mined by precipitation with trichloracetic acid,² and the total nitrogen was estimated in the precipitates by the micro-Kjeldahl method.³ Simultaneous estimations were made of reduced and oxidized glutathione, the former being estimated by the iodometric method, using nitroprusside as end-point indicator, the latter being titrated after reduction with bismuth tartrate and H₂S. The total SH (SH glutathione, plus protein SH groups) was determined by iodine titration, and the pH by the glass electrode.

We found that oxygenation of digests of the muscle results in a relatively rapid protein synthesis, representing an increase of from 10 to 20 per cent. of protein nitrogen in terms of total nitrogen. Similar results were obtained with digests from the living portion of the Jensen sarcoma and Walker carcinoma, and likewise in a digest composed of papain plus fibrin and SH glutathione. Our results with these extracts indicate that the conditions favoring synthesis of proteins are (1) a relatively high oxygen tension; (2) a hydrogen-ion concentration not far removed from neutrality; (3) a relatively high initial concentration of SH groups attached to protein or glutathione which can give rise to a relatively high concentration of disulfides; (4) a sufficient concentration of suitable protein split products.

Small amounts of CuSO₄ added to the digests immediately before the oxygenation is begun may cause a temporary acceleration of protein synthesis, which, however, is followed by a resumption of protein cleavage.

These results obtained in vitro are of interest, as they suggest that variation in the oxygen supply of the tissues under physiological and particularly pathological conditions may exert a controlling influence on the equilibrium between cleavage and synthesis of tissue proteins. It may be that an inadequate O. supply to certain portions of malignant tumors favors necrosis and digestion of intracellular protein. the other hand, in portions of tumors which are well supplied with oxygenated blood, conditions may be favorable for protein synthesis and therefore tissue growth. Further work is required before these deductions can be applied to other normal and malignant tissues. A detailed report of this investigation will be published in the Journal of Pharmacology and Experimental Therapeutics.

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THE UNIQUE NUTRITIONAL ORGANS IN THE EMBRYOS OF THE TOP MINNOWS OF THE MEXICAN PLATEAU

During Cretaceous times the Mexican Plateau was isolated to some extent by the development of high volcanic ridges on the east, south and west and by desert on the north and the northwest. A fauna has developed here which is quite unique in some animal groups, and this is especially true in the case of the teleost fishes. While there has been some migration into and out of the plateau, for the most part the species upon the plateau do not exist elsewhere. In one instance, a family which is represented in North America elsewhere by a single species has bloomed in the isolation provided by plateau into many species.

It has been pointed out by Hubbs that most of the top minnows of the Lerma Valley should be separated into a single family, the Goodeidae. The classification is based principally upon the structure of the teeth and fin peculiarities. The family, as revised by Hubbs, contains the Genera Skiffia, Lermicthys, Charcodon, Balsadicthys, Goodea, Chapalicthys, Zoogoneticus and Girardinicthys. A study of the reproduction of seven of these genera reveals peculiarities that separate the group radically from all other top minnows and several features which are unique and undescribed. These features are the subject of this brief article, which is to be followed later by a more extensive illustrated description.

The course of reproduction in the lowland type of killifish (Gambusia, etc.) is characterized by the following features:

- (1) Fertilization is accomplished by the transfer of spermatophores from male to female by means of a gonopod. Fertilization is internal. The sperm are stored by the female and serve for several succeeding broods of young.
- (2) The ovarian follicle builds up a large yolk mass which is the sole means of nourishment for the embryo.
- (3) The embryo remains within the ovarian follicle until hatched. It is then extruded into the intraovarian space and is born almost immediately. The yolk mass is almost entirely absorbed at birth.
- (4) The gonads of the embryo are quite undeveloped at birth, no differentiation of ovary or spermary being discernible.

The contrast as shown in the reproduction of the highland type, Goodeidae, is indicated as follows:

- (1) There are no spermatophores and there is no true gonapod in the male. In all genera, however, the anal fin has some short stiff rays at the anterior margin. Fertilization is internal, but apparently there is no storage of sperm.
 - (2) The ovarian follicle builds up a very small yolk.

² Seibert, Jour. Biol. Chem., 70: 265, 1926.

³ Koch and McMeekin, Jour. Amer. Chem. Soc., xlvi: 2066, 1924.

There is an extensive degeneration of eggs after they have reached the maximum size.

- (3) The embryo remains within the ovarian follicle until the small yolk mass is almost absorbed, in the meantime developing a group of nutritional filaments. A yolk sac also develops far past the limits of the yolk mass. The ovary develops a system of interfollicular canals and clefts.
- (4) The embryo hatches and is left by a withdrawal of the ovary in the intra-ovarian space. The nutritional filaments develop into long finger shaped processes which are attached to the embryo at the proctodoeum. In at least four genera a wide space in the sub-mucosa of the embryonic gut extends out into the nutritional processes as far as the distal ends. processes become liberally supplied with tissue spaces, sinuses and small blood vessels. While the embryos of all genera possess the nutritional processes there are structural differences that are constant in each
- (5) The embryo increases many times in volume while lying in the intra-ovarian space and after absorbing the small volk mass.
- (6) The embryo is born in an advanced stage with the gonads differentiated into recognizable ovaries and The nutritional processes are still at spermaries. their maximum point of development at birth. They are either broken off or absorbed within a few hours.

Reproduction in the lowland type of killifish is classified as simple ovo-viviparity, the egg, and later the embryo being retained in the ovarian follicle up to the time of birth. Reproduction in the highland type of top minnow is a two-phase process. The first phase, extending from fertilization to the point of the extrusion of the embryo from the follicle into the ovisac is an ovo-viviparity comparable to but shorter than the entire process in the lowland type. second phase is a superimposed and genuine viviparity which is not comparable with any stage in the reproduction of the lowland type. The reproduction in the highland top minnows is therefore classified as ovoviviparity with a superimposed viviparity.

The highland type of top minnows apparently arose from an ancestor much like the lowland type after viviparity had been established in the latter. yolk sac developed in the latter type beyond the limits of the yolk mass is testimony that the ancestral type possessed a large yolk sac. The evolution of the reproductive peculiarities in the highland type from the condition which existed in its ancestral type would involve the following changes:

- (1) Changes in the ovary to prohibit the full development of the yolk or to produce degeneration in the egg before it reached its full size.
 - (2) Earlier hatching of the egg.

- (3) Retention of the embryo for a longer period in the intra-ovarian space.
- (4) Development by the embryo of processes for absorbing food and oxygen from maternal ovary.
- (5) Extensive changes in the somatic portion of the ovary, especially in vascularity, to furnish nourishment for the embryos.
- (6) Interruption of the regular course of the germ cell cycle within the ovary due to the diversion of nutrition from the developing germ cells to the growing embryos.

The material for the above study was secured by an expedition financed by Northwestern University and the National Research Council. The writer was able to collect and to transport alive specimens of seven of the genera of the Family Goodeidae. All have proved to be suitable aquarium fishes and two (Goodea bilineata and Zoogoneticus cuitzeoensis) have produced an excellent breeding stock. Grateful acknowledgment is made to Dr. Isaac Ochoterena, of the National Institute of Biology of Mexico, and Mr. Quintin Rosas, of the Department of Forestry and Game, for courtesies in connection with the securing of collecting permits and to Mr. F. M. Riveroll, of the Department of Express, and Mr. F. C. Lona, of the National Railways of Mexico, through whose efforts shipments of live specimens were greatly facilitated.

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