ON THE BREEDING AND DEVELOPMENT OF INDIAN "MOSQUITO-FISH" OF THE GENERA *APLOCHEILUS* McCLELLAND AND *ORYZIAS* JORDAN & SNYDER.

By T. J. JOB, *M.Sc.*, *Lady Tata Memorial Research Scholar.*

(From the Laboratories of the Zoological Survey of India, Calcutta.)

CONTENTS.

INTRODUCTION.

Mosquito-control by the use of fish has long been recognized, though much controversial literature has appeared in regard to the practical efficiency of individual species. Of the numerous Indian fishes listed as mosquito-consumers, the most important are the indigenous Killifishes[1] of the genera *Aplocheilus* McClelland[2] and *Oryzias* Jordan and Snyder. The former genus is represented in India by three or four

[1] Jordan (1923, pp. 158, 159) includes under the term "Killifishes" the oviparous Cyprinodonts with the anal fin not modified in the male, while the term "Top Minnows" is restricted by him to viviparous Cyprinodonts having the anal fin in the male modified into an intromittent organ.

[2] For nomenclature see Smith, 1938, p. 165.

species *viz.*, *A. panchax* (Ham.), *A. blockii* (Arnold), *A. lineatus* (Cuv. and Val.) and *A. dayi* ? (Steind.), and the latter by a single widely distributed species, *O. melastigma* (McClelland). These Killifishes have been found to be of immense value in the biological control of mosquitoes. The bionomics of the mosquitoes in the Tropics have been studied in fair detail, but as Prashad and Hora (1936, p. 643) have observed, our knowledge of the ecology and biology of the Indian fishes is still " very meagre ", and " for control measures to be successful it is essential that not only the ecology and biology of the pest should be known in all possible detail, but similar information should also be available in regard to the natural enemy which it is proposed to employ for control work "

The larvicidal habits of the North Indian Killifish, *Aplocheilus panchax* have been discussed by various workers such as Brahmachari (1909, p. 320), Chaudhuri (1909, p. 36), Sewell and Chaudhuri (1912, p. 34), Southwell (1920, p. 183), Covell (1927, p. 39), Hora (1927, p. 188), Prashad and Hora (1936, p. 639), Sen (1937, p. 358) and Roy (1938, p. 405) ; its practical utility for mosquito-destruction has also been estimated (Hora and Nair, 1938 ; Job, unpublished work). In view of these observations *A. panchax* has been recognized as one of the best of the indigenous larvicidal fish which can be effectively used in natural pieces of water as well as wells, cisterns and other artifical containers. As for its congener, *A. lineatus* of Peninsular India, writers like Aitken (1902, cited by Bannerman, 1910, p. 525), Bentley (1910, p. 417), Southwell (*loc. cit.*), Sewell and Chaudhuri (*op. cit.*, p. 5), Fraser (1938, p. 97) have all testified to its larvivorous propensities. Analysis of the gut-contents and field observations made in the year 1938 by the present writer at Cheranellore and at the Karamana Entomological station in Travancore, in the suburbs of Ernakulam in Cochin, and at Bandra and Mahim in Bombay point to its being of special value in anti-malarial campaigns, especially in the Deccan. Stoye's (1935, p. 84) remark that the fish " will thrive on a heavy diet of earthworms during the breeding season ", Innes's (1939, p. 150) statement that " its favourite live food is fish " and Moody's (1933, p. 187) observation of a fish suddenly disposing " of a full grown male guppy " are with reference to artificial conditions in aquaria only, and in its natural environment it is seen to feed mainly on insects such as ants like *Camponotus compressus, Solenopsis geminata*, etc., water beetles and small quantities of Dipterous larvae, Ostracods, etc. Rai Bahadur Dr. S. L. Hora has informed the writer that this form has recently been studied in detail by Dr. C. C. John of the University of Travancore in collaboration with the Health Officer, Dr. C. T. George, and that these workers have adduced sufficient experimental evidence regarding its value as an efficient larvivore. The other species, *A. blockii* and the related *Oryzias melastigma*, the writer has reasons to believe, both from previous records and from personal observations made in Madras and Calcutta, are also valuable in checking the numbers of larvae.

The utility of the indigenous Killifishes in the biological control of mosquitoes being thus obvious, it was felt that a detailed knowledge of their breeding and development under natural conditions would be of

value for the culture and dispersal of these efficient larvivores in anti-mosquito campaigns. Willey (1910, p. 120) has emphasised that methods of pisciculture "should be based upon a knowledge of the breeding habits of fishes under natural conditions", and Prashad and Hora (*op. cit.*, p. 646) have pointed out the urgent need for definite sources of supply of larvicidal fish in various provinces of India. Further, in the light of Kemp's statement made in 1938 at Cambridge in his presidential address to the Zoological Section of the British Association that "throughout almost the whole of the vast stretch of the Indo-Pacific region there is scarcely a fish whose life history is fully known and whose various stages from egg to adult can be recognized", the academic value of such developmental studies is obvious.

As the breeding habits and development are believed to be more or less similar in allied species, *Aplocheilus panchax*, the *tri-choki* or *three-eyed fish* of Bengal was taken up for study as the type of Killifishes. Important differences wherever present are, however, pointed out in the consideration of the different forms.

ACKNOWLEDGMENT.

I am deeply indebted to Dr. Baini Prashad, Director, Zoological Survey of India for kindly affording me the necessary facilities for conducting this work and for his valuable criticism. To Rai Bahadur Dr. S. L. Hora, under whose direction the work was conducted, I offer my heartfelt thanks for his kind guidance, useful suggestions and constant help. I am grateful to the Lady Tata Memorial Trust for the award of a scholarship to conduct these researches.

BREEDING AND DEVELOPMENT OF INDIAN "MOSQUITO-FISH".

Aplocheilus panchax (Hamilton).[1]

General.—Aplocheilus panchax is an oviparous Cyprinodont, native of India, the Dutch Indies and the Malay Archipelago, and inhabits clear, shallow fresh and brackish waters at low altitudes. The fish has been reported also from the Andamans, and according to Annandale and Hora (1925, p. 34) "it does not appear to have been introduced artificially as its occurrence in the Andamans was mentioned by Day in 1870" Herre (1939, p. 331) also reports it to be native of the Andaman waters. Mukerji (1935, p. 260), however, presumed that it has probably been introduced there along with *Oryzias melastigma* and other fishes by human agency. Since the penal settlement was opened in the Andamans as early as 1858 (Portman, 1888, p. 8) Mukerji's surmise may not be altogether impossible, though there is no positive evidence for artificial introduction. In India the fish has been recorded from Bengal, Behar, Orissa, Assam, Punjab, the United Provinces, Sind, Cutch and the Central Provinces as also from Burma and Siam, and its bionomics reveal that it can easily be introduced into most places at low altitude. The fish is said to grow to a maximum size of four inches

[1] *Panchax panchax* of Cantor (1849, p. 1234); *Haplochilus panchax* of Günther (1866, p. 311).

in length (Mellen and Lanier, 1935, p. 134); but the average size of the adult common in Bengal is only about two inches though individuals of three inches are often met with. Breeding commences when the fish reaches about an inch and a half in length.

Breeding.—No adequate account of the breeding and development of *A. panchax* in nature is available; but casual references have been made by Day (1878), Chatterjee (1934) and the American aquarists, *viz.*, Innes (1935), Stoye (1935), Mellen and Lanier (1935) and Norlund (1936). It may be observed here that the "*Haplochilus panchax*" mentioned by Thomas (1887, p. 112) was actually *Aplocheilus blockii* (Arnold) (=*Panchax parvus* Sundara Raj).[1] Chatterjee (*op. cit.*, p. 13) writes that *Panchax panchax* (=*A. panchax*) "breeds throughout the year in confined waters of tanks" Innes (*op. cit.*, p. 266) remarks that the fish is "fairly peaceful and a good breeder", and puts down the spawning temperature as 70-84° Stoye (*op. cit.*, p. 84) observes that it is "not as prolific as most others of the group, but quite hardy" Day (*op. cit.*, p. 523) refers to the "very large ova" of the species. Mellen and Lanier (*op. cit.*, p. 134) state that the "eggs are not heavy" and add that the "species matures when half grown, in about four months" According to Norlund (*op. cit.*, p. 60) the eggs take 10-25 days to hatch and "when the young are about 1½ cm. a black spot will appear in the dorsal fin of all of them; when they have grown to a size of about 2½ cm. this spot will disappear on some; these fishes are males"

The sexes in *A. panchax* can be distinguished in the adult usually by a slight difference in size and colour. It has to be mentioned that the colour in the species is variable, and the three types formerly recognized as the blue, the red and the yellow varieties have, as Stoye (*op. cit.*, p. 83) observes, been established as not breeding true to colour. Besides, specimens have been found to adapt their colour, within limits, to varying surroundings. However, the ground colour is usually olive green above, passing to milky white below. But it may vary from a greenish grey to a light olivaceous brown above and may be silvery white below. The fins, which are hyaline, yellowish or greenish grey have orange or sometimes bluish margin, and the caudal is clearly edged with black. Besides minute spots of different colours on body and fins, an ocellus on the lower third of the dorsal fin and above all an oval silvery spot on the occiput, so characteristic of these fish make it possible to distinguish the species without difficulty. The colours of large males are, as Stoye (*op. cit.*, p. 84) says, "richer and more vivid" Further, the dark border along the margin of the caudal fin is wider and more prominent in males, and in them the ventrals and even the anal are usually more of a beautiful canary yellow shade than orange. Norlund's (*loc. cit.*) remark about the disappearance of the black spot on the dorsal fin in males when 2½ cm. long does not hold good for the fish observed by the author. The spot, on the other hand, is seen to persist in both males and females, and it occurs, as Chaudhuri (1916, p. 452) has remarked, in all normal individuals irrespective of sex. Usually the males are of a slightly larger size than the females.

[1] *See* Sundara Raj, 1916, p. 269.

As Innes has noted about the genus in general, the males of *A. panchax* "are vigorous drivers, capable of courting several females at one time" In the aquarium, the chased female stops at intervals, and as Norlund mentions "puts herself in a slanting position against leaves" or other submerged objects, and the male "stops at her side and the pair make some vibratory movements" In response to these the female lays an egg, which is usually fertilised immediately by the male. But the male is not always so successful. Sometimes the female shows resistence and repeatedly avoids the male, which thereupon gives up chase in favour of another female. A strong male may, however, chase and overpower the female, and often the partner is won over by caressing movements with the snout and the body. Spawning usually takes place at dawn though it is often continued into the day.

Dissection of gravid females showed that the ovaries, which are of the cystovarian type, are paired, club-shaped structures, the right one being slightly larger than the left. They are suspended by a thin mesovarium, which is continued backward to form a very short oviduct opening out a little behind and independently of the anus. On an average, there are about a thousand well-formed ova, varying in size from 0·2 mm. up to 1·5 mm. in diameter. Of these, however, only about a dozen are fully developed, while the others are seen to be at various stages of growth. About another thousand minute ova below 0·2 mm. are also discernible besides microscopic egg cells. Thus if Stoye's (1935, p. 82) surmise of 500 eggs for three months is taken into account, there are enough eggs in the adult to lay for a whole year or more, and the fish has been found to be a perennial breeder. The ovarian ova (text-fig. 1*a*) are mostly spherical or subspherical, some being pressed out of shape by the adjoining eggs. Surface threads are differentiated even in the 0·2 mm. eggs and are apparent as peculiar sculpturings, the pattern recalling the ovarian eggs of *Tylosurus strongylurus* described and figured by Jones and the writer (Job and Jones, 1938, p. 245). In the egg of *A. panchax*, however, the threads radiate from around a tiny free spot, and on the rest of the egg the threads are small and scanty, though the folds of the major threads seem to cover the entire egg. The pattern is carried through the older stages until in the oldest ovarian ova the filaments are clearly formed, though they ramain in close apposition to the egg-surface, packed in a wavy pattern. The threads, however, can be easily freed and they stand out from the surface on teasing out an egg.

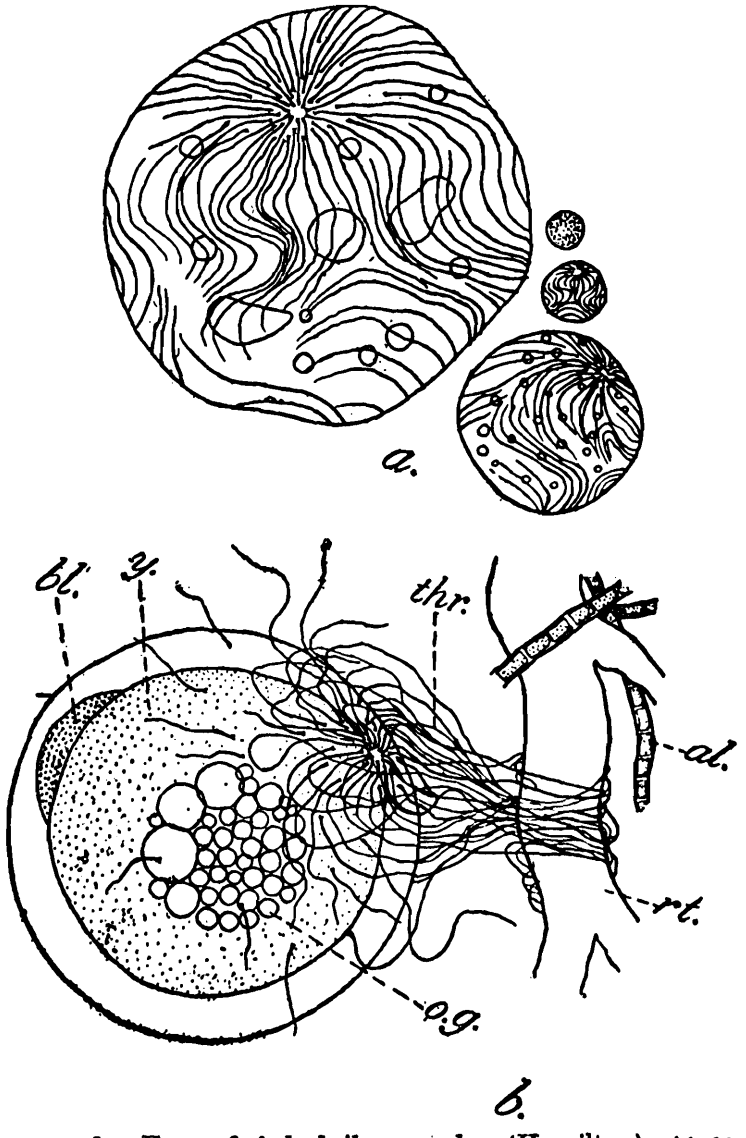
Several eggs of *A. panchax* were collected by the writer on the 1st of May, 1939, long before the monsoon had broken out, from the Municipal drain running along the eastern border of the Superintendent's private compound outside the temple gate of the Pulta water-works in the 24 Perganas. The drain, which is a *semi-pucca* one is practically *cutcha* when overgrown with vegetation as it was in May, but the few inches of water in it was quite clear and there was a very gentle flow. The main source of the water was underground seepage and drainage from a *cutcha* drain running along the north of the compound. Thus while the water was by no means pure, it could still be classed as 'sweet' or fresh. Of the submerged vegetation, the filamentous alga, *Tribonema*

was the most important. Roots of *Mikania scandens* Willd.[1] which crept along the sides, dipped themselves freely in the water. The water supported a rich fauna consisting of larvae of insects such as mayflies, dragonflies, etc., Ostracods and micro-organisms, young of *Gyraulus* and other Molluscs, etc., while ants and other insects moved about among the marginal vegetation. The eggs were found attached (text-fig. 1*b*) singly by the long filamentous threads to the semi-submerged root clumps of *Mikania scandens*, among the meshes of which dark green filaments of *Tribonema* grew. Though solitary, several eggs were generally attached to the same root clump, but these were found to be in different stages of development indicating that they had been laid at different times. Subsequently eggs have been found attached to algal filaments and the fine leaves of aquatic vegetation. During the early stages of development the eggs are transparent, but later become pigmented and appear to be of a dirty brown colour, effectively harmonising with the dark coloured substrata, so that only a very careful examination reveals their presence. Obviously this serves as a protective colouration for the developing eggs. Contrary to the statement of Mellen and Lanier (*loc. cit.*) the eggs are fairly heavy and demersal, and when disengaged from the attachment, sink to the bottom. Subsequently collections were made regularly twice a week throughout the months of May, June and July and afterwards at irregular intervals in the months of August, September and October. The period covered some of the hottest days of the year with the temperature rising to 105°F. on some days in May, with some very stormy and cool days from June to October. Mr. K. K. Nair, who regularly accompanies the Zoological Survey party in connection with the Pulta Survey informs me that he has seen young stages of the fish in the waters round about Pulta in other months as well. It may thus be assumed with Chatterjee (*loc. cit.*) that the fish breeds throughout the year. This does not agree with what has been reported by Norlund (*op. cit.*, p. 60) in regard to the habits of this fish in aquaria in North America. In dealing with *A. panchax* he remarks that the spawning of most of the egg-laying Cyprinodonts is confined to winter. Further, as noted above, Innes puts down the temperature range for spawning as 70 to 84 degrees. But at Pulta, as remarked above, egg-laying is continued even on the hottest day in the year. It is to be noted, however, that the eggs and fry occur in larger numbers during the months of June, July and August when the south-west monsoon rains inundate the low-lying lands and interconnect numerous puddles and pools. The new-born fry at this time wander about the shallow stretches of water over submerged areas, thus effecting their wide dispersal. Hardy as the fish is, it naturally becomes more prolific under favourable conditions. Chatterjee (*loc. cit.*) states that the fish breeds in confined waters in tanks. Observations made at Pulta both in still waters as well as in gently moving waters show that while the fish can live in sweet waters of most kinds, (its occurrence in brackish water has already been recorded), it perennially

[1] The plant was kindly identified for me by Dr. K. P. Biswas, Superintendent of the Royal Botanical Gardens, Sibpur, to whom my thanks are due.

multiplies and flourishes more in shallow waters maintaining a gentle flow and affording suitable substrata for egg-laying.

Some of the early stages of the eggs were examined in the field under a microscope and figures were drawn immediately from fresh material. Others along with the attached root clumps were removed to the improvised laboratory in the Zoological Survey of India camp at Pulta, and the individuals carefully removed along with pieces of attached rootlets to separate aquarium jars containing clear water from the same

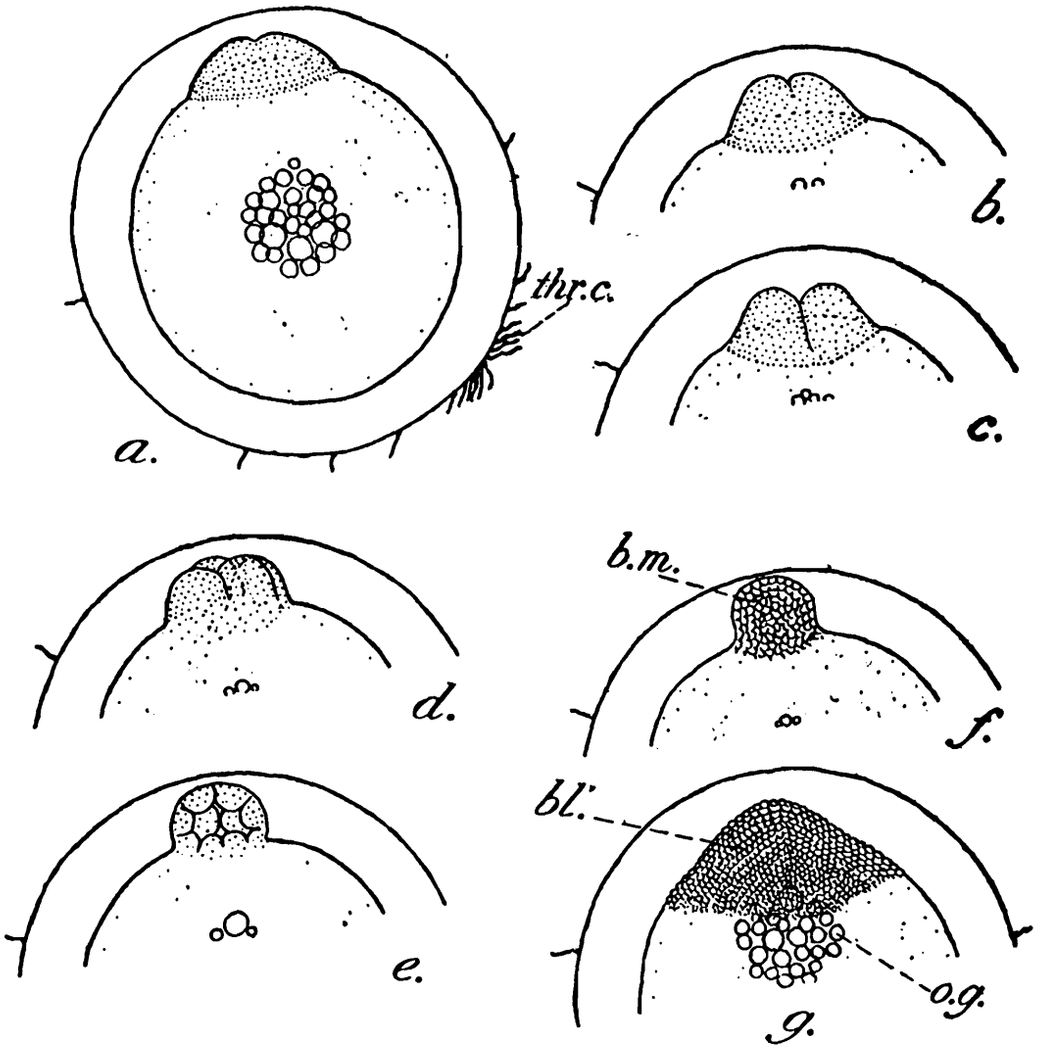


TEXT-FIG. 1.—Eggs of *Aplocheilus panchax* (Hamilton) : × *ca.* 30.
a. Various stages of ovarian ova ; *b*. A fertilised egg anchored to rootlet of *Mikania*. *al*. algal filament ; *bl*. blastodisc ; *o.g*. oil globule ; *rt*. rootlet ; *thr*. adhesive threads ; *y*. yolk.

drain and small quantities of algae so as to aid the natural oxygenation of the water, which was renewed every day. The jars themselves, to keep the contents sufficiently cool, were kept partly immersed in water. The development of the eggs was closely followed, initially at short intervals and later on daily. Care was taken to collect various developmental stages of eggs and larvae from the 'field' and to compare these stages and identical stages in the batches of eggs and larvae allowed to develop in the aquarium jars.

Early Embryonic Development.—The earliest developmental stage collected is figured in text-fig. 1*b*. The egg is fairly large, being highly

laden with yolk, and is more or less spherical, measuring on an average, about 1·4 mm. in diameter. It is transparent and bears adhesive filamentous threads standing out from the surface. At one pole the threads are long and crowded to form a tuft radiating out from around a tiny central spot. The tuft is usually reinforced by a few shorter filaments scattered sparsely over the rest of the egg surface, though most of these " short hairs " stand out free. The long elastic threads can stretch to a length of 20 mm. and readily coil up against any foreign object and effect a safe anchorage for the egg as soon as it is laid. The eggs closely resemble those of *A. lineatus* obtained by Jones and the writer from Travancore (*vide infra*, p. 68).



TEXT-FIG. 2.—Early segmentation and blastoderm formation in the eggs of *Aplocheilus panchax* (Hamilton): × *ca.* 30.

a. Egg, two minutes after first stage (text-fig. 1*b*); *b.* Egg, three minutes after first stage; *c.* Egg, five minutes after first stage; *d.* Egg, half an hour later; *e.* Egg, about two hours after first stage; *f.* Egg, about four hours after first stage; *g.* Egg, about six hours after first stage.

bl. blastoderm; *b.m.* Blastoderm mass; *o.g.* oil globule; *thr. c.* cut ends of adhesive threads.

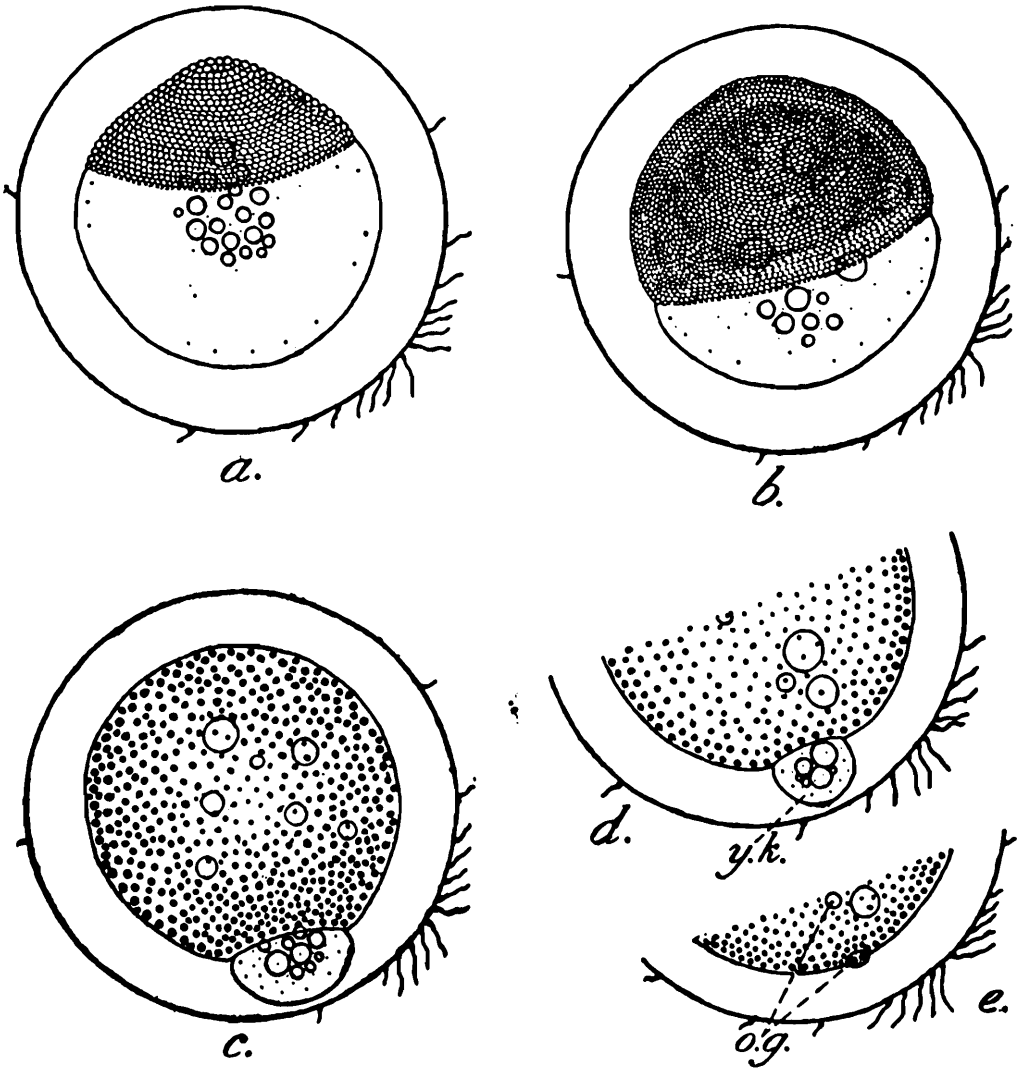
(In *b.* to *g.* only portions of the eggs are shown.)

In a newly fertilised egg the transparent zona radiata is separated from the inner egg mass by a space containing a clear fluid, and the cytoplasm collects together at one pole of the egg and stands out from the yolk surface as a convex dome-like blastodisc which is highly granular and devoid of yolk, while the yolk which forms the major part of the egg is clear and transparent. A group of oil globules, about

thirty-six in number, occurs in the yolk. Their number, however, is highly variable, and even in the same egg it changes in the course of development. They seem to burst and reassemble, but a number of them persist until the larva hatches out. The egg mass being freed from the zona, the blastodisc bears no fixed relation to the pole bearing the adhesive tuft.

Cleavage.

In an egg collected at 9-23 a.m. on the 5th of May 1939, a faint sign of dipping in the middle of the blastodisc was noticed (text-fig. 2*a*) after two minutes, and gradually the dipping advanced (text-fig. 2*b*) and in another three minutes the division was completed (text-fig. 2*c*). Nearly half an hour elapsed before the second meridional



Text-fig. 3.—Invasion of the yolk by the blastoderm and the dispersal of the oil globules in the eggs of *Aplocheilus panchax* (Hamilton): × *ca.* 30.

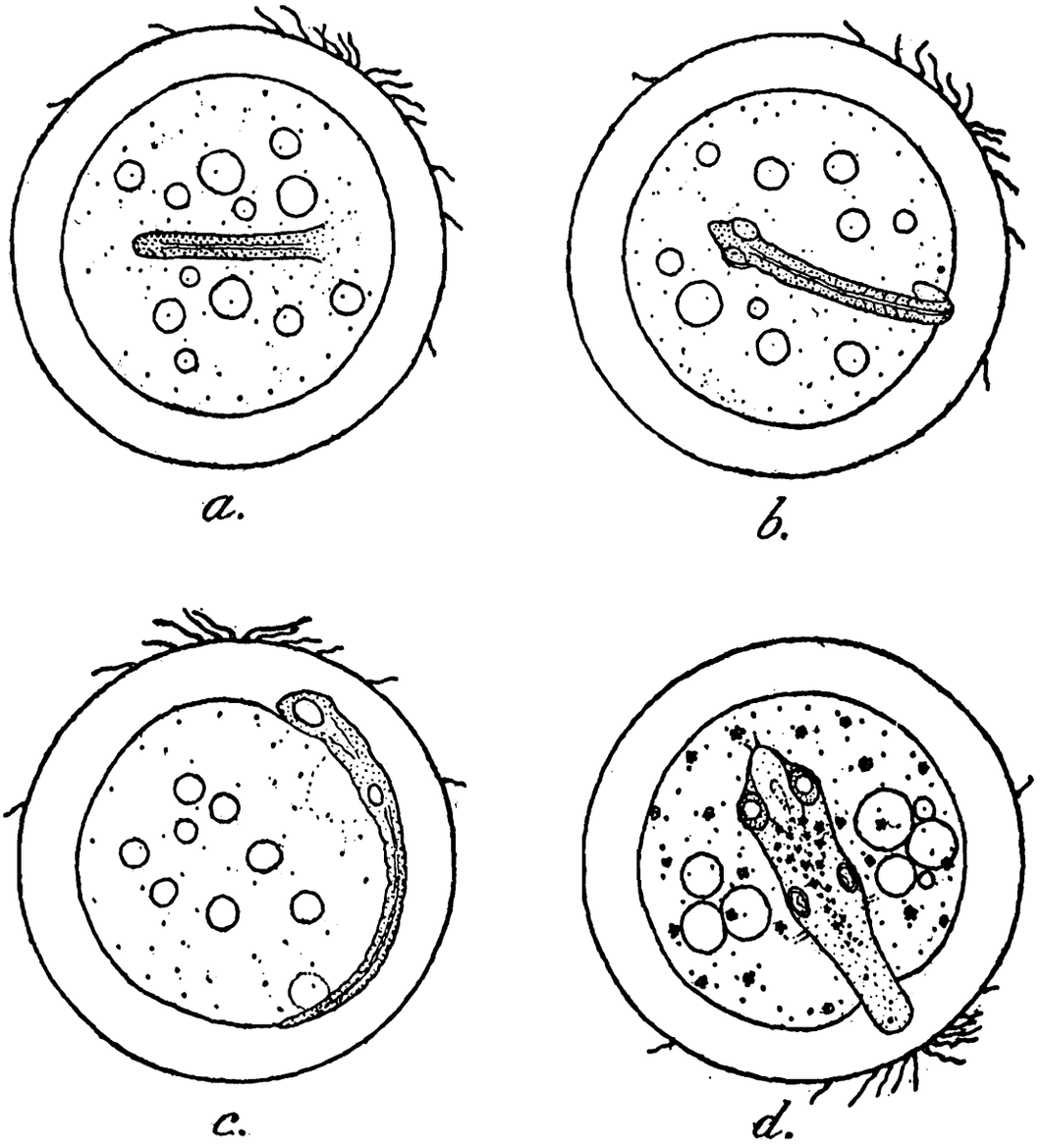
a. Egg, about eight hours after first stage ; *b*. Egg, about ten hours after first stage ; *c*. Egg, about twelve hours after first stage ; *d*. Egg, about 14½ hours after first stage ; *e*. Egg, about sixteen hours after first stage.
o.g. oil globule ; *y.k*. yolk knob.

(In *b*., *d*. and *e*. only portions of the eggs are shown.)

division at right angles to the first was accomplished resulting in four blastomeres (text-fig. 2*d*). In another half an hour the cells doubled their number by dividing horizontally. Subsequent divisions followed in quicker succession in the usual manner and by quarter past eleven the cells formed a projecting button (text-fig. 2*e*).

Yolk Invasion.

In another two hours the button became a mulberry-like mass of minute cells (text-fig. 2*f*) which began to spread at the base over the yolk, and thus the mulberry-like appearance was lost, and the protuberance was reduced as the cells spread out in a thin layer over the yolk with a very faint and indistinct margin (text-fig. 2*g*). In another two hours the cells formed a regular blastoderm layer extending over about one-third of the circumference, with a distinct margin (text-fig. 3*a*). Two hours later, the blastoderm covered more than half of the yolk, and by



Text-fig. 4.—Early embryo differentiation up to the commencement of circulation in the egg of *Aplocheilus panchax* (Hamilton): × *ca.* 30.

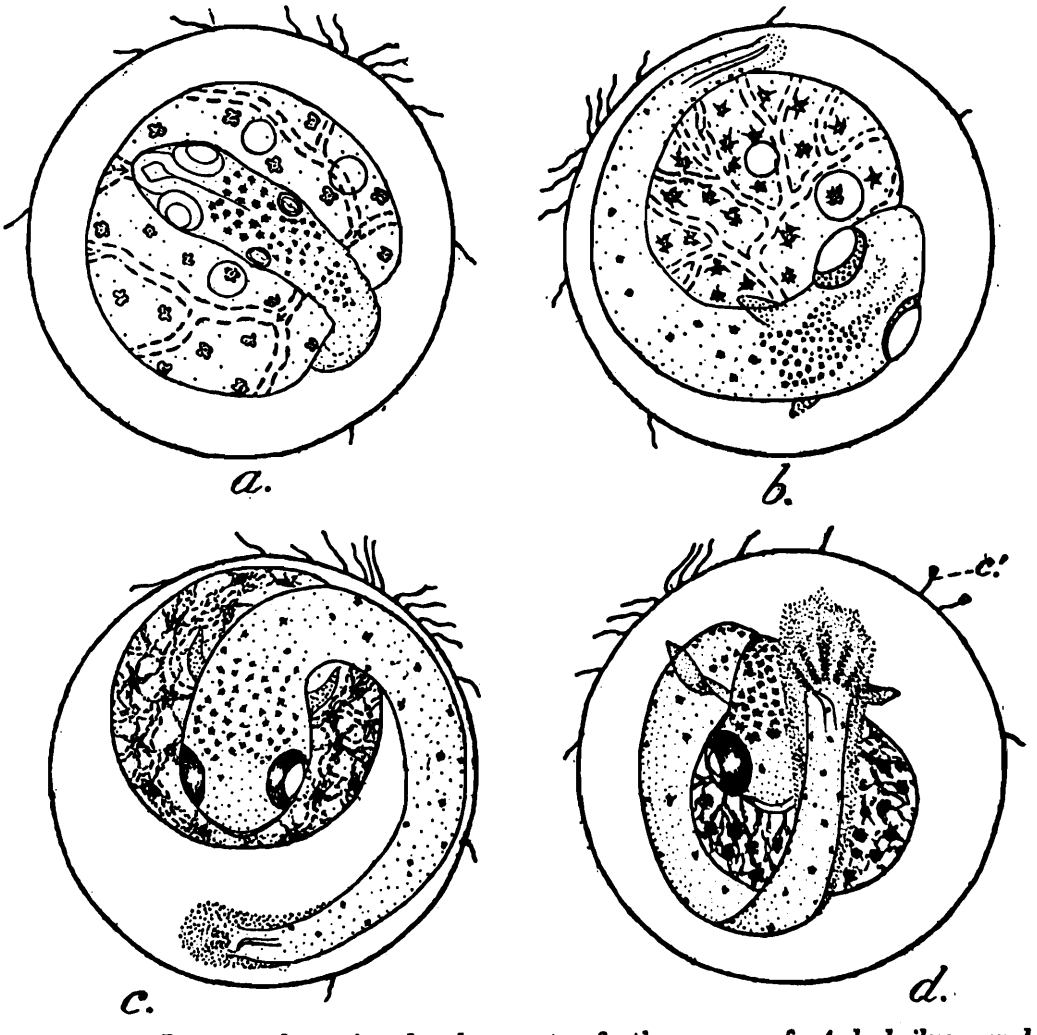
a. Egg, about twenty hours after first stage; *b*. One day old egg; *c*. Egg, thirty hours after first stage; *d*. Egg, about forty-two hours after first stage.

this time the oil globules separated out from the original single group (text-fig. 3*b*). In another two hours the blastoderm extended over more than seven-eighths of the yolk, and the remaining yolk stood out as a large button pinched off as it were by the constricting margin of the blastoderm (text-fig. 3*c*). By midnight this button was reduced to a spherical knob (text-fig. 3*d*) and by 1-30 a.m., *i.e.*, about thirteen hours after the first cleavage, the invasion of the yolk by the blastoderm was

nearly completed (text-fig. 3*e*) and the latter appeared as a thin more or less uniform layer surrounding practically the whole of the yolk.

Formation of the Embryo.

Twenty hours after the first stage the embryonic ridge is formed (text-fig. 4*a*) as a multilayered thickenning in the antero-posterior axis, and in a day old egg (text-fig. 4*b*) the embryo is fairly differentiated, the optic vesicles and about five of the early somites being clear. The embryo now encircles more than one-third of the yolk. In another two hours the auditory vesicles appear and four hours after that they are well marked with about ten somites in the body (text-fig. 4*c*). The whole embryo is embedded as it were in the yolk, and no portion of



TEXT-FIG. 5.—Late embryonic development of the eggs of *Aplocheilus panchax* (Hamilton): × *ca.* 30.

a. Two days old egg; *b*. Three days old egg; *c*. Five days old egg; *d*. Eight days old egg.

c'. Epizoic Ciliate on the egg membrane.

its length is as yet freed from the same. A fair-sized Kupfer's vesicle is visible just ventral to the caudal region. By about the thirty-fourth hour the otoliths are formed in the auditory vesicles, and by the forty-second hour (text-fig. 4*d*) pigmentation is developed. Small rounded black dots at first appearing on the nape and on the yolk-sac develop into star-like blotches, those on the yolk-sac assuming a more diffused form of a brownish colour. The oil globules coalesce and increase in size

while their number decreases. Embryonic circulation just commences with the appearance of vitelline vessels of which there are three main ones, one anterior and two lateral. The embryo shows activity by jerking movements of its tail which is by this time freed from the yolk. The lens appears in the eye, which begins to growfaintly dark.

Late Embryonic Development.—Two days old egg : In a two days old egg (text-fig. 5*a*) the primordia of the pectoral fins begin to appear as tiny conical buds. The heart pulsates rhythmically about 140 times in a minute. The yolk-sac circulation is well established, and a network of vitelline vessels brings blood over the yolk-sac from the posterior region of the body. The side to side movements of the tail are more pronounced. Pigmentation is deeper, the darkest asters being in two paired ovoid groups on the head between the optic and the auditory regions.

Three days old egg : In a three days old egg (text-fig. 5*b*) about 28 somites are recognizable. Oil globules are about 14 in number. The yolk is appreciably reduced. The pectoral buds are well established, but as yet are not mobile. Pigmentation is well defined on the head and nape and sparsely on the body. The yolk-sac bears stellate pigment spots. The heart beats about 150 times a minute. The median fin-fold just makes its appearance as a narrow membranous edging around the caudal end of the embryo.

Four days old egg : In a four days old egg the pectorals are still better developed and begin to make faint flapping movements. The yolk is being reduced markedly by assimilation, and the globules are only about twelve in number. The pigmentation is deeper. The asters on the yolk-sac send out irregular processes, some of which begin to anastomose. Just behind the constriction, a circular area of the yolk-sac is devoid of pigment. The rudiment of the air bladder is faintly visible. The median fin-fold begins to extend as a narrow fringe dorsally and a comparatively broader fringe ventrally. When the egg is four and a half days old the well developed pectoral fins begin to flap actively. The air bladder appears as a clear vesicle. The yolk is further absorbed, and it is now seen to occupy only about half the diameter of the egg. The chromatophores on the yolk-sac begin to form a patchy reticulum. The darkly pigmented eyes on reflection appear pale green.

Five days old egg : In a five days old egg (text-fig. 5*c*) the median fin-fold is fairly well established, and traces of rays already begin to appear in the caudal part. The pigmentation extends to the body as sparsely scattered spots here and there, which, however, appear to form five discontinuous irregular rows, one dorsal, two laterals and two ventrals. The pectoral fins exhibit active flapping movements. The embryo also twists and turns frequently, changing its position inside the egg membrane. The mouth opens and closes at irregular intervals, roughly about thirty-two times a minute. It usually opens slowly, closes incompletely and then snaps tight. Sometimes the whole act of closing is quick and snappy. The embryo has a light greenish yellow hue. Over the yolk, which is now reduced to less than half the diameter of the egg, the vitelline vessels form an irregular network. The air bladder has become elongate. The pigmented areas of the

yolk-sac are connected together into a coffee-coloured patch. Oil globules are reduced to five or six. The hind gut is just discernible. A pale golden sheen is seen all around the cornea in the eye. The pigmentation on the head is denser.

Six days old egg : In a six days' egg the gill arches are clear with the gill rakers. The heart beats about 130 times a minute. Teeth make their appearance as rudiments in the jaws, which latter are now blunt in front. The oil globules are about six in number. Pigment dashes appear on the pectoral fins and three asters are developed on each of the opercles.

Seven days old egg : The next day finds the yolk still further reduced and the embryo larger in size. The pigmentation is deeper, and the Kupfer's vesicle is no longer visible.

Eight days old egg : In an eight days old egg (text-fig. 5*d*) the pigment patches on the yolk-sac anastomose to form a complex web, whose 'stem' is gathered over the yolk constriction. The caudal is rounded and continued as a short narrow dorsal fringe and a longer wider ventral fringe which latter runs up to the anus. About six rays are present in the caudal portion of the fin-fold. The pectoral fins are actively flapped about thirty-two times a minute. The pigment patch on the head is well marked. The eye is blackish with a pale golden green outer ring whose lustre is bright in reflected light. The mouth opens and closes at irregular intervals, about seventy times a minute. The pigmentation of the body is irregular and sparse, but roughly arranged in five discontinuous rows along with some irregularly scattered spots. Epizoic Ciliates such as Vorticellids sometimes become attached to the egg membrane ; for, the accumulation of dirt and traces of decaying matter on the egg surface often promotes the growth of such organisms. But these have not been found to affect the embryo in any way. The egg, however, ceases to be clear and transparent owing to these extraneous organisms, and still more owing to the well developed pigmentation of the yolk-sac.

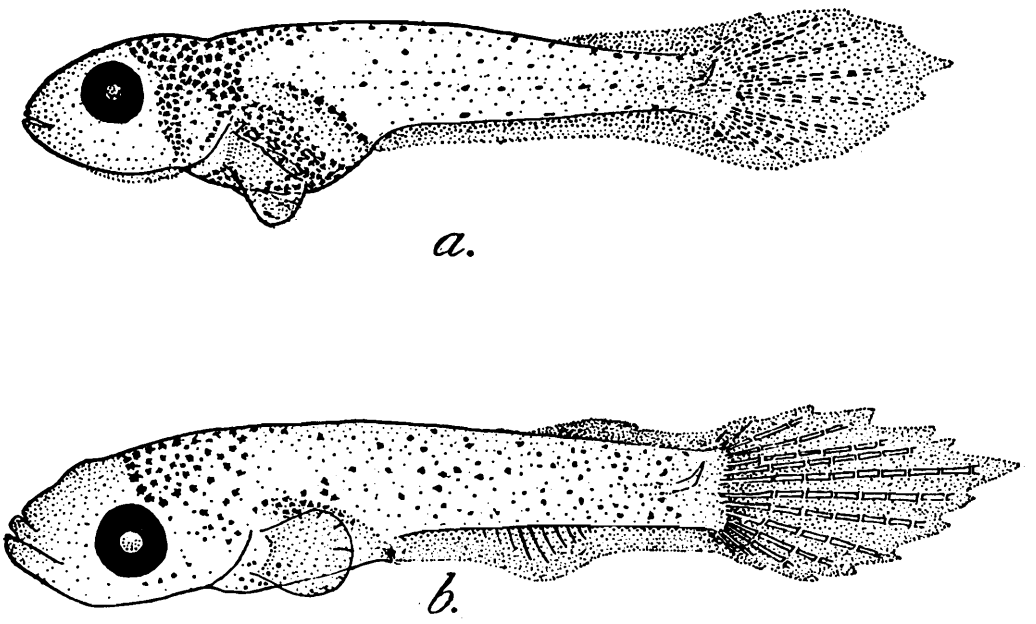
Nine days old egg : In a nine days old egg the yolk is materially reduced, and the embryo, which begins to be remarkably active wriggles furiously inside the egg, while the flapping of the well developed pectorals is strong and regular. Sooner or later hatching takes place, sometimes the head emerging first, sometimes the tail.

Eggs usually take about nine to twelve days to hatch in warm weather and in properly aerated media. But the period may be prolonged in colder weather, and in wates which was not regularly renewed they took eleven to fourteen days to hatch. Norlund (*op. cit.*, p. 60) mentions that in the American winter the eggs of the species sometimes take twenty-five days to hatch.

Newly Hatched Larva.—The exact stage of development of the newly hatched larva varies to some extent according to the period of incubation, those hatching out late being slightly more advanced than early hatchlings.

A larva hatching after ten days' incubation (text-fig. 6*a*) is about 4·4 mm. long and is nearly transparent except for the conspicuous black eyes and dark pigmentation on the nape, and the projecting abdomen

appears more solid and opaque owing to some yolk still present in it. Further, the body is tinged with a light greenish yellow hue. On coming



TEXT-FIG. 6.—Early hatchlings of *Aplocheilus panchax* (Hamilton): *a*. Newly hatched larva: × *ca*. 20; *b*. Five days old larva: × *ca*. 15.

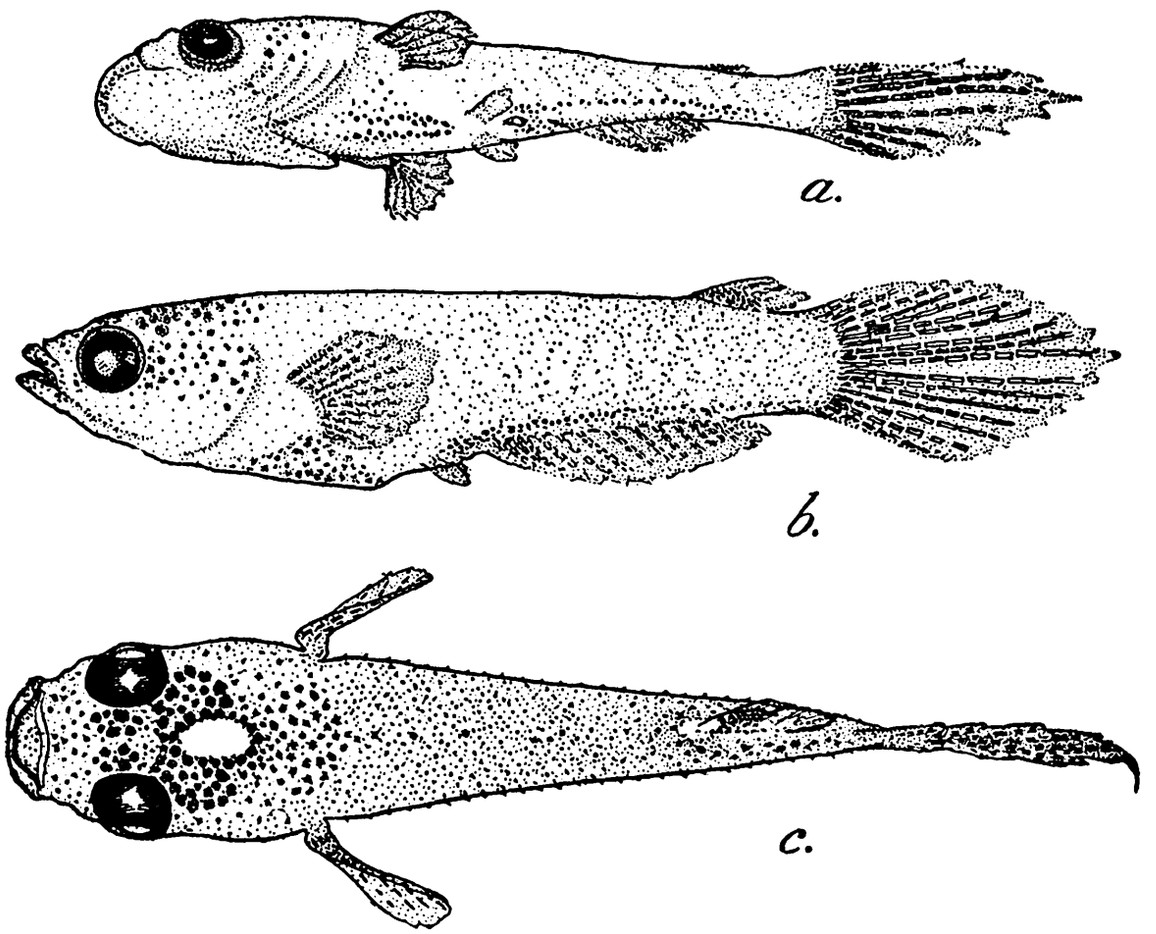
out of the egg the larva usually remains for a minute or two resting on the substratum, with the tiny pectoral fins quivering actively all the while. Having thus acclimatised itself to the new environment it smartly darts up to the surface and swims about hither and thither for some time. When apparently exhausted by these activities it again rests for a while, after which active swimming is resumed. Sometimes, especially in the case of late hatching, the larva swims immediately after liberation from the egg membrane. Often it assumes a nearly vertical pose resting, head downwards, at some spot on the substratum. The head is large, slightly flattened below, but strongly convex above, quite unlike the adult. The eyes are large and blackish with bright green and yellow iridiscent patches. The jaws are sub-equal, the lower being slightly longer than the upper, and the tiny premaxillae are marked off by a shallow groove from the rest of the jaw. The gill region is reddish, and the opercular clefts are well formed. No separate yolk-sac is visible at this stage as the yolk is nearly all absorbed, but it is indicated by a slight swelling of the abdomen. In early hatchers, however, more of the yolk is evident. The vent is apparent just behind the margin of the yolk-sac; from this region the body becomes compressed and gradually tapers towards the posterior end. The pigmentation is characteristic. In most larvae it is blackish, but it may vary from reddish brown to dark brown. In the earliest stage, pigment appears as tiny rounded black spots, these later assume aster-like form and subsequently some of the asters turn into blotchy circles and polygons with irregular margins. A conspicuous patch of asters closely adjoining one another covers the occiput and the nape with a backward prolongation tapering towards the mid-dorsal region and two lateral prolongations on either side over the operculum, the anterior one being directed towards the gular region and the posterior stopping short of the opercular margin, though appearing to merge into the patch on the abdomen behind. In

front the occipital patch of chromatophores is nearly truncate behind the inter-orbital line. On the body at this stage pigment spots are sparse and irregular. They appear to be disposed in five irregular and discontinuous rows, one mid-dorsal, two laterals and two ventrals. Besides, other spots are irregularly scattered. These are few and far between. Chromatophores developing inside the abdominal wall can be seen through the thin wall of the abdomen. The fins excepting the caudal and to a slight extent the pectorals are entirely free from pigment. Each ray in the caudal fin has a double row of tiny pigment dashes. The caudal fin alone is well developed with about nine rays. The urostyle is turned upward. The caudal is fairly large with a few graceful indentations at the terminal region. Dorsally the median fin-fold is continued forward as a thin membranous fold tapering over the middle of the tail. Ventrally it is continued as a similar membrane of more or less uniform width right up to the vent. The pectorals are placed rather low, and though very active, are still hyaline with but slight indications of rays. Since very little of the yolk is left, further growth of the larva depends to a great extent on the quality and quantity of the food available, those obtaining a richer diet and enjoying a free environment growing more rapidly than those kept in small receptacles and supplied with scanty food. The larvae usually feed on diatoms, ciliates and other micro-organisms.

Metamorphosis.—Five days old larva: By about the fifth day after hatching the yolk is completely absorbed, and the length of the larva increases to about 6·5 mm. (text-fig. 6*b*). The dorsal aspect of the head is less convex, and the premaxillae are more pronounced. The pigmentation is deeper and less scanty, but the lateral prolongations of the occipital patch are less marked. The caudal is better developed, having about fourteen well developed rays which appear jointed. The indentations on the margin are more marked. The dorsal fin is beginning to be marked off by a notch in the dorsal fin fold and by the appearance of the rudiments of one or two rays in it. The anal also becomes conspicuous by a slight widening in the middle of the ventral fin-fold, where the rudiments of about nine rays are indicated. The pectorals are still without well formed rays. Though the pectoral fins begin to be functional even before hatching, the fin rays appear only at a later stage. The comparatively dense, though hyaline, appearance of these fins even when inside the egg, shows that even in the absence of rays, they are, with the help of the strong pectoral muscles, capable of active flapping movement.

Ten days larva: By about the tenth day the larva (text-fig. 7*a*) reaches to about 9·5 mm. in length. The premaxillae are now definitely protractile. About twenty rays have appeared in the caudal and eleven in the anal, which is now quite distinctly marked off from the caudal. The dorsal also is similarly demarcated and shows four to five rays. About ten rays have appeared in the pectorals. The pelvic fins are definitely established though no fin rays can be distinguished. A pair of oval patches of pigment spots appear in the inter-orbital region. Pigment has appeared on the jaws, and ventrally on the abdomen the chromatophores are arranged in a pair of irregular longitudinal rows

forking out as it were from the middle of the abdomen and running forward towards the throat. A pair of irregular rows on the ventral aspect



TEXT-FIG. 7.—Late fry of *Aplocheilus panchax* (Hamilton).

a. Ten days old larva, ventro-lateral view : × *ca.* 10 ; *b.* Two weeks old larva, side view : × *ca.* 10 ; *c.* Two and a half weeks old young, dorsal view : × *ca.* 9.

of the tail end in front in a lunar group of pigment round the vent. The pigment patch of the occiput is heart-shaped with a backwardly directed apex ; its posterior continuation appears rather darker.

Two weeks old larva : In a fortnight the larva (text-fig. 7*b*) assumes a length of about 10·8 mm. The ' third eye ' is faintly visible as a slight whitish speck in the middle of the occipital pigment patch. Pigment dashes are formed also on the pectoral fins, the rays of which become jointed by this time. About four rays are seen in the pelvics ; but they are not pigmented. The anal and the dorsal fins are very well demarcated and the membranous fin-fold has completely disappeared except for a narrow adipose remnant on the dorsal side near the base of the caudal. The anal has about twelve rays. About five rays are seen in the dorsal, in which pigment begins to appear towards the base of the three anterior rays as the fore-runner of the ocellus of the dorsal fin of the adult. The caudal has twenty-three rays.

Post larva ; two and a half weeks old : By the middle of the third week, *i.e.*, when nineteen days old (text-fig. 7*c*) the length increases to about 12 mm. The ' third eye ' is established and shines as an oval silvery bright spot. The spot becomes dark being over-run by the chromatophores during fixation. The ocellus on the base of the dorsal fin also is fairly well marked. The mouth appears to be sub-terminal as

the upper jaw looks apparently longer since the premaxillae are now quite protrusible. Larvae of mosquitoes and other insects, if thrown into the aquarium, are consumed by the fish even at this early stage. For all practical purposes the fish is a "young adult", and the assumption of full adult features is now only a matter of further growth.

Aplocheilus lineatus (Cuv. and Val.).[1]

General.—The '*piku*' or '*poochaatie*' is the largest of the Asiatic members of the genus; it has been recorded (Day, 1878, p. 523) as growing to four inches in length; but the average length is not more than three inches. It occurs in Peninsular India, is common in paddy-fields, tanks, canals and even in tidal waters in Bombay, Malabar District, Coorg, Wynaad and the States of Cochin and Travancore, and has been reported also from Ceylon, where it is called '*Irri Nalla Handhaya*' in Sinhalese. Owing to its brilliant hues it is one of the very popular exotic fishes in American aquaria. The habits of this fish are very much like those of its North Indian congener, *A. panchax*, and it is hardy, active and a prolific breeder. In its natural habitat its insectivorous propensities have been fully recognized, as pointed out above, and Prashad and Hora (*op cit.*, p. 640) mention that the fish is reported to be as efficient for mosquito control as *A. panchax*.

Breeding.—No record of the eggs of this fish from natural habitat has appeared so far, and there are but few observations regarding its breeding.

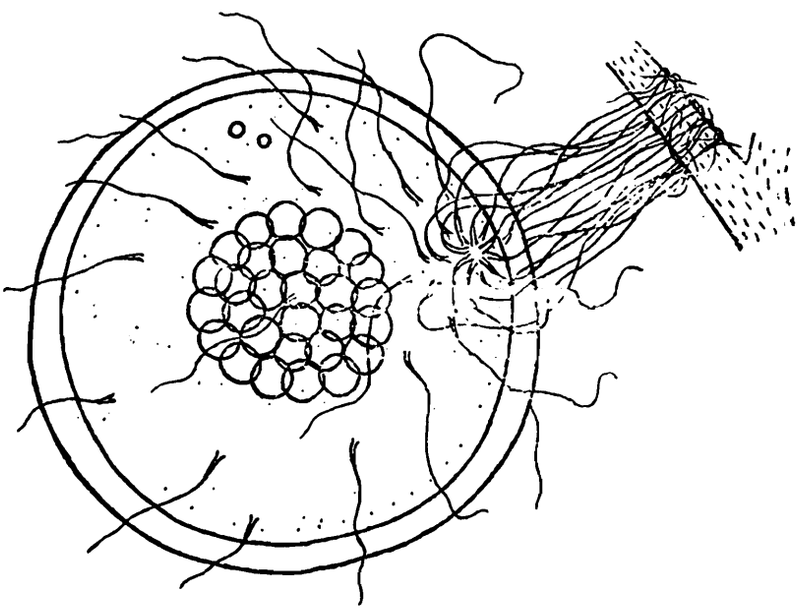
There is a marked sexual dimorphism, and it is noted that the males and females have by some authors been described and designated as two distinct species, *viz.*, *rubrostigma* and *lineatus* respectively. The males far outnumber the females, sometimes comprising, according to Stoye (*op. cit.*, p. 84) ninety per cent. of the offspring. As recorded by various observers the colouration of the species is to some extent variable according to the colour of the surroundings and lighting conditions. The basic colour of the male (*rubrostigma-type*), however, is olive brown, lighter on the ventral region, the chest being bluish white. Rows of metallic golden green and beautiful red dots are scattered on the body. The fins are of a greenish orange hue with reddish fringe. The ventral fins are elongate and with age develop a filose point. About half a dozen faint transverse bars are often present passing across the sides in the hind region of the body.

The female (*lineatus-type*) is darker than the male and has seven to eleven clear black transverse bars, but is devoid of the beautiful red spots of the male. The median fins are paler than those of the male though edged with red. A dark basal spot on the dorsal fin and a shield-shaped silver spot on the occiput are invariably present in both the sexes. The colour differentiation of the male begins with maturity, while the female retains the juvenile colouration throughout life.

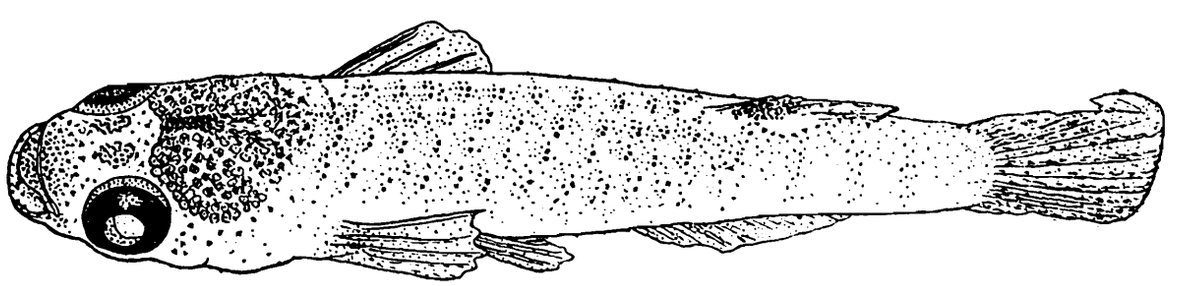
Stoye (*op. cit.*, p. 84) mentions that the fish "seems eager and ready to spawn at all times", and puts down the spawning temperature at 70 degrees with reference to the American aquaria. Mellen and Lanier

[1] *Panchax lineatus* of Day (1878, p. 522) and other authors.

(*op. cit.*, p. 135) state that the spawning habits are similar to those of *A. panchax*. Moody (1933, p. 186) has observed its breeding habits in the aquarium, and writes, " Each morning for a week or more I found them excitedly chasing through the surface plants and pausing every few seconds to deposit an egg. The female was always ahead. She, apparently, picked out the sites. The male's part was limited to coming forward at the proper moment and assisting in the brief ceremony attending oviposition. Both male and female wore their most brilliant colours immediately before and during the egg laying period, and a



a.



b.

TEXT-FIG. 8.—Egg and young of *Aplocheilus lineatus* (Cuv. & Val.).

a. Egg attached by adhesive tuft: × *ca.* 30; *b*. Dorso-lateral view of " young adult ": × *ca.* 7½.

limited amount of courting was observed. The spawning, however, was in no way spectacular, and there was little driving or quarreling " Willey (*op. cit.*, p. 122) refers to the adhesive eggs of *A. lineatus* becoming attached by glutinous threads to water plants. In May, 1938, Mr. S. Jones and the writer collected a few large sized adult fish from the big tank in front of the Entomological Station at Karamana and kept them in an aquarium with a few water plants. The next day eggs (text-fig. 8*a*) were found attached by adhesive threads to the filiform leaf segments and also to algal filaments. The eggs are laid singly as in the case of *A. panchax*.

The eggs are nearly spherical, being about 1·4 to 1·7 mm. in diameter, heavily laden with yolk and demersal, though containing several oil globules. They bear numerous filamentous threads all over the surface, some of which are longer, and these radiate from a tiny central spot as in the eggs of *A. panchax*. There are, however, a larger number of short filaments in the case of *A. lineatus*. The long adhesive threads form an anchoring tuft attaching the egg to the aquatic vegetation. In all essential details the egg resembles that of *A. panchax*.

Fry and Assumption of Adult Characters.—Innes (1939, p. 151) has already observed that the eggs of the species hatch in about a fortnight producing "fry of good size", which "grow fast if supplied with food" Therefore, as Moody (*loc. cit.*) has mentioned, even when no special diet is supplied, "youngsters of respectable size can be netted out at the end of three months" Unlike *Gambusia affinis* the proportion of males among the hatchlings of *A. lineatus* is much greater than that of females. The young of *A. lineatus* closely resembles that of *A. panchax*. An immature fish, about 14·7 mm. long (text-fig. 8*b*) has the adult specific structures more or less established. The body is considerably stout. The pigmentation is very similar to that of *A. panchax*, but the transverse bands are just beginning to be represented, especially in the posterior region. The occipital spot is not shown in the specimen, as during fixation the chromatophores usually "spread" and cover the spot rendering it dark. The fins have developed all the rays, and the protractile premaxillae are well marked as in *A. panchax*. For about three months the colouration of the young continues to be of the "*lineatus-type*" But when about four months old, by which time the fish attains maturity and begins to breed, the male develops his nuptial characters. The brilliant red spots appear on his body, the dark vertical bars grow faint, and the anal fin elongates to form the filose point. The female retains her original appearance except for a somewhat broader abdomen due to the number of eggs.

Aplocheilus blockii (Arnold).[1]

General.—The *dwarf panchax* or *pachai munda kanni* of Madras is the smallest known species of the genus, and attains a maximum size of only two inches. The average length, however, does not exceed one and a half inches. It is said to be a freshwater species inhabiting "stationary and sheltered waters of tanks and rivers overgrown with vegetation" The fish is recorded to be of limited distribution occurring in the East Coast of India near Madras, but Innes (1935, p. 265) lists Ceylon also as its home. It is freely reared in American aquaria as an ornamental fish. *A. blockii* closely resembles *A. panchax*, so much so that Stoye (*op. cit.*, p. 83) treats it as a "variety" of the latter, and refers to it as "*panchax* var *blockii*" It is, however, easily distinguished by its projecting lower jaw, characteristic colouration and the lesser number of scales both in the longitudinal and transverse series. Sundara Raj (*loc. cit.*) has given a good description of the species under the name *Panchax parvus*. The fish is a fairly good larvivore, and its

[1] *Panchax parvus* of Sundara Raj (1916, pp. 268, 269.)

insectivorous propensities have been discussed by writers such as Macdonald (1914), Sundara Raj (1916), Hora (1927) and Gravely (1937). It has, however, been found to be less hardy than *A. panchax* and *A. lineatus*. Further, it is smaller in size and more limited in distribution, and Russell and Jacob (1939, p. 275) who have conducted experiments on the species in the " sandy and scantily vegetated casuarina pits of the Ennore-Nellore coastal area " near Madras, found it to be not as efficient as *Gambusia affinis* for mosquito-control in those pits.

Breeding.—Sexual dimorphism is marked as in the other congeners. The large males looking like " living sparkling jewels ", are more brightly coloured than the females, and have the posterior rays of the dorsal and anal fins drawn out into filaments which reach the caudal fin, while in the female these fins are rounded and do not reach the caudal. The basic colour of the body in both sexes is dark olive with a metallic sheen above, passing to a translucent enamel blue on the ventral side. The fins are light orange. Alternating green and pink dots occur in longitudinal rows over the back and sides, and in the male they extend over the proximal portions of the median fins. A pearly white occipital spot occurs in both sexes. An ocellus at the anterior region of the base of the dorsal fin is less distinct in mature males. The female's colours are merely " a pale suggestion of those in the male " excepting the occipital spot and the ocellus on the dorsal fin.

The first reference to the spawning habit of the species is by Thomas (*op. cit.*, p. 112). He writes, " *H. panchax* (he means *A. blockii*) extrudes one egg at a time and that is disproportionately large, as big as its own eye. This keeps hanging to the vent as the fish is moving and feeding till it is cast and adheres ; and so single eggs are laid and distributed " About the breeding habits of the fish, Mellen and Lanier (*op. cit.*, p. 134) state that its spawning habits are similar to those of *A. panchax*, and Innes (1935, p. 265) notes that it is a good breeder, and puts down the temperature range as 70 to 84 degrees. Russell and Jacob (*loc. cit.*) observe that the fish " does not multiply effectively when confined in casuarina pits or nurseries " According to Sundara Raj (*loc. cit.*) the egg closely resembles that of *Oryzias melastigma* (=*H. melanostigma*), but is slightly larger, the chief difference being the absence of the tuft of long processes.[1]

Oryzias melastigma (McClelland).[2]

General.—This is a delicate little Killifish growing to but an inch and a half, and is widely distributed in the Indian Empire. Though usually regarded as an estuarine and brackish water form, *Oryzias melastigma* is found in large numbers in pieces of fresh water as well, and is common in ponds, lakes, rivers, canals and creeks in Lower Bengal, Orissa and Madras Presidency, and has been recorded from Wynaad, Burma, the Kiangsu Province of China ?, Formosa ?, Korea ?, and Japan ?. Sewell and Chaudhuri (*op. cit.*, p. 4), Fry (1912), Macdonald (1914), Sundara

[1] Mr. S. Jones has informed me that his description of the egg and development of *Panchax parvus* (Jones, 1937, pp. 284, 285) is really that of *Hemirhamphus* sp. and that this correction is in the course of publication.

[2] *Aplocheilus melastigma* of McClelland (1839) and other authors.

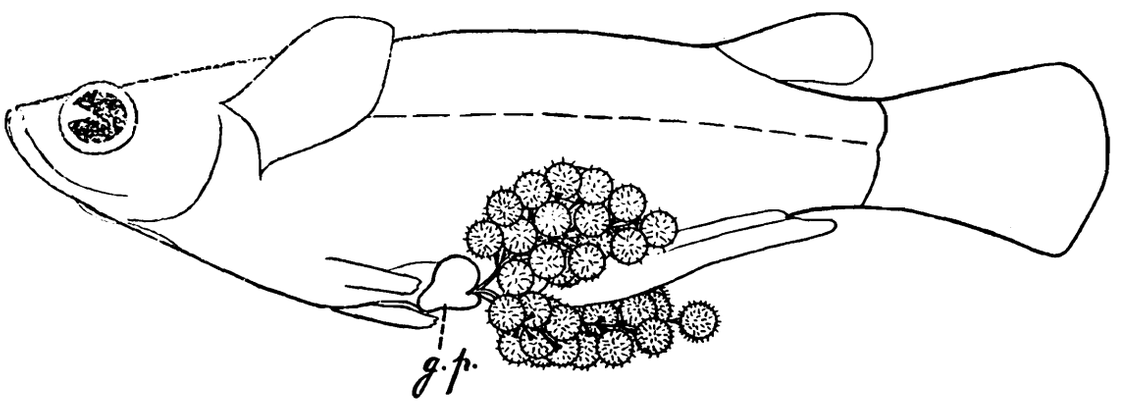
Raj (1916, p. 268) and Southwell (*op. cit.*, p. 182) have commented on the utility of this fish as a larvivore. It is, however, less hardy than the *Aplocheilus* species, and Russell and Jacob (*op. cit.*) found it inferior to *Gambusia affinis* in the matter of mosquito-control in their casuarina pit experiments. Allied in appearance and habits to the *Aplocheilus* species, the fish is easily distinguished by the strongly compressed diaphanous body, small fixed upper jaw, and a thin dark line along the side from the upper angle of the opercle to the middle of the base of the caudal fin, which is truncate or rounded, and the elongated anal fin.

Breeding and Parental Care.—The sexes in the species have the same colouration, the usual ground colour of the diaphanous body being a light olive grey above, passing on to sides of a lemon shade with silvery white abdomen below. The fins are usually faintly yellowish, and in a mature fish the median fins often develop a dash of brilliant yellow along the margin. Minute black spots are scattered over the body apart from the characteristic narrow dark line of the side. A shining silvery spot is usually present in the middle of the head; but it is less conspicuous than in species of *Aplocheilus*. The eyes are bright and greenish in both sexes. As noted by Sundara Raj, the colouration of the fish may vary according to surroundings, "those among green weeds" being "greenish" and "those in clear water translucent white", and in road-side puddles sometimes they show a dirty reddish hue. Mellen and Lanier (*op. cit.*, p. 136) refer to the "Golden Medaka" as an orange gold variation of *Oryzias latipes* (Temminck and Schlegel), a fish which is widely used in genetic studies and which Chaudhuri (1916, p. 455) considers as being synonymous with *O. melastigma*. Although the colours of the sexes are the same, the male can easily be distinguished by its larger and more pointed pectorals, a longer dorsal with the last ray rather separated from the others and a wider anal fin. The female has a deeper abdomen often distended with eggs in the breeding time.

The peculiar method of spawning and parental care exhibited by the fish has attracted the attention of workers such as Jenkins (1910, p. 137), Sundara Raj (1916, p. 267) and Jones (1937, p. 282). Sundara Raj's observations have been supplemented by a detailed description with more accurate figures by Jones of the egg and the development up to hatching.

O. melastigma has been observed to be a perennial breeder in the brackish water area at Adyar (Jones, *op. cit.*, 286). Russell and Jacob (*loc. cit.*) note that "in a special nursery it failed to show much increase in numbers" The writer has observed eggs and gravid females from the fresh waters in the suburbs of Calcutta in all the months from April to October, 1939, but found that intensified breeding occurs from June to August. Thus with the advent of the monsoon rains fry of the fish in crowded associations could be seen wandering over shallow collections of water and even in dirty puddles in the middle of roads and lanes into which drains overflow. Chatterjee (1934, p. 13) has observed that the fish breeds throughout the year in confined waters of tanks. *O. melastigma* may, therefore, be regarded as a perennial breeder both in fresh as well as brackish waters.

Mature males are seen to chase the females during breeding time. As observed by Jenkins, Sundara Raj and Jones, the eggs are carried about by the mother in conspicuous clusters. The eggs, as figured by Jones, are spherical and transparent, each about a millimeter in diameter and containing a single oil globule. The outer egg-membrane bears all over it short filaments, while from one region a cluster of long anchoring threads are given off. As the female extrudes the eggs they remain suspended usually in a pair of clusters ; for, the long anchoring threads of the eggs of each cluster are twisted together to form a common chord which protrudes from the genital opening of the female behind a notch



Text-fig. 9.—Semidiagrammatic ventro-lateral view of a female *Oryzias melastigma* (McClelland) bearing egg cluster : × *ca.* 3½.

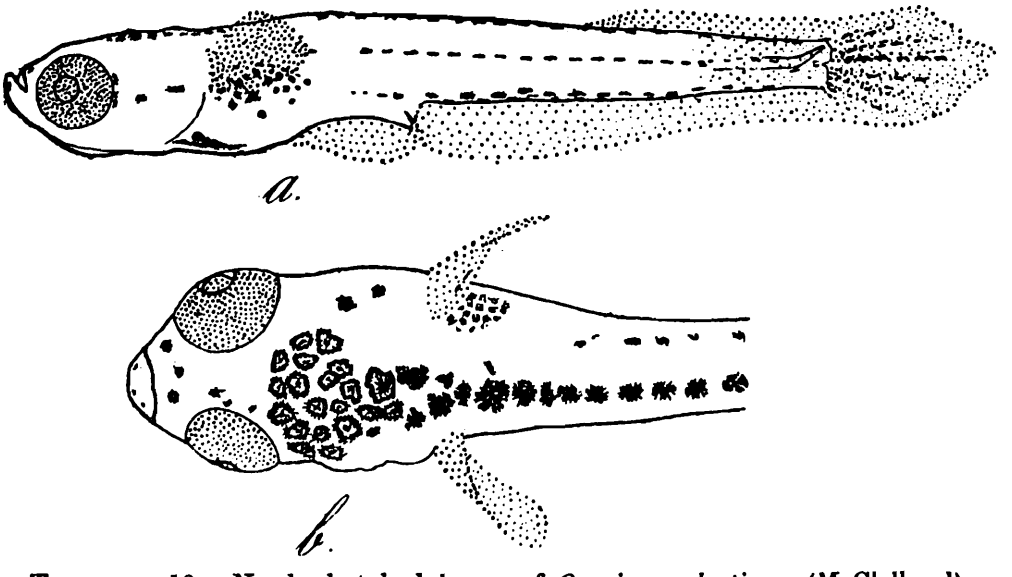
g.p. Genital pad.

in the middle of the flattened genital pad. The two clusters impinging on each other appear as a single bunch of transparent eggs (text-fig. 9). On an average twenty to forty eggs are usually carried by a female. Mellen and Lanier (*op. cit.*, p. 136) record 50 to 225 in the case of *O. latipes* found in Japan. In the position described above the eggs are fertilised by the male, and they may be thus carried by the mother until they are hatched, " a condition highly favourable for their protection and aeration " Often, however, the eggs get detached either singly or in groups and get re-attached to algal filaments or other suitable objects and finally hatch out. The female's " attempts to free herself of her burden of eggs " in the case of *O. latipes* is picturesquely described by Ulmer (1933, pp. 271, 272). According to him " she seeks out some convenient stem and swims slowly across it in such a way that the eggs will catch in a notch formed by the junction of leaf and stalk. Tugging and pulling against this anchor, she struggles valiantly for perhaps a minute or two at a time to free herself of the roe and often becomes quite entangled in the floating plant. If her first effort is unsuccessful, she will rest for a few minutes before returning to her task. Two or three trials are often necessary before she succeeds in attaching the eggs, singly or in clusters, to the plant ; but her persistence invariably arouses the beholder's admiration" In the case of *O. melastigma* observed here, although the parental care, as Jones opines, may not be intentional, the fish exhibits no anxious struggle to throw off the eggs, which on the other hand often continue to be carried about by the mother by means of specialised anchoring " ropes " until they are hatched. As, however, the attachment is not very firm, they easily get detached by accident one after another or in small numbers, and

being adhesive, stick to aquatic plants or other objects as the fish moves about amidst the vegetation. Amemiya and Saburo (1931, pp. 176-178) report rare instances of internal fertilisation and occurrence of developing embryos within the body of the female *O. latipes*. So far the writer has not observed similar instances in the case of *O. melastigma*.

Development.—As Jones has observed, the eggs take from eight days to a fortnight to hatch, though in exceptional cases the hatching period of some eggs may be unduly prolonged. At Calcutta they hatched out in eleven days in water at 88 degrees and in fourteen days at 85 degrees. A few eggs kept in a vessel without change of water hatched out in nineteen days. The average temperature was 85 degrees. Mellen and Lanier assign but ten days for *latipes* at 78 degrees and two weeks at 74 degrees. This shows that temperature is not the sole factor determining the period of incubation. The embryonic development of *O. melastigma*, which has been well described by Jones, is very similar to that of the other Killifishes treated above. Yolk pigmentation to some extent resembles that of the species of *Aplocheilus*.

NewlyHatched Larva.—The newly hatched larva is 4 to 4·5 mm. long according to the period of incubation. It can be easily distinguished from *Aplocheilus* larvae, which it somewhat resembles, by (1) a more slender build, (2) five clear well developed rows of chromatophores



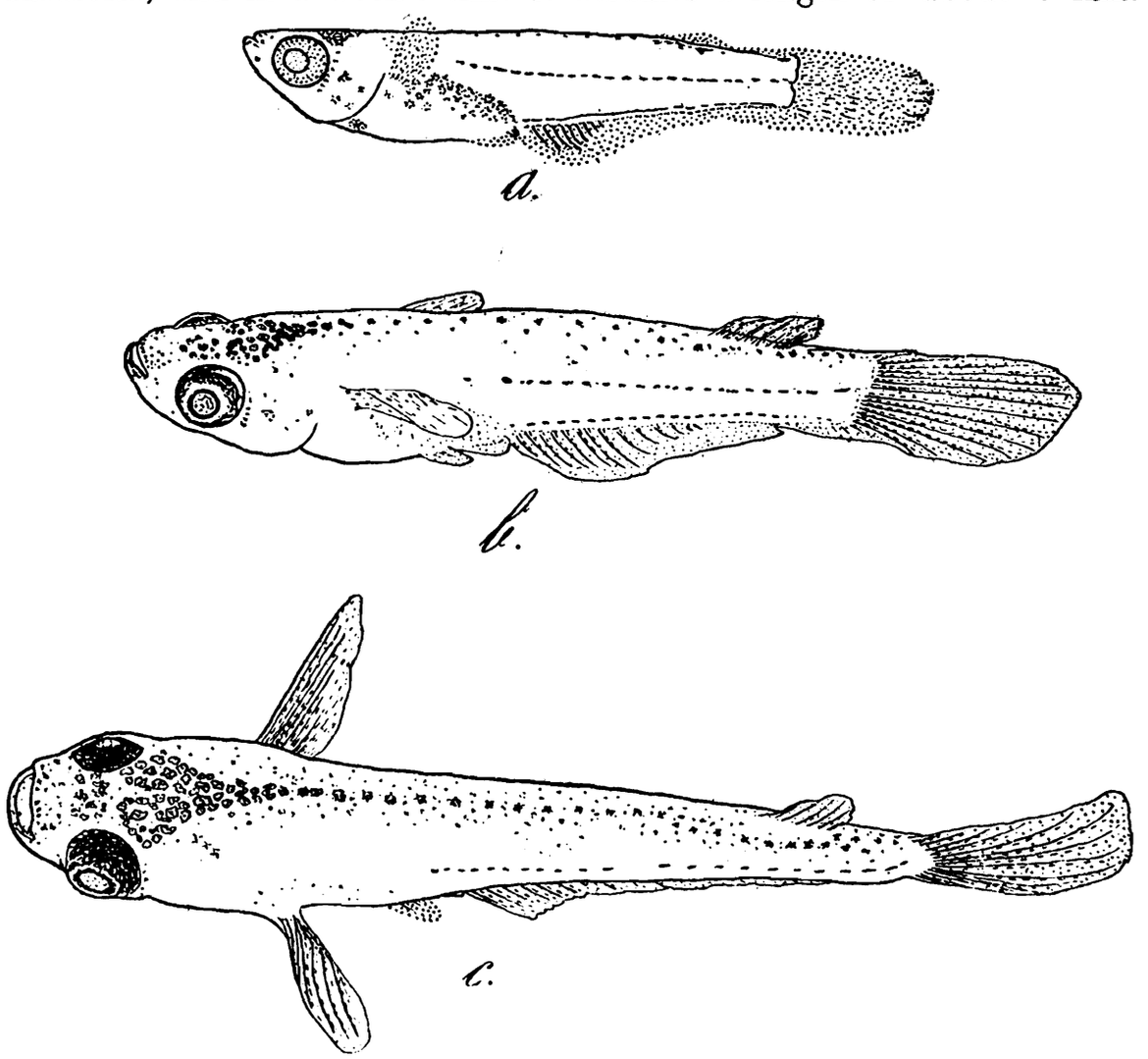
TEXT-FIG. 10.—Newly hatched larva of *Oryzias melastigma* (McClelland).

a. Side view: × *ca*. 24; *b*. Slightly latero-dorsal view of the anterior region: × *ca*. 36.

on the trunk, "one mid-dorsal, two lateral and two ventral, one on either side of the mid-ventral line of the body", (3) highly placed pectorals, (4) large ventral fin-fold with a small pre-anal lobe, the anus being fairly posterior in position and (5) upturned mouth. Sundara Raj has figured the newly hatched larva. Certain points of inaccuracy in the same may be observed. Hatchlings from both Madras as well as Calcutta present in the writer's collection show that the position of the mouth is really more superior than figured by Sundara Raj. Again the dorsal fin-fold is neither so wide nor so anterior as shown by him. It is actually "small and starts far behind" The pigmentation shown also is inaccurate. Actually there is a conspi-

cuous, more or less rounded patch of a variable, but limited number of large chromatophores behind the eyes tapering posteriorly to be continued backward as the mid-dorsal row. Text-figures 10*a* and 10*b* show camera lucida drawings of the side view and dorso-lateral view of the anterior region respectively of a newly hatched larva. " On the head and snout there are a few scattered spots ", and immediately behind the root of the pectoral fin dorsally there is a semicircular patch of close set small asters. On the ventral aspect of the abdomen, which is now practically devoid of yolk, there is an irregular patch of pigment often consisting of two or more anastomosing blotches. Only the heterocercal caudal fin has about seven rays. The other fins are still hyaline. The larva " moves about actively as soon as it comes out and constantly swims along the surface of the water ".

Metamorphosis.—The hatchling begins to grow slowly, feeding on Infusoria, and in a week's time it assumes a length of about 6 mm.



TEXT-FIG. 11.—Metamorphosis of *Oryzias melastigma* (McClelland).

a. Two weeks old larva, side view : × *ca.* 9 ; *b.* Three and a half weeks old larva, slightly latero-dorsal view : × *ca.* 9 ; *c.* One and a half months old " young adult ", latero-dorsal view : × *ca.* 9.

Now the dorsal fin rudiment begins to be separated off by a slight dipping in the dorsal aspect of the median fin fold. The fin-fold also widens slightly behind the anus on the ventral side. When examined under a binocular microscope the eye-balls can be seen to be tilted in opposite directions, *i.e.*, when the right one is turned upward the left one

is turned downward, and when the right one is tilted forward the left one is tilted backward and *vice versâ*. The pupil is black, but the cornea acquires an azure blue and golden green colour and shines with bright iridiscence.

Two weeks old larva (text-fig. 11*a*): Length, 7·5 mm. The snout is triangular, and the lower jaw is more prominent than the upper. The rudiment of the dorsal fin is constricted off from the caudal part of the fin-fold, and the anal becomes pronounced owing to the widening of its anterior region and the appearance of about eight rays in it. The pre-anal fold remains as a small adipose lobe. The pigmentation on the head takes the form of a paired patch united posteriorly. The pelvic buds begin to appear when the larva is about 9 mm. long.

Three and a half weeks old larva (text-fig. 11*b*): Length, 10 mm. The pelvics are clearly formed, and the commencement of two rays can be seen in each of them. About nine rays can be made out in the pectoral, about sixteen in the caudal, six in the dorsal and about fifteen in the anal. The snout is conical. The pigment patch on the head assumes the appearance of a pear-shaped patch distinctly bifid in front. Further, in the inter-orbital space a pair of oval patches appear as in the case of *Aplocheilus* larvae.

One and a half months old post larva (text-fig. 11*c*): Length, 12 mm. Most of the rays are developed in the median fins, and the fish approaches the adult in general appearance. The pectorals, however, has still about ten rays only, and the pelvics only three. The pigmentation of the head consists of a pair of rounded patches adjoining the two sides of a triangular patch behind, the base of the triangle tapering backwards to be continued as the dorsal row. The inter-orbital oval patches are more distinct. Besides, several other irregular rows of small pigment spots appear on either side of the mid-dorsal row, thus making the latter less distinct. The ventral rows also are less pronounced, while the lateral rows continue to persist. Pigment dashes have appeared on the fins also along the course of the rays. From now onward the post-larva is capable of taking in early instars of mosquito-larvae if supplied, besides other tiny insect larvae, Ostracods, etc. With further growth it gradually assumes the typical adult characters and begins to breed when about six months old.

GENERAL OBSERVATIONS.

The Killifishes treated above are widely distributed in the Indian waters, and are seen to be active, hardy and prolific. Most of them are perennial breeders both in fresh as well as brackish waters. The spawning habits of the various species of *Aplocheilus* are found to be very much alike, and even in the case of *Oryzias*, where a sort of parental care is apparent, the presence of the parent is not essential for incubation; for, the adhesive eggs usually get attached to external objects and hatch out in the same way as in *Aplocheilus*. The eggs of these Killifishes can withstand stagnation of water, poor aeration and temperature variation to a remarkable extent, and the hatching period may vary from one week to as much as three weeks or even more. The

yolk is nearly all absorbed by the time of hatching, and active feeding and growth commence very soon. The larvae of *Aplocheilus* species resemble one another very closely with but slight differences, while those of *Oryzias* are more slender and provided with characteristic pigmentation. Whereas *Aplocheilus* larvae are stouter to start with, and feed and grow fairly quickly, taking but three to four months for attaining maturity, the growth of *Oryzias*, which is more delicate to start with, is more slow, and it takes about six months to become mature. Not only in the larval condition, but also in the adult stage, *Oryzias* is found to be a more delicate fish than *Aplocheilus*. Its small size and diaphanous body, however, render it an agile inconspicuous fish which actively wanders about in shallow water. With the advent of the rains, not only tanks and pools, but all drains and shallows into which they overflow also teem with the fry of these surface-feeding Killifishes of Indian waters.

Summary.

The breeding and development under natural conditions of the indigenous Killifishes of the genera *Aplocheilus* and *Oryzias* have been investigated. As a type of the former genus the widely distributed *tri-choki*, *A. panchax* was taken up, and of the latter genus, *O. melastigma*, the only representative of the genus in Indian waters was studied.

The North Indian species, *A. panchax*, unlike what has been observed in aquaria, is, under natural conditions, a perennial breeder, with a spawning maxima in the monsoon months of June, July and August. The rounded, highly yolk-laden, demersal eggs contain several oil globules and bear numerous adhesive anchoring filaments. They are laid singly, and following the typical teleostean type of embryonic development, hatch out in about ten days. The period of embryonic development may, however, vary from nine to fourteen days according to the state of the egg, temperature, aeration and other physical and chemical conditions of the water. The yolk is usually absorbed inside the egg, and on hatching, the larva is fairly large, being about half a centimeter in length and capable of active swimming ; it is provided with efficient pectorals and a well developed caudal fin. At this stage the rest of the median fins are represented by a continuous membranous fold starting from the middle of the tail above and running up to the anteriorly placed vent below. Other characters of the larva are described. Metamorphosis takes about three weeks, in the course of which the median fins are differentiated and develop fin-rays, the " third eye " and the dorsal ocellus appear, the pre-maxillae become protractile, and from this stage the post-larva is capable of consuming larger prey including larvae of mosquitoes and other insects, Ostracods, etc.

The Peninsular species, *A. lineatus*, common in the West Coast districts, is similar to *A. panchax* in its habits. The eggs closely resemble those of *A. panchax*, but usually the transverse bands characteristic of the adult *A. lineatus* begin to appear when the fish is about half an inch long.

The breeding period of the Madras species, *A. blockii*, thriving in fresh water, is said to be January and February.

In the widely distributed diaphanous species, *Oryzias melastigma*, parental care practically ceases as the eggs get detached from the cluster borne by the mother and adhere to external objects where they develop. After an incubation period varying from one to two weeks or more, slim, delicate hatchlings come out. Of a size below half a centimeter, they are distinguished from the *Aplocheilus* larvae by their (1) slender build, (2) five regular longitudinal rows of pigment spots, (3) more highly placed pectorals, (4) a small pre-anal fold and (5) upturned mouth. Metamorphosis is more prolonged than in *Aplocheilus*, since the growth of the dainty hatchlings is more protracted.

The fry of *Aplocheilus* species are very similar to one another, being comparatively stout and marked by dark eyes and pigmented nape and belly. *Oryzias*, both fry and adult are more delicate. All these Killifishes, however, are hardy and enterprising, the adults and fry invading fresh "grounds" during the rains over submerged land.

REFERENCES TO LITERATURE.

Aitken, E. H., 1901.—Notes on Anopheles or the Malaria Mosquito. *Journ. Bombay Nat. Hist. Soc.* XIII, pp. 691-695.

Amemiya, I. and Saburo, M., 1933.—Some Remarks on the Existence of Developing Embryos in the Body of an Oviparous Cyprinodont, *Oryzias latipes* (Temminck et Schlegel). *Proc. Imp. Acad.* (Tokyo) VII, 4, pp. 176-178.

Annándale, N. and Hora, S. L., 1925.—The Fresh Water Fish from the Andaman Islands. *Rec. Ind. Mus.* XXVII, pp. 33-41.

Bannerman, W. B., 1910.—Note on Dr. Bentley's Paper "The Natural History of Malaria (*sic.*)" *Journ. Bombay Nat. Hist. Soc.* XX, pp. 525, 526.

Bentley, C. A., 1910.—The Natural History of Bombay Malaria. *Ibid.*, pp. 392-422.

Brahmachari, B. B., 1909.—Campaign Against Malarial Fevers at Cossipur-Chitpur Municipality. *Calcutta Med. Journ.* III, pp. 312-322.

Cantor, T., 1849.—Catalogue of Malayan Fishes. *Journ. Asiatic Soc. Bengal*, XVIII, pp. 983-1426.

Chatterjee, G. C., 1934.—*Biological Control of Malaria in Rural Areas of Lower Bengal*, pp. 1-28.

Chaudhuri, B. L., 1909.—Mosquito-larvae-eating Propensity of the genus *Haplochilus*. *Journ. and Proc. Asiatic Soc. Bengal*, V, pp. 36-37.

Chaudhuri, B. L., 1916.—Fauna of the Chilka Lake. Fish, Part 2. *Mem. Ind. Mus.* V, pp. 440-458.

Covell, G., 1927.—Anti-Mosquito Measures. *Malaria Bureau*, No. 3, *Health Bulletin*, No. 11, Calcutta, pp. 1-62.

Day, F., 1876-1878.—*The Fishes of India*, London.

Fraser, A. G. L., 1938.—Observations on the Bionomics of *Panchax lineatus* Cuvier and Valenciennes, With Special Reference to its Larvicidal Propensities. *Journ. Bombay Nat. Hist. Soc.* LX, No. 1, pp. 96-99.

Fry, A. B., 1912.—Indigenous Fish and Mosquito Larvae. *Paludism*, No. 5, pp. 71-74. Govt. Central Branch Press, Simla, India.

Gravely, F. H., 1937.—Mosquito-Destroying Fishes. *Curr. Sci.* V, No. 9, pp. 482, 483.

Günther, A., 1866.—*Cat. Fish. Brit. Mus.* VI.

Herre, Albert, W C. T., 1939.—On a Collection of Littoral and Freshwater Fishes from the Andaman Islands. *Rec. Ind. Mus.* XLI, pp. 327-372.

Hora, S. L., 1927.—The Use of Fishes for the Control of Mosquitoes. *Ind. Med. Gaz.* LXII, pp. 187, 188.

Hora, S. L. and Nair, K. K., 1938.—Observations on the Nutrition of *Panchax panchax* (Hamilton). *Proc. Nat. Inst. Sci. Ind.* IV, No. 2, pp. 245-251.

Innes, W T., 1935.—*Exotic Aquarium Fishes*, Philadelphia.

Innes, W T., 1939.—*Panchax lineatus* C. & V *The Aquarium* VII, No. 9, pp. 150, 151.

Jenkins, J. T., 1910.—Notes on Fish from India and Persia with Descriptions of New Species. *Rec. Ind. Mus.* V, pp. 123-140.

Job, T. J.,—Practical Utility of Mosquitocidal Fishes in the Biological Control of Malaria, Yellow Fever, Dengue, Filariasis and Encephalites : *Aplocheilus panchax.* (To be shortly published).

Job, T. J. and Jones, S., 1938.—Studies on the Development of the Indian Garfish, *Tylosurus strongylurus* (Van Hass.) with Notes on the Young Stages of *Hemirhamphus gaimardi* Cuv. & Val. *Rec. Ind. Mus.* XL, pp. 245-253.

Jones, S., 1937.—Observations on the Breeding Habits and Development of Certain Brackish Water Fishes of Adyar, Madras. *Proc. Ind. Acad. Sci.* V, No. 6, Sec. B, pp. 261-289.

Jordan, D. S., 1923.—A Classification of Fishes. *Stanford University Publications, Biological Sciences.* III, No. 2.

Macdonald, W R., 1914.—Note on the Use of Larvicidal Fish in Combating Malaria Fever. *Proc. Third All Ind. Sanitary Conf.* 4, pp. 75-77 (Suppl. *Ind. Journ. Med. Res.*).

McClelland, J., 1839.—Indian Cyprinodontidae. *Asiat. Research.* XIX, Part 2, pp. 301-427

Mellen, I. M. and Lanier, R. J., 1935.—*1001 Questions Answered about Your Aquarium*, New York.

Moody, A. S., 1933.—*Panchax lineatus. The Aquarium.* II, No. 8, pp. 185-187

Mukerji, D. D., 1935.—Notes on some Rare and Interesting Fishes from the Andaman Islands, with Descriptions of Two New Fresh Water Gobies. *Rec. Ind. Mus.* XXXVII, pp. 259-277

Norlund, M. C., 1936.—*Panchax panchax. The Aquarium* V, No. 3, p. 60.

Portman, M., 1888.—The Exploration and Survey of the Little Andamans. *Proc. Royal Geogr. Soc. and Month. Rec. Geogr.* September Number, pp. 1-10.

Prashad, B. and Hora, S. L., 1936.—A General Review of the Probable Larvivorous Fishes of India. *Rec. Mal. Surv. Ind.* VI, (4), pp. 631-648.

Roy, D. N., 1938.—On the Control of Malaria-Mosquitoes in Bengal by the Use of Predaceous Fish and on the Habits of Two of Them. *Journ. Mal. Inst. Ind.* I, No. 4, pp. 405-416.

Russell, P. F. and Jacob, V P., 1939.—Some Experiments in the Use of Fish to Control *Anopheles* Breeding in Casuarina Pits. *Journ. Mal. Inst. Ind.* II, No. 3, pp. 273-291.

Sen, P., 1937.—On the Food Factors of the So-called Mosquito-Destroying Fishes of Bengal.—*Panchax panchax, Barbus stigma, Esomus danricus* and *Trichogaster fasciatus. Curr. Sci.* V, No. 7, pp. 357-361.

Sewell, R. B. and Chaudhuri, B. L., 1912.—*Indian Fish of Proved Utility as Mosquito-Destroyers.* Calcutta, pp. 1-24.

Smith, H. M., 1938.—Status of the Oriental Fish Genera *Aplocheilus* and *Panchax. Proc. Biol. Soc. Washington* LI, pp. 165, 166.

Southwell, T., 1920.—Fish and Mosquito Larvae in Bengal, Bihar and Orissa, India. *Ann. Trop. Med. and Parasit.*, Liverpool. XIV, pp. 181-186.

Stoye, F. H., 1935.—*Tropical Fishes for the Home, Their Care and Propagation*, New York.

Sundara Raj, B., 1916.—Notes on the Freshwater Fish of Madras. *Rec. Ind. Mus.* XII, pp. 249-294.

Thomas, H. S., 1887.—*Tank Angling in India*, Madras.

Ulmer, R. E., 1933.—Breeding the Medaka (*Aplocheilus latipes*). *The Aquarium* I, No. 10, pp. 271-273.

Willey, A., 1910.—Observations on the Nest, Eggs and Larvae of *Ophiocephalus striatus. Spolia Zeylanica* VI, pp. 108-122.