STUDIES ON FISH EGGS AND LARVAE FROM INDIAN WATERS
2. DEVELOPMENT OF EGG AND LARVAE OF *ACENTROGOBIUS ORNATUS* (RÜPPELL)

P. VIJAYARAGHAVAN
Central Marine Fisheries Research Institute, Cochin-18

ABSTRACT

Eggs of *Acentrogobius ornatus* (Rüpp.), attached to *Placenta* shells were collected along with the parent (male) fish guarding it in knee deep water from the Vellar estuary at Portonovo. The embryonic development from the blastoderm stage up to hatching and the larvae up to the seventh day are described.

Besides the present account on *A. ornatus*, the eggs and early development of its congeners, *A. neilli* and *A. viridipunctatus* have been known. The eggs of *A. neilli* is club shaped and smaller than that of *A. ornatus* which is stumpy, broader at the base and is the smallest of all. In *A. ornatus* as in *A. viridipunctatus* the oil globules do not coalesce to form a single one as met with in *A. neilli*. The egg of *A. ornatus* which is the smallest of the three species, has the longest period of incubation. The important features in the embryonic and larval development of the three species are tabulated for comparison.

INTRODUCTION

Typical of most estuaries, the Vellar estuary in Portonovo abounds in gobioid fishes. In all, twenty species representing three in Eleotridae, nineteen in Gobiidae and one in Periophthalmidae have been recorded from this area (Jacob, 1961). The author, during his trips along the banks of this estuary to the fish landing places, had opportunity to collect several gobioid eggs from and near the river mouth and to study their development and larvae, which will be described in this and a few future publications. Some of the material were collected by the combined efforts of the author and the staff of the Marine Biological Station of the Annamalai University at Portonovo. In such instances the parent fish collected were given over to the museum of the latter, after identification, and the eggs retained by the author for observations. All these eggs were found during the years 1958 and 59 in the summer months between March and July before the onset of the south west monsoon which sets the Vellar river in floods. The eggs and larvae were kept in glass troughs containing fresh filtered river water. Descriptions and sketches were made mostly from live material and occasionally from freshly fixed ones. Identification of the eggs was easy when they were collected along with the parent or parents guarding them, though however, the latter also shall be described. The specimens are in the reference collection museum of the CMFRI.

The author is grateful to Professor R.V. Seshaiya, former Director of the Marine Biological Station, Portonovo, and his staff for the various facilities and co-operation.

Earlier observations on the development of eggs and larvae of Indian gobies pertain to *Acentrogobius neilli* (Day) by Raj (1916) and Aiyar (1935), *A. viridipunctatus* (Day) by Jones (1937, 39 and 46), *Glossogobius giuris* (Hamil.) by Alikunhi et al (1947), Chacko (1951) and Bowmick (1966) *Gobiopterus chuno* (Hamil.) by Pillai and Sarojini (1950) and Chaudhuri (1951), *Oligolepis acutipinis* (Val.) by Jones (1946), *Paragobiopsis ostericola* (Chaudhuri) by Bhattacharya (1916) and Jones and Menon (1953), *Stigmatogobius javanicus* (Bikr.) by Nair (1961), and descriptions or records of postlarvae of *Gobius* spp. by Whitehouse (1923), Gopinath (1946), Bal and Pradhan (1945, 46, 47 and 51), Bapat and Bal (1950) and Kuthalingam (1958).

Acentrogobius ornatus (Rüpp.)

Eggs of this species were encountered on two occasions, 15th of March and 13th of May in 1959. On the first instance they were found attached to the inner surface of a dead *Placenta* shell which lay with its inner side facing up in knee deep of fairly clear water about ten feet from the water's edge. The tide was already rising, at 4 pm when the shell was taken. The parent fish, the male, which was remaining almost motionless over the brood was captured with a small hand net. The site of collection was about two furlongs up the river mouth, on its northern bank where the substratum was sandy. The temperature and salinity of water at the time of collecting the specimens were 29°C and ca 30‰ respectively. On the second occasion also the eggs were found on the inner surface of a *Placenta* shell and in the same locality. But this time, the shell was lying exposed on the river bank about five feet away from the water which was receding in the neap tide. Nevertheless, the eggs remained immersed in the small amount of water that was trapped in the shallow saucer like shell. The salinity of water in the river at the time in that place was 1.8‰ and temperature 27°C. To see whether these eggs would have survived till the return of the tidal water the shell with the eggs and the water contained in it was carefully transferred to the laboratory and kept in the same condition. Reexamining them later, at the time when the tide began to rise in the river and the place from where the eggs were collected became submerged, more than hundred and fifty eggs were found to be alive out of over four hundred eggs which were living initially, though the meagre quantity of water that was in the shell had almost completly evaporated and the cluster of eggs remained barely moist by the thin film of water that was present in the interspaces between the eggs.

The hatchlings lived up to seven days in the laboratory.

Description of the egg

The eggs are found arranged close to each other in a single patch of irregular shape, roughly 3 to 4 cm square in area. To the naked eyes they appear like sand

coloured encrustations on the pearly inner surface of the shell. Inclusive of the dead ones there were nearly six hundred in the first collection and about four hundred on the second occasion.

The egg is elongate and stumpy, broader nearer the base. They measure 0.78 to 0.93 mm in length and 0.37 to 0.43 mm in width at the broadest portion. They are attached to the substratum by narrow short hyaline fibers that are fused together forming a sucker shaped ring-like foot. These structures are most often fused with similar ones of adjacent eggs, thus forming a common adhessive base to the entire cluster of eggs. Hence, attempts to detach an egg will usually result in pealing off a number of eggs along with it.

EMBRYONIC DEVELOPMENT

In the earliest stage of development that was met with (Fig. 1) the blastoderm has been formed which covers nearly three-fourths of the yolk mass and the embryonic ridge is clearly discernible. Judging from the growth attained at this stage and the subsequent rate of its progress, fertilization would seem to have occurred about ten to twelve hours earlier. On this assumption the stage represented in figure 1 is taken as twelve hours old and the age of the succeeding stages described below is attributed accordingly. The yolk is filled by a large number of orange coloured oil globules of varying sizes. The egg occupies the distal portion of the egg capsule. Three hours later, that is, in the fifteen hour old embryo (Fig. 2) the blastoderm envelops the yolk mass completly. The head portion of the embryonic axis is conspicuously bulging and has faint indications of developing eyes. Two or three myotomes whose margins are indistinct are present. At this stage the head points towards the base of the egg capsule. The egg remains at the distal end of the latter as in the previous stage.

Embryo: One day old - (Fig. 3). The embryo is semicircular occupying half the circumference and is lifted clearly above the yolk and has eight to nine myotomes. The neural grove is well formed. Eyes are very prominent with the lens in the process of development. Rudiments of the auditory vesicles are visible. The head and tail are adherent to the yolk mass. There is no movement in the embryo. The egg, which had been hitherto orientated with the head facing the base of the capsule now rests with the head pointing towards the distal end of the capsule. This position is maintained all through the rest of the development till hatching.

Embryo: Two days old (Fig. 5). When the embryo is about thirtyfour hours old (Fig. 4) the head which has become unusually large and the tail are detached from the yolk mass. When two day old, the length of the embryo is slightly less than the length of the egg capsule and has fourteen to sixteen myotomes. It may be noted that in this and subsequent stages the myotome counts are given inclusive of urostyle. The most important features of the embryo at this stage are the formation of the heart which is not functional yet, the incipient median fin folds, the advent of two round concretions in the auditory vesicles and the appearence of a few grey stellate chromatophores at the base of the eye balls and on the posterior region of the yolk

sac and an irregular row of black pigment spots along the ventral aspect of the myotomes extending from behind the yolk mass to the beginning of the urostyle.

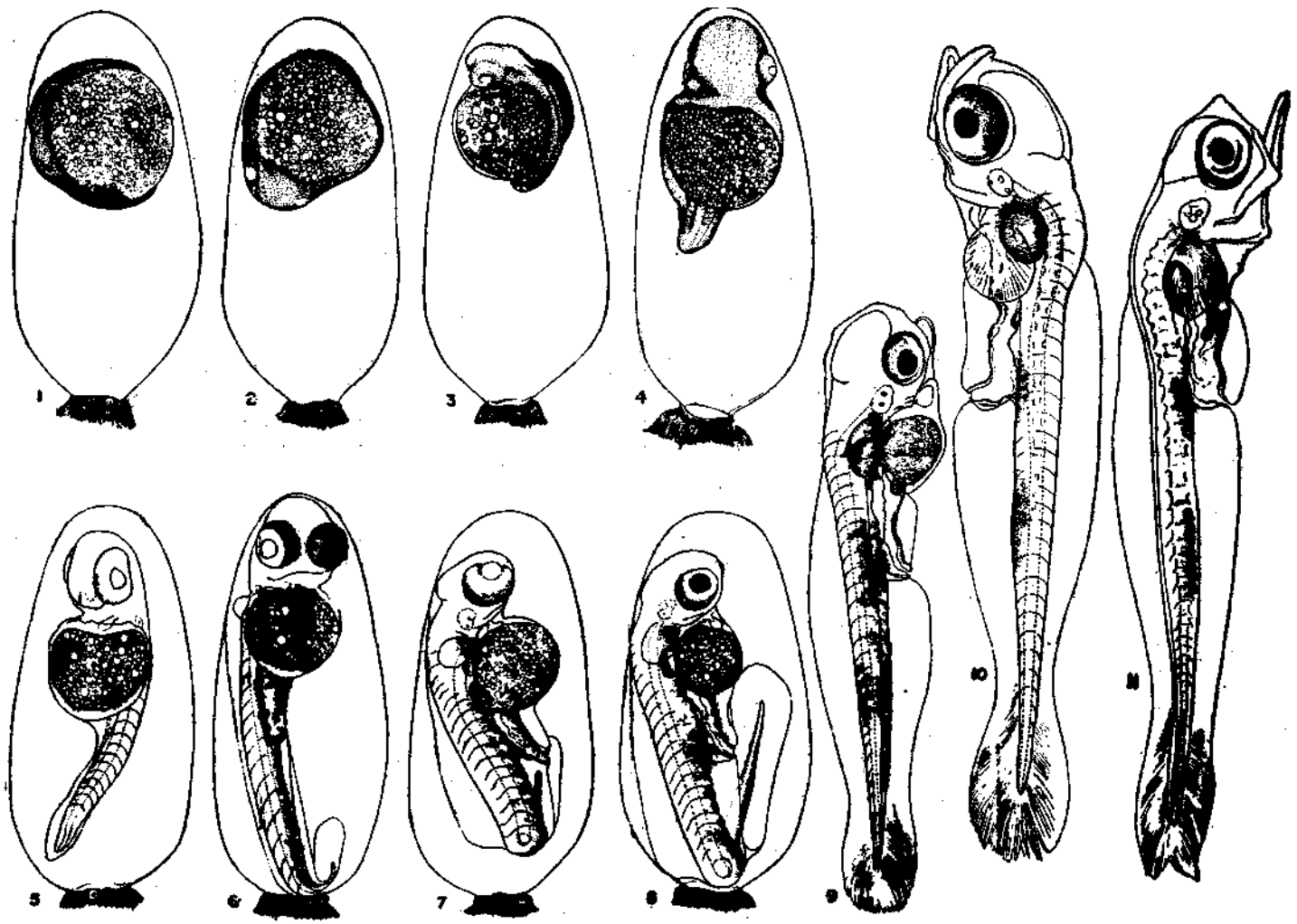
Embryo: Three days old - (Fig. 6). The heart begins pulsating at irregular intervals at first which becomes regular and rhythmic within an hour or two. There is a well defined circulation of blood whose corpuscles are not pigmented. The tip of the tail which is folded upwards for want of space within the egg capsule due to the increase in size of the embryo, changes position occasionally. The rudiments of pectoral fins have developed and the embryo has eighteen myotomes. The black pigment spots along the ventral margin of the body have become stellate. A large branching black chromatophore has appeared laterally just below the pectoral bud. Brownish yellow ramifying pigment cells are scattered on the ventral margin of the body and on the newly developed hind gut. A thin streak is present in the place of the mouth.

Embryo: Six days old - (Fig. 7). But for the increase in size, there has not been any noteworthy change in the embryo during the past two days. By the sixth day, the jaws develop and two gill arches are visible. There are twenty myotomes. The anal pore is clearly visible. The intestine, whose posterior extremity is situated below the eleventh myotome, is not in communication with the mouth and the gall bladder which is pale yellow in colour is visible dorsal to the yolk sac. An air bladder has developed between the body and the yolk sac behind the pectoral origin. The pectoral and the median fin folds have become broader and the continuity of the latter is broken by the hind gut which extends up to the margin of the ventral fin fold, resulting in a small preanal fin fold that was originally part of the ventral fin fold. Some of the black chromatophores have spread over the surface of the yolk sac and are present along with orange coloured pigments beneath the pectoral fin. There are black chromatophores on the inner upper surface of the air bladder. The brownish pigmentation of the earlier stages has become orange in colour.

Embryo: Eight days old - (Fig. 8). There are twelve preanal and fifteen postanal myotomes. The mouth is conspicuous and is connected with the intestine. The volume of the yolk mass is half its original size. Blood corpuscles are pale reddish. Eyes are irredescent, the pupil black and the orange pigmentation seen in the earlier stage is particularly dense on the trunk behind the origin of the pectoral fin, above the anus and midway between the caudal tip and the anus. The black pigments occur all along the ventral margin of the body, along the intestine, beneath the pectoral fin, the yolk sac and in the air bladder. The caudal fin fold has broadened. The pectorals flap frequently and the embryo wriggles quite often.

HATCHING

Hatching of the eggs takes place between the eighth and the tenth days, but in the majority of cases on the ninth day. The head of larva emerges from the egg case through the rupture caused at the distal end of the egg case.



FIGS. 1-11. Developmental stages of *Acentrogobius ornatus* (Rüppell)
1. Twelve hours old blastoderm, 2. Fifteen hours old embryo, 3. One day old embryo, 4. Thirty four hours old embryo, 5. Two days old embryo, 6. Three days old embryo, 7. Six days old embryo, 8. Eight days old embryo, 9. Larva first day, 10. Larva fourth day, Larva seventh day.

TABLE 1. *Main features of development of the embryo and*

A. neilli		*A. viridipunctatus*
Embryonic features	Time after fertilization (hrs./min.)	Embryonic features
2 blastomeres	35 min	
4 blastomeres	1 hr. 5 min	4 blastomeres . . . .
8 blastomeres	1 hr. 30 min	8 blastomeres . . . .
		16 blastomeres . . . .
32-64 blastomeres	2 hr.	32 blastomeres . . . .
Blastoderm with median thickening, head end thickened	8 hr.	Blastoderm with embryonic ridge, head end enlarged. . . . .
Embryo more than halfway round yolk sphere	10 hr.	Embryo more than half way round yolk sphere (from sketch) neuralgrove clear, rudiments of eye (no lens), 4 somites . . .
Heart developing, auditory vesicles with indications of otolith, eyes coloured and has lens, alimantery canal fairly visible, caudal end free from yolk mass (from sketch), 24 somites	18 hr.	Optic and auditory vesicles formed, caudal region free from yolk mass . . . .
		Rudimentary heart, round concretions in auditory vesicles, 22 somites. . . . .
Circulatory system established	22-30 hr.	
		Circulation starts, gentle movements of tail, caudal flexed up (from sketch). . . . .
Pectoral fins	42 hr.	
Caudal flexed up, pigmentation on embryo .	30-48 hr.	
		Circulatory system established, dorsal and anal fin folds, pectorals bud like.
Anal opening distinct, air bladder, caudal fin rays distinct	3 days	Eyes dark, pectorals enlarged, air bladder.
		Eyes shining, gall bladder. . . . .
Hatching on the 3rd day		Gill slits . . . .
		Hatching in 5 to 7 days.

Larval features	Days after hatching	Larval features
Front part of primary dorsal fin present, ventral still as fin fold, caudal end of notochord not upturned, eyes closer in dorsal view, two adhesive structures on ventral surface of lower jaw, lower jaw slightly in advance of upper jaw.	2nd day	Caudal end of notochord turned up . . . .
		Yolk fully consumed, liver developed . . . .
		Anterior end of notochord bent. . . . .

larvae of A. neilli, A. viridipunctatus *and* A. ornatus.

		A. ornatus	
Time after fertilization (hrs/min)	Embryonic features		Time after fetilization (hrs/min)
1 hr. 30 min			
1 hr. 50 min			
2 hr.			
2 hr. 15 min			
10 hr.	Blastoderm with embryonic ridge	. . . .	10-12 hr.
12 hr.	Blastoderm envelops yolk mass completely, head bulging, eyes developing	. . . .	15 hr.
18 hr.	Embryo occupying half the circumference of yolk mass,		
24 hrs.	caudal adherent to yolk mass, eyes prominent and has lens, rudiments of auditory vesicles, 14-16 myotomes.	. . . .	24 hr.
	Caudal region free.	. . . .	34 hr.
36 hr.	Heart formed but not functioning, incipient fin folds, round concretions in auditory vesicles, pigmentation on embryo, eyes with a few black chromatophores.	. . . .	48 hr.
48 hr.	Well defined circulation, caudal region flexed up and often changing position, rudiments of pectorals, mouth as narrow streak, hind gut.	. . . .	3 days
3 days			
4 days	Jaws developing, 2 gill arches, anal pore visible, hind gut extend to margin of ventral finfold, gall bladder, air bladder.	. . . .	6 days
5 days			
6 days	Well formed mouth connected with intestine, eyes irredescent, pupil black	. . . .	8 days
	Hatching in 8 to 10 days		
Days after hatching	Larval features		Days after hatching
1st day	Urostyle straight, anterior end of notochord bent downward, median fin folds continuous up to anus		
2nd day	lepidotrichia seen in caudal lobe.	. . . .	1st day
3rd day	Yolk fully consumed, signs of ossification of notochord.	. . . .	4th day
	12 preanal and 23 postanal vertebrae, rudiments of hypurals, ossification continues, no rays in anal fin, liver developed	. . . .	7th day

LARVAL DEVELOPMENT

Larva: First day - (Fig. 9). The hatchlings measuring 1.1 to 1.3 mm in length come out with small yolk sac and move about near the bottom of the aquaria. The mouth and anus are open, but no feeding has been observed though they were provided with small quantities of river plankton and powdered shrimp. The larva has twelve preanal and sixteen postanal myotomes. The anterior end of the notochord is bent downward. The urostyle is straight. The median fin folds are continuous up to the anus and lepidotrichia are seen as fine striae on the caudal lobe. The margin of the fin fold in this region has frayed appearance. In addition to the pigmentation seen in the eight day old embryo, orange coloured branching pigment cells are present on the dorsal part of the irredescent eye ball. The gall bladder is now pale green in colour.

Larva: Fourth day - (Fig. 10). There is not much change in the larva during the second and the third days except for the reduction in the yolk mass and slight increase in size. By the fourth day, there is no trace of the yolk and the postlarva is 1.8 to 2.5 mm long and has twelve preanal and twenty postanal myotomes. There are signs of ossification of the notochord. Besides the caudal, the pectorals also have distinct striations. The pigmentation is more segregated compared to the earlier stages and the orange and black pigmentations are localised on the ventrolateral aspects of the body immediately behind the head, above the anus and midway between the anus and caudal tip. They are present also on the air bladder. A few black chromatophores are found along the ventral side of the alimentary canal and on the caudal fin where they appear as narrow discontinuous streaks radiating from the base of the fin. The orange coloured pigmentation on the eye ball has become more dense.

Larva: Seventh day - (Fig. 11). There is slight reduction in size of the postlarvae which are now 1.8 to 2.2 mm in length. Twelve preanal and twentythree postanal vertebrae, excluding urostyle, could be counted and the ossification is still continuing. Rudiments of the hypurals are visible. Rays have not yet developed in any of the fins. The liver is visible as a pinkish opaque mass with the pale green gall bladder attached to it. In addition to the pigmentation found in the earlier stage the ventral margin of the body is light greenish from behind the viscera up to three-fourths the distance to the caudal end. Attempts to feed the postlarvae were unsuccessful and it was not possible to keep them alive beyond seven days.

Prior to the present account on *A. ornatus*, the eggs and early development of its congeners, *A. neilli* and *A. viridipunctatus* have been studied by Aiyar (1935) and Jones (1937) respectively. The fertilized eggs of all the three species are distinguishable from each other by their size and shape. The egg of *A. neilli* is club shaped and smaller than that of *A. viridipunctatus* wich is fusiform in shape and that of *A. ornatus* is stumpy, broader at the base and is much smaller than the others. In *A. ornatus* as in the case of *A. viridipunctatus* the oil globules do not coalesce to form a single one as met with in *A. neilli*. The important land marks in the embryo and larval development of the three species are tabulated (Table. 1) for comparison.

In preparing this table an effort that has been made to present the corresponding developmental characters opposite each other in the three species, could be only partially successful due to the difference in the sequence of thier occurrence and the omission or inclusion of certain characters by the concerned authors.

It is interesting to note that while the egg of *A. ornatus* is the smallest of the three species, it has the longest period of incubation, while as a general rule the larger eggs take more time to hatch. It is doubtful whether the slight difference in temperature of the water in which they were developed could be attributed to the observed differences in the rate of development in these species. *A. neilli* was reared in 24 to 26° C, *A. ornatus* in 27 to 29°C and *A. viridipunctatus* presumably in 25 to 33° C. The author has not indicated the temperature in which the eggs of the latter species were reared but has given the temperature of estuarine region during the season from where the eggs were collected as ranging from 25 to 33°C.

REFERENCES

AIYAR, R.G. 1935. Observations on the Development of *Acentrogobius Neilli* (*Gobius Neilli*. Day). *Zool. Anz., III* (3/4): 83-92.

ALIKUNHI, K.H., G. L. RAO AND P.K. JACOB. 1947. Bionomics and development of *Glossogobius giuris* (Hamilton). *Proc. 34th Indian Sci. Congr.* (Delhi), Pt. 3 *Abstracts:* 179 (Also *J. Madras Univ.*, 1951, 21*B*; 238-248).

BAL, D.V. AND L. B. PRADHAN. 1945. *First progress report on "Investigation of fish eggs and fish larvae from Bombay waters" 1944-45.* Govt. Central Press, Bombay.

BAL, D.V. AND L. B. PRADHAN. 1946. *Second progress report on "Investigation of fish eggs and fish larvae from Bombay waters", 1945-46.* Govt. Central press, Bombay.

BAL, D.V. AND L.B. PRADHAN. 1947. *Third progress report on "Investigation of fish eggs and fish larvae from Bombay waters" 1946-47.* Govt. Central press, Bombay.

BAL, D.V. AND L.B. PRADHAN. 1951. Occurrence of fish larvae and postlarvae in Bombay waters during 1944-47. *J. Univ. Bombay, new ser.*, **20** B: 15 p.

BAPAT, S.V. AND D.V. BAL. 1950. Occurrence of fish larvae and postlarvae in Bombay waters during 1948-49. *Proc. 37th Indian Sci. Congr.* (Poona), Pt. 3 *Abstracts:* 255.

BHATTACHARYA, D.H. 1916. Stages in the life-hisotry of *Gobius, Petroscirtes* and *Hemirhamphus. Mem. Indian Mus.*, **5** (4): 383-392.

BOWMIC, R.M. 1966. Studies on some aspects of the biology of *Glossogobius giuris* (Hamilton) with notes on its fishery in the Hoogly estuary. *Proc. Indo-Pac. Fish. Counc. 11th sess.* **1964**. *Sec. II:* 99-115.

CHACKO, P.I. 1951. *Madras rural piscicultural scyeme, Prog. Rept.* (*Period 1st April* 1950 *to 31st March* 1951), Indian Council of Agricultural Research, New Delhi.

CHAUDHURI, H. 1951. Notes on the embryonic development of the 'transparent goby, *Gobiopterus chuno* (Hamilton). *Proc. nat. Inst. Sci. India*, **17** (4): 247-252.

GOPINATH, K. 1946. Notes on the larval and postlarval stages of fishes found along the Trivandrum coast, *Proc. nat. Inst. Sci. India*, **12** (1): 7-21.

JACOB, J. 1961. Scientific and Tamil Names of the Estuarine and Marine Fishes of Porto Novo. *J. Annamalai Univ.* **22** : 1-32.

JONES, S. 1937. Observations on the breeding habits and development of certain brackish water fishes of Adyar, Madras, *Proc. Indian Acad. Sci.*, **5** (6): 261-289.

JONES, S. 1939. On the external gills of *Acentrogobius viridipunctatus* (Day). *Proc. Indian Acad. Sci.*, **9** (5): 256-266.

JONES, S. 1946a Breeding and development of Indian fresh water and brackish water fishes. Part I. *J. Bombay nat. Hist. Soc.*, **40** (2): 317-336.

JONES, S. 1946b. Breeding and development of Indian fresh water and brackish water fishes. Part II. *J. Bombat nat. Hist. Soc.*, **46** (3): 453-471.

JONES, S. AND P.M.G. MENON. 1953. Notes on the breeding habits and developmental stage of some estuarine fishes. *J. Zool. Soc. India*, **5** (2): 255-276.

KUTHALINGAM, M.D.K. 1958. Studies on postlarvae and feeding habits of some fishes found in the Madras plankton. *J. Madras Univ.*, **28** B: 1-11.

NAIR G.S. 1961. On the development and early embryo of *Stigmatogobius javanicus* (Blkr). *Bull. Cent. Res. Inst. Univ, Travancore, Ser. C.*, **8** : 37-54.

PILLAI, T.V.R. AND K.K. SAROJINI. 1950. On the larval development of the Indian transparent goby, *Gobiopterus chuno* (Hamilton) with observations on its bionomics. *Proc. nat. Inst. Sci.*, India, **16** (3): 181-187.

RAJ, B.S. 1916. Notes on the fresh water fish of Madras. *Rec. Indian Mus.*, **12** (6): 249-294.

WHITEHOUSE, R.H. 1923. A statistical study of young fishes from Silvathurai lagoon, Tuticorin. *Madras Fish. Bull.*, **17** (3): 49-103.