

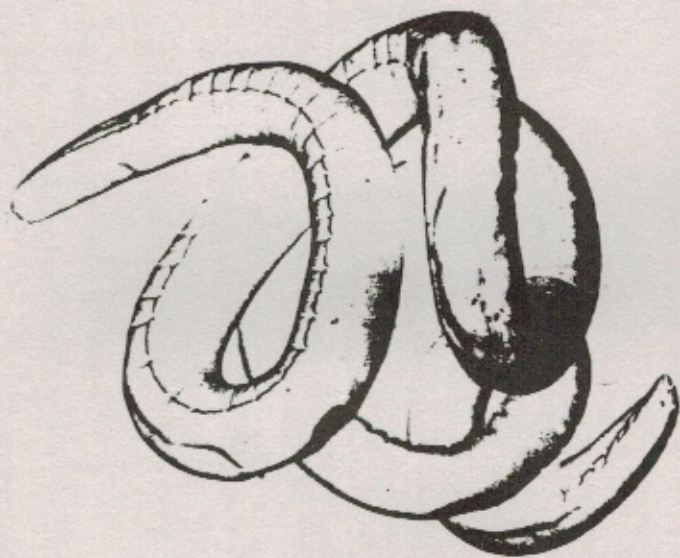
AMCOP XXXVI

RECORDS

ANNUAL MIDWESTERN CONFERENCE
OF PARASITOLOGISTS

Special Topic

IMMUNITY TO HELMINTHS



University of Iowa

Iowa City June 7-9, 1984

AMCOP XXXVI 1984

Affiliate American Society of Parasitologists

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ACKNOWLEDGMENTS

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XXXVI ANNUAL MIDWEST CONFERENCE OF PARASITOLOGISTS

University of Iowa, Iowa City, IA

June 7-9, 1984

Program Schedule

June 7

- 4:00-10:00 p.m. Check-in and Registration, Burge Residence Hall, Clinton Street
- 8:00-12:00 p.m. Party at Cain's, 806 Alpine Drive, Iowa City

June 8

- 8:00-11:00 a.m. Registration, Coffee and Donuts. Main Entrance Lobby, Van Allen Hall Lecture Rooms
- 9:00-9:15 a.m. Welcoming Remarks - D. C. Spriestersbach, Vice President for Research and Dean of the Graduate College
- 9:15-11:30 a.m. General Session - Contributed Papers, Lecture Room 2, Van Allen Hall
- 1:00-3:00 p.m. Symposium--Helminth Immunology, Lecture Room 2, Van Allen Hall

Speakers: Dr. Bert Stromberg, College of Veterinary Medicine, University of Minnesota, "Immune Response to Liver Flukes."

Dr. Robert Grieve, School of Veterinary Medicine, University of Wisconsin, "Studies on Antigens of Parasitic Nematodes."

- 3:00-4:30 p.m. Demonstration Session, Rooms 122, 133, 137, Zoology Building
- 4:30 p.m. Business Session, Lecture Room 2, Van Allen Hall
- 6:00 p.m. Social Hour (Cash Bar), Iowa Memorial Union
- 7:00 p.m. Banquet, Iowa Memorial Union

Speaker: Dr. John Donelson, Department of Biochemistry, University of Iowa, "Genetic Rearrangements and the Basis of Antigenic Variation in African Trypanosomes."

June 9

- 8:00-9:00 a.m. Coffee and Donuts, Main Entrance Lobby, Van Allen Hall
- 9:00-11:00 a.m. General Session - Contributed Papers, Lecture Room 2, Van Allen Hall
- 11:00 a.m. Business Meeting, Lecture Room 2, Van Allen Hall

PAPER PRESENTATIONS

(* In competition for LaRue Award)

It is hoped that each paper can be presented in ten minutes or less

DEMONSTRATIONS

(* In Competition for Herrick Award)

- 1.* ULTRASTRUCTURAL STUDIES ON THE MELANIZATION RESPONSE OF MOSQUITOES AGAINST INOCULATED MICROFILARIAE. K.F. FORTON, DEPARTMENT OF VETERINARY SCIENCE, UNIVERSITY OF WISCONSIN, MADISON, WISCONSIN 53706
- 2.* GROWTH, DEVELOPMENT, AND SENESCENCE OF MICROCOTYLE SPINICIRRUS (MONOGENEA:MICROCOTYLIDAE). JOHN C. MERGO, JR. DEPARTMENT OF ZOOLOGY, OHIO STATE UNIVERSITY, COLUMBUS, OHIO. 43210.
3. THE USE OF THE TECHNIQUE OF CORROSION MODELING FOR THE LACUNAR SYSTEM OF ACANTHOCEPHALA. DONALD M. MILLER AND TOMMY T. DUNAGAN, DEPARTMENT OF PHYSIOLOGY AND PHARMACOLOGY, SCHOOL OF MEDICINE, SOUTHERN ILLINOIS UNIVERSITY, CARBONDALE, IL 62901.
4. PARASITES OF THE FISH FROM OLD WOMAN CREEK NATIONAL ESTUARINE SANCTUARY ERIE COUNTY, OHIO. J. D. STAMPER, J. L. CRITES AND C. E. HERDENDORF, DEPARTMENT OF ZOOLOGY, THE OHIO STATE UNIVERSITY, COLUMBUS, OHIO 43210.
5. MONOCLONAL ANTIBODIES WHICH RECOGNIZE TRYPANOSOMA LEWISI ANTIGENS ON PARASITE SURFACE AND ON HOST ERYTHROCYTES. C. L. WILLIAMS AND D. G. DUSANIC, DEPARTMENT OF LIFE SCIENCES, INDIANA STATE UNIVERSITY, TERRE HAUTE, IN 47809
6. THE ULTRASTRUCTURAL STUDY OF THE CUTICLE OF HAMMERSCHMIDTIELLA DEISINGI (NEMATODA: OXYUROIDEA) AND ITS RELATIONSHIP WITH BACTERIA STREPTOMYCES LEIDYNEMATIS. XIONG YU and JOHN L. CRITES, DEPARTMENT OF ZOOLOGY, THE OHIO STATE UNIVERSITY, COLUMBUS, OHIO 43210.
- 7.* PARTIAL DEVELOPMENT OF CRYPTOSPORIDIUM SP. IN A HUMAN RECTAL TUMOR CELL LINE. D. B. WOODMANSEE, IOWA STATE UNIVERSITY AND THE NATIONAL ANIMAL DISEASE CENTER, AMES, IOWA 50010
- 8.* PLASMODIUM AND AEGYPTIANELLA IN THE RIO GRANDE SUBSPECIES OF WILD TURKEY FROM SOUTH TEXAS. M. D. CASTLE. DEPARTMENT OF VETERINARY SCIENCE, UNIVERSITY OF WISCONSIN, MADISON 53706.
- 9.* HELMINTHS COLLECTED FROM THREE SPECIES OF SOUTH DAKOTA LAGOMORPHS. GLENN E. KIETZMANN, JR., SOUTH DAKOTA STATE UNIVERSITY, BROOKINGS, SOUTH DAKOTA, 57007.
- 10.* MICROHABITAT ANALYSIS OF MICROCOTYLE SPINICIRRUS (MONOGENEA: MICROCOTYLIDAE) AND LINTAXINE COKERI (MONOGENEA: HETERAXINIDAE) FROM FRESHWATER DRUM IN LAKE ERIE. JOHN C. MERGO, JR. DEPARTMENT OF ZOOLOGY, OHIO STATE UNIVERSITY, COLUMBUS, OHIO. 43210
11. THE INCIDENCE AND SEASONAL DISTRIBUTION OF COCCIDIAL PARASITES OF WOODCHUCKS (MARMOTA MONAX) IN SOUTHERN ILLINOIS. THOMAS E. MCQUISTION* AND JERRY M. WRIGHT, DEPARTMENT OF BIOLOGY, MILLIKIN UNIVERSITY, DECATUR, ILLINOIS 62522.
12. EIMERIA SPP. IN RACCOONS IN ILLINOIS. D. C. SNYDER. DEPARTMENT OF VETERINARY PATHOBIOLOGY, UNIVERSITY OF ILLINOIS, URBANA, ILLINOIS 61801.
13. PARASITES OF LEPOMIS MACROCHIRUS FROM MISSOURI, WITH SPECIAL REFERENCE TO THE PATHOGENESIS OF POSTHODIPILOSTOMUM MINIMUM IN THE HEART. LAURITZ A. JENSEN* AND STEPHEN M. PAYSON, THE UNIVERSITY OF HEALTH SCIENCES, COLLEGE OF OSTEOPATHIC MEDICINE. KANSAS CITY, MISSOURI 64124
14. COMPARATIVE EXCYSTMENT RATES OF PHILOPHTHALMUS MEGALURUS AND P. GRALLI METACERCARIAE AT DIFFERENT TEMPERATURES. PAUL M. NOLLEN AND ROBERT K. MACNAB, DEPARTMENT OF BIOLOGICAL SCIENCES, WESTERN ILLINOIS UNIVERSITY, MACOMB, ILLINOIS 61455.
15. DEER MICE AND THEIR CHIGGER PITS: A HISTOLOGICAL VIEW. OMER R. LARSON AND WILLIAM J. WRENN, DEPARTMENT OF BIOLOGY, UNIVERSITY OF NORTH DAKOTA, GRAND FORKS, NORTH DAKOTA 58202.
16. EVOLUTIONARY RELATIONSHIPS AND SPECIATION WITHIN THE GENUS ACANTHOCEPHALUS (ACANTHOCEPHALA: ECHINORHYNCHIDAE) FROM NORTH AMERICAN FRESHWATER FISHES. O. M. AMIN, UNIVERSITY OF WISCONSIN-PARKSIDE, BOX 2000, KENOSHA, WISCONSIN 53141.

ABSTRACTS OF

DEMONSTRATIONS

1. ULTRASTRUCTURAL STUDIES ON THE MELANIZATION RESPONSE OF MOSQUITOES AGAINST INOCULATED MICROFILARIAE. K.F. FORTON, DEPARTMENT OF VETERINARY SCIENCE, UNIVERSITY OF WISCONSIN, MADISON, WISCONSIN 53706

Microfilariae (mff) of *Dirofilaria immitis* were fixed in Karnovsky's fixative at 37°C at 5, 30 and 120 min, 1 and 5 days postinoculation (PI) into the hemocoel of *Aedes trivittatus*. Specimens were postfixed in 1% OsO₄, dehydrated at room temperature, embedded in Epon, thin-sectioned and examined with a Philips EM 410 at 60 kV. Recovered mff also were processed for examination with a JEOL JSM-U3 SEM. Deposition of melanin on inoculated mff began almost immediately following exposure to the hemolymph environment. Initial melanin accumulation occurred at any sight on the surface of mff and rapidly increased in thickness to encase the entire parasite usually within 24 hr PI. The classic hemocyte encapsulation generally described in insects did not occur, but the hemocytes do appear to be required for activation of the melanization response. Although hemocytes were never abundant, those few cells that were always present seemed to show an active secretion of membrane-bound vacuoles toward mff. Activated hemocytes were in close association with the parasite and often were seen in various stages of lysis. Numerous cell remnants were noted through-out the developing melanin capsule. (Supported by NIH Grant AI-19769.)

2. GROWTH, DEVELOPMENT, AND SENESCENCE OF MICROCOTYLE SPINICIRRUS (MONOGENEA: MICROCOTYLIDAE). JOHN C. MERGO JR. DEPARTMENT OF ZOOLOGY, OHIO STATE UNIVERSITY, COLUMBUS, OHIO. 43210

Microcotyle spinicirrus, a gill parasite of the freshwater drum (*Aplodinotus grunniens*), hatches from an operculate egg as an onchomiracidium bearing 5 ciliated bands. After attaching to its host the ciliated bands are shed and holdfast structures (clamps) arise from a region between the second and third pair of marginal hooklets and the two pairs of large anchors on its opisthaptor. As the organism matures clamps are added causing anchor bearing portion of opisthaptor to be pushed posteriorly. The larval haptor remains attached to adult until the worm produces approximately 21 clamps. The earliest appearance of internal organs occurred in the following sequence: ovary at a stage bearing 10 clamps; vagina, 11; spines of cirrus and atrium, 20; and testes, 28. Earliest maturation of a specimen, indicated by the ability to produce eggs, was observed in a specimen having 55 clamps. Mature specimens are believed to live for a period lasting one or two years, during which clamps are continuously added to the opisthaptor. Afterwards, a senescent period occurs indicated by the swelling of the vas deferens and vitelline ducts, a reduction in ovary size, a decline in spermatogenesis with accompanied degradation of testes, and loss of clamps from the opisthaptor.

DEMONSTRATIONS

3. THE USE OF THE TECHNIQUE OF CORROSION MODELING FOR THE LACUNAR SYSTEM OF ACANTHOCEPHALA. DONALD M. MILLER AND TOMMY T. DUNAGAN, DEPARTMENT OF PHYSIOLOGY AND PHARMACOLOGY, SCHOOL OF MEDICINE, SOUTHERN ILLINOIS UNIVERSITY, CARBONDALE, IL. 62901.

Previously we have utilized the techniques of India ink injection, glycerol clearing, and transmission photography as a basis for the construction of a three dimensional representation of two species of Acanthocephala. This previous method had several disadvantages. Among them was the fact that we had to interpret two dimensional images to construct the model, there was no means of recording the direction of flow and also there was no permanent proof of the system to study. In order to adequately construct a three dimensional model without these and other difficulties we have resorted to a new series of techniques. Firstly, the lacunar system is injected with unpolymerized colored butyrate monomer. The monomer is polymerized. After this the tegument is digested away leaving the three dimensional model in the transparent connective tissues. These techniques produce a model which is a direct three dimensional representation of the system in vivo and one which lends itself to photography and Scanning Electron Microscopy.

4. PARASITES OF THE FISH FROM OLD WOMAN CREEK NATIONAL ESTUARINE SANCTUARY ERIE COUNTY, OHIO. J.D. STAMPER, J.L. CRITES AND C.E. HERDENDORF DEPARTMENT OF ZOOLOGY, THE OHIO STATE UNIVERSITY, COLUMBUS, OHIO 43210

From June 21- October 15, 1983, 220 fish representing nine families and twenty species were collected from Old Woman Creek National Estuarine Sanctuary for the purpose of a parasite survey. The fish were collected bi-weekly by means of seines, electro-shock, gill nets and hoop nets. The fish were examed as soon as possible after capture for internal and external parasites. Nine phyla; Ciliophora, Microspora, Myxospora, Platyhelminthes, Nematoda, Acanthocephala, Arthropoda, Mollusca, and Annelida were represented. The most important pathogenic parasites were the neascus of *Posthodiplostomum minimum* which were found in large numbers in the kidney, liver, spleen and heart pericardium of sunfish (*Lepomis* sp.) and largemouth bass (*Micropterus salmoides*), and the diplostomulum of *Diplostomum spathaceum* which was found in the eyes of eleven of the twenty species of fish examined. The most diverse group of parasites were the monogeneans, parasitizing fish in six of the nine families present in the estuary. The incidence and intensity of parasites in the fish coincide with the general ecology of the estuary.

DEMONSTRATIONS

DEMONSTRATIONS

- 5. MONOCLONAL ANTIBODIES WHICH RECOGNIZE TRYPANOSOMA LEWISI ANTIGENS ON PARASITE SURFACES AND ON HOST ERYTHROCYTES. C. L. WILLIAMS AND D. G. DUSANIC, DEPARTMENT OF LIFE SCIENCES, INDIANA STATE UNIVERSITY, TERRE HAUTE, IN 47809

Hybridomas secreting monoclonal antibodies against Trypanosoma lewisi were produced by the fusion of SP2/0-Ag 14 mouse plasmacytomas with spleen cells from rats previously infected with the Taliaferro strain of T. lewisi. The trypanostatic activities of the monoclonal antibodies were assayed in vitro and agglutinin titers were determined with trypanosomes and erythrocytes collected from immunosuppressed (650 R from a ⁶⁰Co source) and immunocompetent rats. Although immunosuppressed rats had higher parasitemias and greater numbers of dividing trypanosomes, agglutinin titers using trypanosomes from immunosuppressed rats were similar to those obtained with trypanosomes from immunocompetent rats. An IgM and IgA monoclonal antibody agglutinated erythrocytes from infected, immunosuppressed rats, but not from uninfected and/or immunocompetent rats. This IgM monoclonal antibody which agglutinated dividing trypanosomes also inhibited the production of dividing forms in in vitro assays more than an IgG_{2a}, IgM or IgA monoclonal antibody which specifically agglutinated nondividing forms. Agglutination patterns and enzyme-linked immunosorbent assays indicated that the monoclonal antibodies which agglutinated nondividing trypanosomes recognized different antigens than the monoclonal antibody which agglutinated dividing trypanosomes. (These studies were supported in part by NSF Grant PCM-8207344.)

- 6. THE ULTRASTRUCTURAL STUDY OF THE CUTICLE OF HAMMERSCHMIDTIELLA DEISINGI (NEMATODA: OXYUROIDEA) AND ITS RELATIONSHIP WITH BACTERIA STREPTOMYCES LEIDYNEMATIS. XIONG YU AND JOHN L. CRITES, DEPARTMENT OF ZOOLOGY, THE OHIO STATE UNIVERSITY, COLUMBUS, OHIO 43210

Hammerachmidtella deisingi lives in the hind gut of American cockroach (Periplaneta americana). Streptomyces leidynematis often attaches to the cuticle of the nematode. EM study of the bacteria and the cuticle of H. deisingi indicated that there is no penetration of the cuticle by the bacteria. Two layers of unknown material existed between the bacteria and the cuticle. Probably they are secreted by the nematode. The structure of the cuticle of H. deisingi (female) can be divided into three zones, the cortical, median, and basal zones. The cortical zone has two layers, a triple layered membrane and an internal cortical layer. The median zone seems fiber like. The basal zone consists of three layers, BL₁, BL₂, and a basal membrane. The cuticular structure in the alae is somewhat different from other body parts. The internal cortical layer becomes thick at the tip of alae, and the median layer becomes thinner. Basal layer one (BL₁) varies in thickness and it becomes very thick as a major component of the alae. Electron dense structures of irregular shape extend from the border of BL₂ into BL₁, especially in alae. In the other parts of the body, these structures become more even, forming the major component of the BL₁ layer. BL₂ is internal to BL₁ and extends through whole body.

You can still participate
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of abstract to the
meeting!

PAPERS

ABSTRACTS OF
PAPER PRESENTATIONS

PARTIAL DEVELOPMENT OF CRYPTOSPORIDIUM SP. IN A HUMAN RECTAL TUMOR CELL LINE. D. B. WOODMANSEE, IOWA STATE UNIVERSITY AND THE NATIONAL ANIMAL DISEASE CENTER, AMES, IA 50010

Sporozoites of Cryptosporidium were shown to develop into immature and mature schizonts after inoculation into human rectal tumor (HRT) cell cultures. The relationship between parasites and HRT cells appeared ultra-structurally comperable to the in vivo host-parasite relationship. The parasites were visible with Giemsa's stain and interference contrast microscopy, stained acid fast and reacted with anti-Cryptosporidium antisera in an indirect immunofluorescence test. No evidence of development past the asexual stages was found.

PLASMODIUM AND AEGYPTIANELLA IN THE RIO GRANDE SUBSPECIES OF WILD TURKEY FROM SOUTH TEXAS. M.D. CASTLE. DEPARTMENT OF VETERINARY SCIENCE, UNIVERSITY OF WISCONSIN, MADISON 53706.

Blood samples taken from 55 Rio Grande wild turkeys on the Chaparosa Ranch and 67 wild turkeys on the Welder Wildlife Refuge in south Texas and subinoculated into domestic turkey poults revealed two species of hematozoan parasites. Light microscopy and host susceptibility studies have shown that one of the organisms is Plasmodium polare-like, found in 58% and 45% of the samples, respectively. Schizont morphology of this parasite is variable, with 5-11 merozoites produced ($\bar{X}=7$), and gametocytes are elongate and lateral. Experimental hosts shown to be insusceptible include domestic chickens and Japanese quail. Ultrastructural studies indicate that the second organism, found only in 24% of birds from Chaparosa, is a rickettsial agent similar to Aegyptianella pullorum. No positive identification of Aegyptianella has been made previously in North America, although there have been two reports of a similar organism found in domestic fowl. At this time, an additional 156 blood samples are being screened and studies are continuing on the susceptibility and pathogenicity of Aegyptianella in domestic chickens and both domestic and pen-raised wild turkeys. Haemoproteus meleagridis was observed in 75% of 72 direct blood smears from birds at Chaparosa and in 76% of 81 blood smears from birds at Welder.

9. HELMINTHS COLLECTED FROM THREE SPECIES OF SOUTH DAKOTA LAGOMORPHS. GLENN E. KIETZMANN, JR., SOUTH DAKOTA STATE UNIVERSITY, BROOKINGS, SOUTH DAKOTA, 57007.

Between October, 1982 and June, 1983, 35 white-tailed jack rabbits (Lepus townsendii campanius), 18 eastern cottontail rabbits (Sylvilagus floridanus similis) and one desert cottontail rabbit (S. auduboni baileyi) were examined for parasites. The helminths collected from each lagomorph species and their prevalence are as follows: Cittotaenia pectinata americana 8.6%, Multiceps serialis (coenuri) 5.7% and Obeliscoides cuniculi 2.9% from white-tailed jack rabbits, C. variabilis 77.8%, C. perplexa 38.9%, Taenia pisiformis (cysticerci) 33.3%, C. p. americana 16.7%, Dermatoxys veligera 22.2% and O. cuniculi 11.1% from eastern cottontail rabbits and T. pisiformis (cysticerci) 100% and D. veligera 100% from the desert cottontail rabbit. The helminths showing a state-wide distribution included C. variabilis, T. pisiformis and D. veligera.

10. MICROHABITAT ANALYSIS OF MICROCOTYLE SPINICIRRUS (MONOGENEA: MICROCOTYLIDAE) AND LINTAXINE COKERI (MONOGENEA: HETERAXINIDAE) FROM FRESHWATER DRUM IN LAKE ERIE. JOHN C. MERGO JR. DEPARTMENT OF ZOOLOGY, OHIO STATE UNIVERSITY, COLUMBUS, OHIO. 43210

From June 1981 through October 1983, 309 freshwater drum (Aplodinotus grunniens) were collected and examined for Microcotyle spinicirrus and Lintaxine cokeri. Each parasites location as to gill arch, arch region, and filament region were recorded. M. spinicirrus was found to locate more often on the third arch. This arch possessed 29% of the approximately 1900 observed specimens, while the other arches possessed 23% of the total. Analysis of the proximal and distal hemibranches revealed that the distal row of filaments possessed 1121 or 62% of the parasites, in contrast to 683 or 38% on the proximal hemibranch. When the arches were arbitrarily divided into thirds, the middle third possessed 49% of the worms while the dorsal and ventral thirds possessed 27.5 and 23.0% respectively. However, when analyzed by quarters a ventral to dorsal gradient in infestation is observed. Out of 65 specimens of L. cokeri removed from drum, the only gill arch or region exhibiting a significantly higher number of specimens than expected was the ventral half of the gill arches. This region possessed 57 or 87.7% of all specimens collected. The relationship between M. spinicirrus and L. cokeri may be an example of niche partitioning in freshwater monogeneans.

PAPERS

11. THE INCIDENCE AND SEASONAL DISTRIBUTION OF COCCIDIAL PARASITES OF WOODCHUCKS (*MARMOTA MONAX*) IN SOUTHERN ILLINOIS. THOMAS E. MCQUISTION* AND JERRY M. WRIGHT, DEPARTMENT OF BIOLOGY, MILLIKIN UNIVERSITY, DECATUR, IL 62522

Thirty-eight woodchucks captured in Southern Illinois were examined for coccidial parasites from July, 1981 to October, 1983. Eighty-nine percent of the woodchucks had at least one species of *Eimeria* in their fecal contents while 29% of the woodchucks had all four species of coccidia represented. *Eimeria monacis* and *E. perforoides* were the most common; infecting 87% and 89% respectively of the woodchucks. *E. tuscarorensis* was observed in 55% of the woodchucks while *E. os* was infecting 45% of the animals.

The incidence of *E. os* was highest in the Spring and Autumn months of the year and *E. tuscarorensis* had the highest incidence during the Summer months. *E. monacis* and *E. perforoides* were abundant in woodchucks throughout the non-hibernating months of the year.

12. *EIMERIA* SPP. IN RACCOONS IN ILLINOIS. D.E. SNYDER. DEPARTMENT OF VETERINARY PATHOBIOLOGY, UNIVERSITY OF ILLINOIS, URBANA, IL 61801

During November and December of 1980, fecal samples were collected from the terminal portion of the gastrointestinal tracts of 100 hunter shot or trapped raccoons from central Illinois. Each animal was sexed and aged (juvenile vs. adult) at the time of collection. Each of the fecal samples was placed in a 2.5% (w/v) aqueous potassium dichromate solution, mixed thoroughly, and distributed in thin layers in petri dishes for one week at room temperature (22°C) to allow sporulation of the oocysts. At this time each fecal-dichromate solution was strained thru a 60-mesh sieve to remove particulate fecal debris. Equal volumes of the strained fecal-dichromate suspension and Sheather's sugar solution were added to conical centrifuge tubes, thoroughly mixed, a coverslip added to the top of the tube, and centrifuged at 2,000 rpm for 5 minutes. The coverslips were removed, placed on glass slides and the presence or absence of coccidian oocysts noted. *Eimeria* spp. oocysts were found in 67 (67%) of the raccoons examined. The presence of *Eimeria* spp. oocysts in juvenile vs. adult raccoons were 68% and 64%, respectively. The overall prevalence observed is similar to a previous study which examined Illinois raccoons for the presence of coccidia.

13. PARASITES OF *LEPOMIS MACROCHIRUS* FROM MISSOURI, WITH SPECIAL REFERENCE TO THE PATHOGENESIS OF *POSTHODIPILOSTOMUM MINIMUM* IN THE HEART. LAURITZ A. JENSEN* AND STEPHEN M. PAYSON, THE UNIVERSITY OF HEALTH SCIENCES, COLLEGE OF OSTEOPATHIC MEDICINE. KANSAS CITY, MISSOURI 64124

From July to November 1983 8 species of parasites were collected from 241 *Lepomis macrochirus* (bluegill), taken in Northwestern Missouri. Parasites consisted of 1 species of Myxosporida (*Chloromyxum trijugum*), 1 species of Monogenea (*Urocleidus* sp.), 2 species of Digenea (*Posthodiplostomum minimum* and *Ornithodiplostomum pychocheilus*), 1 species of Cestoidea (*Proteocephalus ambloplitis*), 1 species of Nematoda (*Eustrongylides* sp.), 1 species of Copepoda (*Lernaea cyprinacea*), and 1 species of Hirudinea (*Myzobdella* sp.). The digenea, *P. minimum* (metacercariae), was the most prevalent parasite and was recovered in 98.8% of 241 fish. *Posthodiplostomum minimum* infects the outer portion of the heart and is responsible for compression atrophy and necrosis of the myocardium, focal pericardial fibrosis, and marked encapsulation of the metacercariae. Worm burdens in the infected hearts ranged from 1 to 212.

14. COMPARATIVE EXCYSTMENT RATES OF *PHILOPHTHALMUS MEGALURUS* AND *P. GRALLI* METACERCARIAE AT DIFFERENT TEMPERATURES. PAUL W. NOLLEN AND ROBERT K. MACNAB, DEPARTMENT OF BIOLOGICAL SCIENCES WESTERN ILLINOIS UNIVERSITY, MACOMB, IL 61455.

Metacercarial cysts of the eyeflukes of birds in the genus *Philophthalmus* formed on solid substrates are bottle-shaped with an open end in direct contact with the environment. Thus they are relatively short-lived compared to completely enclosed species. Cysts of *P. gralli*, a species primarily tropical in origin (S.E. Asia, Hawaii and South Texas) had a maximum survival of 13 days but most died off after 4-6 days at room temperature (22°C). At 4°C, maximum survival was 5 days with most unable to excyst at 4 days. Cysts of *P. megalurus*, a northern temperate species (Michigan, Indiana, Ohio, and Oregon) showed comparable survival rates at room temperature to those found for *P. gralli*. Maximum longevity was 9 days with most cysts losing their ability to excyst from 4-6 days. However, at 4°C, *P. megalurus* cysts were from 75-100% viable for up to 16 days. At 20 days the excystment rate was 38.5%, after which the experimentation was terminated due to a seasonal dearth of infective forms. Viability of cysts seems to be correlated to the habitats from which the eyeflukes are taken, at least for cold temperature survival.

PAPERS

DEER MICE AND THEIR CHIGGER PITS: A HISTOLOGICAL VIEW.
OMER R. LARSON AND WILLIAM J. WRENN, DEPARTMENT OF BIOLOGY,
UNIVERSITY OF NORTH DAKOTA, GRAND FORKS, ND 58202

Larval mites (chiggers) in the family Trombiculidae are parasitic on a variety of terrestrial vertebrates. An unusual characteristic is their production of a stylostome or feeding tube which penetrates the host's epidermis. Several types of stylostomes are known. These appear to represent variable interactions between chigger secretion and host tissue. In the present study, skin of the eyelids and genital area of Peromyscus maniculatus from north-central Minnesota were often found to possess epidermal pits. Most pits housed a single chigger identified as Euschoengastia setosa (Ewing). Paraffin embedded tissues were serially sectioned, histochemically stained, and examined for chigger/host relationships. Well developed, cup-shaped pits possess interiors about 0.4 mm deep x 0.25 mm wide. The acellular, non-fibrous, hyaline pit lining stained negatively for keratin and keratohyalin. Its thickest portion is at the pit apex, through which an 80-100µm long stylostome passes. The pit wall also includes the host's epidermis. In parasitized eyelids, the stratified squamous is 2-10X thicker than normal. Infected genital skin shows little hyperplasia, but dermis adjacent to pits possess leucocytic infiltrations. In either location, host tissues just beyond the stylostome show disintegration consistent with known histolytic action of chigger saliva. Although pit development has not been traced, evidence suggests a host tissue response sufficient to extrude and detach the pit lining.

6. EVOLUTIONARY RELATIONSHIPS AND SPECIATION WITHIN THE GENUS ACANTHOCEPHALUS (ACANTHOCEPHALA: ECHINORHYNCHIDAE) FROM NORTH AMERICAN FRESHWATER FISHES. O. M. AMIN, UNIVERSITY OF WISCONSIN-PARKSIDE, BOX 2000, KENOSHA, WI. 53141

Populations of the three known species of the genus Acanthocephalus from North American freshwater fishes are found in the Mississippi River drainage system, or waters previously connected to it, in the Mobile Bay drainage, Great Lakes, and in New England. Based on geographical and host distributional records, morphological variability, and geological history, it is proposed that A. dirus represents the phylogenetically primary species and that the Wisconsin-Lake Michigan population of A. dirus was geographically isolated from Mississippi River A. dirus-like source less than 15,000 years ago. The New England population of A. dirus may have originated in a like manner. Given sufficient time and continued isolation, the Wisconsin-Lake Michigan and New England populations may diversify enough to achieve a more distinct taxonomic status. Host isolation was probably involved in the speciation of the two southern species, A. tahlequahensis and A. alabamensis. The proposed evolutionary associations provide satisfactory explanation of present geographical distribution, host relationships, and degree of morphological diversification. Cladistic analysis furnishes a framework supporting the above phylogenetic hypothesis.

Report of the 35th Annual
Midwestern Conference of Parasitologists
A M C O P

The 35th AMCOP conference was held in the new Veterinary Medicine Basic Sciences Building on the Urbana campus of the University of Illinois, 2-4 June, 1983. The organization has 151 members this year, an increase of 31. Dr. Charles M. Vaughn of Miami University was Presiding Officer of the meeting and Dr. Kenneth S. Todd, Jr. of the University of Illinois College of Veterinary Medicine was Program Officer and made local arrangements. Eighteen demonstrations and 23 papers were presented by members. The C. A. Herrick Award (and \$200) for the best demonstration by a graduate student was awarded to Kimm J. Hamann from the Department of Veterinary Pathobiology, University of Minnesota (Dr. Bert E. Stromberg, major professor) for "Feather mites: A pictorial review." The G. R. LaRue Award (and \$200) for the outstanding paper presentation by a graduate student was awarded to Kenneth W. Bafundo from the College of Veterinary Medicine, University of Illinois (Dr. Paul R. Fritzgerald, major professor) for "The effects of coccidiosis on lead toxicity and delta amino levulinic dehydratase activity in the chick."

A symposium on "Intestinal Protozoa" dedicated to honor the retirement of Dr. Norman D. Levine from the University of Illinois was presented with talks by Dr. Ron Fayer, "Pathology and diagnosis of Sarcocystis in domestic animals" and Dr. John Ernst, "Bovine coccidiosis--some unsolved problems." Both speakers were from the Animal Parasitology Institute, USDA, Beltsville, MD.

Dr. Harley W. Moon National Animal Disease Center, USDA, Ames, Iowa gave the banquet address continuing the theme of intestinal parasitism with "Speculations on the pathogenesis of Cryptosporidiosis with comparisons to other enteric infections."

Officers elected for 1983-84 were: Presiding Officer, Dr. William H. Coil, Department of Systematics & Ecology, University of Kansas; Program Officer, Dr. George Cain, Department of Zoology, University of Iowa, and Dr. George Garoian was reelected Secretary/Treasurer and Representative to the ASP Council. AMCOP-36 will be held on the campus of the University of Iowa, Iowa City, IA, 7-9 June, 1984.

George Garoian,
Secretary/Treasurer

ljm

Committees appointed by Dr. Vaughn with their recommendations are as follows:

Judging Demonstrations:

Melvin Denner and Kevin Kazacos
Herrick Award to Kimm J. Hamann (U of Minnesota)

Judging Papers:

Donal G. Myer and John H. Greve
LaRue Award to Kenneth W. Bafundo (U of Illinois)

Nominating:

Paul Nollen and Allen D. Johnson
Presiding Officer for 1984 William H. Coil
Program Officer for 1984 George Cain
Secretary/Treasurer for 1984-86 George Garoian

Future Meeting Sites:

George D. Cain and John L. Crites
1984 - University of Iowa
1985 - Iowa State University

Future Programs:

Peter Pappas and Donald Gilbertson
Immunity to helminths
Parasites of zoonotic significance in the U.S.
Experimental ecology of parasites
Reproductive strategies of parasites
Developmental biology of cestodes

Audit:

Frank Etges

Resolutions:

Harry W. Huizinga and Stephen J. Taft

Whereas: Dr. Kenneth Todd, Program Officer, coordinated the many aspects of this meeting at the University of Illinois in an outstanding and commendable manner, and

Whereas: Dr. Dirks, Dean of the College of Veterinary Medicine extended a warm welcome to AMCOP and provided facilities of the College of Veterinary Medicine for this meeting, and

Whereas: Contributions were made by the College of Veterinary Medicine to help defray some of the expenses incurred in providing honoraria for speakers, and

Whereas: Eli Lilly and Company generously provided funding for the Herrick Award, and a number of AMCOP members made contributions for the LaRue Award, and

Whereas: AMCOP Expresses gratitude to the following organizations for their contributions to this 35th meeting to make it enjoyable and meaningful, American Hoechst Corp; Eli Lilly Company; Pfizer, Inc.; Pittman-Moore, Inc.; Smithkline Beckman; and the University of Illinois, and

Whereas: Dr. Ron Fayer and Dr. John Ernst provided a scholarly symposium on intestinal protozoa, and

Whereas: Dr. Harley W. Moon presented an entertaining and informative banquet speech "Speculations on the pathogenesis of Cryptosporidiosis", and

Whereas: Dr. George Garoian provided registration and program information in an efficient and timely manner, and

Whereas: The contributed papers and demonstrations were of high quality, and

Whereas: Dr. Charles M. Vaughn, presiding officer, provided an unquestionably efficient and entertaining format for the presentation of papers.

Be it hereby resolved, that the membership of the 35th Annual Midwestern Conference of Parasitologists express their deep gratitude and sincere appreciation to all of the aforementioned on this occasion.

The motion was moved, seconded and passed unanimously.

Officers elected for 1984 were: Presiding Officer, Dr. William H. Coil, of University of Kansas, Program Officer, Dr. George Cain, of University of Iowa and Secretary/Treasurer, Dr. George Garoian of Southern Illinois University at Carbondale. AMCOP-36 will be held on the campus of the University of Iowa at Iowa City, 7-9 June, 1984.

AMCOP-35 1983
Treasurer's Report (2 June, 1983)

Balance on Hand 2 June, 1982. \$1,163.50

Income

Membership	
94 Regular (\$3)	282
38 Student (\$2)	76
Eli Lilly	200
LaRue Award gifts	163*
Bank interest	66.62
AMCOP-34 Surplus.	54.66
	<u>TOTAL 842.28</u>

(* \$37 additional was donated 3 June, 1983)

Expenses

AMCOP-34.	23.13
Postage (notices & programs)	182.47
Envelopes	7.22
Printing (200 programs)	108.00
Herrick Award	200.00
LaRue Award	200.00
	<u>TOTAL 720.82</u>

Balance on Hand 2 June, 1983. \$1,284.96

Respectfully submitted,

George Garoian

George Garoian, Sec./Treas.

ljm

Financing ICOPA-VI

In light of the resolution passed at the 1982 Conference expressing "our displeasure with the high cost of registration and hotels at ICOPA-V" and to communicate our wishes that there be more careful planning to control expenses for ICOPA-VI attention is called to the First Progress Report for ICOPA-VI which appeared in the International Journal of Parasitology, 14(1):9-11, 1984. The section on "Organization and Finance" is reprinted here.

ORGANIZATION AND FINANCE

ICOPA VI is being organized by the Australian Academy of Science in collaboration with the Australian Society for Parasitology. A budget along the lines outlined in the original invitation to the Executive Board of the World Federation of Parasitologists in 1982 has been submitted by the Organizing Committee to the Australian Academy of Science. The various items in the budget have been categorized as "essential" or "desirable". The former items, including the organization and costs of the Congress and the Congress publications, will be funded from receipts from the registration fees. The later, including assistance to invited speakers and entertainments, will as far as possible, be funded from special grants, donations etc. Our aim is to keep the registration fee as low as possible to facilitate attendance of young parasitologists and those who may find Australian currency difficult to obtain. Professor C. Bryant will be co-ordinating our drive for financial support for these aspects of the Congress. All income received on behalf of the Congress will be payable to and received by the Academy and all expenditure will be authorized by the Congress Treasurer, Dr. A. D. Donald.

THE ANNUAL MIDWESTERN CONFERENCE OF PARASITOLOGISTS

Objectives and Organization

A restatement to incorporate changes approved in 1974. Earlier statements had been approved in 1948, 1953, 1971, 1972, and 1973.

Name

The organization shall be known as the ANNUAL MIDWESTERN CONFERENCE OF PARASITOLOGISTS, hereinafter referred to as the Conference.

Affiliation

The Conference is an affiliate of the American Society of Parasitologists.

Objectives

The Conference is a gathering of parasitologists and students of parasitology for the purpose of informal discussion of research and teaching in parasitology and the furthering of the best interests of the discipline of parasitology.

Members

The Conference is open to all interested persons regardless of place of work, residence, or affiliation in other recognized societies.

Dues

No regular dues are collected, but a registration fee is charged members during registration at annual conferences. The amount of this fee will be decided for each Conference by a committee composed of the Presiding Officer, the Secretary-Treasurer, and the Program Officer, who is to serve as its chairman. Amended by ballot vote 1977. Student dues - \$2.00 and regular dues - \$3.00. Dues are collected by the Secretary-Treasurer.

Meetings

The Conference is held in the general midwestern area in mid-June, unless otherwise specified by a majority vote of the previous Conference or a majority vote of those listed members replying by mail ballot.

Bylaws

1. Simple majority vote of members in attendance at regularly scheduled business meetings of the Conference shall determine the policies of the Conference.

2. The officers are a Presiding Officer, whose term of office is one year or until his successor is elected (normally his term would expire with adjournment of the annual Conference over which he presides); a Secretary-Treasurer, whose term of office is two years or until his successor is elected; a Program Officer whose term of office is one year; and a Policy Committee composed of the last five available retired Presiding Officers plus, ex officio and without vote, the current Presiding Officer and Secretary-Treasurer. The term of office of each full member of the Policy Committee is five years, or so long as he is one of the five most recent, available Presiding Officers. The most recent past Presiding Officer available is the Chairman of the Policy Committee and the Vice Presiding Officer of the Conference.

3. The Presiding Officer, the Secretary-Treasurer, and the Program Officer are elected by a majority vote of those members attending a regularly scheduled business meeting of the Conference or by a majority vote of those replying to a mail ballot of the membership.

4. The Presiding Officer shall preside at all meetings of the Conference and shall arrange for a banquet speaker.

On the first day of a Conference he shall appoint the following committees, which shall serve until they have reported on the last day of the annual Conference: (1) Nominating Committee, (2) Committee to Recommend Future Meeting Places, (3) Committee to Suggest Program Possibilities for Future Meetings, (4) Resolutions Committee, and such other ad hoc committees as may be required.

He shall appoint the Conference Representative for the Council of the American Society of Parasitologists for the year following his tenure of office and serve as a member without vote of the Policy Committee.

5. The Secretary-Treasurer shall issue a call at least four months prior to each Conference for participants in the program for each Conference; inform the new Presiding Officer of his duties and the members of the Policy Committee of their tenure and Secretary of the American Society of Parasitology within three weeks after the annual election; serve as member without vote and Secretary of the Policy Committee; and supervise all funds of the Conference.

6. The Program Officer shall be responsible for the general format of the Conference and for arranging suitable facilities. It shall also be his responsibility to serve as chairman of the special committee to determine the registration fee for the Conference. The format of the Conference may vary, but should include both a demonstration session open to all members and a session of contributed papers limited to graduate students. Occasionally a symposium may also be included, or may replace the session of contributed papers.

7. The Policy Committee shall determine by majority vote all matters of procedure and policy pertaining to the Conference upon which decision must be reached between consecutive Conferences, as well as all matters referred specifically to it by the membership. Such a vote may be requested by any member of the Conference but must be directed through the Secretary-Treasurer. The Chairman of the Policy Committee shall request the approval of the membership for all decisions of the Committee at the earliest subsequent business meeting of the Conference.

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(As of May 10, 1984)

REGISTRATION AND ROOM REQUEST

XXXVI
 ANNUAL MIDWESTERN CONFERENCE OF PARASITOLOGISTS 84-160-01
 The University of Iowa
 Iowa City, Iowa 52242 M283
 June 7 - 9, 1984

Registration forms and payment must be received by May 8, 1984 to guarantee reservations. However, every effort will be made to accommodate late registrants.

Social Security Number (needed to maintain enrollment record)

Name _____

Address _____

Business phone () _____ Home Phone () _____

Dorm Rooms - Burge Residence Hall	Amount
Double: \$14.00 for 2 nights	_____
Single: \$18.00 for 2 nights	_____
Roommate _____	_____

I have no roommate preference; please assign one to me.
 Sex: Female _____ Male _____
 Approximate time of arrival _____

Buffet Banquet, May 8, 7:00 p.m., No. _____ @ \$9.50 _____

Registration Fee (Members \$5.00, Nonmembers \$8.00) _____

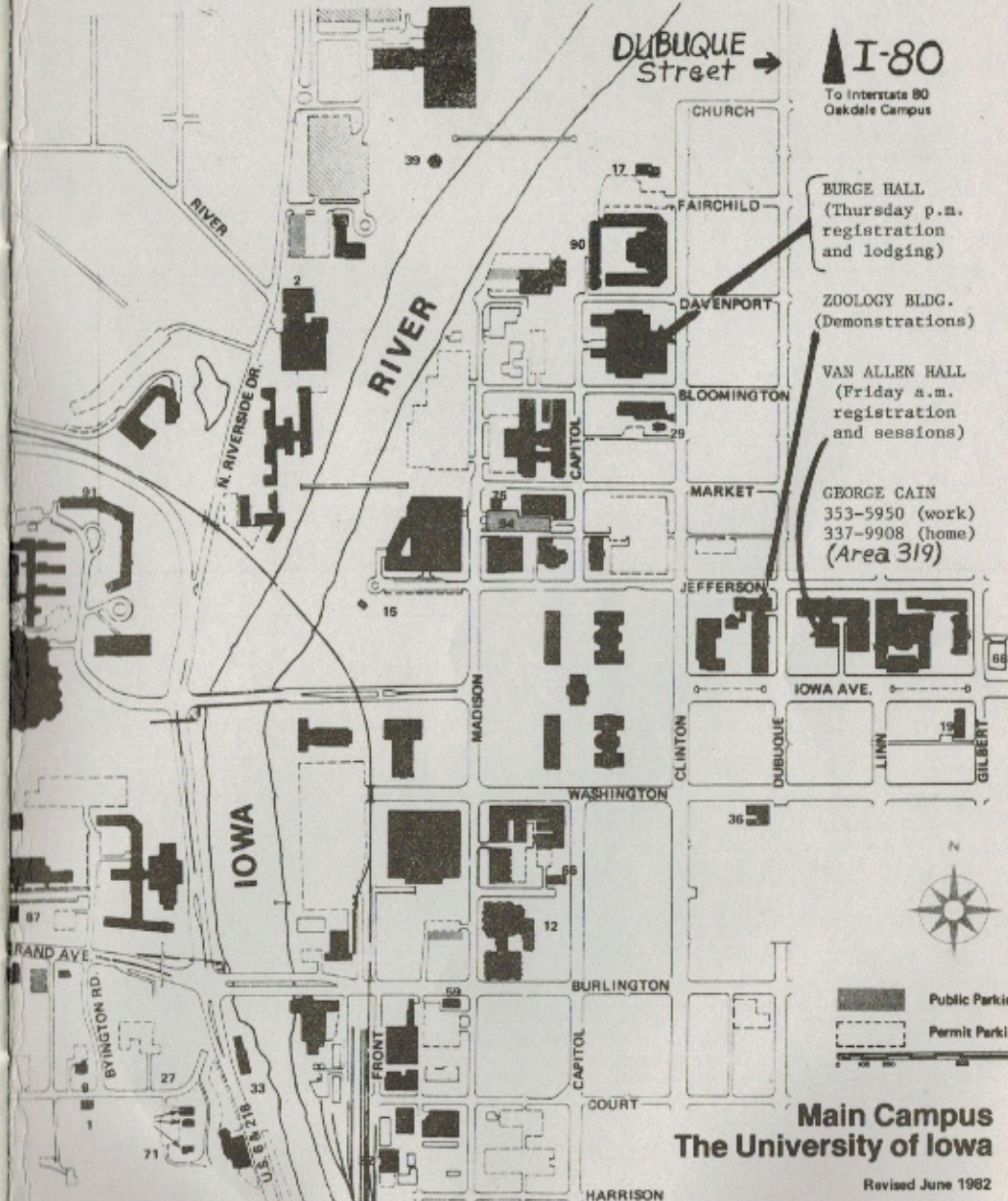
TOTAL AMOUNT ENCLOSED _____

Will you require parking? yes no

PLEASE COMPLETE ONE FORM FOR EVERY INDIVIDUAL

Make checks or money order payable to: The University of Iowa
 Return completed form and check or money order to:
 Center for Conferences and Institutes
 Room 210 Iowa Memorial Union
 University of Iowa
 Iowa City, Iowa 52242 Phone (319) 353-5505

Refunds: All but \$3 of your fee will be refunded if cancellation is made before noon, June 6.



Main Campus
The University of Iowa

Revised June 1982

ADDITIONAL DEMONSTRATIONS

- 6a. Surface Antigen Gene Rearrangements in *Trypanosoma Brucei*. William J. Murphy and Steven T. Brentano, University of Iowa, Iowa City, Iowa 52242

We have cloned and sequenced a previously uncharacterized expression-linked extra copy (ELC) gene of the IaTat 1.2 variable surface glycoprotein (VSG). In addition we have cloned and sequenced the corresponding cDNA and most of the two basic copy (BC) genes for this VSG. A comparison of the four sequences (cDNA, ELC, BC 1 and BC 2) and a series of Southern genomic blots reveal the following properties about this gene system. The two BC gene sequences are very similar although the ELC is clearly derived from BC 1. The 5' boundary of the ELC transposed segment is within a repetitive sequence element located 700 base pairs upstream from the VSG start codon. This repetitive element bears no resemblance to the 76 bp repeat sequence reported to contain the 5' transposition boundaries of the VSG 117 and 118 genes. The 5' transposition boundary is located within an intervening sequence and the 35 bp first exon is identical in sequence to the spliced leader sequence reported for other VSG genes. The 3' transposition boundary is located within the coding sequence for the C-terminal hydrophobic tail. There is no homology between the two boundaries.

The DNA segment located upstream from the transposed structural gene also appears to be duplicated in the IaTat 1.2 genome. This sequence most likely represents a residual portion of a transposed gene copy previously expressed at this site. In support of this, three additional copies of this upstream sequence are found in all genomes tested. One of these extra copies is telomere-linked (as is the ELC) and encodes a potential VSG gene. The terminal sequences of a recombinant clone of this telomere-linked gene are strikingly homologous with the corresponding portions of the ELC. In particular the extreme upstream sequences of the two genes are nearly identical. This includes the repetitive element containing the 5' transposition boundary of the IaTat 1.2 ELC. We infer from this that i) the second telomere-linked gene could have been created via a similar duplication-transposition event, and ii) a transposed copy of this gene may have preceded that of the IaTat 1.2 ELC.

GENOMIC MAPPING AND SEQUENCE ANALYSIS OF DNA FRAGMENTS CONTAINING THE CONSERVED 5' 35 NUCLEOTIDES OF TRYPANOSOME VARIABLE SURFACE GLYCOPROTEIN MESSENGER RNAs, David M. Dorfman, Department of Biochemistry, University of Iowa, Iowa City, IA 52242

laboratory and others have shown that the first 35 nucleotides of the mRNAs of different *Trypanosoma brucei* variable surface glycoproteins (VSGs) are identical. In each case this conserved sequence is not adjacent to the expression of the VSG coding region in the genome. This suggests that different genes are expressed from the same or similar genomic expression sites defined by the location of the 35 nucleotide sequence. To test this hypothesis we have synthesized a 21-mer oligonucleotide probe containing a portion of the nucleotide sequence for genomic Southern analysis and to screen genomic bacteriophage libraries. Southern analysis confirms recent reports that the conserved 35 nucleotide sequence is present in many copies in the genome that are arranged in tandem repeat units of ~1.4 kilobases. Using genomic subcloning and library screening we have isolated genomic restriction fragments containing copies of the 35 nucleotide sequence and flanking regions. DNA sequence analysis, genomic mapping, and other studies have been undertaken to further characterize and compare these repetitive regions and to ascertain their relationship to the VSG gene expression site(s). We are currently testing several models for VSG gene expression based on these findings.

ADDITIONAL DEMONSTRATIONS

- 6c. Production of Trypanosome VSG Serotype-Specific Antisera Using VSG Gene Fragments Expressed in *Escherichia Coli*. Michael Lenardo and John Donelson, University of Iowa, Iowa City, IA 52242

We have used the expression of a trypanosome variable surface glycoprotein (VSG) in *E. coli* to produce VSG serotype-specific antisera which have none of the cross-reacting specificities characteristic of antisera prepared against purified VSGs. This was accomplished by treating restriction fragments of VSG cDNAs with Bal-31 nuclease to facilitate expression of their open reading frames in the *E. coli* expression vector, pMR100 (4). The resultant VSG- β -galactosidase fusion proteins possess various antigenic regions of the original VSG. This provides a rapid means for producing VSG-specific antisera for reagent use and has the capability of large scale production of antigen for immunological investigation.

- 6d. *Trypanosoma (Nannomonas) congolense* Variant Specific Glycoprotein: Isolation and Cell-free Synthesis. George A. Cook^{††}, B. M. Honigberg*, and Robert A. Zimmermann[†], Departments of *Zoology and [†]Biochemistry; University of Massachusetts, Amherst. ^{††}Present Address: Department of Biochemistry, University of Iowa, Iowa City.

Two *Trypanosoma (Nannomonas) congolense* stocks, 1/148 Fly and TREU 921, were cloned in A/J strain mice immunosuppressed with cyclophosphamide. The cloned populations, checked for antigenic homogeneity by the indirect fluorescent antibody test (IFAT) using six-day antisera developed in rabbits against the living parasites, were designated as AmNat 1.1 and AmNat 3.1. The variant specific glycoproteins (VSGs) from both AmNats were purified to homogeneity, as revealed by SDS-polyacrylamide 5-20% gradient gels, by combining the best aspects of two published methods (Onodera, *et al.*, 1981, *Exp. Parasitol.* 52:427; Reinwald, *et al.*, 1981, *Biochim. Biophys. Acta* 668:119). The apparent molecular weights (M_r s) of the two VSGs were 51,700 for AmNat 1.1 and 49,900 for AmNat 3.1. Monospecific antisera prepared in rabbits to each VSG were used to confirm the homogeneity of the clones by IFAT. The VSGs were susceptible to endoglycosidase H (Endo H) digestion indicating the presence of dl-N-acetylchitobiose linkages on these mannose glycoproteins. The apparent M_r s of the Endo H digested VSGs were 48,800 and 46,900 for AmNat 1.1 and 3.1, respectively. The oligosaccharides cleaved from the VSGs and the Endo H resistant oligosaccharides, if present, were not analysed.

Total RNA was extracted from each clone and the enriched poly (A⁺) RNA obtained by oligo-(dT)-cellulose chromatography was assayed for template activity using the mRNA-dependent rabbit reticulocyte lysate as the *in vitro* protein synthesis system. The [³H]- and [³⁵S]-labeled polypeptides were initially characterized by SDS-gradient acrylamide gels and visualized by fluorography. Accurate characterization of these translation products was facilitated by the use of IgGs obtained from the monospecific antisera by sodium sulfate precipitation and purified on a Sepharose CL-6B column. The VSG precursor polypeptides, immunoprecipitated as an IgG/Pansorbin complex, were analysed on SDS-acrylamide gradient gels, visualized by fluorography and the M_r s for AmNat 3.1 and 1.1, estimated at 43,000 and 39,000, respectively.

ADDITIONAL DEMONSTRATIONS

- 6e. SEQUENCE ANALYSIS OF cDNA'S FOR SURFACE GLYCOPROTEINS FROM METACYCLIC TRYPANOSOMES. Gregory Kelly, Michael J. Lenardo, Allison C. Rice-Ficht, Klaus M. Esser,* and John E. Donelson, University of Iowa, Iowa City, Iowa 52242, and Walter Reed Army Research Institute,* Washington, D.C., 20012.

African trypanosomes evade their mammalian host's immune system by the sequential expression of alternative surface glycoproteins. We have examined several cDNA sequences for the surface glycoproteins expressed on metacyclic trypanosomes, the final developmental stage in the tsetse fly. One of us (K.M.E.) has recently shown that metacyclic surface glycoproteins are still the predominant surface antigen of trypanosomes five days after infection in rats.

We have prepared a cDNA library using trypanosomes isolated from rat blood five days after infection. Clones containing sequences for metacyclic surface antigens have been identified using a plus/minus screening in conjunction with Grunstein colony hybridizations and RNA dot blot experiments. Eight putative metacyclic surface glycoprotein cDNA sequences have been identified. In each of the clones sequenced to date the predicted amino acid sequence shows that the metacyclic surface glycoproteins can be classified into the same two C-terminal homology subsets established earlier for the bloodstream trypanosome's variable surface antigen. The expression of these metacyclic surface glycoproteins in trypanosomes will be discussed.

* Denotes eligibility for Herrick Award.

ADDITIONAL PAPERS

- 10a. Production of Trypanosome VSG Serotype-Specific Antisera Using VSG Gene Fragments Expressed in *Escherichia Coli*. Michael Lenardo and John Donelson, University of Iowa, Iowa City, IA 52242

We have used the expression of a trypanosome variable surface glycoprotein (VSG) in *E. coli* to produce VSG serotype-specific antisera which have none of the cross-reacting specificities characteristic of antisera prepared against purified VSGs. This was accomplished by treating restriction fragments of VSG cDNAs with Bal-31 nuclease to facilitate expression of their open reading frames in the *E. coli* expression vector, pMR100 (4). The resultant VSG-8-galactosidase fusion proteins possess various antigenic regions of the original VSG. This provides a rapid means for producing VSG-specific antisera for reagent use and has the capability of large scale production of antigen for immunological investigation.

- 12a. Utilization of Heme and its Precursor Aminolaevulinic Acid by *Schistosoma mansoni* In Vitro. Lewis A. Foster* and Burton J. Bogitsh.

Schistosoma mansoni schistosomules were able to incorporate from mouse reticulocytes portions of the heme moiety of hemoglobin which had been labeled *in vitro* with radiolabeled aminolaevulinic acid (ALA) as well as lyophilized, ALA-labeled hemoglobin in solution. The label was incorporated into proteins and non-proteinaceous material. The labeled, non-proteinaceous material may represent phospholipids as determined by ethanol extraction and thin-layer chromatography. Schistosomules and adult worms also incorporated the label from the radioisotope presented to them in solution. Distribution of label taken up from solution by the worms was comparable with its distribution in schistosomules fed labeled reticulocytes or lyophilized, labeled hemoglobin. Pretreatment of the worms with puromycin partially inhibits incorporation of the label.

NOTE: Paper 10a will be given on Friday morning; Paper 12a will be the first paper of the Saturday morning session.