ON THE SYSTEMATIC POSITION, STRUCTURAL MODIFICATIONS, BIONOMICS AND DEVELOPMENT OF A REMARKABLE NEW FAMILY OF CYPRINODONT FISHES FROM THE PROVINCE OF BOMBAY.

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Introduction.

Early in 1937, in the course of my observations on the breeding of fishes in the creeks of the Bombay Province, I collected a small, almost transparent fish from a backwater near Uttan in the Thana District. In the field the fish was taken to be the young of some Gobioid or Cyprinoid fish, but a few relatively larger specimens, all of which, subsequently proved to be females, were preserved for identification in the laboratory. Later, on dissection, ripe eggs were found in some of them showing thereby that they were adult individuals. A systematic study of the material showed that the fish was closely allied to *Oryzias*

Jordan and Snyder (=Aplocheilus McClelland of authors), but differed from the known species of the genus in having a larger number of rays in the anal fin. Later, males of the species were collected which possessed a remarkable, large male organ formed by the modification of the anterior rays of the anal fin. Not being familiar with the literature on Cyprinodont fishes, the species was provisionally described under Aplocheilus and Dr. S. B. Setna, Fisheries Officer, Bombay, very kindly undertook to have my results verified by sending my description and drawings to Dr. S. L. Hora of the Zoological Survey of India. After going through my account, Dr. Hora informed Dr. Setna that my fish was a perfect novelty and asked for a series of specimens of the new species with a view to study it more closely and to obtain views of other ichthyologists interested in this group of fishes. Accordingly fresh material, comprising males and females, was sent to him, but as he was unable to express any definite opinion on the systematics of the fish, he sent the material to Dr. G. S. Myers, Stanford University, California, and Dr. C. L. Hubbs, University of Michigan, Ann Arbor, for opinion. At the same time, Dr. Hora advised that detailed observations should be made on the morphology, ecology and bionomics of the species, with special reference to mating and breeding habits, for he believed that such a massive male organ could not possibly be introduced in the female genital opening for the internal fertilization of the eggs. Accordingly, from the very beginning he was of the opinion that in spite of the presence of the male organ, the fish will be found to be oviparous. His belief was further strengthened by the presence of long filamentous processes round the egg which could only be used for the attachment of the eggs to foreign objects. Both Dr. Hubbs (vide infra, pp. 383-385) and Dr. Myers not only agreed that the fish belonged to a new genus but suggested that it should be assigned to a distinct subfamily or even a family.

I was greatly encouraged by the above reports and spent a considerable part of my time in making observations on the fish both in the field and under artificial conditions in an aquarium. During all this period I was helped by Dr. S. B. Setna with his valuable suggestions. Accounts of further work done on the fish were sent to Dr. Hora from time to time who very kindly made suggestions regarding new lines of work and helped me with his criticism. He suggested to Dr. Setna that I should be allowed to complete my account of the new fish under his guidance at Calcutta in the laboratories of the Zoological Survey of India. This suggestion was readily accepted but unfortunately owing to the exigencies of service I could not visit Calcutta till August 1939, when I was able to devote four weeks to this work. I took with me a few living specimens of the fish also for making further observations in case the previous observations did not prove sufficient for elucidating any of the important points. My stay at Calcutta enabled me to revise the whole of my morphological work in the light of the literature available in the extensive library of the Department, and, after discussing the various problems with a number of workers on fishes in the Indian Museum, to elucidate the functions of the complicated external reproductive organs,

I take this opportunity to thank Dr. Baini Prashad, Director, Zoological Survey of India, for his kindness in permitting me to work in the laboratories of his department. To Dr. S. L. Hora, Dr. S. B. Setna and Dr. C. J. George, Wilson College, Bombay, I am greatly indebted for encouragement, ready help, valuable criticisms and suggestions. My sincerest thanks are also due to Dr. C. L. Hubbs and Dr. G. S. Myers for their comprehensive notes on the systematics of the fish. Mr. K. S. Misra and Mr. T. J. Job helped me in various ways during my stay at Calcutta, and for this I am obliged to them. Babu R. Bagchi, an artist of the Zoological Survey of India, very kindly made drawings Nos. 2-5, 7-9 and 11-15 under Dr. Hora's and my supervision, and I am thankful to him for his accurate delineations. The remaining drawings were made by Babu A. Mondal either from my rough sketches or specimens with his usual skill and care.

SYSTEMATIC ACCOUNT.

Classification of the order Cyprinodontes.

Classification of fishes of the order Cyprinodontes Agassiz (equivalent to Microcyprini of Regan) is somewhat complicated and has been revised from time to time in recent years. Papers dealing with these revised schemes are scattered in various journals, several of which are not always readily obtainable. As a clear idea of the position of the different groups now classed under the order Cyprinodontes is desirable, a bare skeleton of a recent classification scheme is given below for ready reference. Only such genera as are referred to in this paper are mentioned under their respective tribes.

Order: CYPRINODONTES (Microcyprini).1

Suborder: AMBLYOPSOIDEA. Family: AMBLYOPSIDAE.

Genera: Chologaster Agassiz.

Amblyopsis Dekay.

Suborder: POECILIOIDEA.

Family: CYPRINODONTIDAE.

Subfamily: FUNDULINAE.

Tribe: Fundulini.

Genera: Fundulus Lacépède.

Plancterus Garman.

Cubanichthys Hubbs.

Lucania Girard.

Tribe: Rivulini.2

Genus: Panchax Cuvier (=Aplocheilus McClelland).

Tribe: Aplocheilichthyni.
Tribe: Aplocheilini.2

Genus: Oryzias Jordan & Snyder (=A plocheilus melastigma McClelland of authors).

¹ The classification followed here is mostly according to Hubbs (1924 and 1926) and Myers (1931 and 1938).

² Myers (1938), in a foot-note on p. 137, suggests that in future the tribe Rivulini (1931) should be called Aplocheilini, and that Aplocheilini of 1931 should be known as Oryziatini.

Subfamily: LAMPRICHTHYINAE.

Subfamily: ORESTIATINAE.

Genus: Orestias Cuvier & Valenciennes.

Family: GOODEIDAE.

Subfamily: Zoogoneticinae.

Genus: Zoogoneticus Meek.

Subfamily: CHARACODONTINAE.

Genera: Characodon Günther.

Goodea Jordan.

Family: POECILIDAE.

Subfamily: GAMBUSIINAE.

Tribe: Gambusiini.

Genera: Gambusia Poey.

Belonesox Kner.

Tribe: Heterandriini.

Genus: Pseudopoecilia Regan.

Tribe: Girardini.

Genera: Girardinus Poey.

Glaridichthys Garman.

Tribe: Cnesterodontini.

Genera: Cnesterodon Garman.

Phalloceros Eigenmann.

Phallotorhynus Henn.

Subfamily: POECILIOPSINAE.

Genus: Xenophallus Hubbs.

Subfamily: POECILIINAE.

Tribe: Poecilini.

Genus: Lebistes de Filippi.

Tribe: Alfarini.

Genus: Alfaro Meek.

Subfamily: Tomeurinae.

Genus: Tomeurus1 Eigenmann.

Family: JENYNSHDAE.

Family: ANABLEPIDAE.

Genus: Anableps (Gronow.) Bl. & Schn.

Family: ADRIANICHTHYIDAE.

Family: Phallostethidae.2

Genera: Phallostethus Regan.

Gulaphallus Herre.

Family: Horaichthyidae, nov.

Genus: Horaichthys, nov.

Family Horaichthyidae, nov.

Diagnosis of the family Horaichthyidae.—The new family³ Horaichthyidae comprises small, translucent, elongate and more or less strongly

¹ This fish has recently been found to be oviparous by Dr. Myers (vide Dr. Hubbs' letter, infra pp. 383-385). Drs. Hubbs and Myers, therefore, propose to place Tomeurus in a different family.

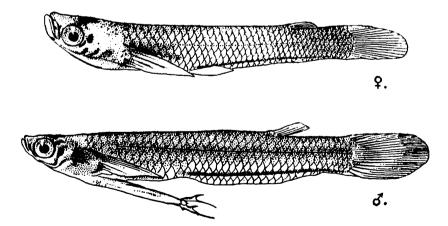
² This family has generally been regarded as belonging to the order Cyprinodontes, but Myers (1928) is of the opinion that it is not a member of that order.

³ The diagnostic characters of this fish family will be amplified on the completion of the study of its osteology.

compressed fishes. The head is depressed; the head and the anterior part of the body are flattish above. The body is covered with thin, cycloid, scales which are rather large, about 32 in a longitudinal series along the lateral line. The head is scaly. The mouth is comparatively large and the premaxillaries are not protractile. Bones of both the upper and lower jaws are loosely connected. The teeth are sharp, conical and arranged in a single series in the jaws. The premaxillaries are produced below into an arm which lies outside the mouth and is provided with teeth. The pectoral fins are set high. The anal fin is long, with about 28 to 32 rays. In the males, six anterior rays of the anal fin are separated from the rest of the fin and modified into a large gonopodium. The dorsal fin is short and situated behind the anal. In the females the right pelvic fin is usually absent. The caudal fin is rounded. The genital opening of the female is asymmetrically situated, and the region surrounding it is strengthened by the development of callosities or genital pads. The gill-membranes are broadly united and are free from the isthmus.

The family is being erected to accommodate the new genus *Horaichthys*; it appears to be allied to the Tribe Oryziatini (=Aplocheilini of the above list) of the family Cyprinodontidae, but in the latter the anal fin of the male is not modified into a gonopodium, and the female does not possess the characteristic genital pads. Moreover, the number of anal rays is much greater than that in *Oryzias*. It is probable that *Horaichthys* is evolved directly from *Oryzias*, but as the development of an elaborate male organ (gonopodium) is of outstanding significance, it is proposed to emphasize the difference by giving a distinct family rank to the new fish.

Both Drs. Myers and Hubbs noted the superficial resemblance (convergence) of this fish to *Tomeurus* Eigenmann from South America (text-fig. 1.) Dr. Hubbs, in his letter to Dr. S. L. Hora, dated May



Text-fig. 1.—Lateral views of a male and a female of *Tomeurus gracilis* Eigenmann (after Eigenmann).

10, 1938, made the following remarks about the similarity and differences in the structures of the two fishes:—

"On comparing your fish with Tomeurus, we note the same minute size, the same flattened upper surface and posterior dorsal fin, and subvertical dorsal mouth, all

indicative of the habit of swimming against the surface film (which habit has been observed by aquarium fish collectors for Tomeurus). Both fish have a similar disposition of fins, including the very posterior dorsal (still further back in your fish than in Tomeurus), the rounded caudal, the high pectoral fin with oblique axis, the main part of the anal fin far forward in the female and extremely far forward in the male (in this respect Tomeurus is the more extreme) and the pelvic fins far forward and much reduced in the female. The resemblance in fins is most striking in respect to the clasper-like modification of the anal. The resemblance goes into very considerable detail, for in both genera the modified organ is very large and has movable, pronged processes near its tip. When we examine the anal fin in more detail, however, we find striking differences. In your fish the modified tip is asymmetrical, the first prolonged ray being on the opposite side from the modified tip of the next prolonged ray and of different form, whereas the organ of Tomeurus is bilaterally symmetrical.

- "An even more striking difference in the anal fin lies in the fact that this organ in Tomeurus is very short; it corresponds almost perfectly in size and in sexual dimorphism and in regard to position as well as structure with the anterior lobe of the anal fin of the Indian genus. The long posterior portion of the anal fin of your fish is represented in Tomeurus by a sharp keel formed from the scales. No doubt the functional relations are identical but the very distinct structure is suggestive of independent origin. This point assumes particular significance when we recall that practically all American Cyprinodonts of all families have a short-based and generally few rayed anal fin, whereas the old world types tend to run to long anal fins. This circumstance leads me to believe that Tomeurus and your new genus probably represent the end products of an independent but almost identical course of evolution—possibly the most extreme example among fishes of convergent evolution.
- "Another difference between the genera confirms the view of independent origin. The premaxillaries in *Tomeurus*, as in American forms in general, are very protractile whereas as you have noted, the Indian fish has non-protractile premaxillaries strongly indicating an origin from the 'Aplocheilini'. I believe that we can accept the view that the two genera are of independent origin.
- "The discovery that the Indian fish has an egg provided with filaments obviously for attachment is even paralleled in Tomeurus, for Dr. Myers has discovered that just such an egg is produced in the South American genus. This egg is so large and the body cavity so small that only a single egg can ripen at one time. There can be little question but that the highly modified anal fin in both genera is a clasper. The excessively modified anal region of the female in your form with cartilaginous ridges and grooves would seem to be modified as a part of a 'lock and key' mechanism. I note that only the left pelvic fin is developed in both \mathfrak{P} at hand.
- "Various ideas regarding the relationship of Tomeurus have been promulgated, but all with the idea that this genus was a member of the large, viviparous family Poecilidae. In view of Myers' discoveries regarding the egg, it has seemed advisable to remove Tomeurus from the Poecilidae, and Myers and I in a joint paper are proposing to do this. Your discovery of an independently derived but almost identical type fits nicely into the view so that I believe we should not now hesitate to remove the Tomeurinae from the Poecilidae. I think the wisest course would be to erect a new family for the genus rather than to refer the subfamily to the Cyprinodontidae. By similar reference, your form should be made the type at least of a distinct subfamily and I would rather prefer to recognise its extreme modification by family separation. To do this would emphasize the remarkable example of convergent evolution which is involved.
- "Although we remove Tomeurus from the Poeciliidae, we still have in that family a form which somewhat approaches your new genus in structural features and no doubt in habits. This is the genus Alfaro, which has the anal fin, especially in the male, very far forward and the ventral edge of the body running into a scaly keel much as in Tomeurus. Our most recent views as to the position of Alfaro as related to other Poeciliidae and to Tomeurus are given in a paper by Howell Rivero and myself (Occasional Papers of this Museum, No. 339, 1936).
- "There is still another very close parallel to your new genus, and that parallelism again extends to details of size, appearance, structures, and, no doubt, habits. I refer to the extremely close resemblance of your fish to *Phallostethus*. I suppose it is remotely possible that your genus is more or less ancestral to *Phallostethus*. This view seems almost impossible in view of the extremely slight possibility that the priapium of the *Phallostethus* can be homologized with the claspers of your genus. I would assume that your new genus and *Phallostethus* represent still another example of convergent evolution. We have in mind Myers' reference of *Phallostethus* to the Percesoces (*American Museum Novitates*, No. 295, 1928). I assume of course that you have Regan's chief paper on the *Phallostethus* in *P. Z. S.*, 1916.

"In many respects the genus Anableps forms another parallel in structure and habits. This genus lives strictly on the surface and as you no doubt know is remarkable for having the eye divided so as to permit atmospheric and aqueous vision simultaneously. It is a larger and stronger fish, but also has the anal fin of the male modified and inserted anteriorly, and has the other fins rather similar to those of Tomeurus, your genus, and Phallostethus."

Genus Horaichthys, nov.

For the description of the genus reference may be made to the account of the family given above. For further details the description of the single species known (vide infra) may be referred to.

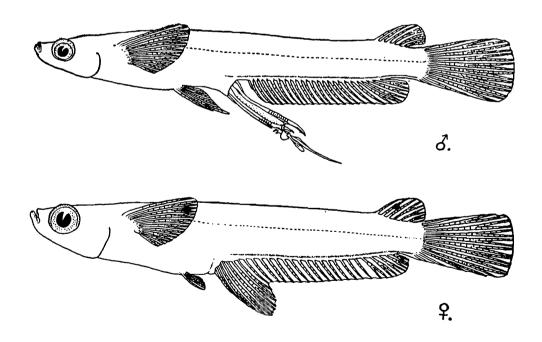
Genotype.—Horaichthys setnai, gen. et sp. nov.

Horaichthys setnai, gen. et sp. nov.

Male.—B. IV; D. 7; A. 22-26; V. 5; C. 9; Ll. 32-34.

Female.—B. IV; D. 6-7; A. 28-32; V. 5; C. 8-9; Ll. 32-34.

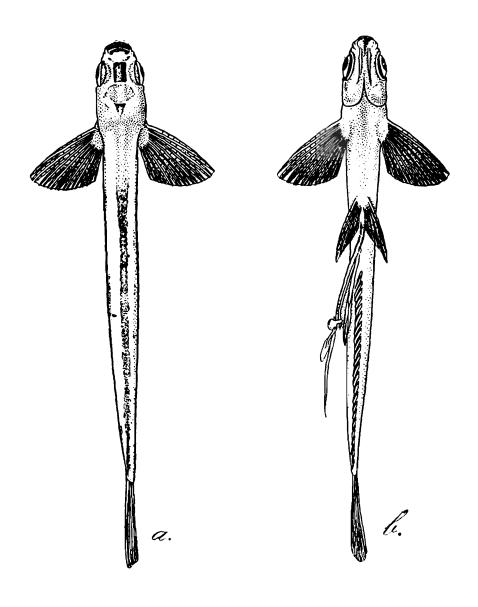
The body is elongate and narrow. The head is slightly depressed, while the rest of the body is somewhat compressed. Both the profiles



TEXT-FIG. 2.—Lateral view of a male and a female specimen of Horaichthys setnai, gen. et sp. nov.: ×4.

are slightly arched. The snout is broad and truncated by the subvertical mouth. The length of the head in the male is contained from 6.0 to 6.8 times and that of the female from 5.5 to 6 times in the standard

length. The width of the head of the male is contained nearly 1.5 times in its length, while that of the female is 1.2 to 1.4 times. The length of the snout is about half the diameter of the eye. The eyes are slightly oblong and lateral in position: they are visible both from the dorsal and the ventral surfaces. The diameter of the eye in the male is contained from 2.0 to 2.2 times and that of the female from 2.4 to 2.6 times



Text-fig. 3.—Dorsal and ventral views of a male specimen of *Horaichthys setnai*, gen. et sp. nov.: ×4.

a. Dorsal view.

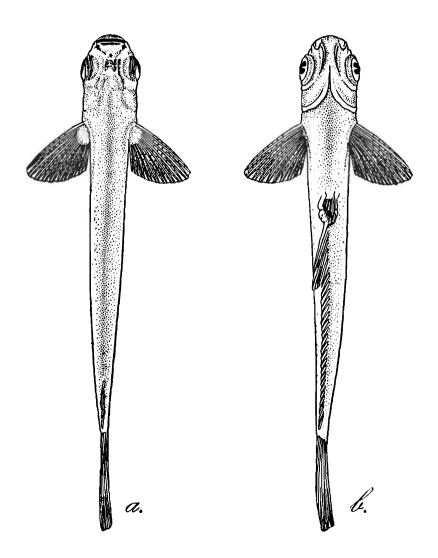
b. Ventral view.

in the length of the head. The interorbital space is raised as a broad ridge, and is about $\frac{2}{3}$ the diameter of the eye in the male and $\frac{3}{4}$ in the female. In front of the interorbital space and just behind the upper jaw there is a transverse oval depression. The nasal openings are inconspicuous and are situated dorsally between the anterior margin of the orbit and the dorso-lateral corner of the mouth.

¹ Though the lower jaw forms part of the dorsal profile and as such is included in the total length, the length of the head is recorded from the tip of the snout to the posterior margin of the operculum (Hora and Mukerji, 1938).

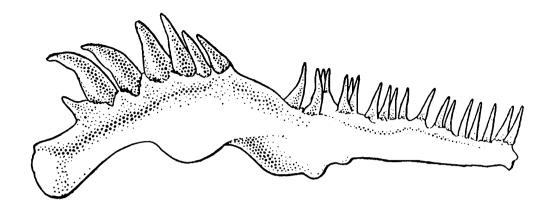
The mouth is large, non-protractile and transverse, extending across the entire width of the snout; it is so much upturned that the lower jaw forms a part of the dorsal profile. The cleft of the mouth extends downwards almost vertically, and for masticating purposes the mandibles, therefore, work backwards and forwards. The premaxillaries of the two sides do not fuse into a single bone in the middle of the upper jaw, but remain apart to make the mouth extensible. The dentaries of the lower jaw also are similarly separate in the middle.

Each jaw is provided with a single row of sharp conical teeth, but on account of congestion a slight overlapping of teeth occurs at times. The premaxillaries possess a stout, free and downward projecting arm on each side which bears teeth, like those of the jaws, but these teeth are comparatively much bigger, thicker and more closely set. They are somewhat obliquely disposed and their number varies between 7 and 9.

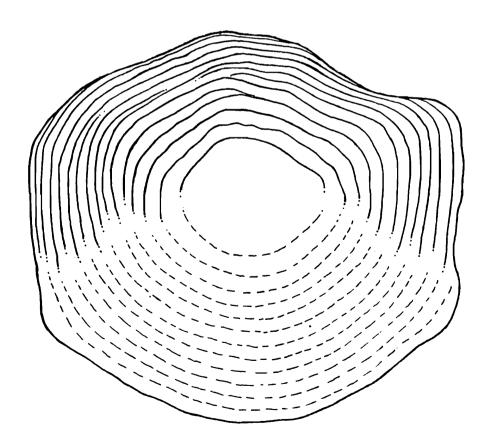


Text-fig. 4.—Dorsal and ventral views of a female specimen of *Horaichthys setnai*, generates et sp. nov.: ×4.

A portion of the premaxilla at the angle of the jaw and the free arm is edentulous and has on its posterior face an articular facet for the attach-



Text-fig. 5.—Premaxilla of the right side of *Horaichthys setnai*, gen. et sp. nov.: ×78. ment of the maxilla. The teeth of the upper jaw and its free arm are directed anteriorly, instead of downwards, in order to suit the working of the lower jaw, which has its teeth directed posteriorly. The toothed free arm of the premaxilla, remaining almost outside, does not, however, enter in the formation of the mouth and has no corresponding teeth in the lower jaw. The tongue is covered with fine tubercles visible only



Text-rig. 6.—A scale of Horaichthys setnai, gen. et sp. nov. from above the lateral line: ×108.

under high magnification. The pharyngeal teeth are simple and conical, like the teeth in the jaws, and are closely set into four groups.

The gill opening is of moderate size and extends slightly above the middle of the body between the base of the pectoral and the orbit of the eye. The gill membranes do not overlap, though they lie close together. The branchiostegals are four in number. There are four thick gills with short gill rackers.

The scales are thin moderately large and cycloid; there are 32-34 scales in a longitudinal series between the upper margin of the gill-opening and the base of the caudal fin. The lateral line appears to be rudimentary. In the male the region from the anal opening up to the base of the modified group of the anal rays is devoid of scales. In the female the region between the right ventral fin, the anal opening and the insertion of the anal fin is devoid of scales. The skin of the female in this region is thickened into irregular ridges and grooves, which I have termed "genital pads"; these are mostly round the urino-genital opening. One of these ridges is invariably continued towards the anal fin to form a thickening at its insertion. The thickened skin immediately posterior to the urino-genital opening is produced into a small conical process which probably serves as an accessory genital papilla.

The urino-genital opening in the majority of females is on the left side, somewhat anterior to the insertion of the anal fin. Sometimes, however, it lies on the right side, or is situated medially. The urino-genital opening in the male is shifted posteriorly from its normal position near the anal opening to the base of the modified group of the anal rays (the gono-podium). The anal opening in the female is deflected somewhat anteriorly to the right side from its normal position between the ventral fins, as in the male.

The dorsal fin is short and placed far back near the caudal; some of the rays almost reach the base of the caudal fin, but the two are quite The length of the base is smaller than the longest ray of the The caudal fin is large and rounded. The caudal peduncle is broad. In the male the anal fin of the young fish becomes divided into two parts The first six rays are separated off into an independent group and from the gonopodium, which is contained about 2.5 times in the standard length of the fish and remains folded on the right side. parallel to the anal fin. (The gonopodium in text-fig. 2 is shown at an angle, though actually it is nearly parallel to the body.) The longest process of this organ almost extends to below the base of the dorsal fin, The rest of the anal fin is long, it commences midway between the gill opening and the caudal base and ends below the last ray of the dorsal fin. All the rays, except for the last six or seven, are undivided and are of nearly uniform length. Anterior to the anal fin, there is a small longitudinal notch in the mid-ventral line in which the basal portion of the gonopodium is lodged. The pelvics are paired, of normal size in the male, abdominal in position and are placed at a distance slightly more than one-third of the standard length from the snout. One branch of the second ray in each fin is more elongated than the rest, has a slightly swollen tip and reaches the base of the gonopodium. In the female generally the pelvic fins are not paired, only a comparatively

small left fin being present. It is abdominal in position and is approximately in the same position as in the male. The anal fin in the female is long, commences in the anterior half of the body and extends up to a short distance from the base of the caudal fin; its second to the sixth rays are more elongated and somewhat more bent backwards than the rest. The fourth and the fifth rays are the longest. Most of the rays, except from the third to the sixth and a few at the end, are undivided; the entire fin is continuous.

The pectorals are large and well developed in both sexes. They are inserted obliquely behind the gill opening, high up on the body at the same level as the eyes; they are longer than the head and their bases are thick and muscular. The ventral edge of the caudal peduncle is not keel-shaped but has a narrow, short, fleshy, adipose edge which gradually protrudes to unite with the procurrent rays of the caudal. Dorsally, this adipose edge is still narrower.

The living specimens are nearly transparent. The head has a prominent dark occipital spot behind the eyes and a number of minute black spots scattered all over as well as on the upper margin of the jaws, the pre-orbital region and on the ventral surface. Similar spots are found on the bases of the pectoral fins, in their axils and on the thoracic region. In the position of the lateral line there is a thin black streak which is at times somewhat discontinuous. In living specimens this line consists of a number of black, elongate, closely-set dots. Above the base of the anal fin on each side there is another similar black line (made up of somewhat bigger dots), which in the male starts midway between the base of the gonopodium and the origin of the anal fin. The dorsal surface of the body and some parts above and below the lateral line are covered with fine black dots. In males the margins of one row of scales on each side of the mid-dorsal row have black pigment. There are pigment spots on the abdomen as well as between the bases of the pelvic fins and the gonopodium. Unlike the males, the whole of the dorsal surface in the females is uniformly covered with black pigment spots. ventral surface of the head, thorax and the abdomen of female specimens on the other hand have comparatively fewer black spots than in the Minute elongate, black dots may be detected under a microscope along the fin rays of most of the fins in both sexes; those on the caudal and the pectoral fins being more pronounced. A slight yellow pigment occurs by the side of the base of the anal fin in both sexes.

Sexual dimorphism.—From the foregoing description it will be noted that Horaichthys exhibits remarkable differences in the structure and appearance of the body in the two sexes. The male differs from the female in the build of the body, possession of a gonopodium and certain details in colouration. The female is characterised by the possession of the genital pads, the absence of the right pelvic fin and the asymmetrical position of the urino-genital opening.

Distribution.—Backwaters along the coast about 100 miles north and south of Bombay.

Type-specimens.—No. F13203/1, Zoological Survey of India (Indian Museum), Calcutta.

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Measurements in millimetres.

				Males.			Females.							
Total length without cauda	ı	••	••	23.4	19.7	19.6	19.0	19.0	18.8	21.3	20.0	20.0	20.0	18· 6
Length of head	••	••	••	3.3	3.2	2.9	3.1	2.9	3.2	3.9	3.6	3.7	3.7	3.1
Width of head	• •	••	• •	2.5	2.2	2.2	2.3	2.4	$2 \cdot 2$	3.0	2.8	3.0	3.0	2.8
Height of head	• •	••	• •	2.3	2.1	2.0	2.0	1.9	2.2	2.5	2•3	2.5	2.6	2•4
Depth of body	• •	••	• •	$3 \cdot 2$	3.0	2.9	3.2	3.0	3.0	3.8	4 ·0	4.0	4.0	3.4
Width of body	••	••	••	$2 \cdot 2$	1.6	2.0	2.0	2.1	1.8	2.8	2.8	3.0	2.6	2.5
Diameter of eye	• •	••	••	1.6	1.4	1.4	1.5	1.5	1.4	1.5	1.5	1.5	1.5	1.4
Interorbital distance	••	••	••	1.0	0.9	0.9	0.8	0.9	0.9	1.3	1.2	1.3	1.3	1.1
Length of snout	••	••	• •	0:8	0.7	0.75	0.7	0.7	0.7	0.7	0.65	0.55	0.7	0.55
Least height of caudal pedu	ncle	••	••	1.7	1.4	1.5	1.6	1.5	1.5	1.9	1.7	1.6	1.8	1.5
Length of dorsal fin	••	••	••	2.0	1.7	1.9	1.8	1.8	1.7	2.0	1.8	2.0	2.0	2.0
Length of pectoral fin	·• •	••	••	3.6	3.6	3.5	3.2	3.7	3.3	3.5	3.8	4.0	3.5	3.6
Length of pelvic fin	••	••	••	3.6	3.2	2 ·8	3.5	3.8	3.4	1.4	1.3	1.4	1.5	1.3
Length of anal fin	• •	••	••	1.9	2.3	$2 \cdot 2$	2.1	2.3	$2 \cdot 2$	3.6	3.5	3.2	3.1	3·1
Length of anal base	••	• •	••	8.7	7.2	7 ·2	7·1	7.1	7·0	9.6	8.5	9.1	9.5	8•2
Length of gonopodium	••	••	• •	8.1	7-4	7-4	7.3	7· 3	7·2					

STRUCTURAL MODIFICATIONS.

In the systematic account of Horaichthys, reference has already been made to certain remarkable modifications, particularly in the structures connected with sexual functions, undergone by Horaichthys setnai. Attention has also been directed to parallelism between this fish, the South American Tomeurus gracilis and a few others. In Horaichthys, not only have the anal rays attained remarkable complexity of organisation to form the gonopodium, but several other modifications of a peculiar kind are also noticeable. For instance, the genital opening of the female has shifted from its normal mid-ventral position to one side and callous pads have been developed round it. The right pelvic fin is absent in most of the females. It is, therefore, proposed to describe these remarkable modifications in greater detail in this section and to indicate their probable significance in the light of our present knowledge of the bionomics of the species.

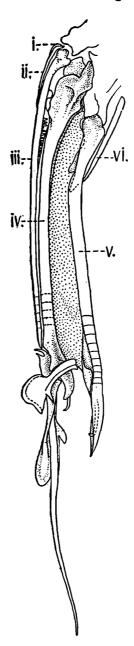
The gonopodium.—In the genera Zoogoneticus, Characodon and Goodea (now included in the family Goodeidae), first five or six rays of the anal fin in the male are shorter, thinner and stiffer and are separated by a notch from the rest of the fin. Langer (1913) coined the term 'spermatopodium' to distinguish these fin rays from the modified anal fin rays which were elongated and thickened as in the Poeciliidae, and for which Philippi (1908) had already suggested the name 'gonopodium'. The significance of the use of the two different terms, spermatopodium and gonopodium, is doubtful when the criteria of distinction are merely the comparative length and thickness of the differentiated fin rays. Langer's standard of differentiation is applied, the modified group of anal rays in Horaichthys deserves to be called a gonopodium. It is, in fact, much more complex than the gonopodium in the Poeciliidae, even though the fish is oviparous, and it functions as an organ of transference of spermatophores to the body of the female and not as an intromittent organ. In view of the above-mentioned considerations, I have used the term gonopodium for the modified group of anal fin rays forming the male organ in Horaichthys.

The gonopodium (text-fig. 7) of *Horaichthys*, as stated before, is composed of the first six rays of the anal fin. It is about 7-8 mm. in length and is contained about 2.5 times in the length of the fish. In its natural position, it lies along the right side of the anal fin and is not easily visible. The third, fourth and fifth rays of the organ are thicker and longer, and all of them are modified in diverse ways. In order to follow the modifications undergone by them, the various structures of this organ are briefly discussed below.

The First Ray.

The first ray of the gonopodium is very short and is represented only as a nodule attached to the base of the second ray. It is normal in form in the early stages but becomes gradually smaller as development pro-

ceeds. It retains, however, its right and left halves intact, even in the adult stage, as may be seen in the view of its posterior face in text-fig. 8a.



Text-fig. 7.—Gonopodium of *Horaichthys setnai*, gen. et sp. nov.: ×16.

i. First ray; ii. Second ray; iii. Third ray; iv. Fourth ray; v. Fifth ray; vi. Sixth ray.

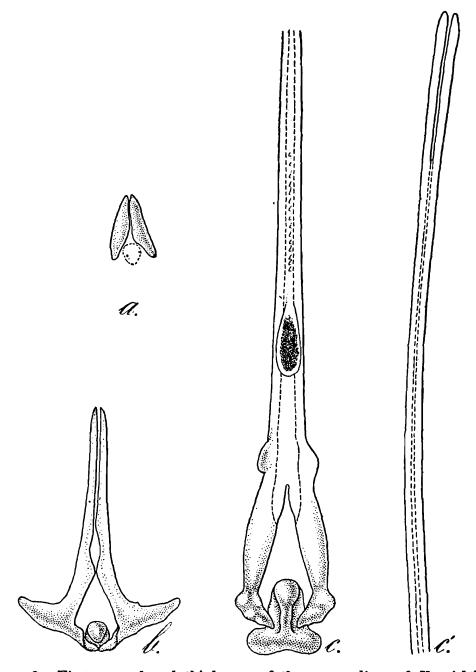
The space between their bases has a comparatively large cartilage, which represents distal segment of the first radial (pterygiophore).

The Second Ray.

The second ray (text-fig. 8b) is about three and a half times as long as the first ray. It is split to the base, though the two halves lie close together at the distal end. The basal part of each half is greatly flattened and produced sideways into curved processes which are directed posteriorly so as to embrace the base of the following ray.

The Third Ray.

The third ray is split at the base as well as at the extreme tip, and has the appearance of a pair of scissors (text-fig. 8 c, ć.). The proximal



Text-fig. 8.—First, second and third rays of the gonopodium of *Horaichthys setnai*, gen. et sp. nov.: ×82.

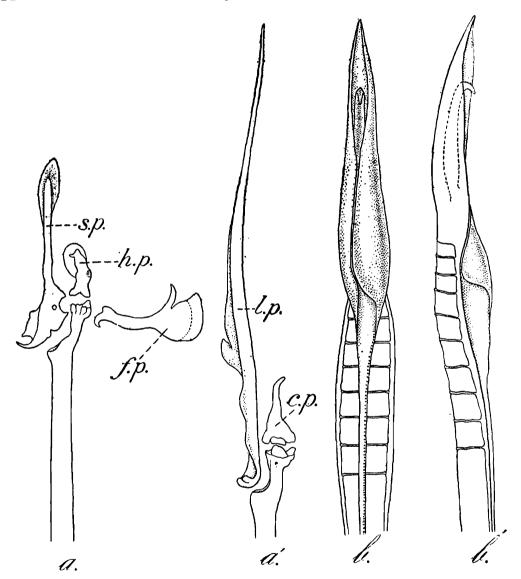
a. First ray. b. Second ray. c. c'., Proximal and distal parts respectively of third ray.

portion of the ray has a few short knobs, the most prominent of them being the one on the posterior face near the fusion of the two halves. Beyond this prominent knob on the same posterior face there are fine denticles which progressively become smaller towards the distal region of the ray. The base of the ray has a cartilagenous nodule between the two halves. This cartilage is formed of two pieces and represents the distal segments of the pterygiophores probably of the third and also of the second ray.

The Fourth Ray.

The fourth ray is made up of several pieces and is the longest of the rays composing the gonopodium. Its proximal portion consists of two laterally flattened unfused halves, and the distal of five hyaline movable pieces. Though the two halves lie side by side close to each other, they are figured separately (text-fig. 9a, \acute{a}) and are termed the right and the left components of the ray.

The proximal portion of the two components are almost identical in structure and have articular surfaces on their terminal portions, where the five pieces forming the distal part of the ray are articulated. In the basal region, the components are considerably flattened, each having three or four short knobs or processes and presenting an irregular appearance as shown in text-figs. 7, iv and 10, iv.



TEXT-FIG. 9.—Distal portions of fourth and fifth rays of the gonopodium of *Horaichthys setnai*, gen. et sp. nov.

a., a'. Right and left components respectively of 4th ray. : $\times 23$. b., b'. Side and front views respectively of 5th ray. : $\times 39$.

h. p., horn-like process; s. p., spoon-shaped process; c. p., Conical piece; l. p., long process; f. p., fifth piece.

In the distal portion, the right component of the ray has at its extremity a short, somewhat horn-like structure, as shown in text-fig. 9a, (h.p.)

This structure is bent on one side and has a short process or knob on the other side. There is also a thin roundish membrane around the bent point. Another structure, somewhat spoon-shaped in appearance (text-fig. 9a, s. p.), is attached slightly behind the first piece on the same component of the ray. The basal portion of this spoon-shaped piece has two small hooks. The remainder of the basal portion is thick and has one or two articular facets. The distal portion, which resembles the tip of a spoon, has a central hyaline rod and a thin membrane around it. The whole piece thus resembles, to some extent, a small spoon—the appearance of which varies slightly in different specimens.

The extremity of the left component also has a pointed conical piece (text-fig. $9\acute{a}$, c. p.) rotating on the articular facet. Its point is also slightly bent and the whole piece corresponds to the horn-like piece of the other component. Behind the attachment of this piece there is, as shown in text-fig. $9\acute{a}$, l. p., a long pointed process articulated to the same component of the ray. The piece has a moderately thick base and at about the second-third of its length it is produced on one side into a thin membrane which is provided with a somewhat fleshy hood-like growth. After this hood-like growth the process gradually terminates into a long stiff point. This is the longest process of the gonopodium and corresponds to the spoon-shaped structure of the other component.

In addition to these somewhat paired structures, there is a fifth piece (text-fig. 9a, f. p.) which is jointed to the articular facets of both components at their distal ends. Unlike the other pieces, it has a curved stem and appears to encircle the basal region of the other pieces obliquely. As shown in the figure, it resembles a single pointed, fleshing knife of a cobbler with an additional pointed thumb-like process on a long curved handle. The broad terminal portion of the piece is leaf-like and has a thin membranous margin. This piece is remarkable in being movably articulated to both the components of the ray.

The Fifth Ray.

The fifth ray (text-fig. 9 b., b'.) is not so complex as the fourth, though it is also considerably modified. It is not so much flattened as the fourth, and the two components are more or less fused along their entire length. Moreover, this ray unlike the former, is asymmetrical at the basal region and the number of short processes predominates on the right component. Other differences from the other rays of the gonopodium consist in an angular bend in the first-third of its length and the retention of the segmented (articulated) nature of the ray in its middle portion. Though the components are more or less fused, they are treated separately in the following description.

The right component has three short processes on its basal portion. The first is near the base and directed backward; the other two are a little away near the angular bend of the ray and are directed sideways. The anteriormost process is the most prominent. The left component has only one process near the base, the other two being undeveloped. The sides of both the components of the ray in their distal region grow into membranous, slightly concave lateral flaps. They are at their widest of the length of the ray from the base, but gradually narrow down dis-

tally as shown in text-fig. 9 b. At about $\frac{1}{3}$ of the length of the ray from the base, the inner edge of the right component grows into a narrow vertical fold and makes the ray a channel-like structure. The vertical fold, bounding the channel, is continued forward, and grows over obliquely to form an opening of a tubular portion in which the ray terminates. This tubular portion is pointed at its extremity. Presumably, it is formed by the fusion of the distal portions of both the components of the ray, their sides being rolled into a tube-like structure which remains incomplete on the anterior face, for it is formed merely by a close apposition of the two lengthwise edges of the tube. At the extremity (text-fig. 9 b) these edges remain slightly apart and give rise to a slit-like oval opening in which a hyaline recurved hook is developed on the inner wall of the tube.

The Sixth Ray.

The sixth ray (text-fig. 7, vi), though not so greatly modified as the preceding ones, differs from a normal anal ray. It is rod-like, osseous and uniformly thickened all along its length. Being comparatively short in length it reaches only up to the basal portion of the fifth ray.

General structure and function of the gonopodium.—The first four rays of the gonopodium lie close together, while the fifth is placed somewhat apart. The fin membrane between the fourth and the fifth rays thickens into a fold of skin; the fifth ray, as it thickens during development, tends to extend more towards the right side of the fold of the skin noted above. Due to this one-sided extension and the groove-like structures described above, the fifth and the fourth rays by their apposition enclose a canal.

The movement of the gonopodium is effected by the muscles on the lateral sides of the body. These muscles are inserted at the basal regions of the fourth and fifth rays. In addition to these, there are other short muscles between the bases of these two rays and their respective components for co-ordination of their actions. A more detailed study of the musculature of the gonopodium will be published at a later date.

The function of the gonopodium, as observed so far, is exclusively to transfer the spermatophores of the male (vide infra p. 410) to the female genital opening during copulation. The organ, as described above, is so complicated a structure that it is not possible to assign definite functions to its individual parts. It is clear, however, that the modifications described result in assuring a safer and surer transference of the sperms in the highly specialised spermatophores, to the female genital opening. The main burden of this task falls on the fourth and the fifth rays, which form a canal for the passage of the The first three rays and the sixth serve merely spermatophores. as supports; their unsegmented osseous nature seems to give more strength to the gonopodium at the base. The fourth ray is ossified except for a small segmented part in the middle. The fifth is ossified only in the proximal portion. From the arrangement of the supporting rays and the thickened ossified nature of the basal portion of the organ, it seems that the organ is meant to be held out firmly at an angle to the

body. Only the middle portion of the fifth ray characterised by the presence of the lateral flaps is, owing to its segmented nature, flexible. The articular surfaces and the movability of the various structures, associated with the fourth ray indicate that they are designed for independent movement while the rest of the body of the gonopodium is rigid. However, it has not been possible to assign any definite functions to these structures or to elucidate the nature of their movements.

Comparative account of the structure of the gonopodium.—The gonopodium, as an organ of the male for the transference of sperms to the female, has not so far been recorded in the oviparous Cyprinodonts. Dr. Hubbs' letter quoted above (vide supra, pp. 383-385) shows that Phallostethus and Tomeurus have recently been found to be oviparous, thus we have a few instances parallel to Horaichthys. Phallostethus is somewhat different for two reasons; firstly because it has been proposed by Myers (1928) to remove the fish from the order Cyprinodontes and secondly because the peculiar male organ of that fish, "the priapium", is not homologous with the gonopodium of Horaichthys. The former is formed by a modification of the ventral fins, whereas in Horaichthys and other Cyprinodontes gonopodia are developed from the anal fin and are thus homologous. For elucidating further the similarity of the gonopodia of various Cyprinodontes, the structures found in Horaichthys may be compared, from an evolutionary point of view, with those found in other groups of the Top-Minnows and Killifishes.

In Cyprinodontidae, in the males of Oryzias melastigma McClelland a few of the anal rays are prolonged into filiform processes beyond the fin membrane. These are not the first six as in Horaichthys, but they are generally from near about the 8th to the 11th. Moreover, they are merely simple elongations to which no function has so far been assigned. In the American Plancterus kansae Garman of the same family, the male possesses "small slender contact organs hooked forward on the anal fin and the adjacent parts of the body" These are obviously to hold the female during copulation and they serve merely as claspers. Further, as they are not restricted to the anal fin only (being found on the adjacent parts of the body as well), they may not be homologous with the modifications of the anal fin in the male of Horaichthys.

There is no doubt that the gonopodium in *Horaichthys*, described above, is homologous with the gonopodia of the family Poeciliidae. In both the families, a few of the anterior rays of the anal fin are modified to form the sperm emission organ. In Poeciliidae this organ (for figure see Hubbs 1926, pl. ii, fig. 1), formed by the specialisation of the third, fourth, and fifth rays of the anal fin is continuous with the rest of the fin while in *Horaichthys* the six anterior rays of the anal fin, forming the organ, are separated from the rest of the fin at an early stage of development and are folded back along the right side of the anal fin, though, as in Poeciliidae, the actual important rays are only the 3rd, 4th and 5th.

Though the hyaline movable structures of different shapes found in the distal region of the gonopodial rays of *Horaichthys* are not to be met with in such complexity in the Poeciliidae, a few simple structures

do, nevertheless, occur among its different genera. Tomeurus gracilis¹ (vide supra, Dr. Hubbs' letter, pp. 383-385) has a few movable processes near the tip of its male organ (text-fig. 1), which are somewhat similar to those found in Horaichthys, but they cannot be further compared as their detailed structures have not yet been described. Besides the structures in Tomeurus, the two horn-like appendages in Xenophallus (Hubbs, 1924), "antrorse appendage" in Phalloceros (Regan, 1913), long curved appendage in Cnesterodon (Regan, 1913) and the pair of curved horn-like processes of Girardinus (Regan, 1913), all to be found in the distal region of the respective gonopodia, though simpler and fewer in number, are probably homologous with the structures found at the distal end of the gonopodium of Horaichthys. Other comparable structures include the retrorse hook at the tip of the posterior branch of the fourth and the fifth rays of Gambusiini, which may possibly be homologised with the recurved hook in the tubular terminal opening of the fifth anal ray of Horaichthys.

In spite of these similarities, there are certain noteworthy differences between the gonopodium of Horaichthys and those of the Poeciliidae, the most obvious being the almost unsegmented (unarticulated) nature of the rays of the male organ in the former. All the rays of the gonopodia of the Poeciliidae are segmented right up to the extremities, whereas in Horaichthys only the fifth ray is segmented in the middle, the other rays are osseous. The middle portion of the fourth, though it retains a few marks of segmentation, is, in fact, thickened and osseous like the others. This condition shows that the gonopodia of the Poeciliidae are much more flexible than the similar structure of Horaichthys. Another outstanding difference is that the "serrae", "elbows" and the "spines" occurring as out-growths of the distal segments of the gonopodial rays in Poeciliidae are conspicuous by their absence in Horaichthys. Moreover, in the latter, the rays involved in the formation of the organ are unbranched² in contrast with the corresponding rays which bifurcate into anterior and posterior branches in the Poeciliidae; even the third ray which is sometimes unbranched in the Poeciliidae is branched in such species as Gambusia holbrooki and many others. Another difference is the asymmetrical growth of structures on the two halves of the gonopodial rays. Except in such genera as Cnesterodon, Xenophalus, etc., such asymmetry is not very common in the Poeciliidae.

Asymmetry of the nature noted above is probably not present (vide Dr. Hubbs' letter, pp. 383-385) even in Tomeurus gracilis, which, in the complexity of structure of the male organ, approaches very closely Horaichthys setnai. Moreover, neither the trowel-shaped scoop of the Phallotorhynus nor the prepucelike hood of the Poecilinae are represented in the gonopodium of Horaichthys.

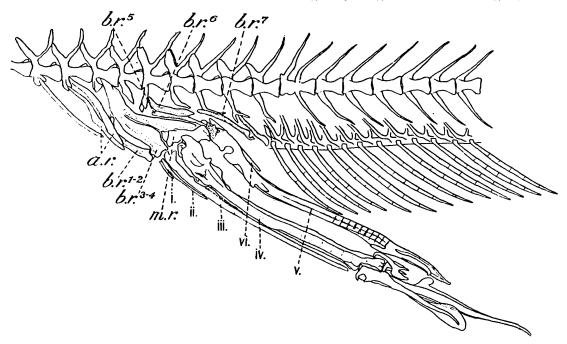
The members of the viviparous Poeciliidae are generally regarded as more specialised than the oviparous Cyprinodontes, but in the case of *Horaichthys*, an oviparous fish, the gonopodium is a very complicated

¹ Drs. Hubbs and Myers propose to separate *Tomeurus* into a new family (vide supra, p. 384).

² Presence of right and left components in a ray is not to be taken as evidence of branching, because even in early development they appear as two components. In a normal ray, they fuse into one in the adult.

structure and much more specialised than that of any member of the Poeciliidae.

Skeletal supports of the gonopodium.—The skeletal supports of the gonopodium of Horaichthys are the usual fin supports, the radials or the pterygiophores, being considerably thickened and modified. Kuntz (1913) characterised these supports in Gambusia as 'interhaemals', but Goodrich (1930, p. 64) names the support of the median fins as the radials or the pterygiophores. As the gonopodium in both the abovementioned fishes is derived entirely from the modified anal fin rays, the supports of this organ are the same as the ordinary fin supports and hence are described as radials or pterygiophores in this paper.



Text-fig. 10.—A part of the skeleton of *Horaichthys setnai*, gen. et sp. nov., to show the nature of the skeletal supports for the gonopodium.: $\times ca$ 10.

 $a.\ r.$, last pair of abdominal ribs; $b.\ r.^{1-2}$, basal radials of the first and second rays; $b.\ r.^3$, fused basal segments of the radials of the third and fourth rays; $b.\ r.^5$, basal radial of 5th ray; $b.\ r.^6$, basal radial of 6th ray; $b.\ r.^7$, basal radial of the 7th ray; $m.\ r.$, fused median segments of the 3rd and 4th rays; i-vi., 1st to 6th rays of the gono-podium.

Just as in normal fin supports, the radials of *Horaichthus* are divided into basal, median and distal segments. The basal radials of the first and second rays are united into a somewhat thicker straight bone (textfig. 10, b. r.1-2), and are in contact with the last pair of the abdominal ribs. These ribs instead of being the thinest, as is usually the case, are much thicker than the other ribs. The median segment of this radial is a small piece which fuses with the end of the basal portion of the third The distal piece of the first radial is rounded and lies at the base of the first ray; and that of the second radial appears to be fused with the corresponding segment of the third. The basal segments of the radials of the third and fourth rays are elongated, flattened and thicken-They are fused all along their length to form a large flat bone (textfig. 10, b. r.3-4), extending from the vertebral column to the base of the gonopodium. The median segments of these radials also fuse to form a separate bone with several articular facets and cavities on it. distal cartilages of these radials, as well as those of the following rays

are separate and lie at the base of each ray. The basal radials of the fifth and the sixth (text-fig. 10, b. r.5, b. r.6) are separate, thickened and rod-like, tending to form a common medial segment. The basal radial of the sixth ray, which is thicker than that of the fifth, is prolonged anteriorly and meets the haemal process of one of the vertebrae. The association of these thickened radials with the vertebral column, either directly, as in the case of the third, fourth and sixth rays, or indirectly through the vertebral ribs, as for first and second rays, is evidently to obtain an additional support and strength from the vertebral column during the movement of the organ.

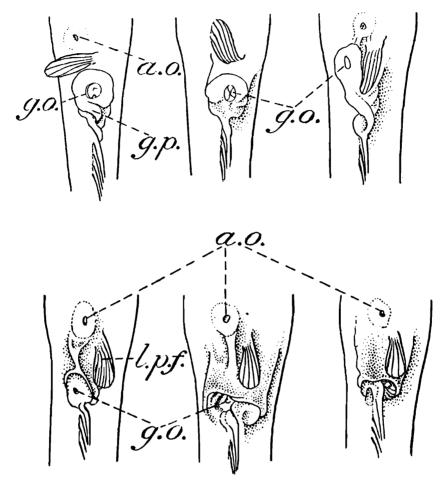
Thus, in Horaichthys the skeletal supports of the elaborate male organ are mainly the thickened radials described above. There are no gonapophyses, as in Gambusia and several other Poeciliids for the support of the muscles of the gonopodium. In Gambusia, only one fin support (called interhaemal by Kuntz, 1913) of the third ray has undergone a remarkable modification, the rest remaining normal. In Belonesox belizens also, the basal radial of the third is well developed and that of the fourth is attached to it. The radials of the third and fourth rays in Horaichthys, in addition to mere thickening, as in Gambusia and Belonesox, unite to form a large bone for supporting the gonopodium. Anableps anableps (Müll.) and Glaridichthys januarius (Phil.) have also two or three basal radials somewhat similarly fused for the support of the gonopodium.

Deflection of the genital opening in the female.—A great majority of females of Horaichthys are remarkable in that their genital openings are shifted from the usual median position to the right or left side. Examination of about 210 specimens collected in different seasons of the year showed that about 60 per cent. of them had the genital pores on the left, 20 per cent. on the right and the rest medial. It is clear, therefore, that the position of the genital pore varies in different specimens and is sometimes intermediate in position between the three types described above. This intermediate position is at times temporarily due to the gravid nature of the specimens, though in many it is a permanent feature.

The above proportions of different types of individuals show that the deflection towards the left dominates over the other two positions. If one takes into consideration the right-sided situation of the gonopodium of the male, the left-sided position of the genital opening of the female would appear to be a natural adaptation for the successful transference of the spermatophores. These adaptations on the part of both sexes obviate the necessity of bending and twisting the gonopodia so as to reach the medially placed opening of the female, as has been observed in the Poeciliidae. Even such female specimens of Horaichthys as have the apparently disadvantageous variation of having the genital opening on the right, do not fail to lay fertilised eggs in the normal Examination of preserved specimens of a few females (text-fig. 14) showed that even though the genital openings were to the right, spermatophores were found to be correctly attached to the edges of the openings. Thus it appears that being a right-sided female is not a serious bar in the way of propagation of the species. Nevertheless, it

seems that females with the genital opening on the leftside have a better chance for a successful transference to them of the spermatophores.

Asymmetrical position of the genital opening though rare among fishes, is by no means unique, as a somewhat similar characteristic is recorded in *Anableps* where the females have the genital opening either



Text-fig. 11.—A part of the ventral surface of 6 females of *Horaichthys setnai*, genet sp. nov., showing the asymmetrical position of the genital opening, the absence of the right pelvic fin and the presence of genital pads round the genital opening.

a. o., anal opening; g. o., genital opening; g. p., genital pad; l. p. f., left pelvic fin. on the right or on the left side. This difference in its situation is, however, due, as Garman (1895) believed, to "a large scale to cover the opening of the uterus and the ovary" He further adds that "it (the scale) is at one side or the other so as to open to the right in some and to the left in others" Thus a single large scale makes the females right or left-sided, in reference to the position of the female genital opening. The males also are of two types with the gonopodium either on the left or on the right side. In Horaichthys, however, all the males are of one type (right-sided), but are presumably capable of meeting the exigencies in case of wrong-sided females.

Another curious example of the unusual location of the genital opening is afforded by the blind fish Amblyopsis, in which the opening of the oviduct is placed far forward near the gill opening, so that the eggs may easily pass into the gill chamber for incubation (Henn, 1916), The genital opening in Phallostethidae also is removed far anteriorly, almost below the throat. The genital openings of both these fishes are, however, in the same position in all the individuals, but in Horaichthys,

as described above, the position of the genital opening is extremely variable in different female individuals. The occurrence of these variations in specimens from the same environment is certainly of great ecological interest and would require more extensive field work to understand its significance, if any.

Absence of right pelvic fin in the female.—Another remarkable feature of Horaichthys is that the females, instead of having paired pelvic fins, possess only a single, comparatively small fin on the left side; the right being invariably absent (text-fig. 11). A fully developed pelvic bone (the basipterygium) supporting the pelvic fins is also present on the left side only and is entirely absent on the right side. Even in the early stages, when the fins just begin to grow out only the left fin makes its appearance and neither the right fin nor its basipterygium could be traced in any stage of development. This shows that the fin normally does not develop at all. However, in five out of 210 specimens examined, both fins were found to be present. These cases in which both the basipterygia were normally developed certainly appear to be abnormal.

It is difficult to explain satisfactorily why such reduction should occur in the case of only a single fin. Even if the Lamarckian theory is brought in to explain the disappearance of the fin, there is no reason why only the right fin should run into such a disuse. The absence of pelvic fins in such families as the Muraenidae, Symbranchidae and Rhynchobdellidae, might be assigned to their habits of lurking in holes and crevices and also of burrowing in soft mud. But Horaichthys is a perfect surface dweller, rarely descending even to the bottom of shallow pools. Curiously enough, in genera like Chologaster, Orestias and Tellia¹, which resemble Top-Minnows in their habits, both the ventral fins are absent, thus differing materially from Horaichthys, in which only one The only probable explanation, therefore, is that the right fin must somehow come in the way of the movement of the gonopodium during copulation. Against this explanation, it may be urged that the male usually approaches the female from left, and that if any obstruction is felt during copulation, it must be at the left fin. does not, however, hold good if the structure of the gonopodium and its movement during copulation (vide infra, p. 410) are taken into consideration. During this act the long process of the fourth ray which is longer than the fifth ray (from the tubular extremity of which the spermatophores are presumed to issue forth) may be the first to impinge against the right fin when the gonopodium is lashed over the left side obliquely from behind. This possibility becomes clearer when it is remembered that during the aforesaid movement, the long processes of the fourth ray (being longer than the fifth) must be going across the body of the female every time when an attempt is made for the tip of the fifth ray to reach the genital pore and then to dart away. Hence it may be presumed that by such constant striking, the right fin has in the course of time been reduced and ultimately lost.

Another possible view is to consider the absence of the right ventral fin as an indifferent genetical variation (mutation). Even if, according

¹ Females of *Tomeurus* were also supposed to have no ventral fins, but recently **Dr.** Myers informed Dr. S. L. Hora that both sexes of *Tomeurus* have these fins.

to this view, the mutant had no definite advantage over the double-finned specimens, there is no reason why the original form should have died out or run into such insignificant minority after the appearance of this new form. Logically, therefore, some survival value can be safely attached to the single-finned condition as at present.

Genital pads round the female genital opening.—Another adaptive feature of the female Horaichthys to which a reference may be made is that the area near the genital opening is covered by certain callosities of the nature of grooves and ridges and which I have termed "genital As stated already, the skin in the region between the left ventral fin, the anal opening and the insertion of the anal fin is thickened and hardened to form folds and ridges in the area round the genital opening (text-figs. 11 and 14). These ridges are always irregular in disposition, transverse or longitudinal; one of them, however, is invariably continued to the anal fin to embrace the first ray. Moreover, the thickened skin at one place in this region assumes the form of a small conical process, which is probably a genital papilla; this is found always posterior to the genital opening. If the opening is to the extreme right or left, these thickenings are prominent only in the middle region. All the same, a small thickened ring does exist round the opening. These ridges are not present in the very early stages of the fish, but begin to appear when the larva is about 10 mm. in length, the anterior thickening of the anal fin being followed by the other thickenings.

Casually these thickened grooves and ridges may appear as modifications which serve as an interlocking arrangement for the hooks and process of the gonopodium, so as to enable the male to hold the female during copulation (Dr. Hubbs, vide supra, p. 384, in his letter mentioned a similar possibility). But in view of the quick process (vide infra p. 410) by which the spermatophores are transferred to the female, this explanation does not appear feasible. The probable function of these genital pads, therefore, may be for the protection of the genital area against the strokes which it receives during copulation. When the gonopodium is lashed during the momentary copulation against the body of the female, there may be, in addition to direct strokes, some piercing and scratching action of the genital area by the hooks and pointed processes of the terminal portion of the organ. In order to safeguard against such injury the development of genital pads in the female appears to be a useful Moreover, the tips of the abdominal ribs below this region, instead of ending in points as in other ribs, have their extremities slightly bent at times. Though this modification occurs in a limited number of specimens, it may also be regarded as an additional modification which has come about to counter the powerful strokes of the gonopodium during copulation. Similar protective modifications are also found in Cubanichthys, in which "the genital opening is protected by a pair of specialised scales which in the female become enlarged" The single scale on the genital pore in Anableps may be regarded as having a protective function. In Lucania "the uro-genital opening is surrounded in the female by a membranous pouch which embraces the front of the anal fin" these instances to support the surmise made above, it may confidently be stated that the genital pads in Horaichthys are protective in function.

BIONOMICS.

Habitat.—Horaichthys setnai is found in the backwaters and tanks within tidal limits along the western coast of Peninsular India about 100 miles north and south of the city of Bombay; it is a typical backwater species and has not hitherto been found in open sea or in flowing waters. It is most frequent in puddles of stagnant brackish water and in pools resulting from artificial enclosures set up by fishermen, and its range of distribution extends to waters under tidal influence. For instance, during summer the fish was taken from a creek near Mahad in the Kolaba district as far inland as nearly 35 miles from the sea. During the monsoon, when vast areas of low-lying land near creeks are flooded, the fish becomes wide-spread, and breeds in sheltered places along the edges of the creeks. After the monsoon, when the water level is lowered, large numbers become isolated in puddles, and when even these dry up towards the approach of summer, almost all of the fish perish. Large numbers, however, still remain in perennial pools within tidal limits to propagate the species. Being of a very small size and therefore of little food value, it invariably escapes the depredation of

A remarkable characteristic of *Horaichthys* is that, like *Panchax*, it occurs invariably near the surface of the water and mostly in the midst of such aquatic plants as *Enteromorpha*, *Ruppea*, etc., which usually abound along the edges of pools. It is generally found in swarms which move about in close formations. Though invariably swimming near the surface, it can not always be spotted because of its small size and transparent body.

Adaptability to different salinities of water.—In common with most estuarine fishes, Horaichthys is able to withstand a wide range of salinity. During summer, owing to intensive evaporation, isolated puddles of sea water, not always within reach of tidal influence, become highly impregnated with salt, and the fish thrives there. In one of such pools, the salinity, according to Kundson's method, was found to be 4.363 per cent. On the other hand, during the monsoon it is found in puddles with salinity as low as 1.348 per cent.

Acclimatization to fresh water.—In 1939 a few experiments were conducted with a view to determining whether the fish would thrive in fresh water. Specimens collected from a creek in May were kept in a pot of salt water for four hours in order to eliminate those which might have accidentally suffered from any injuries during collection. Out of 15 fish two died at the end of the four hours. Out of the survivors half a dozen were directly introduced into a freshwater aquarium tank in which the fish survived for only two days. In another experiment a dozen fish were introduced in a basin of brackish water in which a few marine weeds had been placed. Every day six ounces of this water was removed from the basin and 10 ounces of freshwater was added This process was continued for 12 days after which the fish were transferred to a freshwater aquarium. There were only two casualties (both males) during the 12 days, and the rest lived in the aquarium for about 2½ months. The fish, however, did not show their characteristic vigour or active habits in fresh water.

Another attempt to acclimatize the fish to fresh water was made in July 1939, when there was heavy rain-fall for a fortnight and the water of creeks and puddles had been considerably diluted. At this time half a dozen specimens were introduced directly into a freshwater aquarium in which they, with the exception of two males, which died on the 8th and the 10th day respectively, survived for about two months:

Some of the gravid females, when introduced into fresh water, laid eggs on the second day. The eggs were removed to another jar of fresh water. Out of the 15 eggs thus treated, only four hatched into larvae which died after three days.

Adaptations of the body structure for surface life.—The upturned mouth of the fish is adapted for seizing its prey by pouncing on it from below. The head, which is flat dorsally, enables the fish to swim just below the surface film of water. The mouth is comparatively large and both the jaws are provided with conical, prominent and closely-set teeth. Owing to the premaxillaries and the dentaries not being rigidly fixed the jaws are extensible. A peculiar feature is the presence of powerful teeth on the free arm of the premaxilla; these remain outside the gape of the mouth and may be helpful in holding the prey if, owing to size, it cannot be wholly swallowed. The eyes, which are large as compared to the size of the head, no doubt help the fish in locating its prey and watching the approach of any enemies.

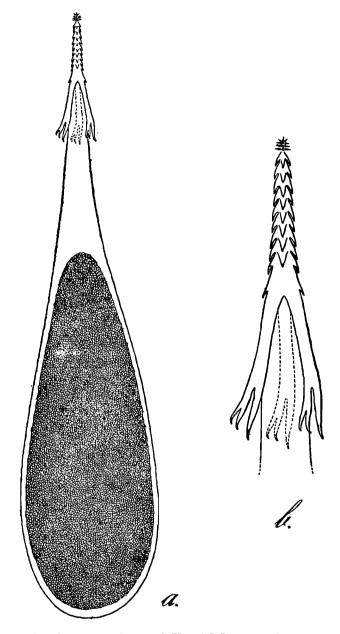
Feeding habits.—Horaichthys lives mainly on copepods, diatoms, minute crustacean larvae, etc. It seldom descends very much below the surface of the water for feeding purposes. In addition to its usual food mentioned above, it appears to devour voraciously whatever particles, whether of food or otherwise, may fall on the surface. Thus among the gut contents of several fish were found fine particles of sand, pieces of grass, leaves and other debris, which evidently do not form a part of their diet.

Like other allied carnivorous species, *Horaichthys*, too, has a short intestine. From an examination of the stomach contents, the fish does not appear to be cannibalistic in habit, but it was observed that hungry females, in captivity, at times devour their young ones.

Breeding season.—The fish breeds throughout the year, as gravid females were collected during every month of the year. However, the peak period of breeding appears to be during July and August, as the proportion of gravid females was found to be the highest in these months; at this time the water of the creeks is less saline and possesses more planktonic food. The eggs are laid in weeds along the edges of the pools in groups of about 20 to 30 at a time. They remain attached to the weeds and hatch out in about 8 to 10 days.

Spermatophores of the male.—A special feature of Horaichthys is that the testis of the male instead of giving rise to semi-fluid milt, with suspended spermatozoa, just as in other oviparous fishes, produces sperm capsules or spermatophores in each of which innumerable spermatozoa are enclosed. About 250-280 spermatophores are encountered in different stages of development in a healthy male. When fully developed a spermatophore (text-fig. 12) is a hyaline club shaped body, the broad part of which contains sperms in a mass. On an average, it is 0.61

mm. in length and 0.11 mm. in thickness. For some distance towards the tapering end of the spermatophore, there are series of barb-like



Text-fig. 12.—Spermatophore of *Horaichthys setnai*, gen. et sp. nov. a. A complete spermatophore: $\times 205$. b. Tapering end of a spermatophore with barb-like processes: $\times 530$.

structures which point backwards. The three barbs nearer the wider part form a whorl of very stout and conspicuous bifid spines. Some of the bifurcations of these spines divide again into two near their tips. A short distance in front of these spines, there are 8 or 9 whorls of short and thick closely-set barbs. The space between the bifid spines and the upper series of barbs is usually smooth or at times may possess one or two solitary barbs. The barbs near the tip of the spermatophore, consisting of two or three whorls, are very thin and, unlike others, point nearly at right angles to the long axis; and a few at the extreme tip are directed upwards. The rest of the body of the spermatophore is smooth. It is with the aid of these stiff processes and the barbs that

the spermatophores, when they are shot out, get attached near the genital opening of the female. The bifid spines by their outwardly spreading divisions probably serve to limit their penetration to the necessary depth only.

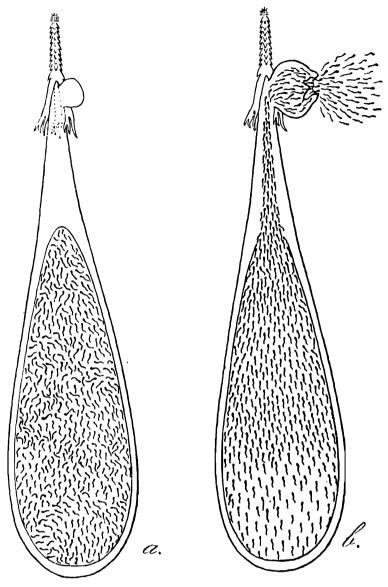
So far as I am aware, this appears to be the first instance when true spermatophores have been noted in fishes. Spermatophore-like bodies have been stated to occur in the family Poeciliidae. Philippi (1907), who first found these bodies in Glaridichthys januarius Hensel and G. decemmaculatus Janyus, called them "Spermozeugmata" (spermmasses) as they had no outer coating. In these two fishes sperms are merely rolled into a thick slightly oblong mass with their heads pointing to the periphery and a sticky cementing substance in the centre (figured by Philippi on p. 107). As described above, in Horaichthys there is a distinct hyaline case (outer cover) in which the inner sperm mass, corresponding to the spermozeugmata of Glaridichthys, is enclosed. (1913, p. 186) refers, to the 'spermozeugmata' of Gambusia as spermatophores and says, "the walls of the spermatophores are exceedingly My observation on the sperm mass of Gambusia affinis holbrookii has convinced me that there is no such delicate wall round the sperm mass as mentioned by Kuntz and that the term spermatophore, in its strict sense, is not applicable to such a structure. it seems that a true spermatophore has not so far been recorded even in Poeciliidae.

Among invertebrates Cephalopoda (Mollusca) possess tubular, well-developed spermatophores; in certain species they may be as long as 2 cms. The hectocotylised arm or the intromittant organ of Sepia is used for the transference of the spermatophores into the mantle cavity of the female, and in consequence there are no structures of attachment corresponding to the barbs and stiff processes as in the spermatophores of Horaichthys. Each spermatophore of Sepia has, however, a spring mechanism at the end of the structure by the action of which the wall of the spermatophore bursts to liberate the sperms whereas in Horaichthys this purpose, as is explained below, is attained by quite a different mechanism.

Escape of sperms from the spermatophores.—There is no permanent opening on the spermatophore for the liberation of sperms. The way in which the sperms are liberated in nature is clear only when a spermatophore from a live specimen is observed in normal saline solution. After a lapse of about 10 minutes a small rounded bulging appears between the basal portions of any two bifid spines and begins to enlarge (text-fig. 13a); this region of the spermatophore is presumably provided with somewhat weaker walls and some sort of osmotic pressure may be responsible for the bulging noted above. At a time the number of such protuberances may be even as many as three, each occurring in the interspace between two adjoining spines, but in such cases only the biggest becomes enlarged while the others remain stationary.

¹ According to Henn (1916), Ballowitz in the case of insects first proposed the term "spermozeugma" (plural "spermozeugmata") for aggregations of sperms which had no external investment.

When the protuberance is sufficiently large a semifluid substance from above the sperm mass slowly moves up and passes into it. This



Text-fig. 13.—Diagrammatic representation of the escape of sperms from the spermatophore.

a. An early stage with a bulging between the basal portions of two bifid spines.
b. Later stage showing direction of movement of sperms, the rupturing of the distended wall and escape of sperms.

action is followed by the activation of a few adjoining sperms which slowly pass into the cavity. Other sperms do likewise with greater speed, and a large number of these concentrate into the bulging part and ultimately escape when the distended wall ruptures. By the time this opening is formed the movement of the sperms in the spermatophore is greatly accelerated and numbers of these are ejected at a time. Eventually the whole sperm mass inside the spermatophore is activated and there is a continuous movement up and down in the entire cavity of the spermatophore and innumerable sperms rush about in all directions to find an exit. After about an hour or so the whole of the spermatophore becomes empty.

Act of mating.—Breeding takes place all the year round and evidence of this may be gathered from the fact that males are often seen following the females. At the time of mating, the male, with his eyes always directed towards the female, swims below and behind her at a distance of about 2 to 3 cms. He then darts towards her on the left with almost lightning speed. As he approaches his mate he lashes out the gonopodium sideways almost at right angles to his body and strikes its terminal end against her genital opening. The spermatophores are transferred to the female in this momentary contact, and become attached by their distal hooks.

The male does not, however, always succeed in transferring the spermatophores to the female during these momentary contacts. Several attempts are made at times, the male always keeping himself to the left of the female—the side on which her genital opening is usually situated. At times the male merely darts towards the female, and this is probably a sort of courting. The male appears to be always afraid of the female which on occasions chases him away.

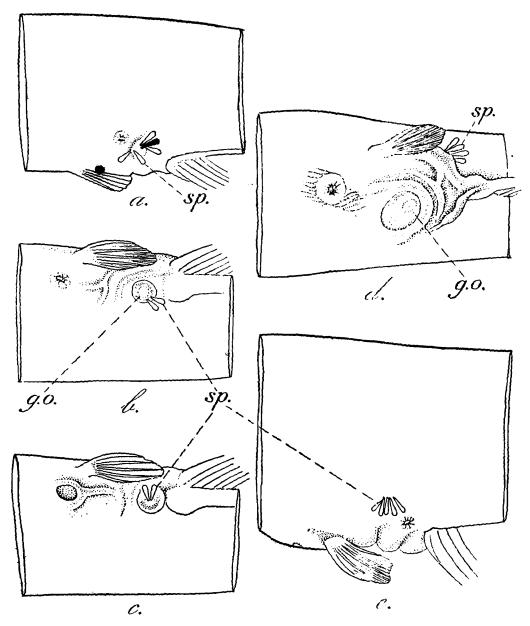
The small size of the spermatophores and the speed with which they are dischared make it almost impossible to observe the actual act of transference, but immediate examination of the female after the male's behaviour described above, may reveal the result of his amorous dashes towards his mate. On one occasion a female with no spermatophores previously attached to her body was immediately killed in formalin after it had been approached by the male. Examination of the female showed that she had 18 undischarged spermatophores attached to the anterior edge of the genital opening. They were so firmly attached, with their pointed ends imbedded in the flesh, that even the violent struggle for life by the fish, when it was put into formalin, did not dislodge them.

The gonopodium which is normally folded back along the right side of the anal fin, is capable of a forward movement when it lies at an angle to the body. The female genital openings of most females are generally situated towards the left. When the gonopodium is darted forwards from its resting position during the aforesaid activity of the male, the sperm duct which opens at the base of the 4th and 5th rays is presumably extended through the canal formed by these rays to their distal end. As the organ strikes against the body of the female during mating, the spermatophores are shot out from the sperm duct, and get attached to the female opening.

After the spermatophores are attached, the sperms are, as described above, liberated from the spermatophores, and are believed to reach the oviduct either by chance or under the chemical influence of the oviducal fluid. Their entry into and presence in the folds of the oviduct have been ascertained by dissecting live females in normal saline. Females separated from the males for about five days and having no spermatophores attached to them were dissected and the distal parts of the oviducts teased on a slide. Sperms in incredible numbers came out of the folds of the oviducal walls which seem to serve as a receptaculum seminis. Henn (1916), also observed innumerable sperms in the oviducal walls of *Lebistes* and *Pseudopoecilia*, and found that the effect of the oviducal secretion on the sperm mass of the fish

was the immediate dissolution of the mass and the activation of the sperms. It is likely that the oviducal secretion in *Horaichthys* may have some chemical attraction for the sperms, but it has not been possible for me to verify this experimentally.

In some females the genital openings were found on the right side, and even with these the males do not seem to have had any difficulty



Text-fig. 14.—Genital region of 5 females (a.-e.) of Horaichthys setnai, gen. et sp. nov. showing attachment of spermatophores in different positions: ×12.

g. o., genital opening; sp., spermatophore.

in transferring the spermatophores to the genital opening, as spermatophores were noticed to be correctly placed in a few preserved specimens as shown in text-fig. 14 b and c. A detailed examination of these specimens showed that the exact place of attachment varied in different individuals. In some the spermatophores were fixed on the inner edge of the opening, while in others on the outerside. From both situations, the sperms, when liberated from the spermatophores, would seem to be

capable of travelling the distance from the spermatophores to the oviduct. In exceptional cases they were attached rather away from the genital opening as shown in text-fig. 14d and it seems doubtful whether they are capable of covering so much distance in order to reach the oviduct.

Though the gonopodium of Gambusia is situated medially, the mode of mating is strikingly similar to that of Horaichthys in which the gonopodium is distinctly placed on the right side. Seal (1911), who was the first to observe the mating of Gambusia holbrooki, describes it as follows :-

"The male follows incessantly and warily after the female, on the left side and to the rear, the female turning and making savage dives at him, causing him to turn and flee, but to return immediately and follow, watching for a moment when her attention will be distracted, when he will make a sudden dash, sometimes succeeding in inserting the intromittent organ² into the genital pore, but oftener apparently missing it because of a quick turn of the female from which he flees in apparent terror. The contact is so sudden and brief that it required many observations to verify it. In these movements the male organ is thrust forward and to the right towards the female."

Philippi also gives a similar account of mating in *Phalloceros* and Cnesterodon.

It is thus clear that the mode of mating in Horaichthys is strikingly similar to that of the American Poeciliid fishes, though they are only very distantly related.

Longevity of sperms and fertilisation of eggs.—To study their breeding three gravid females collected from the field were kept in a jar. After a day each of them laid eggs and after 11 days another lot of eggs was laid. All the eggs were seen to develop normally. In the case of another batch,3 females were observed to lay eggs 14 days after their separation from the males. Some eggs, examined immediately after being laid, showed no trace of segmentation, the blastoderm being just in formation.

These observations make it clear that the eggs are fertilised within the body of the female and that the sperms are capable of living in the oviduct for at least 15 days. There are several records of such longevity of sperms in the oviduct in Poeciliidae. Hildebrand (1917) stated that "a female (Gambusia) kept under close observation produced five broods after she had been separated from all other fish " Henn (1916) states that Zolotnisky raised broods of Phalloceros more than ten weeks after separation from the male. It is not surprising, therefore, if the sperms of Horaichthys are found to live in the body of the female for more than a fortnight and are potent enough to impregnate even a third or fourth brood of eggs. The above observations also seem to show that in Horaichthys fertilisation occurs probably when the eggs are just being extruded as freshly laid eggs do not show any traces of segmentation. According to Villadolid and Manacop the eggs

¹ The account of mating in Gambusia published by Ryder (1885) from the details

given by A. A. Duly is contradicted both by Philippi and Seal.

2 It is doubtful whether the organ is in reality inserted in the genital pore as stated here. According to Philippi's account (vide Henn, 1916) the male "with extraordinary speed places the tip of the anal bearing a sperm capsule upon the urogenital papilla of

³ In this experiment, females at the time of separation from the males were found to have no spermatophores attached to them. Similar observations were not made in the first experiment,

of Gulaphallus are also believed to be fertilised internally in a similar manner.

Mode of laying eggs.—When the eggs are to be laid, the female moves over weeds, bends her caudal region sideways and ejects the eggs either towards the right or left, according to the position of the genital opening. The anal fin is pressed forwards, the abdomen being perhaps pressed with it, so that the fin which usually points posteriorly has its long fin rays pointed anteriorly while the eggs are being liberated. The solitary ventral fin is also pressed backwards. The eggs are shot out obliquely to a distance of about 5-6 mm., they are ejected quickly one after another, at intervals of less than a minute. When an egg is not completely ejected, but adheres to the genital opening, the next egg is ejected more forcibly, with the result that both eggs leave as it were in one conglomerate mass.

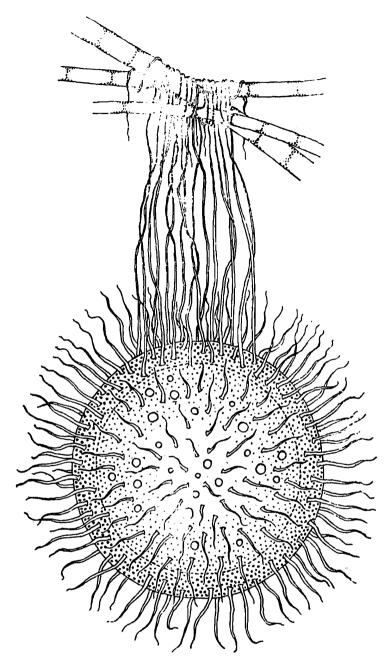
All the eggs are not laid at one spot. The female, while ejecting the eggs, is constantly on the move, though within a limited radius, evidently searching for fresh places to deposit the eggs. In this respect she manifests a preference for a place with filamentous algae, so that the eggs may get attached to them. A female which had ejected 23 eggs in 18 minutes was observed, on one occasion, to have deposited 21 more eggs after about 17 hours, but the interval at which each of these eggs was laid was not recorded. In most of the cases egg-laying occurred in the afternoon.

EMBRYOLOGY AND LARVAL DEVELOPMENT.

The Egg.—The egg of Horaichthys, which is almost transparent, spherical and demersal, usually measures about 1 min. in diameter (text-fig. 15). Like the eggs of certain oviparous Cyprinodonts and Gar-fishes, it bears numerous hair-like, hyaline processes or filaments all over the surface. A group of these processes is exceptionally elongated and appears as a long tuft on the egg. It is by these processes that the eggs are attached to the weeds. These long processes are very elastic and invariably become coiled to ensure attachment of the egg as soon as it is laid. The short filaments are mostly of uniform length and vary from 0.21 to 0.35 mm. in length in different specimens. Thus, the egg of Horaichthys, with its two types of filaments, appears in many respects to be very similar to that of the Philippine Gulaphallus (Villadolid and Manacop, 1934). The egg of Oryzias (=Aplocheilus) melastigma (Jones, 1937) is also similar, though the shorter filaments, from Jones' figure, appear to be much smaller and the longer ones rolled into a thicker cluster than in Horaichthys.

The egg membrane in *Horaichthys* is transparent and has a granular texture. Between the egg membrane and the delicate vitelline membrane covering the ovum, there is a small perivitelline space. The ovum contains a large amount of yolk with a number of oil-globules in it. The globules are numerous, small and scattered in eggs just removed from the ovary of a female, but in those in which the development has partially proceeded the oil globules are large, fewer in number and concentrated at one pole. Even in respect of oil globules, the egg

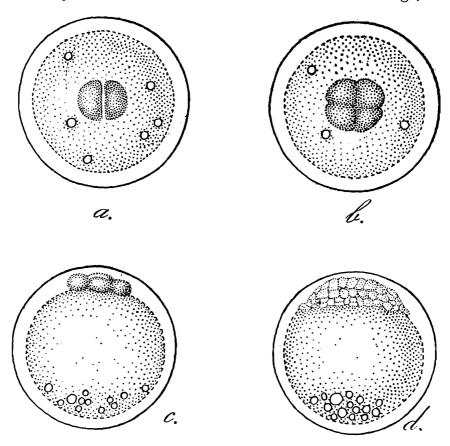
resembles that of Gulaphallus and also slightly that of Aplocheilus blokii (=Panchax parvus).



Text-fig. 15.—An egg of *Horaichthys setnai*, gen. et sp. nov. attached to some filamen tous algae. Diagrammatic.

Development of the egg.—Soon after the eggs are laid, the oil globules which are small and scattered in the yolk, begin to move towards the upper pole. While they are thus moving two or more of them come together and unite to form bigger globules, which, in turn, continue the same upward movement. The globules thus diminish in number, but simultaneously increase in size. By the time the second cleavage of the egg occurs the globules reach the upper pole and remain there till the entire development of the embryo is completed. No more coalescence of the neighbouring globules occurs after they have reached the upper pole. The number of those which persist varies from 15 to 30, and they have an average diameter of about 0.06 mm. During the movement of the oil globules towards the upper pole a circular blastodisc forms at the lower pole as a result of the concentration of the

protoplasm of the egg and presents a dome-like appearance on the mass of the yolk. The blastodisc before the first cleavage, which is

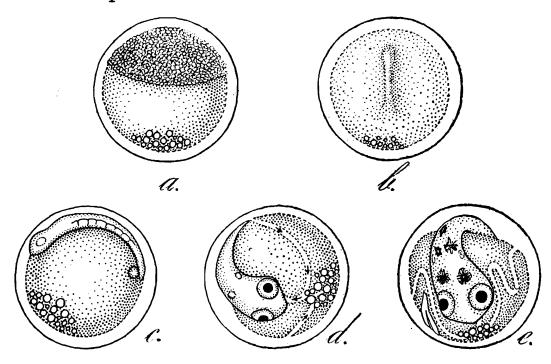


Text-fig. 16.—Stages (a.-d.) in the cleavage of egg during the development of Horaichthys setnai, gen. et sp. nov. Diagrammatic.

about two hours after the egg is laid, becomes slightly elongated. The first cleavage divides the blastodisc into two equal blastomeres by a division along the axis of the egg (text-fig. 16a). The line of the second cleavage is also meridional and at right angles to the first, the division taking about two hours to complete. Thus at the end of about four hours the first four blastomeres are clearly visible and stand out on the lower pole as shown in text-fig. 16c.

The four blastomeres are divided later by a plane of division at right angles to the axis of the egg and give rise to eight cells. The process of division then continues in the usual manner, so that at the end of about eight hours a cap of large cells forms the blastoderm. The peripheral cells of the blastoderm at this stage have no distinct cell limits and divide rapidly to give rise to the periderm. As the division proceeds the cells of the blastoderm become smaller and spread outwards on the yolk. The blastoderm thus becomes thin and forms a uniform layer of small cells covering a larger circular area on the yolk (text-fig. 17). This process of expansion continues till the great part of the yolk is covered by the blastoderm cells. After about 26 hours a linear thickening of the cells, a few layers deep, is observed to develop in the blastoderm. This linear thickening (text-fig. 17b) is the embryonic axis, which later gives rise to the embryo. After the completion of the axis, the terminal portion, which is generally away from

the oil globules, gives rise to the cephalic region; this takes about 40 hours to develop. The caudal end, at this stage, merges imperceptibly



Text-fig. 17.—Stages (a.-e.) in the formation of the embryo of Horaichthys setnai, general sp. nov. Diagrammatic.

into the blastoderm layer. A few hours later the outline of the embryo becomes visible, with a faint outline of the head, eyes, about six or seven posterior somites and the terminal caudal end (text-fig. 17c). A small vesicle, probably the Kupfers' vescicle, appears under the caudal fin at a time when the cephalic region is faintly perceptible, grows larger and gets absorbed after a day or two. The egg membrane and the embryo being quite transparent, all the stages of development are clearly observable.

On the third day (after 54 hours) the outline of the embryo is more distinct, with its structural features clearly defined externally. The head now lies near the oil globules. The head and the caudal portion of the body are free from the yolk, while the rest of the ventral portion is continuous with it. The embryo at this stage has grown over nearly three-fourth the circumference of the yolk and has about 12 visible somites.

The outstanding feature of the development of the embryo on the fourth day is the appearance of three vitelline vessels bringing blood from the yolk to the newly formed auricle and the ventricle of the heart, which has just started pulsating rhythmically. The blood current in the three vessels, two lateral and one in the middle, is clearly visible and the vessels unite below the oil globules to join the heart, which seems to lie below the cephalic region at this stage (text-fig. 17d). The auditory pits can be easily distinguished, but possess none of the chromatophores which appear later. A pair of small protruberances, which later give rise to the pectoral fins, are visible behind the auditory region.

The pigment in the eyes first manifests itself on the fifth day. The outer edge of the choroidal layer is the first to get dark, this being followed by the pigment spreading inwards. A pair of large chromato-

phores appears on the head between the eyes and the auditory region, and a solitary chromatophore on the yolk to the right of the embryo. Three or four minute black chromatophores are also seen at the bottom of the auditory pits. The vitelline vessels, which are now more developed, are not straight as before, but assume a zigzag course and tend to spread over a larger area on the yolk. A small additional vessel joins the middle one in its posterior region. The blood corpuscles appear distinctly reddish. The embryo shows jerky movements at intervals, and changes its position within the egg.

On the sixth day the choroidal layer of the eyes is completely black (text-fig. 17e). In addition to the first, a second pair of medium sized chromatophores appear on the dorsal side, one or two on the mid-dorsal line and a pair on the yolk. The embryo is now coiled within the egg-membrane and completely encircles the yolk.

After the seventh day all the characteristic pigment spots on the body are developed. Both the jaws are formed and are capable of slight movement even inside the egg. The pectoral and the caudal fins are well developed and occasionally move while still in the egg. The embryo escapes from the egg membrane about the tenth day, and after lying still for about ten minutes begins to swim freely.

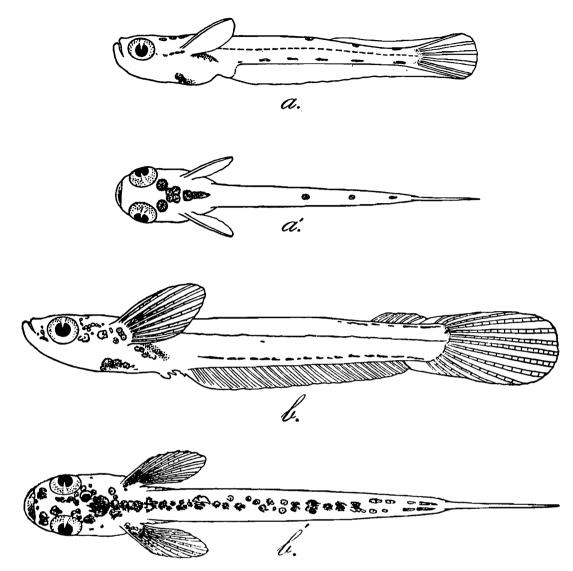
The time required for the eggs to hatch and for the various intermediate stages of development vary greatly according to the temperature and the oxygen content of the water. Even in the same room eggs laid at the same time and kept in water of the same temperature took 22 days to hatch, when the water of the vessel contained no aquatic plants. This shows that oxygenation of the surrounding water is essential for the normal development of the eggs.

Larva.—The newly hatched larva measures about 3.5 to 4 mm, from the tip of the snout to the tip of the caudal fin (text-fig. 18a, a'). There is no trace of the yolk sac appended to the body at the time the larva is hatched, but the bulging of the abdomen shows that a portion of the original yolk still persists. Even some of the oil globules of the yolk are faintly visible under the skin in some specimens. The head of the larva is about 0.75 mm. in length and 0.5 mm. in breadth. The rest of the body behind the pectoral fins is laterally compressed and gradually tapers to a thin membrane in the caudal extremity. The eyes and the mouth are well developed. Minute teeth are present in the lower jaw. The auditory capsule is clearly seen.

The pectoral fins are fully developed, have a broad base and are placed high up at the sides of the body. Each fin is about 0.5 mm. in length. The vertical fins appear as a continuous vertical fold starting dorsally at about 2/3rd of the body from the anterior end and pass round the caudal region of the body. No other fins are present. The dorsal, caudal and the anal fins originate from the ventral fold, while the others grow independently. The fin-rays have already developed in the caudal region of the vertical fold, while in other regions of this fold no such development is traceable.

The free living larva seems almost transparent to the naked eye, except for the prominent black eyes and a few dark spots on the head. The size as also the number of these large chromatophores (spots) on

the head are not constant. Generally they are two to four in number. The second pair of the chromatophores (vide p. 417) on the head fuses



TEXT-FIG. 18.—Two larval stages of Horaichthys setnai, gen. et sp. nov. in lateral and dorsal views.

a. a'. Newly hatched larva: $\times 20$. b., b'. A larva 10 mm. in length: $\times 10$.

at times to form a large chromatophore in the centre of the head. is a whitish central area in each chromatophore, while there are two or three such areas in the one that is fused. As shown in text-fig. 18a', there are two or three smaller (fused) chromatophores behind the larger ones and then follow three or four still smaller, slightly oblong black spots on the mid-dorsal side of the body. These spots are absent in some larvae. There is a row of dash-like black spots in the middle of the body on the lateral line. They are so closely set that they form an almost continuous line. There is another row of five or six dots similar to those on the dorsal side and situated just above the base of the ventral vertical fold on both sides. A number of small black dots are visible in the axilla of the ventral fins. On the abdomen there are four or five irregular chromatophores. These appear on the yolk sac as one or two chromatophores during development and are now visible on the abdomen. A few tiny spots exist on the caudal rays also.

The larger as also the other smaller pigmented spots (chromatophores) are clear and well defined in outline in living specimens, while the outlines of those preserved in formalin spread outwards and they appear less dark in colour.

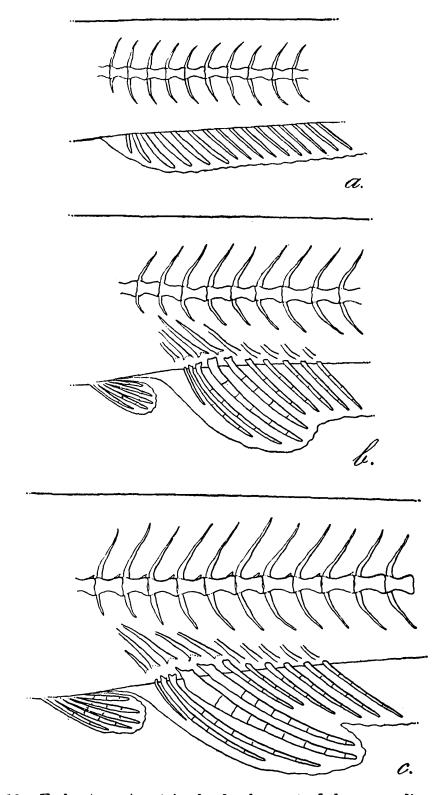
Development of the larva.—Several attempts to rear larvae in an aquarium were made, but under artificial conditions the specimens did not survive for more than a week. Those which survived till the seventh day did not exhibit many developmental changes. Only the caudal fin was slightly more demarcated, though continuous with the ventral fin fold, and traces of fin rays had appeared in the anal fin. The larvae did not thrive in the artificial surroundings probably owing to the absence of their normal food and other conditions, which have not been elucidated so far.

Even larvae of about 7 mm. in length, collected from creeks, show no development of pelvic fins. In these the fin rays are developed only in the anal portion of the vertical fold which is still continuous round The pigment spots on the head, the mid dorsal line and the caudal. by the side of the ventral vertical fold, have increased in number and size. In larvae of about 8 mm. mere outgrowths of the pelvic fins are marked and in those of about 10 mm. (text-fig. 18b), pelvic fins are clearly visible. These fins are paired in some and single in others. Individuals with paired fins are, in a majority of cases, males, while those with only one fin are definitely females. Thickenings of the skin in the genital area which later on give rise to the genital pads also begin to appear at this stage in the female larva. Except for the above-noted difference, both the sexes are superficially identical in all respects at this stage. The anal fin, which varies so greatly in two sexes in the adult form, is yet quite similar. The number of fin rays, about 28 to 32, and their sizes are also the same in both the sexes. Each ray of the anal is double with its right and left components having separate bases. Both dorsal and the anal fins have developed rays in them and are quite distinct from the caudal. The urino-genital opening of the male is seen near the base of the first anal ray. There are four pairs of teeth in the lower jaw and two pairs in the upper, but the free arm of the premaxilla (upper jaw) is so far edentulous. Some of the black pigment spots on the body of the larva have now become smaller, but their number has increased considerably on different parts of the body.

The 3rd, 4th and 5th anal rays of the male in a larva of about 12 mm. (text-fig. 19b) and the 3rd to 6th in a female larva of the same size are more elongated and the fins in both appear somewhat similar. At this stage the external morphological development of the female is almost complete, except for the teeth on the free arm of the premaxilla and slight modifications in the snout. The differentiation of the first six anal rays of the male to form the gonopodium starts only in the subsequent stages.

Development of the gonopodium.—In a male larva of about 14 mm. in length (text-fig. 19c) the 3rd, 4th and 5th anal rays increase in length, become thickened and bend towards the body. In later stages bending, thickening and lengthening continue, but the 4th and 5th rays are not

yet disconnected from the anal fin. When the larva grows to about 16 mm. in length (text-fig. 20a), the first six rays are separated from the

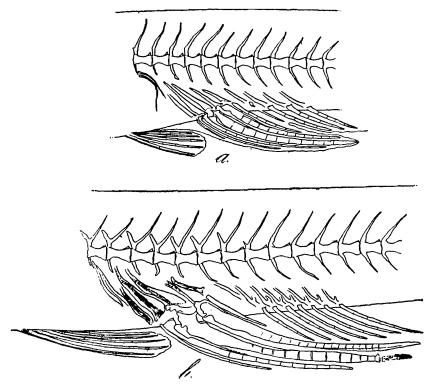


Text-Fig. 19.—Early stages (a.-c.) in the development of the genopodium of Horaickthys setnai, gen. et sp. nov.: ×22.

a. 10 mm. stage. b. 12 mm. stage. c. 14 mm. stage.

rest of the fin and the growth of the 3rd to the 5th fin rays is continued further. It is suspected that the seventh ray atrophies at this stage and disappears completely. The sixth ray remains short throughout the growth of the fish. Thus the first six rays of the anal fin of the male form a separate group. The bending, thickening and lengthening of the 4th and 5th rays continue till ultimately the rays come to lie side

by side with the anal fin on the right side (text-fig. 20b). Each ray of the anal fin, which is double in origin during development, fuses to



Text-fig. 20.—Later stage (a.-b.) in the development of the gonopodium of Horaichthys setnai, gen. et sp. nov. ×13.

a. 16 mm. stage. b. A later stage.

form a single ray, but the 4th and 5th rays retain their double nature, and their halves thicken simultaneously to produce well-developed double rays, each having two condyles for its movement at the base. The component parts of the fourth are flatter and appear to be thicker than those of the fifth ray. Later on other structures which are at the distal end develop simultaneously on both the rays till they assume the form of the fully developed gonopodium (text-figs. 9 and 10).

BIBLIOGRAPHY.

Day, F., 1889.—Fauna of British India. Fishes I, London, pp. 414-417.

Eigenmann, C. H., 1890.—On the Egg Membranes and the Micropyle of some osseous fishes. Bull. Mus. Comp. Zool. Harv. XIX, pp. 129-154.

Garman, S., 1895.—The Cyprinodonts. Mem. Mus. Comp. Zool. Harv. XIX, No. 1, pp. 5-179.

Goodrich, E. S., 1930.—Studies on the Structure and Development of Vertebrates, London, p. 94.

Harvey, H. W., 1928.—Biological Chemistry and Physics of Sea Water, Cambridge.

Henn, A. W., 1916.—On various South American Poeciliid Fishes. Ann. Carneg. Mus. X, pp. 93-142.

- Hildebrand, S. F., 1917.—Notes on the Life History of the Minnows Gambusia affinis and Cyprinodon variegatus. Rep. U. S. Comm. Fish. Appendix VI, pp. 1-15.
- Hildebrand, S. F., 1922.—Notes on Habits and Development of Eggs and Larvae of the Silversides *Menidia menidia and Menidia beryllina*. *Bull. U. S. Bur. Fish.* XXXVIII, pp. 111-120.
- Hildebrand, S. F. and Louella, E. C., 1930.—Development and Life History of Fourteen Teleostean Fishes at Beaufort, N. C. Bull. U. S. Bur. Fish. XLVI, pp. 383-488.
- Hora, S. L. and Mukerji, D. D., 1938.—Table for the Identification of Indian Freshwater Fishes, with descriptions of certain families and observations on the relative utility of the probable Larvivorous Fishes of India. *Health Bull.* No. 12, *Malaria Bureau* No. 4, Delhi.
- Hubbs, C. L., 1924.—Studies of the Fishes of the Order Cyprinodontes I.—IV Misc. Publ. Mus. Zool. Univ. Michigan. No. 13, pp. 1-31.
- Hubbs, C. L., 1926.—Studies of the Fishes of the Order Cyprinodontes VI. Misc. Publ. Mus. Zool. Univ. Michigan, No. 16, pp. 1-87.
- Jones, S., 1937.—Observations on the Breeding Habits and Development of certain Brackish Water Fishes of Adyar, Madras. *Proc. Indian Acad. Sci.* (B) V, pp. 261-289.
- Jordon, D. S., 1905.—Guide to the Study of Fishes. New York.
- Jordon, D. S., 1923.—A Classification of Fishes. Stanford Univ. Publ. Biol. Sci. III, pp. 157-160.
- Kuntz, A., 1913.—The Embryology and Larval Development of Bairdiella chrysura and Anchovia mitchelli. Bull. U. S. Bur. Fish. XXXIII, pp. 1-20.
- Kuntz, A., 1913.—Notes on the Habits, Morphology of the Resproductive Organs, and Embryology of the viviparous fish Gambusia affinis. Bull. U. S. Bur. Fish. XXXIII, pp. 177-190.
- Kuntz, A. and Radcliff, L., 1916.—Notes on the Embryology and Larval Development of twelve Teleostean Fishes. Bull. U. S. Bur. Fish. XXXV, pp. 87-134.
- Langer, W F., 1913.—Beitrage Zur Morphologie der viviparen Cyprinodontiden. Morph. Jahrb. Leipzig XLVII, pp. 193-307.
- Myers, G. S., 1928.—The Systematic Position of the Phallostethid fishes with Diagnosis of a new Genus from Siam. Amer. Mus. Nov. No. 295, pp. 1-12.
- Myers, G. S., 1931.—The Primary Groups of Oviparous Cyprinodont Fishes. Stanford Univ. Publ. Biol. Sci. VI, No. 3, pp. 7-14.
- Myers, G. S., 1938.—Studies on the Genera of Cyprinodont Fishes. Copeia, pp. 136-143.
- Norman, J. R., 1931.—A History of Fishes, London.
- Parker, T. J. and Haswell, W. A., 1938.—A Text-book of Zoology I, London.

- Philippi, E., 1908.—"Spermatophoren" bei Fischen. Verh. Deutsch. Zool. Ges. Leipzig XVII, pp. 105-108.
- Regan, C. T., 1913.—A Revision of the Cyprinodont Fishes of the Subfamily Poecilinae. *Proc. Zool. Soc. Lond.*, pp. 977-1018.
- Ryder, J. A., 1895.—On the Development of viviparous osseous Fishes. *Proc. U. S. Nat. Mus.* VIII, pp. 128-155.
- Seal, W. P., 1911.—Breeding Habits of the viviparous fishes Gambusia holbrooki and Hetirandria formosa. Proc. Biol. Soc. Wash. XXIV, pp. 91-96.
- Sundara Raj, B., 1916.—Notes on the Freshwater Fish of Madras. Rec. Ind. Mus. XII, pp. 249-294.
- Villadolid, D. V. and Manacop, P. R., 1934.—The Philippine Phallostethidae; a description of a new species, and a report on the biology of *Gulaphallus mirabilis* Herre. *Philip.* J. Sci. LV, pp. 193-220.
- Weber, M. and de Beaufort, L. F., 1923.—Fish. Indo-Austral. Archipel. IV, pp. 370-383.