility trial established significant differences between rabbits fed high or low fiber alfalfa diets. The higher fiber diet appeared to cause decreased severity of coccidiosis. Preliminary findings in a second digestibility study showed with even higher coccidia levels, high fiber diets decreased the pathogenicity of the infection. A long term growth trial showed the effects of high coccidia infections during the 5 to 9 week growth period on growth, morbidity, and feed conversions.

39. **IN VITRO CULTIVATION OF THE SWINE COCCIDIA ISOSPORA SUIS**


*Isospora suis* oocysts obtained from the feces of experimentally infected pigs were sporulated, cleaned from fecal debris by density gradient centrifugation, and subjected to in vitro excystation techniques to release sporozoites. Thirty minutes after initiating excystation, sporozoites were suspended in culture medium and inoculated into Leighton tube cultures of Madin-Darby bovine kidney, bovine colon, embryonic bovine trachea, and porcine kidney cells. Sporozoites became intracellular within 1 hour and underwent development by endodyogeny within 48 hours in all cell types examined. As many as 20 progeny were found in a single cell at 120 hours. Periodic acid Schiff (PAS) staining revealed changes in the carbohydrate content of sporozoites. Those within sporocysts were strongly PAS-positive. At 30 minutes after initiating excystation there was a significant decrease in the number of PAS-positive granules present. In smears of sporozoites incubated for 120 minutes in excystation fluid and in intracellular sporozoites examined 60 minutes after inoculation into cell cultures, few or no PAS-positive granules were present. In intracellular sporozoites examined after 24 hours in culture and in progeny after 48 hours several granules were present. Thus, PAS-positive granules appear to decrease concomitant with energy-intensive processes such as excystation and host-cell penetration and then to increase within intracellular sporozoites and progeny.

40. **PRESIDENT'S ADDRESS**

J.F. WILLIAMS, Department of Microbiology and Public Health, Michigan State University, East Lansing, Michigan.

41. **AUTORADIOGRAPHIC TRACING OF PARASITE MIGRATIONS**

J.R. GEORGIES, Department of Preventive Medicine, New York State College of Veterinary Medicine, Cornell University, Ithaca, New York.

Radioselenomethionine was incorporated into the infective larvae of such diverse parasites as *Schistosoma mansoni*, *Fasciola hepatica*, *Brugia malayi*, and *Haemonchus contortus* by exposing the infected intermediate host (*S. mansoni*, *F. hepatica*, and *B. malayi*) or culture medium (*H. contortus*) to $^{75}$Se-methionine during
development. No effect of gamma radiation arising from decay of label $^{75}\text{Se}$ in the range 130 to 1311 pC/larva could be measured in terms of survival or sex ratio of $H.\ contortus$ worms recovered at 17 days after infection. Autoradiography of flattened, dried organs permitted quantification of larva distribution in the bodies of mice for up to 21 days after infection with $^{75}\text{Se}$ labeled schistosomula of $S.\ mansoni$. Autoradiography of fourth and fifth stage $H.\ contortus$ worms arrayed on cards permitted differentiation of labeled and unlabeled individuals for 35 days after infection with a mixture of $^{75}\text{Se}$-labeled and non-labeled infective larvae. It was concluded that autoradiography offers the specificity and sensitivity required to quantify weakly radioactive migrating parasites in the presence of spurious radioactivity from catabolized label and to distinguish between labeled and non-labeled individuals when these can be isolated from the host's tissues.

42. ASSESSMENT OF PULMONARY MIGRATION OF ASCARIDS (BAYLISASCARIS, ASCARIS) BY BRONCHOPULMONARY LAVAGE
W.K. WYAND-OUELLETTE, K.R. KAZACOS, and D.B. DENICOLA, Department of Veterinary Microbiology, Pathology and Public Health, Purdue University, West Lafayette, Indiana.

The responses to pulmonary migration of ascarids [$Baylisascaris\ procyonis$ (BP), $Ascaris\ suum$ (AS)] were studied in mice by sequential cytologic and biochemical analysis of bronchopulmonary lavage fluid. Parameters evaluated included total erythrocyte (RBC) and nucleated cell (NCC) counts, differential counts, total lactate dehydrogenase (LDH) activity and total protein levels in the lavage fluid. Two groups of 130 mice each received 250 (BP-L) or 1000 (BP-H) BP eggs; 150 mice each received 1000 (AS-L) or 5000 (AS-H) AS eggs; and 40 controls (CON) received 0 eggs. Ten mice per group were killed at 6 and 12 hours then every 1 to 2 days, and their lungs lavaged with two aliquots of 0.7 ml sterile saline. A marked increase in RBC's was seen on day 1 in BP mice, peaking on d 2 to 3 at up to 130X CON (BP-H); levels returned to normal by d 7 to 8. In AS mice, RBC's rose slowly to peak at d 7 to 8 at 30X CON (AS-H). A biphasic NCC response was seen in BP mice, peaking on d 3 and 8, at 7 to 9X CON (BP-H); the early peak was due to PMN's (210X) and macrophages (3X) and the latter to eosinophils (209X) and macrophages (4X). AS mice had a steady rise in NCC, peaking on d 13 at 12X CON (AS-H); PMN's peaked d 10 (34X), eosinophils d 15 (639X), and macrophages d 20 (3X). LDH and protein levels paralleled RBC responses, to 32X CON in BP-H and 5X CON in AS-H. Bronchopulmonary lavage proved valuable for evaluating and comparing pulmonary responses to ascarid migration, and clearly showed differences in the two species; BP was far more aggressive and caused more severe lung damage much earlier than AS.
43. STUDIES ON PRENATAL AND LACTOGENIC TRANSMISSION OF TOXOCARA CANIS AND ANCYLOSTOMA CANINUM I. EXPERIMENTAL INFECTION OF THE BITCH BEFORE PREGNANCY

E.L. ROBERSON and T.M. BURKE, Department of Parasitology, College of Veterinary Medicine, The University of Georgia, Athens, Georgia.

Breeding of 10 Beagle bitches (5 parasite-free and 5 which had been previously experimentally infected with 6000 Toxocara canis eggs orally and 2500 Ancylostoma caninum larvae subcutaneously) was arranged so that pairs of bitches (1 uninfected, 1 infected) whelped simultaneously. Pups born to an infected bitch were removed at birth and nursed by the paired uninfected bitch until 4 weeks of age when pups were necropsied to determine the number of parasites they had acquired prenatally from their infected mother. Pups born to the parasite-free bitch were nursed by the infected bitch until necropsied at 4 weeks of age to determine the number of parasites passed via the lactogenic route. Of 680 ascarids transmitted to pups by either route, 98.5% were transmitted prenatally, 1.5% lactogenically. Transmission of 2746 hookworms to 22 pups occurred solely by the lactogenic route; prenatal transmission of this parasite did not occur in any of the 25 pups born to infected bitches.

44. STUDIES ON PRENATAL AND LACTOGENIC TRANSMISSION OF TOXOCARA CANIS AND ANCYLOSTOMA CANINUM II. (a) EXPERIMENTAL INFECTION OF THE BITCH AT MIDPREGNANCY AND AT PARTURITION (b) LONGEVITY OF LACTOGENIC TRANSMISSION

T.M. BURKE and E.L. ROBERSON, Department of Parasitology, College of Veterinary Medicine, The University of Georgia, Athens, Georgia.

The degree and longevity of lactogenic transmission of Toxocara canis and Ancylostoma caninum to pups were studied in Beagle bitches which were experimentally infected with both parasites (10,000 eggs of T. canis orally, 10,000 larvae of A. caninum subcutaneously) at either midpregnancy (2 bitches) or within 48 hours following parturition (2 bitches). Pups born to infected bitches were removed at birth and raised by a surrogate parasite-free mother. At the beginning of each week of lactation, a new group of 4 parasite-free pups was placed with each infected bitch. Parate-free pups were provided by 15 uninfected laboratory Beagle bitches which were bred to whelp simultaneously with the 4 infected dogs. After a group of 4 pups had nursed an infected dog for 1 week, the group was transferred to an uninfected bitch for another 3 weeks of nursing after which the pups were necropsied to recover and tabulate parasites acquired lactogenically from the infected bitch. If a pup died at <1 week after nursing, its liver, lungs, and GI viscera were subjected to pepsin-HCl digestion to recover larval stages. Each of the 15 uninfected bitches retained one of its own pups as a control to substantiate the bitch's parasite-free status.

With midpregnancy infections, 95% of 5842 ascarids were transmitted prenatally by 2 bitches to their own offspring and 4.5% or 263 ascarids were transmitted lactogenically to weekly rotated pups. Ascarids were transmitted via the milk during each of 5
weeks of lactation—consistently higher numbers during weeks 1 to 3 and lower numbers during weeks 4 and 5. Infection of 2 bitches at parturition denied ascarids the prenatal route of transmission but resulted in greater numbers of ascarids being passed through the milk—388, 612, 373, and 214 ascarids for each of 4 consecutive weeks of lactation.

Regardless of when infection of bitches occurred, hookworms were transmitted only by the lactogenic route. With midpregnancy infections, 63% of 610 hookworms were transmitted by 2 bitches during the 1st week of lactation; decreasing numbers (17%, 12%, 7.5%, and <1%) were passed in subsequent weeks 2 to 5, respectively. With 2 bitches infected at parturition, relatively even numbers of hookworms were transmitted through the milk during the first 3 weeks of nursing (27.9% of 655 worms, 36.9%, and 26.7%, respectively) and a lower number (8.4%) was passed during the 4th week.

45. INFLUENCES OF SELENIUM INTAKE ON INFECTIONS BY NEMATODES AND COCCIDIA IN RATS AND GOATS
J.C. FRANDSEN, U.S. Department of Agriculture, ARS, Regional Parasite Research Laboratory, Auburn, Alabama.

The periodic injection of a commercial sodium selenite and vitamin E preparation into goats fed a diet low in Se, protein and energy, and infected with Trichostrongylus colubriformis and mixed species of coccidia reduced mortality caused by coccidiosis and was associated with better weight maintenance as compared with control animals injected with the vehicle only. Experiments conducted on rats fed a diet adequate in vitamin E but congenitally deprived of Se and born to dams deprived of Se since the onset of gestation demonstrated that (1) in infections by Nippostrongylus brasiliensis there was no effect produced on response to challenge infections or the development of DTH to KLH by 0.1 ppm Se in the diet or the injection of the same preparation (at the same basic rate) used in the goats; and (2) in infections by Eimeria nieschulzi there was no effect produced on oocyst production, response to challenge infections, feed:gain ratios, or the development of DTH to KLH produced by 0.1 ppm Se in the diet.

46. PATHOPHYSIOLOGY OF FASCIOLOIDES MAGNA IN SHEEP
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The pathophysiologic responses of seven (7) sheep infected orally with 100 metacercaria of Fascioloides magna were monitored for four (4) months post-infection and compared with parameters established for an equal number of uninfected control animals. Sheep were routinely weighed and bled. A CBC was run weekly for five weeks and then bi-weekly to the end of the experiment. Serum was collected on alternate weeks and examined for alkaline phosphatase, aspartate aminotransferase, alanine aminotransferase, y-glutamyl transferase, total bilirubin, blood urea nitrogen,
creatinine, inorganic phosphorous, calcium, albumin, chloride, potassium, and sodium. Four months after infection, all sheep were necropsied and flukes recovered.

There were no differences in weight gains between the two groups during the experiment. No significant differences were observed in the CBCs of the two groups until week 13 post-infection, when infected animals began to show eosinophilia. No differences in serum parameters were observed. Gross lesions attributed to fluke migration were found, in the liver and portal lymph nodes, diaphragm, lungs, kidney, and spleen. Microscopically, principal liver lesions occurred in the portal areas, veins, and capsule, and were characterized by both active and chronic inflammation. Abundant infiltrates of eosinophils and plasma cells often marked the portal areas.

47. **TAENIA HYDATIGENA INFECTIONS IN OLD WORLD MONKEYS**


Ten of 52 African Green monkeys (Cercopithecus aethiops) and 10 of 26 African Red monkeys (Erythrocebus patas), wild-trapped for use in medical research in Sudan, were found to be infected with abdominal cysticerci. Some of the animals had grossly enlarged, protruding abdomens. In all cases the cysticerci were within the peritoneal cavity or attached to the serosal surfaces of viscera or omentum. Parasites varied from 0.5 to 5 cm in diameter, bore a single scolex and the rostellar hook measurements and form were typical of those of *Taenia hydatigena*. This parasite is common in small ruminants in the Sudan. Although few records exist, isolated instances of *T. hydatigena* have been described in sub-human primates in East and West Africa. Infections of the intensity and frequency seen in these Sudanese monkeys are unusual and are difficult to explain in terms of the normal sheep/dog transmission cycle which characterizes this parasite elsewhere.

48. **EXPERIMENTAL VISCERAL LEISHMANIASIS IN DOMESTIC CATS**

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Ten laboratory-reared domestic cats were inoculated intravenously with amastigotes of *Leishmania donovani infantum* (strain WR 438). Pairs of cats were killed at 1 hour and at 1, 2, 4, and 8 weeks of infection and examined for parasites and evidence of parasite-induced pathology. Examination of liver and spleen impression smears revealed modest growth of the parasites in these tissues. Bone marrow was cultured and found to be positive for *Leishmania* in all cats, while whole blood was culture-positive in a few individuals. *Leishmania* antibody titers, as measured by an indirect fluorescence technique, rose ten-fold over the eight-week observation period. Animals remained afebrile throughout infection, and no signs of illness or gross pathology were seen.
The results of this experiment suggest a possible role for domestic cats as inapparent reservoirs of human visceral leishmaniasis.

49. DEVELOPMENT OF IMMUNITY TO ASCARIS SUUM IN SWINE AFTER NATURAL INFECTION
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The development of protective immunity to infection with Ascaris suum in pigs was determined after natural exposure to eggs on contaminated lots. Protective immunity was defined as the percent reduction in the recovery of larvae from the lungs of principals, compared to control, seven days after challenge with \(10^4\) infective eggs. In addition, the number and intensity of liver lesions were recorded. Two litters, each containing eight pigs, were split into four groups as follows. Litter #1 was farrowed and weaned six weeks later on a contaminated lot; four pigs (group 1) were then transferred to another contaminated lot while the remaining four pigs (group 3) were transferred to helminth-free stalls. Litter #2 was farrowed and weaned six weeks later in helminth-free stalls; four pigs (group 2) were transferred to the contaminated lot while the remaining four pigs (group 4) were maintained in helminth-free stalls (groups 3 and 4 were anthelmintic-treated four weeks after weaning). Seventeen weeks later pigs from groups 1, 2, 3, and 4, along with a group of six week old normal pigs (group 5), were challenged. No significant differences were observed in the recovery of lung larvae from groups 3, 4, and 5; however, groups 1 and 2 were >98% protected from challenge. In addition, markedly fewer liver lesions were seen in groups 1 and 2 than groups 3 and 4. These results indicate the development of a strong internal immunity after chronic natural infection and that resistance to challenge infection is not simply related to age.

50. IMMUNODIAGNOSTIC ANTIGENS FOR SWINE TRICHINOSIS ISOLATED USING MONOCLONAL ANTIBODIES

The usefulness of serologic tests such as the enzyme-linked immunosorbent assay (ELISA) for the diagnosis of swine trichinosis has been hindered by the occurrence of a significant rate of "false-positive" reactions in swine found negative by direct methods. In an attempt to identify antigens for diagnosis that are unique to Trichinella spiralis, monoclonal antibodies were prepared against the parasite. Many of the antibodies generated recognized antigenic determinants also present in other swine parasites, particularly Trichuris suis. One monoclonal antibody recognized an antigen unique to T. spiralis, present in excretory-secretory (ES) products of the worm, and apparently elaborated through the oral opening of the worm. This antigen was isolated by monoclonal antibody-affinity chromatography and used in a triple-antibody ELISA procedure. The antigen effectively eliminated false-positive
reactions in sera known to produce these reactions when tested with crude worm extracts, and reliably detected all true positive (infected) pigs.

51. LYMPHOYCYTE REACTIVITY TO OSTERTAGIA OSTERTAGI L₃ ANTIGEN IN TYPE I OSTERTAGIASIS
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To better understand the immune response of calves infected with Ostertagia ostertagi, studies were conducted to examine cell-mediated immune (CMI) responses to L₃ antigen and phytohemagglutinin (PHA) by lymphocyte reactivity assay (LRA). Consecutive multiple-dose inoculated calves showed marked increases in stimulation indices (SI) to L₃ antigen over naturally inoculated calves (56.5 and 25.8, respectively). Negative SI were obtained in calves of the uninoculated control group (-1.0). There was significant (p<0.01) agreement between positive SI and evidence of patent infection (development of Type I ostertagiasis). Kinetic studies of lymphocyte reactivity to L₃ antigen and PHA showed that responses were briefly suppressed to both of these stimuli in prepatent calves. It was concluded that transient nonspecific suppression of CMI responses occurs during prepatent periods of Type I ostertagiasis. This immunosuppression phenomenon may play a role in the pathogenesis of O. ostertagi and in development of immunity to Type I ostertagiasis.

52. ANTIGEN-ANTIBODY COMPLEXES FROM THE SERA OF DOGS WITH FILARIAL INFECTIONS
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Dogs naturally infected with Dirofilaria immitis or experimentally infected with Brugia pahangi furnished sera to be studied for the presence of immune complexes typically associated with microfilaremic filarial infections. Serum fractions eluting in the void volume from column chromatography over BioRad A-50m (exclusion molecular weight, 50,000,000) contained IgM, IgG, and C3. Dissociation of the complexes with 8M urea followed by ultrafiltration to separate immunoglobulins and complement molecules greater than 100,000 in molecular weight from possible parasite antigens less than 100,000 MW yielded a substance which has been incompletely characterized at this time. Electron microscopy of microfilariae freshly collected from sera or re-exposed to dog sera in vitro demonstrated immunoglobulin binding to cuticle and sheath surfaces and suggested potential sources of antigen-antibody complexes in microfilaremic filarial infections.
53. ACQUIRED IMMUNITY TO TRICHINELLA SPIRALIS IN SWINE: INFLUENCE OF PARASITE DENSITY

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The population dynamics of Trichinella spiralis in naive and chronically infected swine is of interest because of their relevance to the epidemiology and immunology of swine trichinosis. A series of experiments are in progress to ascertain the influence of infection level on primary worm expulsion and on host response to secondary exposure; the initial findings are reported here.

Hogs inoculated with either 500 or 10,000 muscle larvae (L₁) exhibited nearly identical intestinal parasite population changes. The intestinal worm burdens were 15 to 18% of larval inoculum size and remained at that level until about 6 weeks postinoculation (PI); at that time, the adult worm numbers fell until reaching a level of about 2% at day 79 PI.

Hogs inoculated initially with 10,000 L₁ and challenged with 55,000 L₁ on day 55 PI did not exhibit significant reduction of challenge intestinal worms at day 12 postchallenge (PC), but did expel 69% of the adults by day 18 PC. Hogs inoculated initially with 500 L₁ and challenged with 28,000 L₁ on day 51 PI had expelled 96% of challenge adult worms 22 days later. Muscle larvae recovered from these previously infected pigs were 98% fewer than the controls at day 22 PC. Experiments are now in progress to determine if the muscle larvae reduction is due to immune effects on the intestinal adults or to a protective response specific to newborn larvae. Similar experiments are underway to investigate the population dynamics and immune response to T. spiralis in swine inoculated with as few as 10 or 50 L₁; the resulting muscle larvae densities at this exposure level should closely approximate the densities observed in farm-raised swine.

54. THE ANTIBODY RESPONSE TO SARCOCYSTIS OF EXPERIMENTALLY INFECTED CATTLE AND SHEEP DETERMINED IN AN ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA)

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An enzyme-linked immunosorbent assay (ELISA) system was developed to assess the humoral immune response of cattle and sheep experimentally infected with Sarcocystis. A soluble protein antigen was obtained by mechanical disruption of zoites from intramuscular cysts of Sarcocystis boviscanis or S. ovicanis. The test for bovine antibody was an indirect ELISA using subclass-specific anti-bovine reagents. Experimentally-infected calves had measurable serum Sarcocystis-specific IgM antibodies 3 to 4 weeks after infection, and measurable IgG₁ responses as early as 5 to 6 weeks after infection. IgM reached a peak 7 to 8 weeks after infection and then decreased; IgG₁ levels continued to rise until 9 to 10 weeks after infection, and remained high for at least 30 weeks. Sarcocystis-specific bovine IgG₂ and IgA response were not detected by the ELISA. The test used in sheep was a direct ELISA using class-specific anti-sheep reagents. Sheep serum had
measurable IgG antibody responses at 9 to 10 weeks postinfection, with the levels of Sarcocystis-specific IgG antibody remaining high in excess of 20 weeks. Sarcocystis-specific IgM responses were not demonstrable in sheep. S. ovicanis or S. bovicanis antigens worked equally well in the ELISA system for detecting anti-Sarcocystis antibody from animals infected with the homologous or heterologous parasite species.

55. ANTIBODY DEPENDENT CELL MEDIATED ADHERENCE OF STRONGYLUS VULGARIS THIRD STAGE LARVAE IN VITRO
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The adherence of equine circulating white blood cells to Strongylus vulgaris L3 was studied in vitro using parasite-free pony sera, hyperimmune pony sera, buffy coat cells and enriched populations of neutrophils (PMN), eosinophils (EOS), and mononuclear cells. A nonspecific heat labile factor and species specific antibody were shown to mediate adherence by all cell populations. The antibody mediated adherence could be reduced by absorption with adult antigen. Adherence of EOS enriched populations with immune sera produced encapsulated larvae which coiled tightly and became immobile. This phenomena was not observed using other cell types including EOS from parasite free ponies.