

ON THE REPRODUCTIVE PROCESSES AND DEVELOPMENT OF *PILA GLOBOSA* (SWAINSON).

PART I.—COPULATION AND OVIPOSITION.

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(Plates I, II.)

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1. INTRODUCTION.

In a recent monograph on the "Anatomy of the common Indian Apple-Snail *Pila globosa*, Prashad (4, 1925) writes, "No observations have hitherto been published regarding the copulation or oviposition of any Ampullarid and I have not been able to observe these phenomena myself." He, however, quotes the following paragraph from a manuscript of Mr. V. V. Ramanan preserved in the Connemara Library at Madras :—

"The *Ampullaria globosa* (*Pila virens*) Swainson invariably goes to the banks of the pond, puddle or river in which she lives for the purpose of spawning. She selects a place a few inches above the water margin, burrows underground till her shell is just visible above the mud and deposits her ova. So to speak, she incubates, after gathering the bunch of eggs within the folds of her mantle and foot and till the rather soft and transparent jelly-like substance of her spawn turns in a few hours considerably thicker, and each egg becomes invested with a pure white calcareous shell. After the operation is over, she returns to the water."

This account of oviposition is unfortunately very incomplete as well as incorrect in several details, while no published account exists of the copulation of an Ampullarid.

The embryology of this type has also been incompletely worked out. Carl Semper's work (7, 1862) on the embryology of an allied species, *Pila polita*, was published in 1862 and although, considering the fact that the work was carried out sixty-six years ago, it is an admirable piece of research, yet it is inadequate and out of date for our present day requirements, while the illustrations leave a great deal to be desired. Later, Brooks and McGlone

(1, 1908) worked out the origin of the lung of *Ampullaria depressa* but unfortunately their work is very fragmentary and needs amplification.

Since *Pila* is now studied as a type of the Gastropoda in almost all the universities of India, Burma and Ceylon, it is necessary to have as complete and correct a knowledge as possible of this form. I, therefore, undertook to study its breeding habits and development and this paper is the result of my observations and work both in the field and in the laboratory extending over a period of about a year.

I am describing copulation and oviposition in this first part of my paper and will publish the embryology of this type in part II at a subsequent date.

I am deeply indebted to my friend Dr. Bains Prashad for acquainting me with the literature on the subject and sending me reprints of several memoirs from his private library. He has also kindly read through my manuscript and made helpful criticism.

2. BREEDING SEASON AND THE INFLUENCE OF WARMTH AND MOISTURE.

The Apple-snail, *Pila globosa*, breeds at the beginning of the rainy season. In a hot and dry country like India, it is *moisture* that makes the greatest difference in the life of aquatic and semi-aquatic animals, and *Pila*, like many other typical Indian forms, is adapted for life in surroundings where there is a prolonged period of drought alternating with three to four months of heavy rains. The monsoon in Northern India blows, off and on, for about four months, *i.e.*, June, July, August, and September, and it is only in these months that ponds and puddles are formed in large numbers. In the upper Gangetic valley October to June are practically dry, there being very little rain during these nine months. Of these, May and June are the hottest and driest, and the hot winds (the *loo*) blow during these two months for the greater part of the day; the land gets absolutely parched and baked, as it were, and the temperature goes up to between 110°F. and 118°F. in the shade. In Lucknow, the moisture-bearing current breaks in towards the end of June or the beginning of July and brings life to the parched land. The first few inches of rain are rapidly absorbed by the dry ground, but later, as the monsoon continues, innumerable ponds and puddles are formed all over the country-side and animal life shows its activity at its best.

Pila globosa is found in large numbers in the ponds during the monsoon and in bigger ponds hundreds of specimens can be obtained right up to December. But as the ponds dry up, the animal burrows underground and I have dug up specimens of *Pila* as deep as four feet under the ground in March and April. Whenever specimens are needed for class-work in March and April, they are always obtained by digging the ground in places where ponds and puddles are formed during the rains. This aestivation or summer-hibernation of *Pila* is an adaptation not to *temperature* but to *drought*.

Warmth and moisture are the two essential conditions for the development of the eggs of this animal and it is only during the monsoon that these conditions are fulfilled and hence breeding takes place at this time of the year. In Europe the land is moist during the greater part of the year and there is a good deal of humidity in the air at all times, but it is the warmth that is absent in autumn and winter and hence animals hibernate during these months to tide over the intense cold of the winter months. In Northern India and also in other parts

of the Indian Empire, on the other hand, it is warm during the greater part of the year but practically eight to nine months are dry and there is little moisture either in the air or on land. All ponds and puddles, teeming with life during the monsoon, dry up during the succeeding months to be filled up again with water and accompanying animal and vegetable life at the onset of the next monsoon. In the tropics, therefore, it is the *moisture* that largely determines the conditions of animal life and it is not at all surprising, therefore, that animals ordinarily passing a certain period of their life in water (fresh-water or amphibious animals) make the most of the monsoon months and breed at this time of the year, when both moisture and warmth are available.

Very little is known of the physiological condition of these animals at the time they aestivate in hot and dry seasons.

3. THE COPULATORY ORGANS AND THE PROCESS OF COPULATION.

Immediately at the onset of the rains, the apple-snail comes to the surface of the ground after its prolonged aestivation and sexual activity begins at once. Copulation takes place either in water or on land at the edge of the pond. The male which is, as a rule, smaller than the female and has the body-whorl of its shell less swollen, creeps up on the top of the shell of the female and takes up its position on the right side of the head of the female. In this position, the foot of the male is firmly attached to the outer side of the shell of the female and the two shells are set in such a way that the aperture of one is opposed to that of the other (Pl. I, fig. 1). The female has her foot and head generally fully extended and protruded out of the shell, so that her mantle-cavity is wide open and the left trumpet-shaped siphon is kept constantly working. While the greater part of the foot of the male rests ~~on~~ the body-whorl of the shell of the female, a small anterior portion of the foot extends beyond the margin of the shell and the male thus takes a secure position on the shell of the female. The pair having been set in this position, the male thrusts out its white or light blue penis and inserts it into the mantle-cavity of the female.

Apple-snails readily copulate in an aquarium in the laboratory, either floating in the water (Pl. I, fig. 1) or attached to the glass-side of the aquarium (Pl. I, fig. 2). My observations on copulation are based mainly on snails pairing either in the aquarium or on the muddy ground in the frog-pond of my laboratory. The female creeps along the glass-side of the aquarium and comes up near the surface of the water, while the male mounts on the female, takes up its proper position and inserts its penis-sheath and penis into the mantle-cavity of the female. The whole process can be easily observed through the glass-side of the aquarium and Pl. I, fig. 2 shows the copulating snails attached to the side of the aquarium. Pairing specimens either floating in water or lying on the ground can be picked up and their copulatory mechanism examined. The mouth of the shell of the female is easily kept open by pressing the operculum against the shell with the thumb of the left hand and, by doing so, the white or light blue penis-sheath is easily seen inserted into the mantle-cavity of the female. Further, by moving aside the foot of the female with a section-lifter in the right hand, it is seen that the inserted penis-sheath is a flattened structure measuring about half an inch in width. It is curved into an arch and is inserted

more than an inch into the mantle-cavity of the female. There the rectal opening is readily seen, while the female generative opening, lying close to it, is completely surrounded by the penis-sheath. The penis itself is not visible in copulating specimens, as it is completely covered on the underside by the penis-sheath. A fact of practical interest, however, is that the male continues to stick to the female and does not usually retract its penis-sheath and penis when pairing specimens are handled during the copulatory act. In fact, all the details mentioned above can be easily observed by holding the copulating pair in the left hand and manipulating the different structures with a section-lifter in the right hand.

During copulation, which lasts for several hours (I have seen pairs copulating for three hours), the female, with its head and foot generally extended, feeds freely on water-weeds in the pond or the aquarium, but the male keeps its head generally retracted and puts out only its penis-sheath and inserts it into the mantle-cavity of the female.

While the penis-sheath can be seen enveloping the female generative aperture, the rôle of the penis in the process and the exact mode of transmission of the spermatic fluid from the male generative aperture to the female opening cannot be made out by a direct observation of the copulating pairs, and consequently I resorted to other methods of research. I tried several methods. The first was to break portions of the shell of several males in order to expose the penis-sheath and keep such males with females in an aquarium. The shells of the females were also broken so as to observe more easily the terminal portion of the vagina and the female generative aperture. I expected to see the exact mode of transference of the seminal fluid from the male genital aperture to the base of the penis and also the insertion of the penis into the vagina. But unfortunately my expectations were not fulfilled as the snails with partially broken shells refused to copulate, probably because the males could not secure a good position on the female, the shell of the latter having been broken at the place where the male takes his seat.

Attempts were then made to kill and fix rapidly copulating pairs in sexual congress and these were partially successful. I dropped several copulating pairs in strong formalin in order to fix them at once but they always contracted and separated. I next tried to narcotise several pairs with chloroform and chloral hydrate respectively, but the male and the female invariably separated and no observations on the mechanism of copulation could be made. The last method I tried was electrocution of copulating pairs. This quick method was successful and I was able to kill a couple of pairs in a condition of sexual congress and thereby make out the exact details of the process of copulation. One such electrocuted pair is shown in Pl. I, fig. 4, and with the help of this figure we can follow the condition of the copulating organs in pairing specimens and the mechanism of copulation.

The structures of the male animal directly concerned in copulation are (1) the terminal papilla of the vas deferens bearing the male generative aperture, (2) the penis-sheath, (3) the hypobranchial gland, and (4) the penis. In the female, on the other hand, the generative aperture, together with the terminal portion of the vagina, are the only structures that take part in copulation. Prashad (4, 1925) has described these structures in non-copulating specimens, and it is interesting to note that, at the time of copulation, the disposition of the structures and their proportions are markedly different from what they are in a non-copulating individual.

Figs. 5 and 4 on Pl. I show these structures in non-copulating and copulating male specimens respectively. Fig. 5 is taken from Prashad's paper (4, 1925) and fig. 4 is drawn from copulating specimens killed by electrocution and fixed in formalin.

The last part of the vas deferens terminates in a prominent structure, the genital papilla, lying close to the rectum. The genital papilla bears the male generative aperture at its free end and consists of two parts,—(1) the basal part, which is opaque white in colour and has thick walls and (2) the apical part, which is thin and transparent and consists of two membranous folds closely pressed together. It is the apical part which opens into the mantle-cavity through the male generative aperture. The whole papilla (Pl. I, fig. 4) bears a strong resemblance to the claw of the Carnivora : the basal thick-walled portion resembling the bony lamina around the base of the claw and the apical part resembling the horny claw itself.

This genital papilla is a very important structure as it serves to establish a communication between the male generative aperture and the base of the penis, thereby conveying the spermatic fluid from the male genital aperture to the penis. Prashad (4, 1925) just mentions this papilla :—“ The terminal glandular part of the vas deferens runs along the rectum to terminate in a small papilla, about a quarter of an inch behind the anus. The papilla is free and lies over the rectum.” But he gives no details of this structure and does not even show it in his diagram (Fig. 15, 4, 1925). Evidently he could not realise its importance in the act of copulation. Fig. 6 on Pl. II shows a transverse section passing through the basal part of the genital papilla and the rectum, and it is clear from this diagram that the genital papilla is not free but is closely attached to the rectum and its connective tissue envelops the latter structure. The thick connective tissue surrounding the vas deferens in the genital papilla consists of a close net-work, the meshes of which are formed of blood-spaces and blood-vessels, while the trabeculae of connective tissue contain elastic fibres and bundles of plain muscle-fibres. The vas deferens itself is a thin-walled tube with an inner lining of ciliated epithelium but it is surrounded by a thick coat of connective tissue, which reminds one of the spongy and erectile tissue of the mammalian penis. In fact, the genital papilla is a kind of miniature second penis.

The penis-sheath and penis proper are developed as outgrowths of the mantle and in the adult *Pila* are fixed to the inner surface of the mantle-edge, being absolutely independent of the male generative aperture. This fact characterises the whole family of Ampullariidae and must be emphasised here. In fact in *Pila*, as in marine Prosobranchiate Gastropods, the male generative aperture is *not* in direct continuation with the penis but opens separately into the mantle-cavity. The question, therefore, in *Pila*, as in marine Prosobranchiate Gastropods, is, “ How is the spermatic fluid conveyed from the male generative aperture to the base of the penis ? ” In marine Prosobranchiates it has been ascertained that there is a ciliated canal running from the genital opening to the base of the penis along the right side of the body and this canal serves to connect the male genital opening with the penis (3, 1927). But in *Pila* such a ciliated canal is absent and the transference of the spermatic fluid from the male pore to the base of the penis takes place in the absence of an intervening canal or channel. A careful examination of specimens, fixed at the time of copulation, reveals the fact that the transference of spermatic fluid in *Pila* is effected by an adjustment of the male generative opening and the base of the penis, in such a way that the apical portion of

the genital papilla is directed towards and is received into a pit formed at the root of the penis, between the hypobranchial gland and the penis-sheath (Pl. I, fig. 4). We have already seen that the genital papilla consists in large part of erectile tissue and that its apical part crosses over the rectum to approximate the base of the penis. The attachment of the penis-sheath and penis to the inner surface of the mantle brings their basal ends in close apposition with the male generative aperture. In fact, a mid-dorsal incision of the mantle-fold of a snail fixed at the time of copulation leaves no doubt whatever as to the close approximation of the apical end of the genital papilla with the base of the penis during copulation. The genital papilla, or the miniature second penis, therefore, serves to transfer the sperms from the generative aperture to the base of the penis proper, and from there the spermatic fluid is conveyed along the deep groove of the rod-like penis to the female generative aperture. The penis itself is a comparatively long and stout structure measuring about 3.8 cm. or one and a half inches in length in a copulating specimen. The relative length of the penis in copulating and non-copulating specimens can be judged from the fact that Prashad gives the length of the penis as *half an inch* only. The penis extends right up to the vaginal aperture and its terminal end is thrust into the vagina. In my electrocuted pairs, the base of the penis with the hypobranchial gland and the proximal portion of the penis-sheath are contained in the male, while the distal portions of the penis-sheath and the penis lie in the mantle-cavity of the female and the free end of the penis is inserted into the vagina (Pl. I, fig. 4). The penis-sheath is shaped like an elongated spout-like structure and envelopes the penis along its whole length. There is no doubt that it acts as a protective covering for the penis and may at the same time prevent waste of the spermatic fluid.

In a copulating male, in which the penis-sheath and the penis are fully extended, the hypobranchial gland forms the root of the penis. Its histological structure, as described by Prashad (4, 1925), lends colour to the view that its secretion is poured directly on the surface. What the nature of the secretion is and what function it subserves in the reproductive processes are questions still unsolved.

Lastly, we may refer to the question of the possible advantage of having the male generative aperture independent of the penis-sheath and the penis. The only alternative situation for the penis-sheath and penis would be in continuation with the vas deferens and the genital gland. In this position, they would undoubtedly be an obstruction in the mantle-cavity at all times and would not receive an adequate supply of blood for the efficient discharge of their function. The origin of the penis from the inner surface of the mantle close to the so-called lung or pulmonary chamber ensures a copious supply of blood for its erection, and at the same time leaves the mantle-cavity relatively free to accommodate the head and foot when the animal retracts itself into the shell and closes it with its operculum.

4. OVIPOSITION.

Although *Pila* is a member of a completely amphibious clan, it shows an advance towards a terrestrial habit in its breeding activities. The typical amphibia almost always lay their eggs in water, while *Pila globosa* invariably lays her eggs on land¹ and never in water.

¹ Another species of *Pila*, *Ampullaria (Pila) depressa*, studied by Carl Semper (7, 1862) deposits her eggs in vertical rows on the stems of tall reeds and grasses above the surface of the water but close to it.

The female *Pila* selects a place in the bank of a pond a few inches to a few feet above the water margin, where she makes a hollow¹ in the ground to deposit her eggs. As a rule, egg-masses are deposited in sheltered places or hollows not only to protect them from enemies like rats and squirrels but also to protect them from the sun and consequent desiccation. The shell of the eggs is porous and unless evaporation is avoided by keeping the eggs sufficiently damp, the albumen inside dries up and the embryo cannot develop.

Oviposition takes place in a day or two after copulation, but it can be delayed by preventing the snails, which have copulated and have been fertilized, from going on land and keeping them instead in water in an aquarium in the laboratory. As soon as they are taken out of water and placed on land they commence oviposition. After keeping several fertilized females in water in a glass aquarium for a week, I took them out and placed them on moist ground in the frog-pond with the result that there was, so to speak, an "epidemic" of oviposition since all of them had been waiting to lay eggs.

During oviposition, the foot is thrown into a number of folds so as to discharge efficiently the function of depositing and collecting the eggs. In fact, it plays such an important part in the whole process of oviposition that it may rightly be termed the ovipositor² of the water-snail.

At the time of egg-laying, the greater part of the body of the female is extended out of the shell. The foot is fully protruded out, while the head and tentacles and also the left siphon are moderately extended. The base of the foot, instead of lying flat on the ground as in locomotion, contracts a little all round the edge forming a wavy border, and is arched in such a way as to form a saucer or dome-shaped structure. The dome-shaped foot receives the eggs as they are laid, works up the eggs into a mass and continues to grip the mass firmly during the whole period of oviposition. Further, the right side of the foot is deeply folded at two adjacent places in such a way that one of the two folds—the posterior one—forms a temporary obliquely vertical tube. The upper opening of this tube (Pl. II, fig. 9) lies on the outer sloping surface of the foot just outside the shell-opening, while the lower end opens out near the centre of the saucer-shaped sole of the foot. As shown in fig. 9, this temporary tube is really a deep channel formed by folding a part of the outer surface and edge of the foot. Approximately at an interval of a minute, a soft, flaccid rounded egg comes out of the vaginal opening, rolls out of the shell and passes along the outer surface of the foot into the upper end of this temporary tube. On its entrance into the tube the egg disappears from view for a few seconds until it comes out of the lower end of the tube into the saucer of the sole of the foot. It creeps or rolls along the surface of the sole of the foot and is readily placed along with the other eggs by the muscular contractions of the foot. As the egg comes out of the vagina, a peristaltic wave, no doubt due to muscular contraction of the foot, travels all the way from near the vaginal opening along the right side of the foot to the upper opening of the temporary tube, and from there through

¹ In the frog-pond in my laboratory, several snails laid their eggs on the moist ground without making a hollow at all, while others made shallow pits and deposited their eggs in them.

² The term *ovipositor* in the case of insects is used for "three pairs of chitinous styles at the end of the oviduct which form a strong, powerful apparatus for boring into the ground or into leaves, stems of plants, the bodies of insects or even into solid wood, so that the eggs may be deposited in a place of safety." In *Pila* the foot discharges a similar function; it collects the eggs, works them up into an egg-mass, holds the egg-mass until all the eggs are laid and then deposits the egg-mass into a hollow in the ground—a place of safety. Hence I have applied the term *ovipositor* to the foot.

the tube to the sole of the foot. It is by means of this wave of contraction that the egg is rolled along all the way from the female generative aperture to the sole of the foot.

It will thus be seen that the foot on its right-hand side forms a temporary tube for the conveyance of the eggs from the female generative aperture to the sole of the foot; the sole of the foot rolls the eggs together to form the egg-mass and holds this mass until all the eggs are laid; and that the whole process of oviposition is accomplished by means of the muscular contraction of the foot. The foot, therefore, efficiently discharges the function of oviposition and may justly be called the ovipositor of the water-snail *Pila*.

The body of the animal during this process is very much relaxed and specimens can be handled without their showing the usual movements of contraction. I handled several specimens which were laying eggs and found that none of them would relax the hold on her eggs with her extended foot. The instinct of self-preservation, so evident at other times, had now given place to the motherly instinct of egg-preservation and hence to race-preservation. Since the egg-laying specimens allow themselves to be handled, it was possible to make out the details of the whole process by direct observation.

Egg-laying goes on for several hours and usually takes place in the morning. In the laboratory frog-pond I invariably found snails laying eggs between seven and eight in the morning and they continued to lay for 4 to 5 hours.

As the egg is laid, its covering is very soft and sticky and it is due to this adhesive character of the outer covering that so many eggs are glued together to form the characteristic egg-mass of each snail (Pl. II, figs. 7 & 8).

Ramanan's observation that "the female, so to speak, incubates, after gathering the bunch of eggs within the folds of her *mantle* and *foot*" is, I am afraid, incorrect. In the first place, it is only the dome formed by the sole of the foot that envelopes the eggs and the *mantle* takes no part either in gathering the eggs or, as Ramanan supposed, in incubating them. In the second place, there is no incubation of eggs at all by the snail. For the purpose of development, moisture and warmth are the two requisites and these are admirably fulfilled by the weather during the monsoon months and no process of incubation on the part of the mother snail is necessary. As egg-laying extends over a period of 4 to 5 hours, it is necessary for the snail during this period to keep enveloping the eggs as they are being laid. As each egg is laid it is added to the previous mass, until the snail has laid all its eggs and the mass of eggs is arranged as compactly as possible. When egg-laying is completed, the snail leaves the egg-mass in a sheltered hollow and resorts to the water. There is no doubt that Ramanan was led to believe in the incubation of the eggs by the fact of the prolonged association of the foot with the eggs he observed. In fact, the foot is attached to the egg-mass for kneading together and adding new eggs as they are being laid to the pre-existing egg-mass and not for the purpose of incubation.

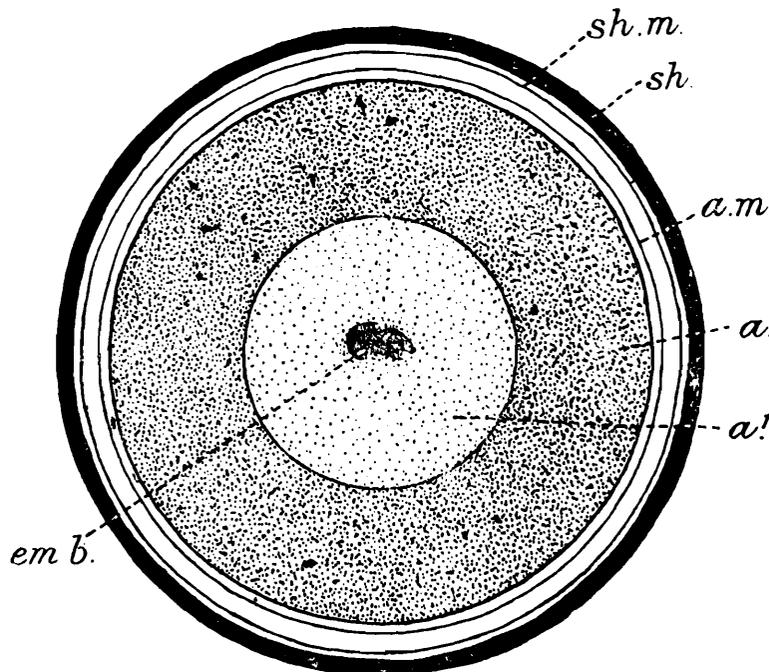
5. THE EGG.

Pila globosa, as Prashad writes (4, 1925), lays its eggs in masses in which the eggs are firmly attached to one another and together form a group of pure white calcareous shells. The egg-masses vary in size and weight and also in the number of eggs they contain. Prashad's estimate of the number of eggs in each mass as being two to three hundred is a very

modest one. The smaller egg-masses, weighing from ten to twenty grams each, do contain two to three hundred eggs, but the larger egg-masses, which one frequently comes across, weigh as much as 36 grams and contain as many as 700 to 800 eggs. Of course, egg-masses vary in size and the number of eggs they contain forms a continuous intermediate series between these two extremes.

In form, each egg is rounded but is slightly flattened at its points of contact with other eggs in the egg-mass. There is a white calcareous shell protecting each egg and forming a thin but opaque covering for the delicate structures within. The shell is lined internally with a very thin transparent shell-membrane, which is closely attached to the shell, and generally comes off in shreds along with pieces of the shell, when the egg is opened. On the removal of the shell, a milk-white sphere formed of thick solid albumen comes out of the egg. This albuminous sphere closely fills up the whole space inside the shell and comes out entire on opening the egg. There is a thin membrane which closely invests this albuminous sphere, and since this membrane lies immediately inside the shell-membrane, we shall distinguish it from the latter by calling it the albumen-membrane. It is difficult to make out this second inner membrane in a fresh egg, but in a boiled egg it can be easily detached from the underlying sphere of albumen. We may note here that while both the shell-membrane and the albumen-membrane are thin and transparent, the sphere of albumen inside is opaque.

On opening the albumen-sphere, it is found that the sphere is a hollow structure and that its cavity inside is filled with a transparent fluid also albuminous in nature. The impregnated ovum or oosperm lies floating in this transparent fluid. In cutting open an



TEXT-FIG. 1.—Diagrammatic representation of a section through an egg of *Pila globosa* showing a three days' embryo. *a.*, thick solid albumen; *a¹*, fluid albumen; *a. m.*, albumen membrane; *emb.*, embryo; *sh.*, calcareous shell; *sh. m.*, shell membrane. ($\times 16$.)

egg, the shell is removed first of all and then the sphere of solid albumen is opened by an incision with a needle. The fluid albumen filling up the cavity of the sphere flows out carrying the embryo with it. The embryo in its early stages is yellowish in colour on account of

the yolk-granules present in it, but as development proceeds the embryo becomes white and translucent.

The egg (Text-fig. 1) is about 5 mm. in diameter and the albumen sphere about 4 mm. in diameter. The wall of the albumen-sphere is a little more than 1 mm. in thickness.

6. MATERIAL AND TECHNIQUE.

In order to examine the details of the process of copulation, several male and female specimens were collected from a pond and brought to the laboratory. They were kept in an aquarium where they copulated either floating in water or attached to the glass side of the aquarium. Details of copulation were observed and recorded and photographs were taken of the copulating pairs *in situ*. Several pairs found copulating in a pond were separated and kept in the aquarium. Since their copulation had been interrupted, they copulated again in the aquarium.

Pairs *in coitus* were successfully killed by electrocution on a metal plate with the help of a battery and a small induction coil. They were fixed in 8 per cent. formalin, and after breaking the shells, the disposition of their copulating organs was examined. The genital gland and the attached portion of the rectum were dissected out and sectioned in order to make out their histological structure.

Impregnated females were kept on muddy ground in the frog-pond of the laboratory where they laid their eggs. They were taken out along with their masses of eggs and were photographed in the act of oviposition. The rôle of the foot in the process of oviposition was carefully determined by observing several females laying eggs. Egg-masses of several specimens were collected and weighed and the number of eggs counted in a few selected masses.

7. SUMMARY.

(1) After a prolonged period of aestivation during the dry months, *Pila* comes out at the onset of the rains and begins breeding at once.

(2) Copulation takes place in the water or on the ground at the edge of a pond. It lasts for three hours or more.

(3) The penis being a structure independent of the male generative aperture, the genital-gland acts as a miniature second penis and effects the transference of the spermatid fluid from the male pore to the base of the penis proper. The ciliated canal, running from the male generative opening to the penis in marine Prosobranchiates, is absent in *Pila*.

(4) Both the penis-sheath and the penis are inserted into the mantle-cavity of the female and the free end of the penis enters the vagina. Impregnation is, therefore, internal.

(5) The genital-gland, like the penis, consists of vascular erectile tissue.

(6) Snails are killed in a copulating position by electrocution.

(7) Oviposition takes place within a day or two after copulation, and the foot acts as a very efficient ovipositor. There is no incubation of eggs by the snail as supposed by Ramanan.

(8) Eggs are laid on the ground and *not* in the water. They are laid in large masses in small hollows in sheltered places. Each egg-mass usually contains 200 to 300 eggs, but I have come across some very large egg-masses containing as many as 700 to 800 eggs.

(9) Each egg consists of a shell, a double shell-membrane, a thick layer of opaque white solid albumen and a core of fluid albumen in which the embryo floats.

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