

ZOOLOGY

Chair: RONALD L. RICHARDS
Indiana State Museum
202 N. Alabama Street
Indianapolis, Indiana 46204 (317) 232-1637

Chair-Elect: ALBERT A. WILLIAMS
Department of Biology
Manchester College
N. Manchester, Indiana 46962 (219) 982-2141 ext. 308

ABSTRACTS

A Pigmented Northern Blindfish, *Amblyopsis spelaea*, and Notes on Pigmentation in Amblyopsid Cavefishes. CLAUDE D. BAKER, Indiana University Southeast, New Albany, Indiana 47150.—An unusual adult specimen of northern blindfish, *Amblyopsis spelaea*, exhibited a striking bicolor pigmentation pattern. Previous investigators have considered pigment loss irreversible and subsequent pigment reappearance unlikely. During normal development of this species, pigment disappears under natural light or laboratory illumination. The gross anatomical and histological characteristics of the pigmentation pattern were studied. Additional work was undertaken to update information on pigmentation in the amblyopsid cavefishes.

Proteolytic Enzymes in Brook Trout and Goldfish Follicles during Final Maturation and Ovulation. AMY K. BERNDTSON AND FREDERICK W. GOETZ, University of Notre Dame, Notre Dame, Indiana 46556.—Follicular proteolytic activity was observed, using substrate-gel electrophoresis, in brook trout (*Salvelinus fontinalis*) and goldfish (*Carassius auratus*) oocytes at different stages throughout final maturation and ovulation. Follicular extracts were resolved on a 12.5% polyacrylamide gel containing 0.1% gelatin as the substrate. Proteolytic enzymes were observed as clear zones of digestion against the blue protein background after the gel was incubated in a reaction buffer and stained with 0.1% Amido Black. Five major enzymes were observed in both brook trout and goldfish follicles. In both species, the activity of two enzymes (28,000 and 80,000 mol wt) decreased before final maturation and the activity of two other enzymes (25,000 and 67,000 mol wt) increased before ovulation. Investigations on goldfish follicles using proteolytic enzyme inhibitors suggest that the 25,000 and 28,000 mol wt enzymes are serine proteases, the 80,000 mol wt enzyme is a Ca-dependent serine protease and the 67,000 mol wt enzyme is a metallo-protease. Supported by a research grant from the Indiana Academy of Science.

A Study of Adenosine Deaminase in the Fruit Fly, *Drosophila melanogaster*. DAVID L. BRAND AND PANG F. MA, Center for Medical Education, Ball State University, Muncie, Indiana 47306.—Adenosine deaminase catalyzes the deamination of adenosine to inosine in the purine degradation pathway. Three forms of this enzyme are known to exist in animal tissues, each with its own characteristic molecular weight. Masses of 200,000 (form A), 100,000 (form B) and 35,000 (form C) have been estimated by gel filtration column chromatography. These enzyme forms are present in various proportions in different organisms and exhibit tissue specificity. The B form is not found in advanced mammals. The enzyme activity level and form distribution patterns are known to change during the process of development of certain organisms. Very little is known about this enzyme

in insects. In this study, adenosine deaminase is isolated from the fruit fly of the genus *Drosophila melanogaster*. The activity level will be measured at various stages of development of the fruit fly. The molecular weight and some kinetic parameters also will be determined.

Studies of Ecology, Ectoparasites, and Other Associates of the Eastern Box Turtle, *Terrapene carolina*. PRISCILLA COSTELLO, Terre Haute South High School, Terre Haute, Indiana 47802.——The purpose of this study was two-fold. The first objective was to analyze four years of data of 20 Eastern box turtles, *Terrapene carolina* confined in a 22' x 19' study area. The data include measurements, records on behavior and hibernation, and maps of home ranges. The second objective was to study the ectoparasites and associates of the confined turtles and to compare these data with those of turtles taken directly from the field. Five basic types of associates were found. They are currently being identified, but apparently none are truly parasitic.

Histopathology of Experimental *Dendritobilharzia pulverulenta* Infections in Avian Hosts. DAVID L. DANIELL, Butler University, Indianapolis, Indiana 46208.——Eggs deposited by *Dendritobilharzia pulverulenta* females in the arterial system of experimentally infected avian hosts frequently are carried to tissues other than the intestinal wall, and thus can not escape from the host. Schistosome eggs were found in kidney, lung, liver, pancreas and brain tissues of laboratory-infected mallards, muscovy ducks, Canada geese, and domestic chickens. Stained sections of these organs reveal lesions containing eggs accompanied by varying degrees of inflammatory reaction and granuloma formation. A typical large granuloma contains numerous multinucleated giant cells, a layer of fibroblasts and fibers, and an irregular outer layer of leucocytes (mainly lymphocytes and granulocytes).

Improved Analysis of Physical Forces and Tissue Rheology in Amphibian Gastrulation. GRAYSON S. DAVIS, Valparaiso University, Valparaiso, Indiana 46383.——Cellular aggregates of deep ectoderm, deep mesoderm, and deep endoderm excised from *Rana pipiens* gastrulae undergo movements in organ culture which often mimic normal gastrulation. Such movements resemble the flow of viscous liquids, suggesting that surface tension, which directs ordinary liquid flow, also may direct similar movements of certain living tissues. Indeed, these tissues demonstrably possess the physically-defined (*i.e.*, area-invariant) surface tensions appropriate to entirely account for their liquid-like *in vitro* rearrangements. Unfortunately, quantification of a living tissue's surface tension requires many measurements including a tedious and somewhat subjective curve-matching step, which makes the result prone to random error. A new method using a microcomputer and digitizing tablet makes this analysis much faster, easier and more reliable. This method: 1) provides the correct value for air-water interfacial surface tension and 2) preliminarily indicates that aggregates of deep ectoderm enveloped with surface ectoderm (coat) are not liquid-like, but instead have area-dependent surface tensions just as do elastic solids.

Localization of Acetylcholine Binding Sites in Prepubertal Mouse Ovaries. LARRY R. GANION AND MARY E. MOHR, Ball State University, Muncie, Indiana 47306.——It has been demonstrated that oocytes of several animal species respond to acetylcholine, which suggests the presence of acetylcholine receptors in the oocyte membrane. To assess the site of acetylcholine binding, ovarian fragments from prepubertal, HS mice were cultured at 37°C *in vitro* in Eagle's medium containing H³ - acetylcholine (68 µc/ml) for 60 minutes. Next the tissues were rinsed, fixed in 3% glutaraldehyde, and embedded in paraffin. Sections were cut, coated with Kodak NTB-2 emulsion, exposed for 20 days, developed, and stained. As revealed from the autoradiographs, the label is primarily associated with

the secondary ovarian follicles. In such follicles, most of the silver grains are concentrated over the follicular envelope and localized between and on the surface of follicle cells. In some follicles, the oolemma is extensively labelled. These data suggest that acetylcholine receptors are present in ovarian follicles. Additional studies are planned to check binding specificity. The possible function of acetylcholine in the biology of the ovarian follicle will be discussed.

Sterilization of Chinook Salmon, *O. tshawytscha*, Fry by 17α -methyltestosterone. FREDERICK W. GOETZ, F.P. BINKOWSKI, P.D. LORSCH AND S.E. YEO. University of Notre Dame, Notre Dame, Indiana 46556 and University of Wisconsin-Milwaukee, Milwaukee, Wisconsin 53204.——Chinook salmon were treated with 17α -methyltestosterone (17-MT) by periodic immersion (400-800 ug/1 wellwater) during the alevin stage. Low frequency (2 immersions) and high frequency (5-10 immersions) treatments were conducted. 17-MT treatment was continued in the diet at 4 levels: 10, 20, 40 and 80 mg/kg diet for 8-16 weeks depending on treatment. Fry were sampled at swim-up, and 4, 8, 12 and 16 weeks of feeding and the gonads assayed histologically. Phenotypic sex (i.e. testes and ovaries) differentiation could be distinguished by 4 weeks in control fry though an accurate sex ratio was not always possible at this time. All fry that received 17-MT by immersion and in the diet, exhibited some sterilization of the gonads. Many gonads had greatly reduced numbers of germ cells and stromal tissue and an increase in connective tissue. There was an increase in the amount of sterilization observed with 1) higher 17-MT immersion frequencies, 2) higher dietary 17-MT concentrations and 3) longer dietary treatments. The results indicate that sterilization of chinook salmon is possible with certain 17-MT treatment regimens. Project funded by the Wisconsin Department of Natural Resources.

Habitat and Food Interrelationships of Shrews in West Central Vigo County. RONALD L. HELMS, West Vigo High School, West Terre Haute, Indiana 47885.——Shrews were trapped using sunken pit traps at both moist and dry locations in a variety of habitats varying from old field to woods.

Differences in food utilization by *Blarina brevicauda* and *Sorex* sp. and shifts in food utilization with differing habitats are being analyzed. Stomach contents of specimens also are being analyzed to determine amounts and kinds of foods eaten. Diets of same or similar species studied elsewhere will be compared with the specimens collected in this study.

Ultrastructural Changes in the Proximal Tubule of the Axolotl. MOHINDER S. JARIAL, Center for Medical Education, Ball State University, Muncie, Indiana 47306 AND MARGARET W. EGAR, Department of Anatomy, Indiana University School of Medicine, Indianapolis, Indiana 46223.——Short finger mutants of *Ambystoma mexicanum* from the Bloomington colony were examined for ultrastructural changes in the mesonephros. Two of these adult animals were expressed ascites as well as characteristic reduced limb and digit length. Upon gross dissection, one showed extreme kidney pathology. The short finger mutation was described by Humphrey (1967) as a recessive lethal trait that results in reduced digit and limb length, absence of the Mullerian duct, and with edema and/or ascites.

The grossly abnormal kidney had pathological changes at the ultrastructural level in the proximal tubule. These tubule cells were massively distended by multiple lysosomes, filled with small membrane bound bodies. Normal mitochondria were not found except in the distal tubule and collecting duct where the other cytoplasmic organelles are like those in the control kidney. The abnormal proximal tubule cells appear to have been involved in an autophagic reaction that left no functional cytoplasm at all.

Winter Fishes in Bayou Creek Drainage. SCOTT A. LEATHERLAND AND THOMAS H. CERVONE, University of Southern Indiana, Evansville, Indiana 47712.—In the winter of 1986, twenty-five species of fish from ten families were collected in Bayou Creek drainage. The most abundant fishes found were the emerald shiner, bluntnose minnow, bluegill, mosquitofish, and creek chub respectively. These five species comprised 80.5% of the entire collection, while disproportionately, the remaining twenty species made up but 19.5%. This exaggerated abundance of but a few fishes compounded with an average species diversity index of 1.5 ± 0.7 indicated a depressed fish fauna in Bayou Creek and its tributaries. The spottail darter, a special concern species in Indiana, was found in six of the twenty-six (23.1%) stations sampled. All collections of this darter were in headwater tributaries. Both the spottail and mud darters found in this investigation were new records for Vanderburgh County.

Coteratogenesis of Caffeine and Acetazolamide in Mice. CHARISSA M. URBANO AND SIDNEY L. BECK, Ball State University, Muncie, Indiana 47306.—Preliminary results suggest the positive coteratogenic effect of caffeine coadministered with acetazolamide to pregnant C57B1/JBk mice on day 9 of gestation. Subcutaneous injections of caffeine with a total dose of 50 mg/kg were followed immediately by injection of acetazolamide at total dosages of 200 mg/kg (low dose) or 1.00 g/kg (high dose) in three equal injections 7 hours apart. Females were sacrificed on day 18 by CO₂ asphyxiation and fetuses were removed and examined for gross malformations. A pronounced increase in fetal malformations, especially forelimb ectrodactyly, was evident in both low dose and high dose groups when contrasted with controls (untreated, vehicle-treated, caffeine, low dose or high dose acetazolamide). Other malformations observed included: hemimelia, anophthalmia, and agnathia.