

ZOOLOGY

Chairman: WILLIAM J. BRETT, Department of Life Sciences
Indiana State University, Terre Haute, Indiana 47809

DOROTHY ADALIS, Department of Biology, Ball State University
Muncie, Indiana 47306, was elected Chairman for 1972

ABSTRACTS

Perception of the Plane of Polarized Light and Its Use by Orienting Salamanders.¹ KRAIG ADLER, Department of Biology, University of Notre Dame, Notre Dame, Indiana 46556, and DOUGLAS H. TAYLOR, Department of Zoology, Miami University, Oxford, Ohio 45056.—Tiger salamanders (*Ambystoma tigrinum*) were trained to move along a specific axis in an indoor tank under a polarized light (produced by a polarizing filter, HN-38 Bausch & Lomb). Orientation tests were performed indoors in a small water-filled arena which was completely enclosed in opaque plastic. Animals were placed individually in the center of the arena beneath the polarized light source, then released and allowed to move to the edge of the arena where they were scored. Directional responses of salamanders in the arena were the same with respect to the plane of polarization as they had been in training. Rotation of the polarization filter by 90° produced corresponding changes in the direction of movement.

Orientation to the plane of polarization can be demonstrated in sighted and blinded animals. However, if opaque plastic is inserted over the skull of blinded animals, movement is at random; oriented movement is restored, however, when clear plastic is substituted in these animals. These studies suggest that the critical receptor for polarized light is extraoptic.

A Population Study of the Ozark Hellbender Salamander, *Cryptobranchus alleganiensis bishopi*. CHARLES E. MAYS, Department of Zoology, DePauw University, Greencastle, Indiana 46135, and MAX A. NICKERSON, Department of Zoology, Arkansas State University, State University, Arkansas.—During the summers of 1969 and 1970, an extensive mark and recapture study of *Cryptobranchus alleganiensis bishopi* was made on the North Fork River, a branch of the White River, Ozark, Co., Missouri. It was estimated that there are 1,142 salamanders in a 1.65 mile study area (N = 439). From length and weight measurements, it appears that the population consists of at least four age groups. According to a biomass estimate, there are 919 pounds of *C. alleganiensis bishopi* in this area. One riffle has a density estimate of one hellbender per 182 square feet.

¹Supported by an Indiana Academy of Science Grant, NSF GB-30647, and NIH FR 07033-05 to Adler; NSF Post-doctoral Fellowship GU-2058 to Taylor.

Observation of feeding activities as well as stomach analysis indicates that crayfish are the principal food source of *C. alleganiensis bishopi*. Cannibalism and egg-eating, which have been observed in the laboratory, may be important factors in maintaining population stability.

Specific Estrogen Binding Sites in the Nuclear Fraction of the Rat Uterus. J. H. CLARK, J. A. ANDERSON, and E. J. PECK, JR., Department of Biological Sciences, Purdue University, Lafayette, Indiana 47907.—Numerous investigations have demonstrated that the interaction of estradiol -17β with the rat uterus is probably a two-step process in which the estrogen first binds with a cytoplasmic component to form a complex which subsequently moves to the nucleus. A method has been developed for the determination of the number of these nuclear binding sites in estrogen sensitive tissues. The method is based on the observation that estradiol, previously complexed with nuclear binding sites as a result of hormonal injection or ovarian secretions, is freely exchangeable with ^3H -estradiol during *in vitro* incubations of the nuclear fraction. This technique has revealed that the injection of estradiol -17β results in an increased number of nuclear receptors in the uterus, vagina and anterior pituitary but has no effect on kidney or muscle. In addition, the injection of estrogenic hormones, but not testosterone or progesterone results in increased quantities of nuclear binding sites in the immature uterus. Fluctuations in the quantities of nuclear binding of the uterus as a function of endogenous hormonal levels were also examined. In mature cycling rats, uterine nuclear receptor concentrations were: proestrus, 2.44; estrus, 0.58; metestrus, 0.42; and diestrus 1.42 pico moles/per milligram DNA. These data reveal a cyclic fluctuation of nuclear receptor concentration which parallels the ovarian estrogen secretory rate during the estrous cycle.

Effects of Differentiated Brain on the Development of the Nervous System of the Explanted Chick Embryo. ABDULLA LAIRJE and NORMAN A. DIAL, Department of Life Sciences, Indiana State University, Terre Haute, Indiana 42809.—One theory of development suggests that differentiated tissue inhibits in some way the differentiation of the same type of tissue in the early embryo. Blastoderms of chick embryos incubated for 24 hours were explanted to an agar-albumin-glucose medium made up in Howard Ringer solution and further incubated in Petri dish moist chambers for a period of 12 to 24 hours. Small pieces of brain from 7-10 day incubated embryos were placed on the blastoderm near the developing brain of the explant or in small depressions in the medium under the anterior portion of the explant. Homogenized brain mixed in the medium was also used. As a control, chick leg muscle from 7-10 day incubated embryos was used in the same manner. Untreated controls were also used. Fifty-seven (40.7 per cent) of 140 explants treated with differentiated brain showed brain abnormalities, while 15 of 74 (20.3 per cent) muscle-treated controls showed such abnormalities. Twenty of 112 (17.9 per cent) untreated controls had brain abnormalities. The usual kinds of abnormal changes observed included rudimentary or absent optic vesicles, reduced size

and rounded or pear-shaped anterior neural structures. These morphological changes were limited primarily to the forebrain.

Coccidial Immunity Studies In The Grey Squirrel¹ THOMAS JOSEPH, Department of Biology, Indiana University at South Bend, South Bend, Indiana 46615.—The response of the grey squirrel *Sciurus carolinensis* to infection with *Eimeria lancasterensis* and *E. confusa* was investigated. All 17 squirrels live-trapped from Tippecanoe County, Indiana, were naturally infected with *E. lancasterensis*. One of the squirrels carried a mixed infection with both parasites, but *E. confusa* disappeared after a week in captivity. The infection with *E. lancasterensis* persisted throughout captivity in all squirrels. This parasite infected the epithelial cells of the distal two-thirds of the villi. When experimentally inoculated with *E. confusa*, the squirrels took the infection with a patent period that ranged from 7-15 days. The patent period was shortened by repeating the infection; however, additional infections proved to be negative. *Eimeria confusa* infected the entire villus and penetrated deeper into the epithelial cells. The results of the study lead to the following tentative conclusions: 1) Grey squirrels do not develop immunity to *E. lancasterensis*, but will become immune to *E. confusa* and remain so for a period greater than 5 months; 2) Repeated infections with an interval of 1 month do not produce immunity; 3) It appears that immunity is more likely to develop against coccidia that penetrate deeper into the host tissues and may be a factor responsible for the low incidence of *E. confusa* in nature.

¹Supported by a grant from the Office of Research and Advanced Studies, Indiana University.

Relationship between Metabolic and Emergency Rhythms in *Drosophila melanogaster*. KAREN BELCHER, Department of Life Sciences, Indiana State University, Terre Haute, Indiana 47809.—Oxygen consumption and lactic dehydrogenase (LDH) activity were measured to determine a metabolic rhythm and this rhythm was then compared with the emergency rhythm and the relationship determined. Oxygen consumption was determined for *Drosophila melanogaster* pupae that had been exposed to 12:12 or 1:23 light-dark (LD) regime and LDH activity was determined for pupae exposed to 12:12 LD regime.

Bimodal circadian fluctuations of oxygen consumption were found to exist in pupae of *D. melanogaster* exposed to 12:12 or 1:23 LD regime. Pupae under both regimes appeared to demonstrate an anticipatory change in oxygen consumption associated with the change in illumination. Techniques employed in the study did not permit detection of any regular fluctuations in LDH activity but a steady decrease in LDH activity during pupal development was evidenced.

The oxygen-consumption trend for the entire pupal stage showed a high at the time of emergence, but the diurnal rhythm for oxygen consumption showed a low at the time of emergence. These data suggest that emergence occurs at a metabolic low in the diurnal cycle.

Emergence data for the population showed an anticipatory change in the level of emergence to a change in illumination. A dark to light change was accompanied by an increase in number of emergents and a decrease in oxygen consumption. Emergence maximum showed a 3-hour lead over the oxygen-consumption maximum. Changing the LD regime from 12:12 to 1:23 and changing the time of the 1 hour of light by 12 hours in the 1:23 LD regime produced similar changes in the phasing of both oxygen-consumption and emergence rhythms.

Sites of Cell Proliferation in a Colonial Hydroid, *Campanularia flexuosa*. ROBERT L. SUDDITH, Department of Zoology, Indiana University, Bloomington, 47401.—The colonial hydroids exhibit diverse and specific patterns which are generated during the normal growth of the colony. Analysis of these growth patterns requires an understanding of the mode of growth. Morphological growth is expressed in the colonial hydroids as terminal elongation concomitant with an increase in colony mass (all hydroids) and pedicels (thecate hydroids). The sources of growth—primarily cell proliferation and secondarily increase in cell mass—have been thought to reside in the site of morphological elongation, the stolon tip, in a situation analogous to an apical meristem in plants.

When bands of cells in an actively growing stolon are marked with Nile blue sulfate, their movement does not correspond with that to be expected if cell proliferation is in the terminal region. We have undertaken a series of pulse-chase experiments using ^3H -thymidine as a marker to determine the site(s) of cell proliferation in an elongating stolon. Colonies were placed in filtered sea water containing ^3H -thymidine at a concentration of 5 micro Curie per milliliter for 1 hour and then incubated in filtered sea water containing 10^{-4} molar cold thymidine for one hour, before fixation. The tissue was fixed in Bouin's, embedded in Paraplast, and sectioned at 6 microns. The slides were coated with Kodak NTB-3 liquid nuclear tracking emulsion and exposed for 29 days. The developed slides were stained with Harris hematoxyline and counterstained with eosin.

Detailed analysis of the position of labeled cells following the 1 hour pulse with ^3H -thymidine reveals that only a few cells are labeled in the terminal 200 microns of the stolon tip. In the region 0.5-1.0 millimeter behind the tip there is a high density of labeled cells; beyond this to a distance 2.4 millimeter behind the tip there are few labeled cells. This is in direct contrast to the concept of a restricted terminal cell proliferation region. Proliferation of cells elsewhere than at the stolon tip is compatible with the results of vital staining experiments of Hale, Crowell, Syttenbach, and Suddith. In the particular stolon studied, the region containing a high density of labeled cells is at the level at which a new hydranth would be expected to develop.

Some Effects of Methylmercury on Early Frog Development. NORMAN A. DIAL, Department of Life Sciences, Indiana State University, Terre Haute, Indiana 47809.—Little is known of the effects of methylmercury on early embryonic development. Preliminary investiga-

tions utilizing embryos of the frog (*Rana pipiens*) have been conducted. Embryos in various stages of early development were treated with various concentrations of methyl mercuric chloride in Holtfreters growing solution. The extremes in concentration ranged from 0.2 to 200 parts per billion (ppb). Solutions were changed daily to maintain a relatively constant exposure to a given concentration. Duration of exposure ranged from 2 days at higher concentrations to 7 days at lower concentrations.

Results indicated that selective developmental defects (poor tail development) may occur at concentrations as low as 1-5 ppb when treatment was begun in the late blastula and neural plate stages. Concentrations of 80 ppb or more inhibited development altogether within 36 to 48 hours when treatment was begun in early cleavage and late neural plate stages. A graded response as a result of time and concentration was evident. For example, midgastrula eggs treated with 10 ppb methyl mercuric chloride resulted in stunted, irregular tails and a slight slowing of body development after 6 days of exposure. On the other hand, eggs treated at the 32-cell stage of cleavage with 20, 40, 60, 80 and 100 ppb methyl mercuric chloride showed the following after 24 hours: Controls—late blastula—early gastrula; 20 ppb—same as controls; 40 ppb—same as controls; 60 ppb—mid-blastula; 80 ppb—mid-blastula; 700 ppb early blastula. After 48 hours controls were in the neural plate stage, while the 20, 40, 60, 80 and 100 ppb treated groups showed the following: Complete dorsal lip, early dorsal lip, late blastula, mid-blastula and early blastula, respectively. No further development occurred in any of the treated groups while controls continued to develop normally. Further investigation is continuing to determine uptake, localization and whether or not stage tolerance exists.

NOTE

Fish Populations in the White River near Petersburg, Indiana. ROBERT S. BENDA¹, Department of Life Sciences, Indiana State University, Terre Haute, Indiana 47809.—A 6-year study of the fish populations in the White River near Petersburg, Indiana, was concluded in October 1970. Two hundred and twenty-seven collections were made in a total of 237 hours. Fifty-three and one-half hours were used in 63 seining collections, 44 1/2 hours were used in 111 AC electro-fishing collections, and 139 hours were used in 53 DC electro-fishing collections. Collections were made in all habitats during the months of May through November each year.

The most frequently captured fish, based upon the per cent of collections containing at least one specimen, are as follows:

Gizzard shad, *Dorosoma cepedianum* (69%); Carp, *Cyprinus carpio* (44%); River carpsucker, *Carpionodes carpio* (37%); Spotted bass, *Micropterus punctulatus* (34%); Longear sunfish, *Lepomis megalotis* (34%); Emerald shiner, *Notropis atherinoides* (34%); Spotfin shiner *N. spilopterus* (31%); Bullhead minnow, *Pimephales vigilax* (31%); Longnose gar, *Lepisosteus osseus* (23%); Bluegill, *Lepomis macrochirus* (22%); Silvery minnow, *Hybognathus nuchalis* (20%); Freshwater drum

Aplodinotus grunniens (18%); Shortnose gar, *Lepisosteus platostomus* (15%); Sand shiner, *Notropis stramineus* (14%); Steelcolor shiner, *N. whipplei* (13%); river shiner, *N. blennioides* (13%); Green sunfish, *Lepomis cyanellus* (12%); Orange-spotted sunfish, *Lepomis humilis* (11%); Black crappie, *Pomoxis nigromaculatus* (11%); and the white crappie, *Pomoxis annularis* (11%). The other 49 species were collected less frequently and can be found listed in two other works (1, 2).

The Mosquitofish, *Gambusia affinis*, was captured twice. A single specimen was collected in a slough area in 1969 and one specimen was collected in the main river in 1970. This is the farthest north this species has been collected in Indiana according to published records (3, 4).

¹Present address, Natural Science Division, Aquinas College, Grand Rapids, Michigan 49506.

Literature Cited

1. PROFFITT, M. A. 1969. Effects of heated discharge upon aquatic resources of the White River near Petersburg, Indiana. Water Resources Research Center. Rep. of Invest. No. 3. 101 p.
2. BENDA, R. S. 1971. Effects of thermal effluents upon the growth and distribution of fish in the White River near Petersburg, Indiana. Unpublished Ph.D. Dissertation. Indiana State Univ. 97 p.
3. GERKING, S. D. 1945. The distribution of fishes of Indiana. Invest. Indiana Lakes and Streams. 3:1-137.
4. NELSON, J. S., and S. D. GERKING. 1968. Annotated key to the fishes of Indiana. Indiana Univ. Indiana Aquatic Res. Unit Proj. No. 342-303-815. 85 p.