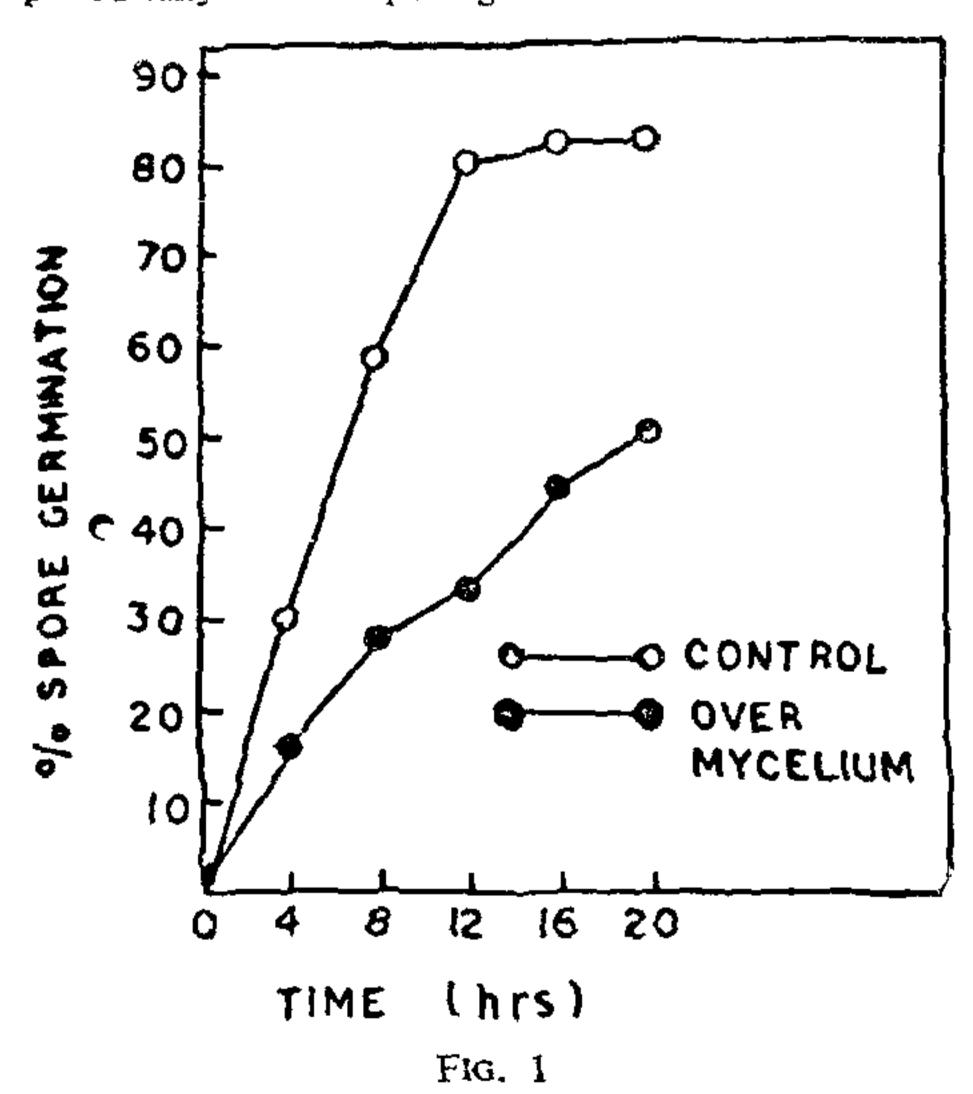
are mostly aldehydes and other miscellaneous compounds (Park and Robinson<sup>3</sup>; Rabinson and Park<sup>4</sup>; Robinson, Park and Garrett<sup>5</sup>) which in low concentration delay the spore germination. Inhibition of spore germination is due to the continuous production of volatile substances from the mycelium.

#### Muterials and Method

The test fungus as well as assay fungus were isolated from the soil, B.H.U. campus. The F solani f coeruleum was tested for its pathogenic nature.

The test fungus was cultured in sterilized czapek's-don broth, after 10 days of incubation at room temperature (27 ± 2°C). When the thick mycelial mat formed in static culture, was separated and placed in sterilized petriplates.

The sporostatic activity was tested by placing two glass rods over the mycelium over which microscope slide with boiled, sterilized cellophane, streaked with spores of the assay fungus (4 × 10<sup>5</sup> macroconidia/ml) was inverted. The lids of the petriplates were replaced by the dish of a similar size (as lower) and sealed with tape to check the drying off of the cellophane and mycelium. The plates were set in duplicate. For control similar slides were inverted in sterilized moist chamber over the glass rods. The slides were observed periodically for the spore germination.



Viability of the spores was tested by transferring the lides showing the inhibition of spore germination, to controlled condition and observing the germination after certain period of incubation when maximum germination was found.

Results and Discussion

The experimental result indicates that the mycelial mat of A. flarus produces volatile inhibitory substances to the spore germination which is confirmed by testing the viability of the spores of the assay fungus.

The A. flavus is known to produce non-volatile vacuolation factors like aspergillic acid, allatoxin and other miscellaneous toxins. These volatile and non-volatile metabolites in soil might be responsible for the mycostasis of several other pathogenic forms of soil inhibiting fungi.

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# CHANGES IN THE AMOUNT OF ASCORBIC ACID IN NORMAL AND REGENERATING BARBELS OF FISH HETEROPNEUSTES FOSSILIS (BLOCH)

VITAMIN C is very necessary for the repair of wounds in mammals, including man<sup>1</sup>. One of the obvious debilitations of scurvy is the strikingly poor repair of wounds. Vitamin C deficiency interferes with collagen fibre synthesis<sup>2</sup> and thus in the repair of wounds.

Though distribution of ascorbic acid has been studied by numerous investigators<sup>3,4</sup>, its distribution in the normal and regenerated barbels of fish has not been studied. In the present investigation, distribution of ascorbic acid has been studied in normal and regenerated barbels and regeneration blastema.

15 live H. fossilis (15-20 cm) were acclimatized in the laboratory aquaria for 9-10 days. Normal barbels from 5 fishes were cut and fixed. The barbels of remaining 10 fishes were amputated leaving half the length of the barbels. Amputated barbels of 5 fishes were removed (after 4 days) in which blastema formation has taken place. In remaining 5 fishes the amputated barbels were allowed to regenerate, and the regenerated barbels (fully formed after 30 days of amputation) were removed and fixed. Silver nitrate

method was applied to the transverse sections of normal and regenerated barbels, and longitudinal sections of blastema, to show the ascorbic acid content of the barbel fissues.

Epidermis of the barbel shows the highest intensity of ascorbic acid. In dermis, only perichondrium of cartilaginous rod and mylin sheath of the nerve show the response. In regenerated barbel silver nitrate accumulations are most prominent in the outer 2-3 cell layers of the epidermis and very much reduced in the inner layers. However, the basement membrane shows the deposition. In dermis, Vitamin C accumulation is seen around the blood vessel and in perichondrium. Blastema shows a high amount of ascorbic acid. Thus the quantity of ascorbic acid present in the normal barbels increases during the blastema formation, and again reduced in the regenerated barbels. Most of the blastema tissue in barbels of *H. fossilis* originate from the perichondrium which is made up of collegenous fibres. This shows that ascorbic acid content increases in the blastema with the increase of the collegenous fibres of periachondrium.

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## AMINO-ACID AND SUGAR COMPOSITION OF THE SILK OF THE WEAVER-ANTS, OCCOPHYLLA

The Tropical weaver-ants, Oecophylla smaragdina (Fab.) live in arboreal tent-like nests. The arboreal nests are made up of large number of leaves separated in space by considerable distances, but are connected by fine silken threads. These silken threads are secreted by the spinning glands of young larvae. During exposition, the larvae expel small droplets of 'silken fluid', exuding from the spinning glands,

and the fluid dries instantly upon exposure to air<sup>1</sup>, as in the commercial silkworm, Bombyx mori<sup>2</sup>, Chrysopa flava<sup>3-5</sup> and the spider, Lycosa sp.<sup>6</sup>. In this way several silken threads are interoven forming a thin 'cloth'. Although considerable chemical analyses have been made on the silk of Bombyx<sup>2</sup>, Chrysopa<sup>3-5</sup> and Lycosa<sup>6</sup>, no attempt has been made to study the chemical nature of the silk of the weaverants, Oeco-phylla. In view of this, the amino-acid and sugar constituents of the silk of Oecophylla have been recently made and the results are presented here.

TABLE I

Amino-acid composition of the silk of the weaverants,

Oecophylla smaragdina (Fab.)

Amino-acid	Amino-acid nitrogen (g.) per 100 g protein nitrogen		
Glycine	23.5		
Alanine	20.2		
Serine	40.7		
Threonine	3.0		
Valine	trace		
Leucine	trace		
Isoleucine	trace		
Lysine	trace		
Tyrosine	0.8		
Aspartic acid	5.7		
Glutamic acid	0.8		
Arginine	1.8		
Methionine	absent		
Cystine	absenf		
Cysteine	absent		
Proline	absent		
Hydroxyproline	absent		
Total nitrogen	16.7 %		

The silk of Oecophylla was collected with a fine forceps from the foliage nests of the mango trees, Mangifera indica in the Alagar Koil Forest Hills Madurai and in the Botanical Gardens of the Agriculture College, Madurai University, Madurai, India. Since the 'silken fluid' exuding from the spinning glands of the lativae dries up instantly, it was impossible to isolate and analyse the chemical nature of the 'silken fluid'. We collected 'silk cloth' from fresh foliage nests in which the leaves were green. The silk collected from such fresh nests is milkish-white in colour, where-

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as the silk obtained from old nests with dried leaves is brown in colour. For the present study we collected 4.5 g of fresh silk from the fresh nests and care was taken not to use the old silk.

Qualitative and quantitative analyses of the aminoacid, qualitative analyses of sugar constituents have been made employing the methods used by Giri and Rao<sup>7</sup> and Goodwin and Martin<sup>8</sup>, and Giri and Nigam<sup>8</sup> respectively. A total of five analyses have been made.

From Table I, it is seen that the amino-acids with two simplest side chains (glycine, alanine and serine) account for 84.5% of the total protein. In the silk fibroin of Bombyx, the total of the same amino acids is closely similar at 86.2%, but this total is made up of 44.1% of glycine, 29.7% of alanine and 12.4% of serine. In this context it is of interest to note that the silk of Oecophylla has a very high content of serine as in Chrysopa silk. Another point to be noted is that sulphur-containing amino acids, methionine, cysteine and cystine are totally absent in the silk of Oecophylla.

Qualitative analysis of the sugar constituents of the silk of Oecophylla shows the presence of larger amounts of glucose, galactose, and rhamnose; mannose and arabinose are present in trace amounts. In this respect, the silk of Oecophylla seems to be different from Tussak silk.

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### AN INTERESTING OBSERVATION ON GNATHOSTOMA SPINIGERUM INFESTATION IN A DOMESTIC CAT

Gnathostoma spinigerum Owen, 1836, a spirutid nematode parasite, is known to occur as adults in large tumours in the stomach of cats, dogs and several wild Carnivora. Its prevalence in cats besides other carnivores has been reported from various parts of India. During an investigation into the endoparasitic infections in domestic cats in Madra, eggs of Gnathostoma weie found in the faeces of a stray, female cat. The latter was maintained in the laboratory as a source of eggs for culture and infection studies. conclusion of the experiments, it was desired to study by roentgenography, the extent of gastric damage due to gnathostomes. Two skiagrams of the stomach of the cat, one with and the other without barium meal were taken respectively on the 13th and 14th week after the capture of the cat. Both the skiagrams showed only a shadow on the greater curvature of the stomach wall but evidence for the presence of a tumour was inconclusive. The cat was sacrificed during the 17th week. Surprisingly no specimen of Gnathostoma could be found either in the stomach or in any other location in the body of the cat, Nevertheless a shrunken, circular, fibrosed tumour-like swelling was noticed on the inner surface of the greater curvature of the stomach. It was about 25.0 mm in diameter and about 5.0 mm in thickness at its elevated centre which showed The lesion was a pinhead sized depression. strongly suggestive of a gastric rumour, the possible abode of the worms, the central punctiform depressilon evidently representing the aperture of the hollow (cavern) of the tumour. It appears that the cat should have had atleast a male and a female worm to pass fertile eggs and that they, had disappeared after the barium meal and X-ray exposures of the cat. It is interesting to note that in Australia, Heydon (1929) (cited by Miyazaki<sup>1</sup>) found as many as 49 parasites (19 males and 30 females) in the single stomach tumous of a cat.

The possibility of expulsion of the parasites, dead or alive from the cat under study, prior to X-ray exposure is remote, as fertile eggs were obtained from the faedes collected for the final egg culture experiment which was carried out just 10 days prior to X-ray studes. On the other hand there was every reason to suspect that barium meal and/or X-rays had some role in the disappearance of the paracites. Nevertheless there was no clear evidence to show how they disappeared—whether they were forced out of their habitat and expelled alive or were killed and subsequently eliminated. Further

investigations would therefore be needed to show if barium meal and/or X-rays exert any inimical effect directly or indirectly on the parasite. The finding, if positive, would be of immense value to provide a novel approach in the treatment of gnathostomosis in animals and possibly in man also, for which no effective treatment is known as yet.

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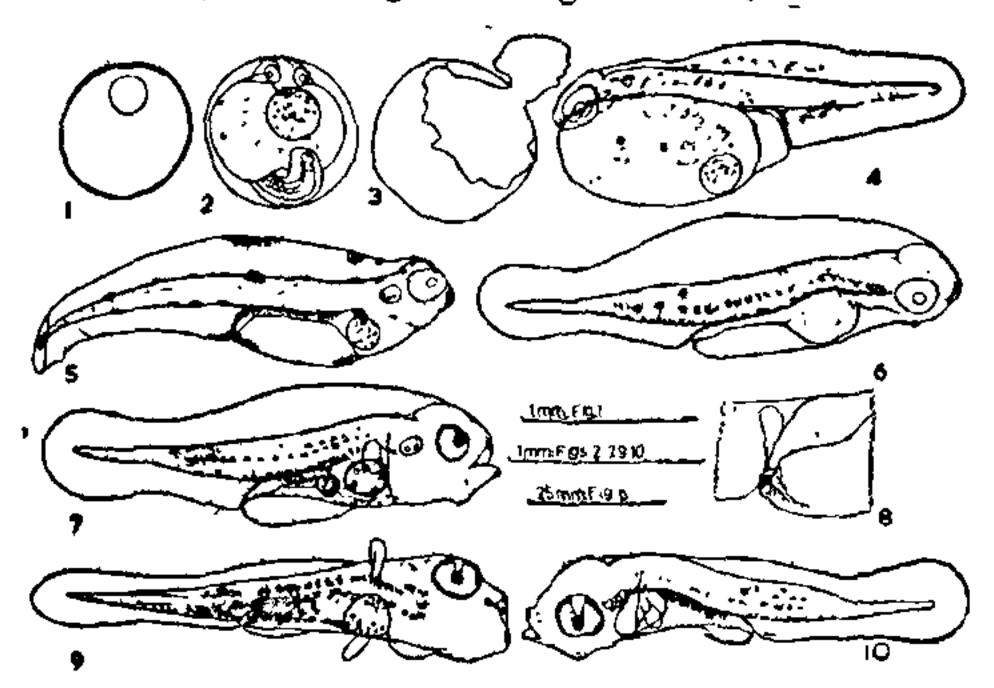
# EGGS AND EARLY LARVAE OF THE INDIAN MACKEREL, RASTRELLIGER KANAGURTA (CUVIER) FROM NEARSHORE WATERS OF PORTO NOVO

The planktonic eggs of the widely distributed and commercially valuable Indo-Pacific species Rastrelliger kanagurta<sup>1</sup> and the larvae hatching out of them have not yet been identified positively and described. Although Delsman<sup>2,3</sup> attributed certain eggs collected from Java coast at first to this species, oater, based on certain vital features, he expressed doubt on the validity of his identification. Subsequently, from the south-west coast of India, a couple of reports on the occurrence of mackerel eggs were made<sup>4,5</sup>. But, in the absence of adequate descriptions and illustrations, these accounts are not considered as reliable<sup>6,7</sup>. As such, the present communication on the eggs identified as of this species, collected from the nearshore waters of Porto Novo, may be of interest.

The Indian mackerel spawns along the south-east coast of India from January to November<sup>8</sup>; and, spawners as well as spent specimens were collected at Porto Novo during February and March, 1978. A search for the eggs in the plankton samples collected from the 10 metres depth area off Porto Novo during these months revealed the presence of mackerel eggs for the first time on 8th March 1978; and a batch of twelve eggs were isolated alive and ten numbers were reared in the laboratory for the early larval stages.

The eggs were spherical and, although pelagic, had a tendency to sink slightly and remain suspended below the water surface. The diameters of five live eggs ranged from 0.731 mm to 0.890 mm, with an average of 0.786 mm. A large oil globule was present in each, ranging in diameter from 0.204 mm to 0.250 mm, with an average of 0.208 mm. The diameter ranges of the planktonic eggs and oil globules agreed with those of ripe unfertilized eggs and oil globules recorded earlier<sup>9,10</sup> and the unpublished data with

one of us (P.B., Fig. 1). The yolk was neither segmented nor vacuolated and the eggs were pale yellolowish owing to the pigementation present on the embryo, yolk-sac and oil globule. In the egg figured at 10.00 hours on 8-3-1978 (Fig. 2) the embryo was well indicated with pale yellow pigmentation on the head, along the sides of the body and a few dark spots on the trunk and posterior region. On the yolk-sac there were a few black branching pigements. Pigmentation on the oil globule was characteristic, with pale yellow xanthophores as well as dendritic melanophores, particularly in the region facing the embryo anteriorly.



Figs. 1-10. Camera-lucida drawings of the eggs and early larvae of R. kanagurta. Fig. 1. Ripe, unfertilized ovum. Fig. 2. Planktonic egg. Fig. 3. Empty egg capsule after hatching. Fig. 4. Newly-hatched larva. Fig. 5. 20 hours old larva. Fig. 6. 26 hours old larva. Fig. 7. 46 hours old larva. Fig. 8. Anal region of 46 hours old larva enlarged to show the melanophore. Fig. 9. 68 hours old larva. Fig. 10. About 73 hours old larva.

The embryo at the time of hatching was observed to split the egg capsule and emerge out of it (Fig. 3) in a partly curved condition. Soon the newly hatched larva straightened out (Fig. 4), measuring 2.09 mm long and 0.75 mm deep at the deepest region. The oil globule was situated at the hind end of the massive yolk-sac. Pale yellow reticulation and dark spots along the dorsal and ventral margins of the body were present in the postanal region. A few pale yellow pigment spots were seen on the larval finfold dorsally and on the yolk-sac. Pigmentation of the oil globule conformed with that of the embryonic condition. The myosepta were clearly discernible, with 13 preanal and 18 postanal myotomes, which number and disposition agreeing with the adult vertebral condition.

By rearing the early larvae in the laboratory, five more stages were obtained, viz., 20h old (Fig. 5), 26h old (Fig. 6), 46h old (Figs. 7 and 8), 68h old (Fig. 9) and about 73h old (Fig. 10) measuring 2·14, 2·47, 2·32, 2·42 and 2·26 mm respectively. The

most salient features in the course of development as may be seen from the figures—were the gradual elongation of the body, pigmentation of the eyes, formation of the mouth, coiling of the alimentary canal, coagulation of the yolk and increase in pigmentation. From 26h old stage onwards, owing to the increase in pigmentation, all the myosepta were not clearly discernible, but in the 73h old stage 13 myosepta could be counted in the preanal region and 9 in the postanal region. As development progressed, highly diffused black pigments have appeared as a series from the postorbital region upto the urostyle, especially along the ventral margin of the body. Laterally also a series has appeared, initially in the postanal region and then extending anteriorly till the crbit. From 46h old stage onwards, a series of black pigements was present ventral to the viscera upto and beyond the isthmus. A pale brownish or pinkish colouration was present in the living and newly-fixed larvae, particularly beneath the mid-lateral line from behind the anus to the snout. A dendritic melanophore was present in the anal region in the 26h, 46h (Fig. 8) and 68h old larvae. Pigmentation in the 68h old stage (Fig. 9) was highly diffused ventrally; but in the 73h old larva (Fig. 10) the pigmentation was more clearly defined, in the form of a band dorsal to the viscera, a series anterior to it and another in the postanal region ventrally. Also, pigments were present ventral to the viscera, on the snout, lower jaw, etc.

The ripe ova of the Indian mackerel ranges in diameter from 0.629 to 0.900 mm<sup>9, 10</sup> and their oil globules from 0.20 to 0.25 mm. The perivitelline space is rather narrow; and, as in the case of the Japanese mackerel<sup>11</sup>, the eggs do not appear to undergo marked increase in size in the course of development. The identity of the present material as those of the Indian mackerel is based on the similarities in the size-ranges of the eggs and their oil globules as well as on the number and disposition of the myotomes, particularly in the earlier stages. The lesser number of myotomes in the 73h old larva may be explained as a feature similarly found in certain stages in the course of development, reported earlier?. Apart from these facts, the general appearance of the larvae and the pattern of pigmentation are similar to those of the Japanese and European mackerels11, 12. The position of the vent as well as the relative lengths of the preanal and postanal components of the body in the larvae of these species are seen to differ from each other and from one stage to another. Also, in both these cases, a forward shifting of the vent takes place in the early developmental stages. Thus, in the newly hatched larva of Scomber scombrus<sup>12</sup> (2.4 mm) the preanal length is 40.3%, but in the 3.7 mm stage it is only 34.6%, showing a decrease of 5.7%. In the present material also, a decrease in the prenal

length is observed, from 55.5% in the newly-hatched larva (2.09 mm) to 50.7% in the 73 hrs old stage (2.26 mm), showing a reduction of 4.8%. Hence, it is reasonable to expect that the preanal length would become further less and postanal length much more than in the earlier stages. This feature is obviously linked with the number and disposition of larval myotomes in the course of development.

Apart from R. kanagurta, two more species of mackerels were reported from the Indian region, viz., R. brachysoma from Andamans<sup>13</sup> and R. faughni from Madras<sup>14</sup>. Among all these species, R. brachysoma is the shortest bodied one and R. faughni the longest bodied. The former species is only confined to Andamans and the latter is rather oceanic and not yet recorded from Porto Novo. During February and March, 1978, mature and spawning specimens of R. kanagurta alone were available off Porto Novo, thus confirming that the present eggs and larvae are only those of R. kanagurta.

Centre of Advanced Study in Marine Biology, Porto Novo, March 25, 1978.

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### SUCCINATE DEHYDROGENASE ACTIVITY DURING ALLOXAN DIABETES IN THE BRAIN OF ALBINO RATS

Considerable information is available on enzymatic activities in rats and rabbits during insulin deficiency/absence<sup>1-5</sup>. An augmentation in the activity of liver and a decrease in kidney pyrophosphatase have been reported in rats during alloxan daibetes<sup>2</sup>. Chung and Kittong<sup>6</sup> demonstrated an elevation in the activity levels of liver aspartate amino transferase and alanine amino transferase during alloxan diabetes in rabbits. Jayashree and Nayeemunnisa<sup>7</sup> detected a general increase in the activities of aminotransferases in the broad compartments of the brain of alloxan diabetic rats. However, no information is available on the changes occurring in the activity levels of succinate dehydrogenase (SDH) in the different regions of the brain of rat during diabetes.

Motivated by the above consideration and keeping in view the importance of SDH in metabolism, the present study was initiated.

#### Material and Methods

Immature albino rats (both sexes) of Wistar strain ranging in weight from  $60-100\,\mathrm{g}$  were used. They were housed in cages at room temperature (25  $\pm$  2° C) and fed a standard pellet diet (Hindustan Lever Ltd., Bombay, India). Water was available ad libitum.

Diabetes was induced by itnravenous injection of a freshly prepared aqueous solution of alloxan-monohydrate (40 mg per kg body weight)<sup>8</sup>. The control group was injected with the same dosage of mammalian ringer<sup>9</sup>. Blood was extracted for glucose analysis<sup>10</sup> from the tail vein by a sterilized syringe. Rats with fasting blood glucose ranging from 200-220 mg/100 ml were considered diabetic and were analyzed 48 hours after alloxanization. Rats with fasting blood glucose less than 190 mg/100 ml were rejected.

The animals were sacrificed by decapitation and the brain was dissected from the dorsal side. The adhering blood vessels were removed and the brain was kept in a petri-dish with ringer at 0° C. The different regions, viz., cerebrum, cerebellum, optic

lobes and medulla oblongata were separated with sterilized instruments, weighed in ringer in an electric balance and immediately used for analyses.

Homogenates (10% w/v) of different regions of the brain were prepared in 0.25 M cold sucrose solution using glass homogenizer and were used for the enzyme assay. The SDH activity was determined following the modified triphenyl tetrazolium chloride reduction method<sup>11</sup>. The incubation mixture contained 0.5 ml of 0.1 M sodium succinate, 0.5 ml of sodium phosphate buffer of 0.1 M of pH 7.4, 0.5 ml triphenyl etrazolium chloride, 0.3% solution at neutral pH and 0.1 ml of the 10% homogenate. The incubation was carried out at 37° C for 45 minutes after which 6 ml of glacial acetic acid and 6 ml of toluene were added and kept in the refrigerator overnight The toluene layer was extracted and the optical density was read at 505 nm in Du<sup>2</sup> Beckman's spectrophotometer.

#### Results and Discussion

The data after statistical analysis is presented in Tables I and II.

The blood glucose levels showed 105 % increase over controls (Table I). Brain weight and body weight decreased considerably in the diabetic state (Table I). The activity level of SDH in general showed an increase in all the compartments of the brain investigated, viz., cerebrum, cerebellum, optic lobes and medulla oblongata during alloxan diabetes (Table II). It is also seen that the increase in the activity levels of SDH showed variation in relation to the region of the brain. The activity levels of SDH was more in the cerebellum and less in the optic lobes in control rats. Such a difference in the activity level of the oxidative enzyme may be related to the differential functional status of the concerned region. Further optic lobes and the brain stem exhibited marked response for the changes in the activity levels of SDH (Table 11). It is therefore evident that the optic lobes and brain stem are the regions which are highly susceptible to the effects of alloxan diabetes, thus safeguarding the cerebrum and cerebellum with their highly significant functional assignments.

TABLE I

Blood glucose levels and body and brain weight of control and alloxan diabetic rats\*

Blood glucose* mg/100 mi		Body weight* in gm		Brain weight* in gm	
Control	Diabetic	Control	Diabetic	Control	Diabette
100	305	77.5	68.0	1.58	1.40
±: 2·5	± 2·2	士 3·2	± 2·8	0÷06	± 0.1

<sup>\*</sup> Values are means ± S.D. of 6 observations,

TABLE II

Succinate dehydrogenase activities in different regions of the brain of normal (controls) and alloxan diabetic rats

	Control		Test	Level of Significance
Ccrebrum	5·2±2·1	+95·7*	10·2±1·8	P<0.01
Cerebellum	$5 \cdot 3 \pm 0 \cdot 03$	<del>+</del> 104·6*	10·7±2·7	P<0.001
Optic lobes	4·5±1·7	十171·9*	12·1±1·9	P<0.001
Medulla oblongata	5·6±2·7	+140.3*	$13 \cdot 3 \pm 3 \cdot 6$	P<0.001

Values (Means  $\pm$  S.D. of 5 observations) are expressed as milligrams of NTC reduced/gram wet weight/hour. \* Percentage change, + signs indicate increase in the activity level with respect to the control.

The general increase in SDH activity in all the four regions of the diabetic brain could be due to a constant production of succinate as it is known that lipid cata, bolism is accelerated and fatty acids get catabolised at an enhanced rate in the diabetic state<sup>12</sup>. This may account for the increased production of acetyl coA via enhanced beta oxidation of fatty acids ultimately leading to the augmentation in the activity level of succinate dehydrogenase.

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### PHYSALOPTERA FUNAMBULI SP. N. (NEMATODA, PHYSALOPTERIDAE) FROM FUNAMBULUS PENNANTI

ORTLEPP<sup>1</sup>, while dealing with the nematodes of the genus *Physaloptera* Rudolphi, has described six species as parasites of rodents. Schulz<sup>2</sup> has dealt with *P. varani* as *Abreviata* (*Abreviata*) varani. The two are now considered as synonyms. Ortlepp<sup>3</sup> has laid emphasis on the number and mode of origin of uterine branches as the main taxonomic character and concluded that "the whole genus is so compact that its individual species have so many characters in common, that at the present state of our knowledge of *Physaloptera*, I am compelled to retain all in the same genus".

Morgan<sup>4</sup>, while emphasizing the host specificity of *Physaloptera* of rodents for Rodentia, has described eight species from rodents and Johnson<sup>5</sup> four species from mammals out of the 11 known from India. So far, no species of *Physaloptera* has been reported from the present host, *Funambulus pennanti*, the five-stripped squirrel.

Adult worms, (13), excluding 434 larvae collected from the body cavity, have been recovered from the stomach of the five-stripped squirrel, Funambulus pennanti at Jodhpur, India, and designated as Physaloptera funambuli n. sp. This study is based on eight specimens (3 males and 5 females) which were cleared in lactophenol for morphological details and measurements (mean in parentheses).

Physaloptera funambuli sp. n. (Fig. 1 A-E).

Body tapering at two extremities except in male where the posterior end is provided with expanded caudal alae. Cuticle thick and transversely straiated. The head bears two large massive fleshy pseudolabia, each of which has two papillae, one amphid and a large prominent pointed median tooth with a small lateral tooth and six denticles on either side,

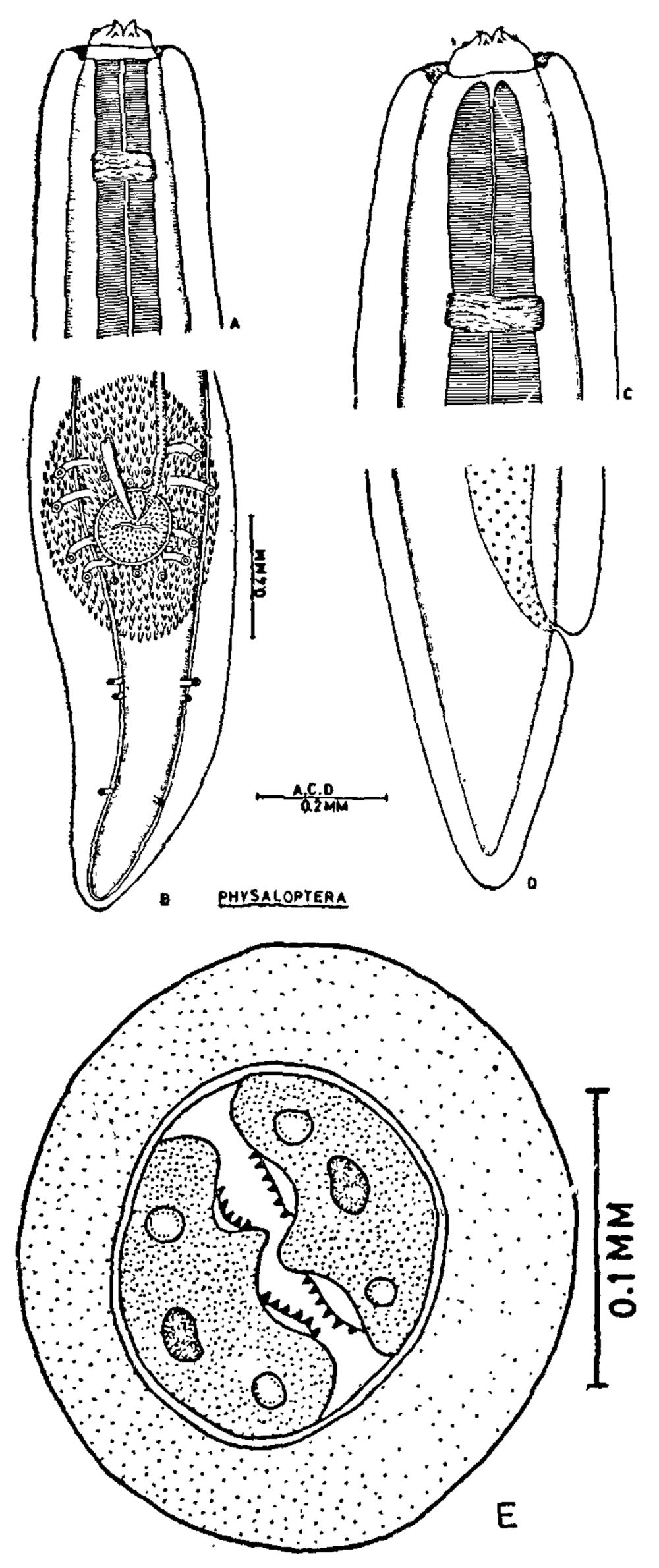


Fig. 1. Physaloptera funambuli sp. n. A. anterior end of male; B, Posterior end of male—ventral view; C, Anterior end of female; D, Posterior end of female—lateral view; E, En face view.

Male: Body  $21 \cdot 15 - 23 \cdot 19$  ( $22 \cdot 28$ ) mm long and  $0 \cdot 60 - 0 \cdot 66$  ( $0 \cdot 63$ ) mm maximum wide. Oesophagus

3.45-3.60 (3.53) mm long and 0.30-0.33 (0.31) mm maximum diameter. Nerve ring at 0.36-0.40 (0.37) mm from anterior end. Spicules unequal, dissimilar, the left larger, 2.72-2.88 (2.80) mm, than the right, 0.19-0.20 (0.193) mm. Caudal alae 1.96-2.12 (2.03) mm long and 0.81-0.87 (0.83) mm wide. Papillae 10 pairs, 7 pairs of pedunculated (2 pairs precloacal, 1 pair addeloacal and 4 pairs postcloacal) and 3 pairs of sessile (3 precloacal and 3 postcloacal). Tail blunt, 0.92-0.96 (0.94) mm long

Female: Body 27.55-49.20 (35.12) mm long and 0.74-1.20 (0.86) mm maximum wide. Oesophagus 3.88-6.41 (4.47) mm long and 0.34-0.50 (0.38) mm maximum diameter. Nerve ring at 0.45-0.60 (0.49) mm from the anterior end. Vulva 4.67-9.26 (5.88) mm from the anterior end leading into a thick walled muscular vagina which communicates with the main trunk of the uterus, the latter branching dichotomously into four uterine branches. Eggs  $0.02-0.045 \times 0.02-0.04$  ( $0.028 \times 0.024$ ) mm. Tail 0.40-0.73 (0.53) mm.

Host: Funambulus pennanti

Location: Adults in stomach and larvae in cysts in body cavity.

Locality: Jodhpur, India.

Specimen: Holotype, male No. UDZ 218; allotype, female No. 219; paratype males and females No. 220; deposited in the Department of Zoology, University of Jodhpur, Jodhpur.

P. funambuli sp. n. is distinguished from all the known species reported from rodents, viz., P. circularis Linstow, 1897, P. citelli (Rud., 1819), P. inermis Linstow, 1906, P. murisbrasiliensis Dies. 1861, P. sciuri Parona, 1898, by Ortlepp<sup>1</sup>; P. aduensis Baylis, 1928, P. bispiculata Vas et Pereira, 1935, P. brasiliensis Morgan 1942, P. getula Seurat, 1917, P. massino Schulz, 1926, P. spinicauda McLeod, 1933 and P. torresi (Travassos, 1920) by Morgan<sup>4</sup>; and from P. hispida Schell, 1950 by Schell<sup>6</sup>; besides the host, by the size of the spicules (left largest and the right smallest), number of papillae (10 pairs, 7 pedunculated and 3 sessile) and size of the eggs (smallest except that of P. sciuri).

The new species can also be separated off from the four species reported from mammals out of the 11 spp. known from India (Johnson)<sup>5</sup>, viz., brevispiculum Linstow, 1906, P. masoodi (Mirza, 1934), P. praeputialis Linstow, 1899, by the absence of prepuce-like cuticular sheath—and from P. musculi Thwaite, 1927, by the size of the spicules (left larger and the right smaller), the number of the papillae (10 pairs—7 pedunculated and 3 sessile vs. 12 pairs—4 pedunculated and 8 sessile with a single controversial subventral one), absence of longitudinal ridges and the size of the eggs (smaller vs. larger).

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## A CASE OF AMBISEXUALITY IN AN AIR-BREATHING FRESH-WATER TELEOST, CLARIAS BATRACHUS (L.)

HERMAPHRODITISM is often found in lower phyla of the animal kingdom; but it is occasional in higher vertebrates. Teleostean fishes, in general, are dioecious in nature. However, instances of monoecious specimens have also been reported, but only in a few species.

In teleosts, hermaphroditism may occur either as a result of teratological variation or during the course of sex reversal, except in certain species where it is a regular feature. Teleostean fishes providing instances of teratological hermaphroditism in India include—Hilsa ilisha<sup>1</sup>, Cirrhina reba<sup>2</sup>, Barbus stigma<sup>3</sup>, Polynemus heptadactylus<sup>4</sup>, Rastrelliger canagurta<sup>5</sup>, Kotsuwonus pelamis<sup>6</sup>, Mystus vitatus<sup>7</sup>, Sardinella longiceps<sup>8</sup> Clarias batrachus<sup>8</sup>, Eleutheronema tetradactylum<sup>18</sup>, Gerres oyena<sup>11</sup> Heteropneustes fossilis<sup>12-13</sup>, Channa striatus<sup>14</sup>, Labeo fimbriatus<sup>15</sup>, etc.

The present observation is made on a specimen of Clarias batrachus belonging to zero-year class. The specimen stemmed from a paddy field catch in the Ranchi plateau region in September, 1977. The specimen had a total length of 130 mm weighting 15 gm. The smooth external appearance of the gonads gave the usual impression of being ovaries, The left gonad was smaller than the right one, measuring 11.0 mm and 11.5 mm in length respectively. The gonoduct measured 7.0 mm in length (Fig. 1).

The material was fixed in Davidson's fixative and sectioned at 10 micron. The sections were stained with Biebrich scarlet and FCF green, according to

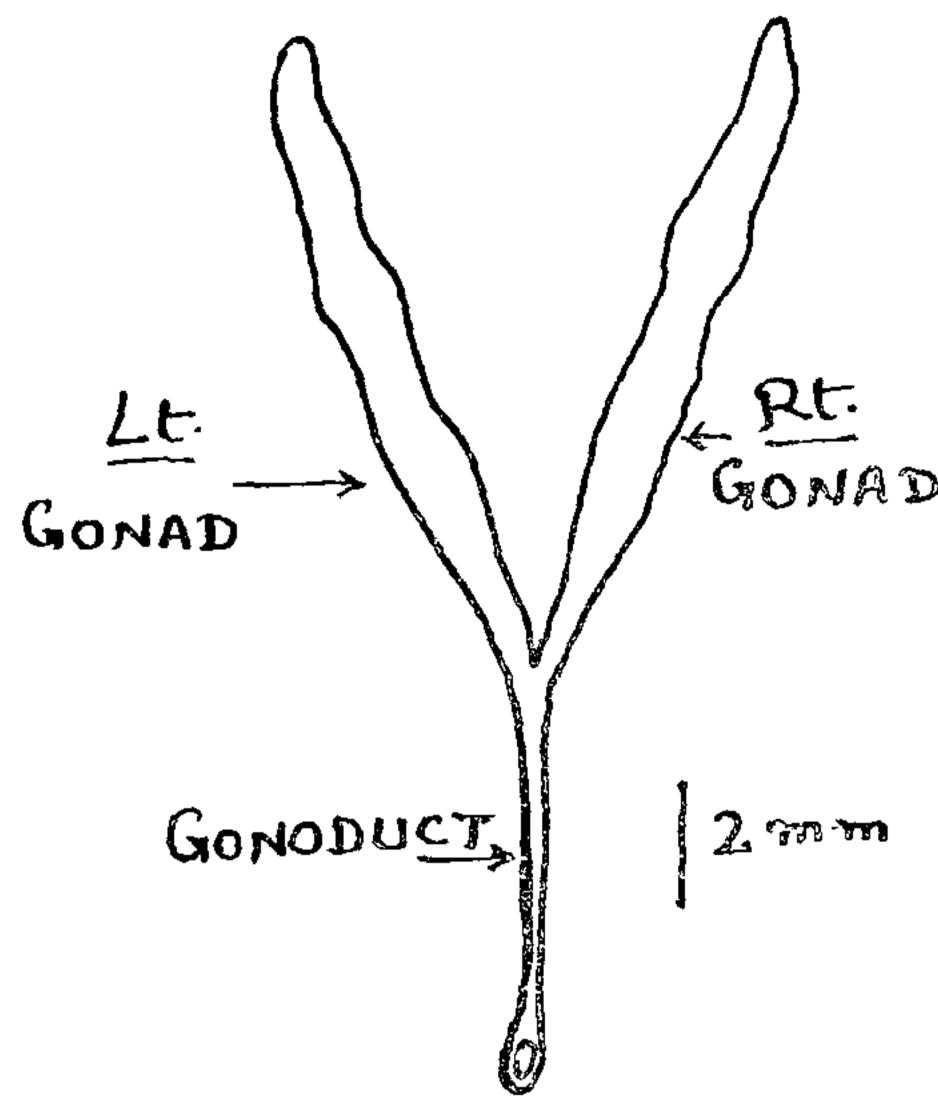


Fig. 1. Outline drawing of the gonads of the examined Clarias batrachus (Total length 130 mm).

the procedure of Beckert and Garner<sup>17</sup> Other stains such as haematoxylin-eosin and iron-alum haematoxylin were also used. Examination of the sections revealed that the gonad is ovotestis, contrary to its external anatomical appearance as ovary. The major part of the gonad was composed of testicular tissues (Fig. 2). Hermaphroditism has been observed by Lehri<sup>9</sup> in the mature specimen of Clarias batrachus, He observed that the testicular and ovarian tissues were enveloped by a common sheath, and there was a clear line between testicular parts, which lie on the anterior side, and ovarian region, on the posterior side of the gonad, connected by a narrow connection. In the present case, however, the ovarian tissues were found strewn in between the testicular lobules (Fig. 2 A). The germinal epithelium which gave rise to the oocytes, were found scattered in between the testicular lobules. The oocytes were not found to be intermingled with the spermatogonial cells as has been observed by Belsare18 in the four months old specimen of C. batrachus.

The maturing oocytes were characterized by basophilic cytoplasm and a nucleus with diffuse chromatin. Nucleoli were arranged on the priphery of nucleus. The testicular part was composed of testicular lobules containing spermatogonia. Spermatocytes and sperms were not seen in the lumen of the tubule.

To assign it as a case of sex reversal, needs further study, as the specimen in question was in the first

preparatory phase (for breeding) of its life. At the same time, it is premature to assert it as a normal hermaphrodite.

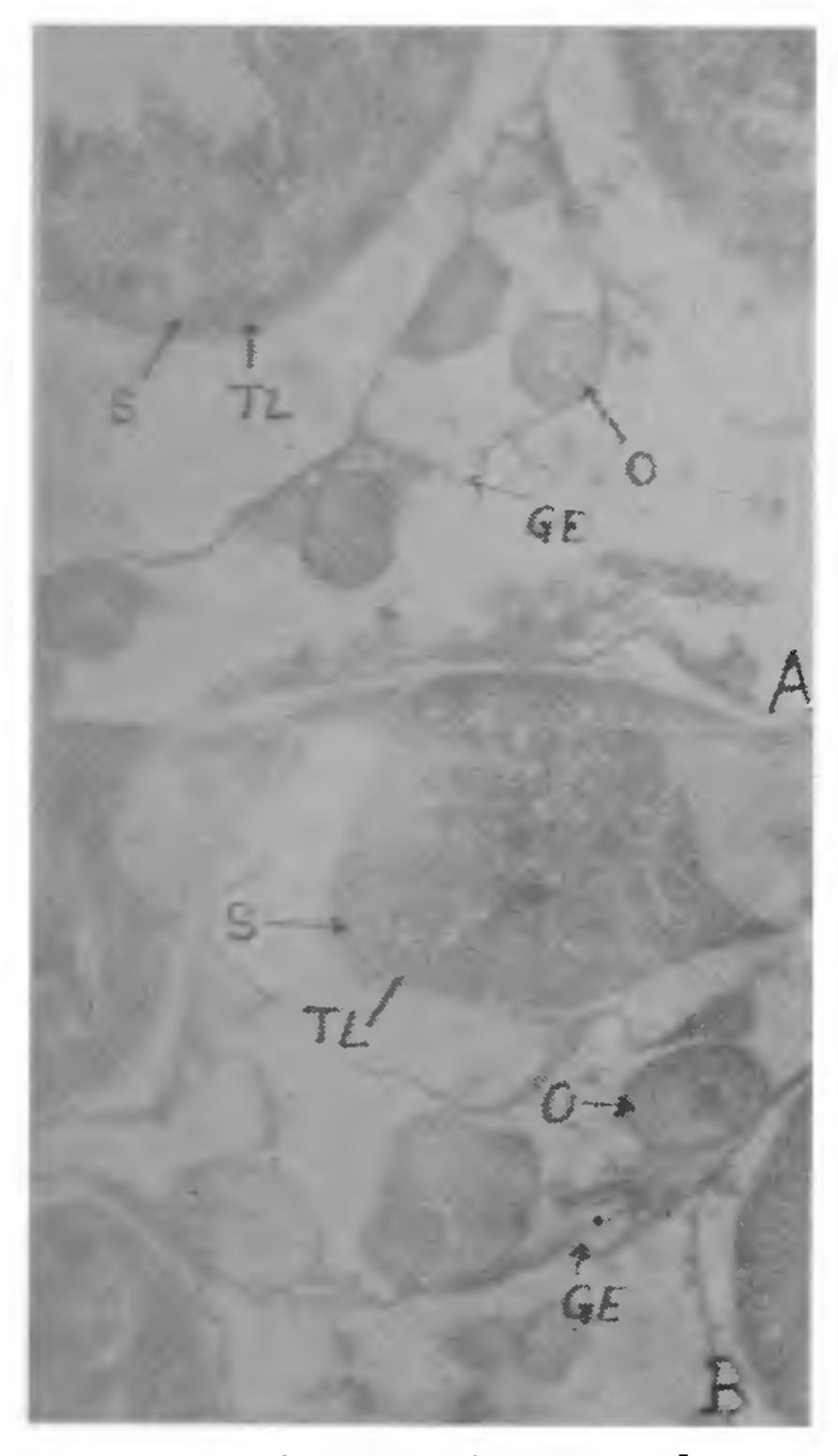


Fig. 2 A-B. Sections of the gonad of the examined *Clarias batrachus*. A. Showing scattered ova in between testicular tissues. B. Showing maturing occytes arising from the germinal epithelium.

(TL, Testicular lobule, O, Ovum; GE, Germinal epithelium; S, Spermatogonia).

The somatic substratum of the gonad has its origin from the peritoneal wall (D'Ancona)<sup>10</sup>. In the course of development, the somatic substratum of the gonads gets differentiated either into male or female under the influence of inducing factors/morphogenetic operators. The development of abnormal gonad reported in the present paper, may be as a sequel to the simultaneous effects of both male and female determining factors.

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### PARASITIZATION OF ISOPARORCHIS INVPSELOBAGRI BILLET IN CHANNA PUNCTATUS BLOCH

A REVIEW of the literature reveals that Isoparorchis hypselobagis Billet (Order, Prosostomata; Family, Isoparorchidae; Genus, Isoparorchis, Species, I. hypselobagii) usually infests the swim bladder of siluroid fishes. However, a few reports are available about its occurrence in nonsiluroid species.

These reports too, have recorded only the juvenile forms of this parasite in the nonsiluroid fishes, resulting in what is commonly known as 'ink spot disease' due to the presence of metacercaria stage.

The present investigation reports the widespread infestation by this parasite in fully developed sexually adult state in Channa punctatus Bloch, a non-siluroid fish of the order ophiocephaliformes from water bodies around Jaipur. The adult parasites found infesting C. punctaus during this investigation varied in size from 14 to 20 mm in length and 8 to 12 mm in width (c.f. 2.01 mm × 1.05 mm reported by Rai and Pande<sup>6</sup>). It is also interesting to record that the average length of the ovarian tube alone was 2.48 mm which exceeds the total length of the parasite itself reported by Rai and Pande<sup>6</sup>.

In a single fish usually two to five adult parasites were found with a number of juveniles showing exceptional variation in number and size. The degree of infestation can be realized from the fact that these parasites weighed 3.13% of the host's body weight. Further the adult parasites were not limited to the swim bladder alone but attacked almost every visceral organ such as liver, spleen, ovary, body musculature, intestinal muscles and mesentery.

Detailed studies on the effect of parasitization on the physiology of the fish per se and its possible effect as food for man are being reported elsewhere.

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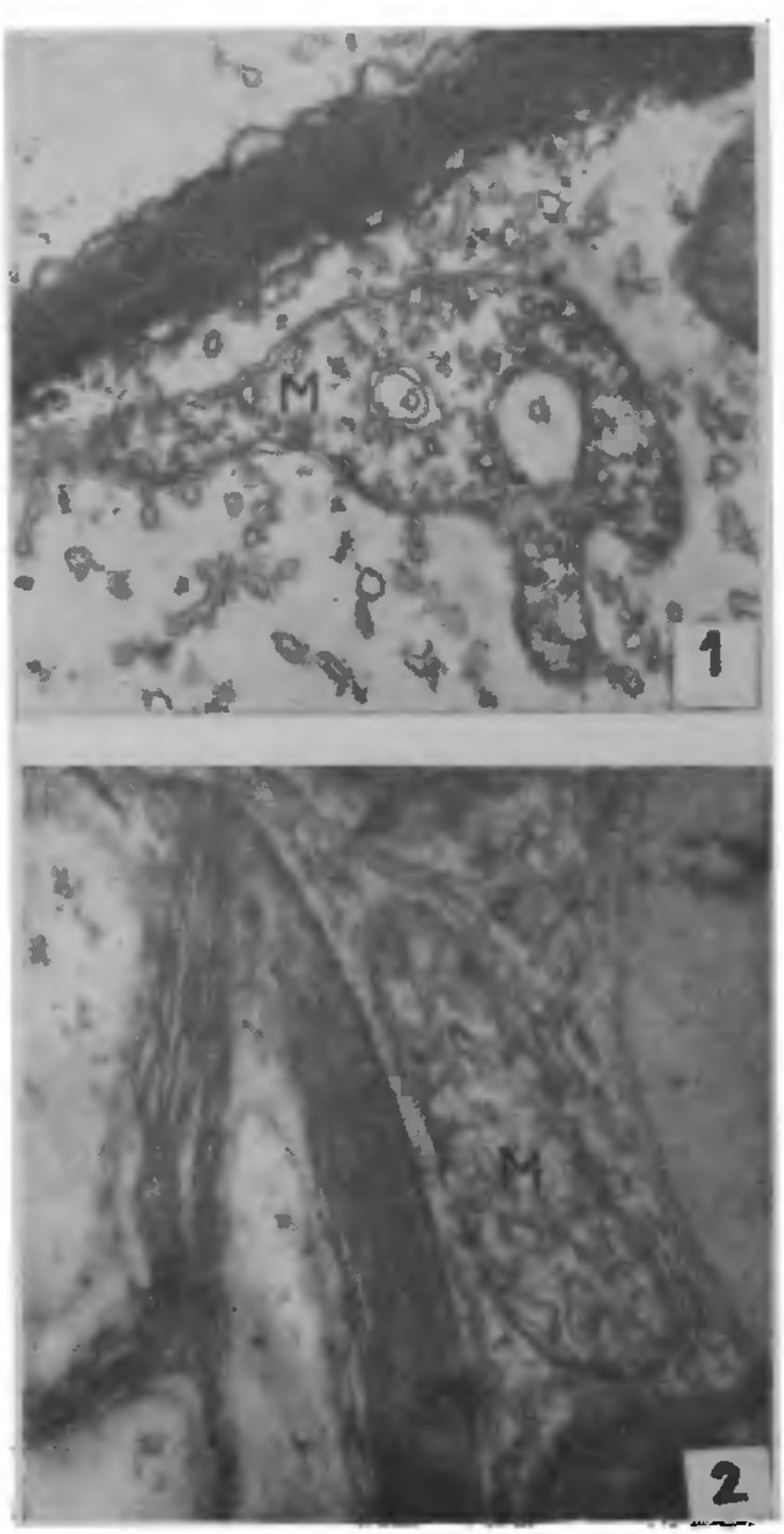
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### OCCURRENCE OF GIANT MITOCHONDRIA IN THE CELLS OF GAMETOPHYTES OF LYGODIUM FLEXUOSUM (L.) SW. EXPOSED TO GAMMA RADIATION

SPORES of L. flexuosum were sown in 2 separate petridishes, each having approximately 500 spores in 1% Knop's solution. Various stages of gametophrees of L. flexuosum, viz., ungerminated spores, 2 to 3 days old and one month old were exposed to doses of gamma

radiation from cobalt source ranging from 8,000 rad to 1,20,000 rad. The radiated gametophytes as well as controls were kept in cultures at 26±2° C with diffused daylight augmented with 600 ft.c. light for 8 hours.

It was observed that irradiated plants show delay in germination, slowing of cell division and other abnormalities in growth patterns. Gametophytes with interesting abnormalities at sublethal doses were chosen for ultrastructural studies. Such gametophytes were fixed in Caulfield's fixative and were kept at 4° C for 4 hours, then they were processed for ultrathin sections by ultratome. Finally, sections were studied in Hitachi H.U.—11 E and photographed py transmission electron microscope.



FIGS. 1-2. Fig. 1. Micrograph of a portion of the cell of gamma irradiated gametophyte of Lygodium flexuosum showing giant (or complex) mitochondrion (M) (× 32,000). Fig. 2. A portion of another cell of gamma irradiated gametophyte of L. flexuosum showing another giant mitochondrion (M) (× 1,920).

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The cytological effects observed included many alterations as well as damages in various organelle such as mitochondria, chloroplasts, endoplasmic reticula ribosomes etc. Their membrane system was severely disturbed. The most interesting findings are the giant mitochondria with changed configuration of cristae which were observed in several cells of many gamma irradiated gametophytes.

The giant mitochondria (Figs. 1,2) have irregular shape, they have swollen and irregularly arranged cristae and are many times larger than the normal ones. Such giant mitochondria are probably formed by the fusion of several mitochondria or they may be proceeding towards fission and final disintegration.

Underbrink et al.<sup>2</sup> while studying the effects of X and gamma radiations on a green alga Brachiomonas noted that the mitochondria of this alga were somewhat swollen and their cristae disturbed. But the giant mitochondria of the kind produced as a result of itradiation observed by us have not been reported in plant material.

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### AN AIR-DRY METHOD FOR CYTOLOGICAL PREPARATIONS OF APHIDS (HOMOPTERA: APHIDIDAE)

UNTIL the middle of the present century, most studies of aphid chromosomes were made from sectioned material. Earlier, Smith! used the acetocarmine technique for squash preparations. Colling<sup>2</sup> reported on a squash method using osmid acid fixative for the somatic chromosomes that apparently gives satisfactory results. Other squash techniques using acetocarmine, acetoorcein, Feulgen and Gomori's hematoxylin do not give satisfactory results. Dionne and Spicer<sup>3</sup> gave another squash method using the fixative acetiq alcohol and chloroform in place of osmic acid. They hydrolyzed the material with normal hydro-

chloric acid and then stained it with Gomori's hematoxylin. MacDonald and Harper<sup>4</sup> and Sun and Robinson<sup>5</sup> used an improved squash technique using Feulgen stain for aphid chromosomes. Sun<sup>6</sup> made further improvements in the squash method for the study of somatic chromosomes of aphids. Robinson and Chen<sup>7</sup>'8 adopted the same method for squash preparations as of Dionne and Spicer<sup>3</sup> and MacDonald and Harper<sup>4</sup> in more or less a similar way except using Feulgen stain in place of Gomori's hematoxylin. Blackman<sup>9</sup>'<sup>10</sup> also used a rapid Feulgen squash technique<sup>4</sup>. For the first time Blackman<sup>11</sup> used Giemsa stain for the dytogenetical studies of aphids after squashing the material.

The present method is based on the air or heat drying and tapping technique which takes less time and gives better spread of the chromosomes than by other earlier squash methods.

Best results can be obtained by using embryos either from apterous viviparous females or the fourth instar nymphs. In India it is easier to find cells in late prophase and metaphase stages during the winter when the range of temperature is 15°-20° C. At this time the growth and development are most rapid and fat bodies scanty. An aphid (of the above mentioned morph) is taken on a clean slide in a drop of 0.5% sodium citrate solution. Under a binocular microscope, the terminal part of abdomen is snapped so that the gut and reproductive organs come out leaving the rest of the body behind. Fat bodies can be removed by adding another drop of 0.5% sodium citrate solution and shaking the material with a needle. This will also spread the embryos can now be removed to a cavity block having 1-3 ml of the sodium citrate solution in which the material may be kept for half an hour. The solution being hypotonic, the treatment will swell the cells. Individual ovarioles can now be separated, from which very young embryos can be sorted out as they are distinguishable from the more advanced ones having noticeably developed eye pigmentation. The advanced stage embryos are not suitable for dytological preparations. The ovarioles along with germarium are transferred to the fixative consisting of 3:1: methanol: Glacial adetic acid. Leave them in fixative for 15-30 min with a maximum period of one hour. The fixed embryos (3 to 4 to be taken) are now transferred to a drop of 60% acetic acid on a microscope slide (already boiled in chromic acid, washed and stored in 90% ethyl alcohol). The embryos are crushed by gently tapping for 30-50 seconds so as to disperse the cells in the drop using the flat end of a metal rod (10 cm long and 0.3 cm dia). The cells of the embryos get separated and float free in

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the drop so that a cell suspension is formed. The cell suspension is dispersed over a slide by adding a few drops of acene methanol. After this, the separated cells can be dried in air at room temperature or hear-dried on a warm (not hot) brass plate by moving the drop at intervals to a new site on the slide until the liquid has evaporated.

The dried slide can be examined under phase contrast microscope to ensure that different stages of cell in divisions are there. These slides may be stored dry vertically in a dust proof box. It keeps well up to a few months. The slides may however be stained immediately and made permanent by the following method:

Place the slide in 0.1 M Sorenson's phosphate huffer of PH 6.8 for 5 min. Prepare 5% Giemsa stain in 6.8 pH phosphate buffer from Giemsa stock stain which should be thoroughly shaken for five min before mixing. The diluted Giemsa stain should also be thoroughly mixed for 5 min. It should be emphasized that the addition of Giemsa to the buffer does not alter the pH in the system. Only freshly diluted and well shaken Giemsa stain must be used for separate set of slides because the stain is precipitated after about half an hour when it becomes effete. Place the slide horizontally on a watch glass and put 5% freshly prepared Giemsa stain covering the area of spread material on to the slide. Stain for 10-30 min. Fifteen min staining

gives best results by using 5% Giemsa in phosphate buffer of pH 6.8. Differentiate the slide by a dip in phosphate buffer of pH 6.8. Wash the slide with distilled water and dry in air at room temperature by placing it vertically so as to avoid the patches of water drops. Dry the slide overnight in a dry place free from dust. Immerse in xylene for 5 min. Mount in D.P.X. using rectangular coverslips. Dry in oven at 60° C for nearly 24 hr.

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### AWARD OF RESEARCH DEGREES

Kakatiya University, Warangal, has awarded the Ph.D. degree in Chemistry to Sri. D. Nageswara Rao; Ph.D. degree in Mechanical Engineering to Sri. P. Narasimha Reddy.

Karnatak University, Dharwad, has awarded the Ph.D. degree in Botany to Sri. S. B. Dandin; Ph.D. degree in Chemistry to Sri. M. B. Adi and to Smt. S. S. Sangapure.

Sri. Venkateswara University, Tirupati, has awarded Ph.D. degree in Physics to Sri. B. Srinivasulu Naidu.

Utkal University, Bhubaneswar, has awarded Ph.D. degree in Anthropology to Sri. S. K. Ghosh Maulik; Ph.D. degree in Botany to Sri. K. V. Janardhan, Sti. Brnayak Das, Sri. Rabindranath Padhy and Sri. J. Veeraraghavan.

University of Cochin has awarded the Ph.D. degree in Science to Sri. K. T. Mathew; Ph.D. degree in Marine Science to Sri. Kuruvilla Mathew.