

The Journal of Infectious Diseases

PUBLISHED BY THE MEMORIAL INSTITUTE FOR INFECTIOUS DISEASES

VOL. 18

February, 1916

No. 2

THE MODE OF INFECTION IN PULMONARY DISTOMIASIS *

CERTAIN FRESH-WATER CRABS AS INTERMEDIATE
HOSTS OF PARAGONIMUS WESTERMANII

PLATES 2 TO 5

KOAN NAKAGAWA
Formosa, Japan

Paragonimus westermanii Kerbert, the distome of the human lung is rather widely spread in the Far East. Ringer was the first to discover it, in 1879, in the lungs of inhabitants of the city Tamsui in Formosa. Since then it has been found in the various parts of Japan proper. In some places the distomes affect so large a number of the inhabitants that pulmonary distomiasis assumes almost an endemic nature. Consequently, the distome has been very carefully studied by various Japanese investigators. Altho it is now over 30 years since the discovery of the distome, the developmental stages of the worm, except its miracidia, have not as yet been clearly understood. For 10 years I have been carrying on an investigation¹ of pulmonary distomiasis in the locality of Formosa, where the disease is most prevalent, and finally

* Received for publication August 25, 1915.

¹ On the human lung distomes in the region Shinchiku, Jour. Taiwan Med. Assn., No. 138. Preliminary report on the discovery of the intermediate host of the human lung distomes, Tokyo-iji-shinshi, No. 1910. Study on the development of human lung distomes, Chugai-iji-shimpo, No. 1843; No. 1845.

I have been fortunate enough to discover the second intermediate host of the lung distomes. Recently the mode of infection has been experimentally determined.

THE DISTRIBUTION OF PULMONARY DISTOMIASIS IN FORMOSA

Before entering into details it seems proper to record here briefly the distribution of the cases of pulmonary distomiasis in Formosa; for it will show how I came to the discovery presently to be described.

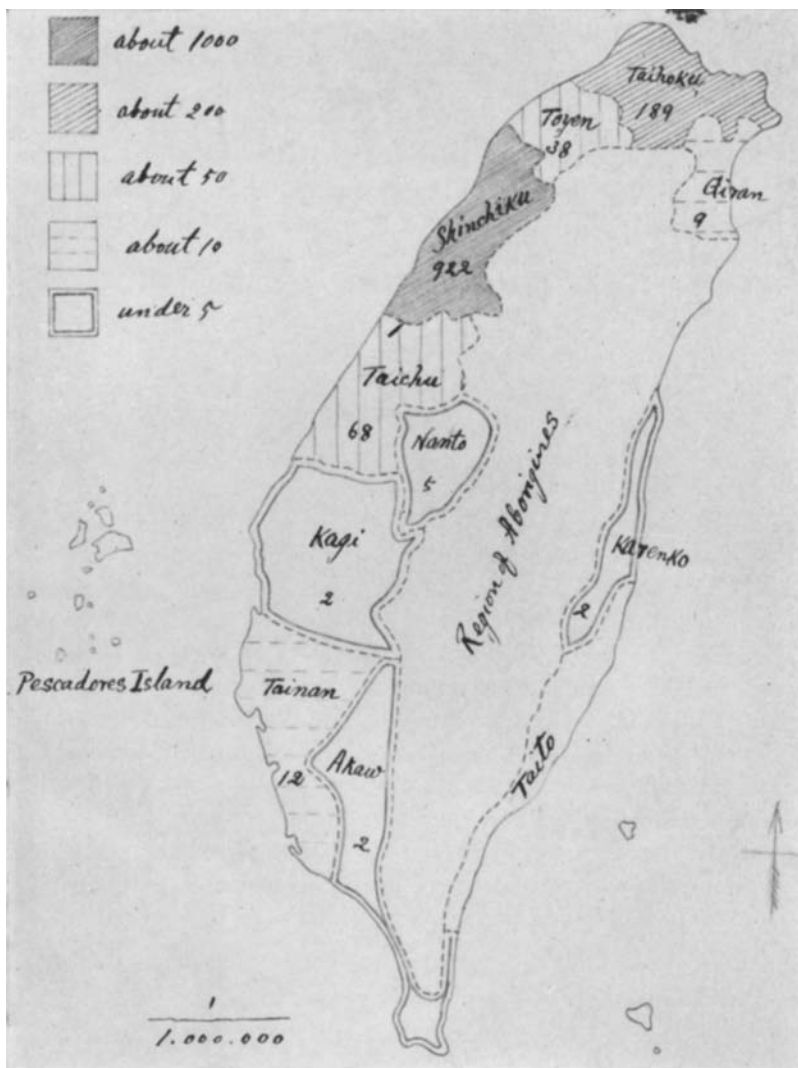
Pulmonary distomiasis in Formosa was reported by Ringer in 1879. Nothing as to the distribution of cases was known until Nagayo reported in 1910 that cases were found in the northern region around Shinchiku. In the course of 1913-1914, I made a careful study of the disease throughout the whole island of Formosa, and found that of 1,249 patients 922, or about 73%, were residents of the Shinchiku Prefecture. This clearly shows that this prefecture is the center of infection (Map A). The distribution of patients in the central region of infection has been made clear by Matsuo, Yokokawa,² and others. They all agree that about 10% of the inhabitants of Shinchiku are affected by this disease. My microscopic examination of the expectorations of the boys and girls of the public schools revealed that 4.3% of all pupils were infected. This percentage means that not less than 1,300 cases must be present in Shinchiku Prefecture alone; the percentage may be somewhat larger for the whole population.

Altho nothing is known as to how the matter stands among the aborigines of Formosa, my frequent visits to their villages in 1914, convinced me of the fact that the nearer the valley they dwell, the more they are affected by distomiasis. Indeed, 50% of the inhabitants of such places were found to be infected. Among the people of the mountainous regions, the cases seem to occur far less frequently, tho my examinations were not numerous enough to warrant any definite conclusion.

INVESTIGATION OF THE SECOND INTERMEDIATE HOST OF PARAGONIMUS WESTERMANII

Appointed head of the Public Hospital at Shinchiku in 1912, I was placed thus under favorable conditions for carrying out an investigation of the lung distome. My attention was first directed to the search after the second intermediate host of the distome so that I might trace its developmental cycle. When I started the investigation, all that was known of the life-history was contained in Nakahama's and Manson's observations, that the miracidia hatched from the eggs begin to swim in about 4 weeks from the time of entering the water after they come out of the patients. As to what becomes of them afterwards, nobody had any idea.

² On the transmission path of the human lung distomes in the final host, Tokyo-iji-shimpo, No. 1910.



Map A. The distribution of patients with pulmonary distomiasis in Formosa. The map shows the number of patients in each prefecture during the years 1913-1914.

Collecting as many fresh-water molluscs as possible from the streams or ponds in the region where distomiasis exists as an endemic disease, I made careful search after the cercariae. I succeeded in obtaining 17 different kinds, but I could not distinguish the cercariae of the lung distomes from others. So I put various molluscs into the water in which miracidia of the lung distome were kept, to see what kinds of molluscs the miracidia would infest. It resulted that they infested *Melania libertina* Gould and *M. obliquegranosa* Smith most abundantly. From this it may be assumed that these two species of fresh water molluscs are the first intermediate hosts of the lung distomes. Notwithstanding the utmost care taken in keeping the molluscs infested with miracidia alive in an experimental pond, all of them died within a few weeks. They were found to contain no grown cercariae. Altho this experiment turned out to be a failure, I found that a certain kind of cercaria is found in all *M. libertina* living in the rivulets and creeks of the mountainous regions where the aborigines are infected in an enormous percentage.

The cercaria has a tadpole-like appearance, the body being 0.12 mm. long and 0.09 mm. wide, with a tail 0.054 mm. long. Attached to the oral sucker (0.036×0.032 mm.) are 2 pear-shaped bodies, the apices of which point towards the median plane of the body. The sucker has spines, each provided with a ring along its anterior edge. The ventral sucker is much smaller than the oral one, being 0.018 mm. in diameter. Within the parenchyma are 3 pairs of poison glands. The excretory vesicle is heart-shaped. Besides this kind of cercaria, the liver of the molluscs contains a good many sporocysts. Some melaniae have a number of half-grown cercariae. From the fact that *M. libertina* is found abundantly in the region where distomiasis of the lung prevails most widely, it may not be unreasonable to conclude that these cercariae are those of the lung distome. However, we have not as yet any experimental proof.

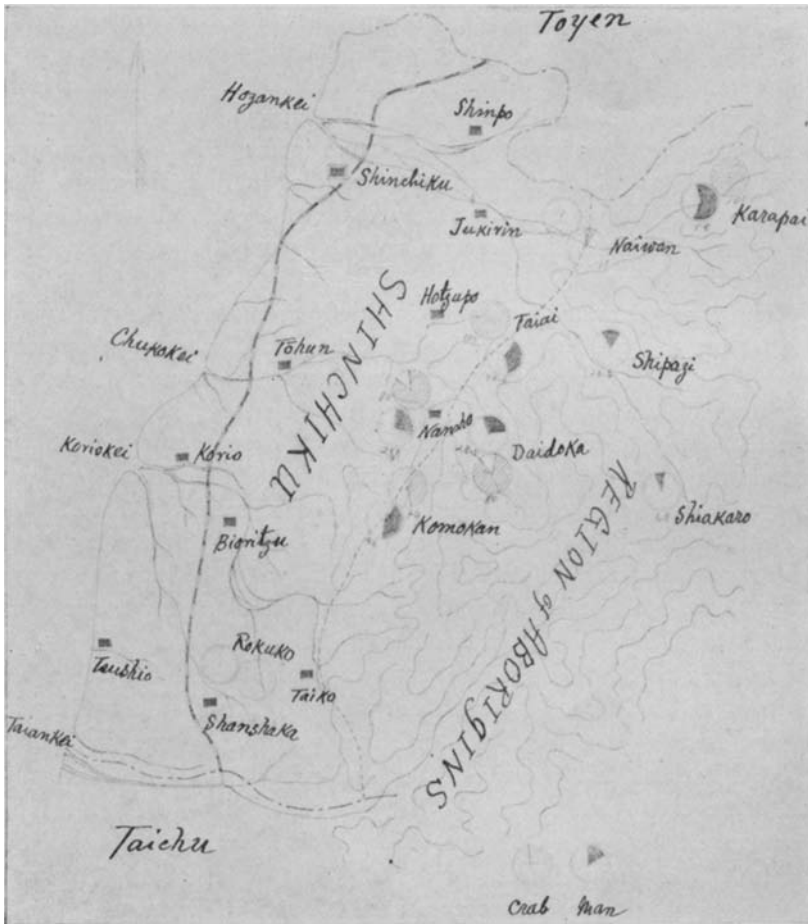
ENCYSTED LARVAE OF THE DISTOME OF THE HUMAN LUNG IN CRABS

Kobayashi's³ interesting work on the discovery of the encysted larvae of *Chlonorchis sinensis* Cobbold in certain fishes gave me an impulse in the study of the question of the second intermediate host of the distome of the human lung. At first I selected a region of the Shinchiku prefecture where patients are abundant and made several difficult excursions to collect all molecules, fishes, amphibians, and

³ A preliminary report on the second intermediate host and encysted larvae of the human lung distomes, Tokyo-iji-shinshi, No. 1918.

insects that I could. My microscopic examination showed that *M. libertina* Gould only has a certain kind of cercaria. In September, 1914, I captured in the rivulet in Kalapai Village a kind of crab which had not been caught before. My microscopic examinations were rewarded with the discovery of numerous encysted larvae in the liver. (Plate 3, Fig. 7; Plate 4, Fig. 8). They were all half grown, but they were unmistakably larvae of certain trematodes. At first I had no idea that they were the young distomes of the human lung. However, as the result of further investigation I found in the gills full-grown ones with all the morphologic structures peculiar to the distome of the human lung. Furthermore, I observed that the encysted larvae, when they were introduced into certain animals by mouth, grew into *Paragonimus westermanii*.

The young encysted larvae infest chiefly the liver of the crab, while the farther advanced ones are found in the gill. Sometimes they penetrate the muscle. The young encysted larvae are round, 0.2 mm. in diameter (Plate 2, Fig. 4). The young distomes in the cysts lie straight. They have a conspicuous, large, black excretory vesicle and comparatively large oral (provided with a spine) and ventral suckers. They do not have a distinctly developed alimentary canal. The encysted larvae found in the gill are well developed, measuring 0.3 to 0.4 mm. in diameter (Plate 2, Fig. 5). The young distome in the cyst has a short and thick body and lies straight, unlike others, which are contorted. The oral sucker has a spine, and is 0.08 to 0.11 mm. in diameter. The short esophagus is connected with the bifurcate intestine. Each branch of the intestine is thick and undulating, running parallel to, and outside of, a long, thick excretory vesicle. The ventral sucker is a little larger than the oral one, measuring 0.07 to 0.12 mm. in diameter. The entire surface of the body is provided with short spines. The wall of the cyst is 0.01 mm. thick—a characteristic feature of the species. The young distome rotates sluggishly in the cyst. The full-grown encysted larvae sometimes reach 0.5 mm. in diameter (Plate 2, Fig. 6), and to the naked eye look like a white speck. Sometimes they assume an elliptical shape, 1.0 mm. long and 0.4 mm. wide. They can readily be removed from the gill. Twenty percent of larvae thus freed were observed to float on the surface of the water. Under natural conditions the full-grown larvae would leave the crab's gills, get into the water, and be taken up by the dwellers on the banks of the lower stream.



Map B. Showing the correlation between cases of pulmonary distomiasis and infested crabs.

FRESH-WATER CRABS WITH THE LARVAE OF PARAGONIMUS WESTERMANII

Potamon obtusipes (Stimpson)—“Red crab” (Plate 2, Fig. 1). This was the first of the three species of crabs in which the encysted larvae of the human-lung distome were found. The carapace is coarse, flat, and provided with teeth along the lateral margins; the pterygostomian region is granular; the dorsal surface is a deep-chestnut color; the abdomen and legs are somewhat reddish. One large specimen has a carapace over 38 mm. in diameter. The native name of this crab is “shahoi” or “chahoi,” which means red crab. Sometimes it is eaten. This species has never been found in any place in Japan other than the mountainous region in Formosa. I have found that the crab is most abundant in rivulets or creeks running through the mountainous regions of Shinchiku Prefecture. The number of encysted larvae in the gills increases in correlation with the number of cases of distomiasis of the lungs; for instance, in the region where 30-50% of the inhabitants are found to harbor the parasites in their lungs, 100% of the crabs carry the encysted larvae, while 5 miles down the river, where comparatively few patients are met with (I have had no opportunity to make sure of the percentage), only 11% of the crabs have the encysted larvae. In the crabs of Sansaka region no encysted larvae were found (Map B).

Potamon dehaanii (White).—This species (Plate 2, Fig. 3) occurs in the same locality with the species mentioned, but not so abundantly, and it is a little smaller in size. The carapace is somewhat round and smooth. There are no teeth on the lateral margin. The pterygostomian region is smooth. The dorsal surface is grayish-black, or sometimes slightly reddish. The ventral side and the legs are grayish-white. This species also occurs in the rivers of the mountainous regions in Japan proper. The natives, who call it “sai-hoi” (dung crab), do not eat it. Encysted larvae of the distome of the lung were also found in this species, tho less numerous than in the former.

Eriocheir japonicus De Haan.—Unlike the other two species, this one (Plate 2, Fig. 2) never occurs in the streams of the mountainous region, but occurs in the rivers flowing across the plain. This species can easily be discriminated from the other two by its chelae, which are provided with a thick growth of hair. Very big specimens have a carapace over 76 mm. in diameter. The native name is “mon-hai,” or “mun-hai” (hairy crab). This crab is eaten by the natives. The

encysted larvae were found in only 2 out of 330 specimens (300 large and 30 smaller ones) which I collected and examined. I am not certain whether or not the attachment to the crabs is merely an accident. The solution of the problem awaits further investigation.

In conclusion I may say that of the three species of fresh-water crabs the encysted larvae of the human lung distomes are found in the first and the second species, the occurrence in the third being problematical. In view of the fact that the second species occurs outside Formosa, this may be the second intermediate host of distomiasis in Japan proper.

EXPERIMENTAL PULMONARY DISTOMIASIS

In order to verify experimentally the conclusion reached in the last section, I secured some dogs that had been brought up in a place where no cases of distomiasis of the human lung are known. I used dogs because they are known to be easily infected by the lung distomes. The liver, the gills, and other organs of the crab that harbored a large number of encysted larvae were given to 2 puppies on September 23 and October 10, 1914. One of the dogs died December 9; that is, 60 days after feeding. Postmortem examination showed the lungs to have a number of cysts. Within each cyst 2 or 3 distomes were present. They had no eggs in the uterus at this time. They measured 4 to 5 mm. in length and 2 to 3 mm. in width. The other puppy died on December 27 after a lapse of 90 days from feeding. His lungs contained numerous cysts, in which adult distomes with eggs ready to be discharged were found. These distomes measured 6 to 7 mm. in length and 3 to 4 mm. in width—not larger than half the ordinary size of the distomes found in the lungs of man, the cat, and the dog. Their morphologic features, however, tallied well with those of *Paragonimus westermanii*. The worms thus raised in the lung of a puppy have a short, thick body, dark in color in the living stage. They assume various irregular shapes while in physiologic salt solution. After being fixed in formalin they are grayish-white and ovoid, the anterior end being rounder than the posterior. The transverse section is nearly circular.

The experiment was repeated at the Shinchiku Public Hospital with 3 other puppies that had been brought from a region free from lung distomiasis. Two were fed with a large quantity of the internal organs of crabs, while the remaining one was left untreated as control.

The two animals died 50 days after feeding; that is, on February 7, 1915. A number of distomes were found in their lungs. The control animal, however, was found to be free from them when I examined its lungs on the same day. Again, another puppy, to which water containing some encysted larvae was given, contracted the disease.

MODE OF TRANSMISSION OF THE DISTOME OF THE LUNG IN THE
FINAL HOST

In order to ascertain how the distome finds its way into the final host, and to see what pathologic changes the lungs undergo, I made examination of puppies at various intervals after infection.

On the fifth day from feeding no macroscopic changes were visible in the lung. On the seventh day 2 or 3 fresh petechiae, not larger than a pinhead, were seen. On the fourteenth or fifteenth day hemorrhage was obvious on the surface of the lung, especially on the lower lobes. From the twenty-first to the twenty-seventh day small nodules made their appearance, besides hemorrhagic spots. Generally the nodules were in connection with the hemorrhagic spots, but sometimes the former appeared quite independent of the latter. On the thirty-fifth day, hemorrhagic spots as big as linseeds were visible. On the fiftieth day, large nodules or cysts of *Paragonimus westermanii* appeared, which were as big as the end of the little finger. Some of the nodules bulged out a little on the surface of the lungs. They were rather hard and of a dark-red and gray color. The section was of a dark-red color, which might have been due to the hemorrhage. In the center of such nodules the distomes were embedded. Sometimes the worm lay embedded in surrounding tissues. On the ninetieth day, the cysts sometimes assumed a bluish-gray color. On section they were found to have a cavity as large as a pea, filled with a porridge-like substance in which one or two fully grown worms were present. The walls of such cysts were formed of a thin layer of connective tissue.

Two puppies were fed with grown encysted larvae from April 15 to April 27, 1915 (both died of emaciation); likewise 3 kittens—No. 1 from April 18 to May 2; No. 2 from April 18 to May 16; No. 3 from April 25 to May 16. I chloroformed these animals and took out all the internal organs. The abdominal cavities were filled with comparatively large quantities of serous secretion. The livers were generally congested. The diaphragms had numerous small perforations

like passages made by small distomes. Over the surface of the liver and the omentum major in each animal were seen many distomes attached. On the mucous membrane of the jejunum and spleen one petechia, or two, or sometimes more, was seen of the size of a pinhead or sometimes larger. Hemorrhagic spots were present on the pleura; on the thoracic side of the diaphragm were minute petechiae and small perforations; the lung had many hemorrhagic spots and small nodules.

On examination of microscopic sections through a petechia of the jejunum, the worms were found to have penetrated through the mucous membrane and inner muscle layer, and to be about to bore through the external layer (Plate 4, Fig. 9). It is obvious that these worms had come out of the cysts and penetrated the intestinal wall. Moreover, the passage through the muscle layer was indicated by the interruption of the muscle layer and by a marked infiltration of leukocytes, especially of eosinophile cells. Preparations of omentum major showed that it had some worms embedded in the net-tissue and also in the fat tissue near the region (Plate 5, Fig. 10) where it is united with the large curvature. In the sections of the petechia of the diaphragm worms were found just penetrating the tendon (Plate 5, Fig. 11). The penetration path had the same microscopic features as that in the intestine. In the petechiae of the spleen and the kidney no worms were seen. In the sections of the lung of a puppy killed 30 days after being fed with encysted larvae, were numerous hemorrhagic spots, but no worms were found embedded in the tissue.

The distomes that had reached the thoracic cavity had not as yet penetrated the lung. It may be conjectured that they live for some time floating in the serous excretion of the thoracic cavity or under the pneumopleura, and attached by suckers to the lung-parenchyma, thus causing small petechiae there. Of the worms in the thoracic cavity only a small percentage seemed to get into the lung; for the number of such nodules was far less than that of the worms in the serous liquid. The sections of the nodules showed a trace of loss of tissue and a high degree of infiltration of leukocytes (especially eosinophile cells) around it. This was undoubtedly the path through which the worms had penetrated. But the worms had not developed into cysts there; in more than 50 days from the infestation, the cysts would have been completely formed.

The wall of a newly formed cyst consisted of young connective tissue with an infiltration of great numbers of polymorphonuclear

leukocytes and small cells, while that of an old one was formed of a strong, fibrous connective tissue. The wall, however, did not present a uniform structure; in some places the cyst was completely lacking in walls, being directly apposed to the alveolar tissue of the lung, the small bronchi, or veins, thus causing local bleeding and pneumonic infiltration of the alveoli. Within each cyst lay one or two worms. Sometimes one or two of the worms that lay in the same cyst, would be found decomposing. The eggs were observed to have been laid first on the ninetieth day after feeding. They lay in the cyst or the parenchyma of the lung around the cyst, and generally were found mixed with erythrocytes, leukocytes, epithelial cells, and their products. Near the pathologic regions dilatation of bronchi was present, the cavity being filled with erythrocytes, eggs, etc.

From what has been described it may be seen that the lung distome will form a cyst in the lung of the host, live only for a while in it, and then leave it, finding its way through the tissue, bronchi, blood-vessels or the wall of the cyst by virtue of a vigorous motion that causes local bleeding and pneumonic infiltration. The degenerative products will be discharged from the lung through the trachea, carrying eggs with them.

To sum up, the encysted larvae of *Paragonimus westermanii* that have been taken into the alimentary canal of the host, creep out of the cysts and making their way through the intestinal wall near the jejunum, reach the abdominal cavity. They then penetrate the diaphragm and reaching the thoracic cavity, scatter all over the space under the pleura, whence they can easily find their way to the lung. They pierce the lung parenchyma, and there the cysts are formed until they become fully grown. These parasites can bore through various tissues and may reach other organs than the lung, where they form their regular cysts; but the lung seems to be the most favorable place for their development and the laying of their eggs; in other organs they can never reach the perfect growth.

EXPLANATION OF PLATES

PLATE 2. FRESH-WATER CRAB

- Fig. 1. *Potamon obtusipes* (Stimpson) nov. sp., natural size.
Fig. 2. *Eriocheir japonicus* De Haan nov. sp., natural size.
Fig. 3. *Potamon dehaanii* (White), natural size.
Fig. 4. Young encysted larva in the liver of a crab (*Potamon obtusipes*).
× 20.
Fig. 5. Half-grown encysted larva in the liver of a crab. × 20.
Fig. 6. Full-grown encysted larva on the gill of a crab. × 20.

PLATE 3

- Fig. 7. Encysted larvae of *Paragonimus westermanii* in the liver of a fresh-water crab (*Potamon obtusipes*).
Fig. 8. Encysted larvae of *Paragonimus westermanii* in the liver of a crab.
× 15.

PLATE 4

- Fig. 9. Cross-section of the jejunum of an experimentally infected kitten (No. 1). Young distome is seen passing through the intestinal wall. × 20.
Fig. 10. Adipose tissue of the omentum of an experimentally infected kitten (No. 1). a = worm. × 50.

PLATE 5

- Fig. 11. The diaphragm of an experimentally infected kitten (No. 1). a = young worm in the tendinous part of the diaphragm. × 50.
Fig. 12. Section through the lung of an experimentally infected puppy (No. 1) 60 days after feeding. a = passage of the worm. × 20.

PLATE 2

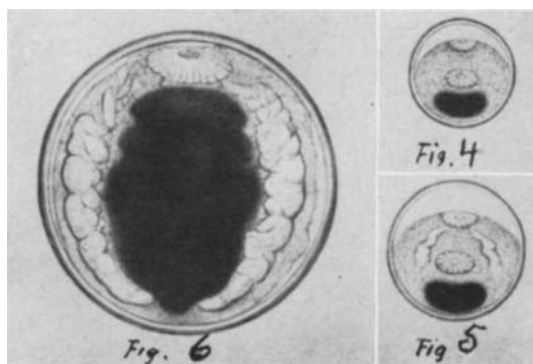
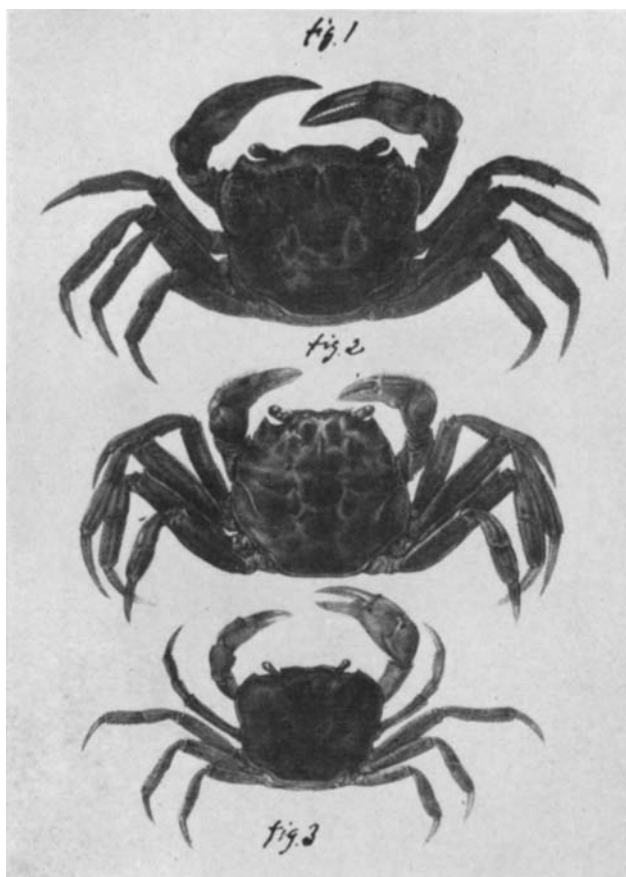


PLATE 3

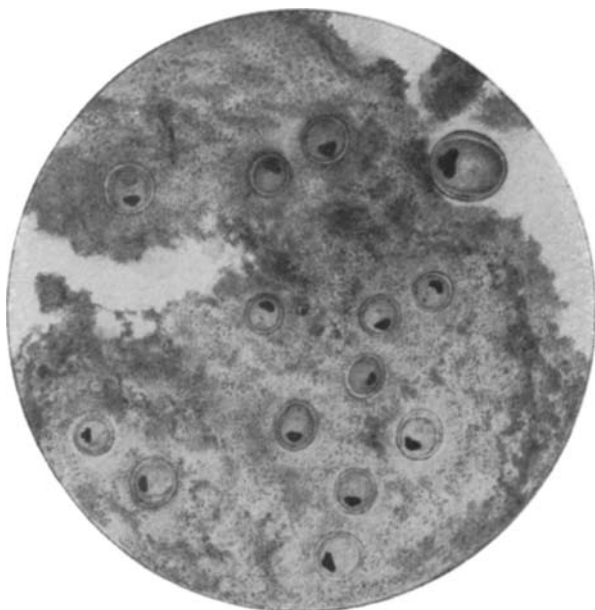


Fig. 7

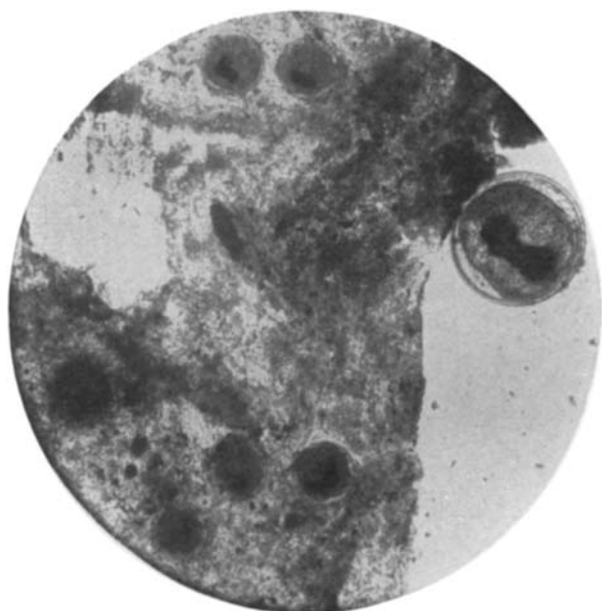


Fig. 8

PLATE 4



Fig. 9

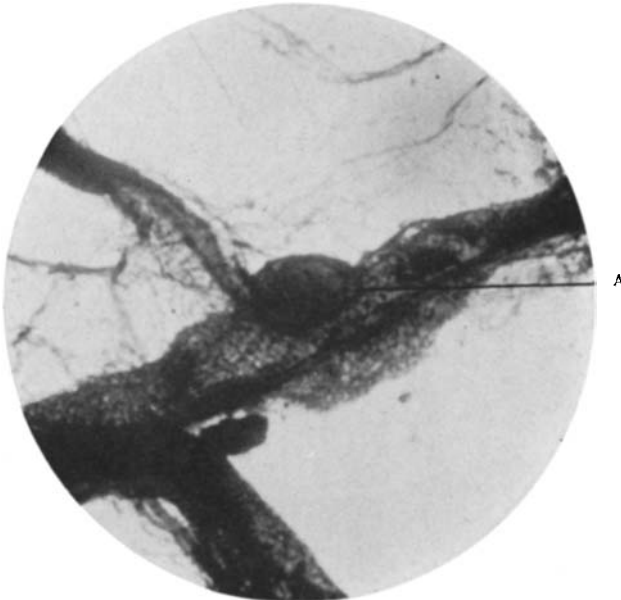


Fig. 10

PLATE 5



Fig. 11



Fig. 12