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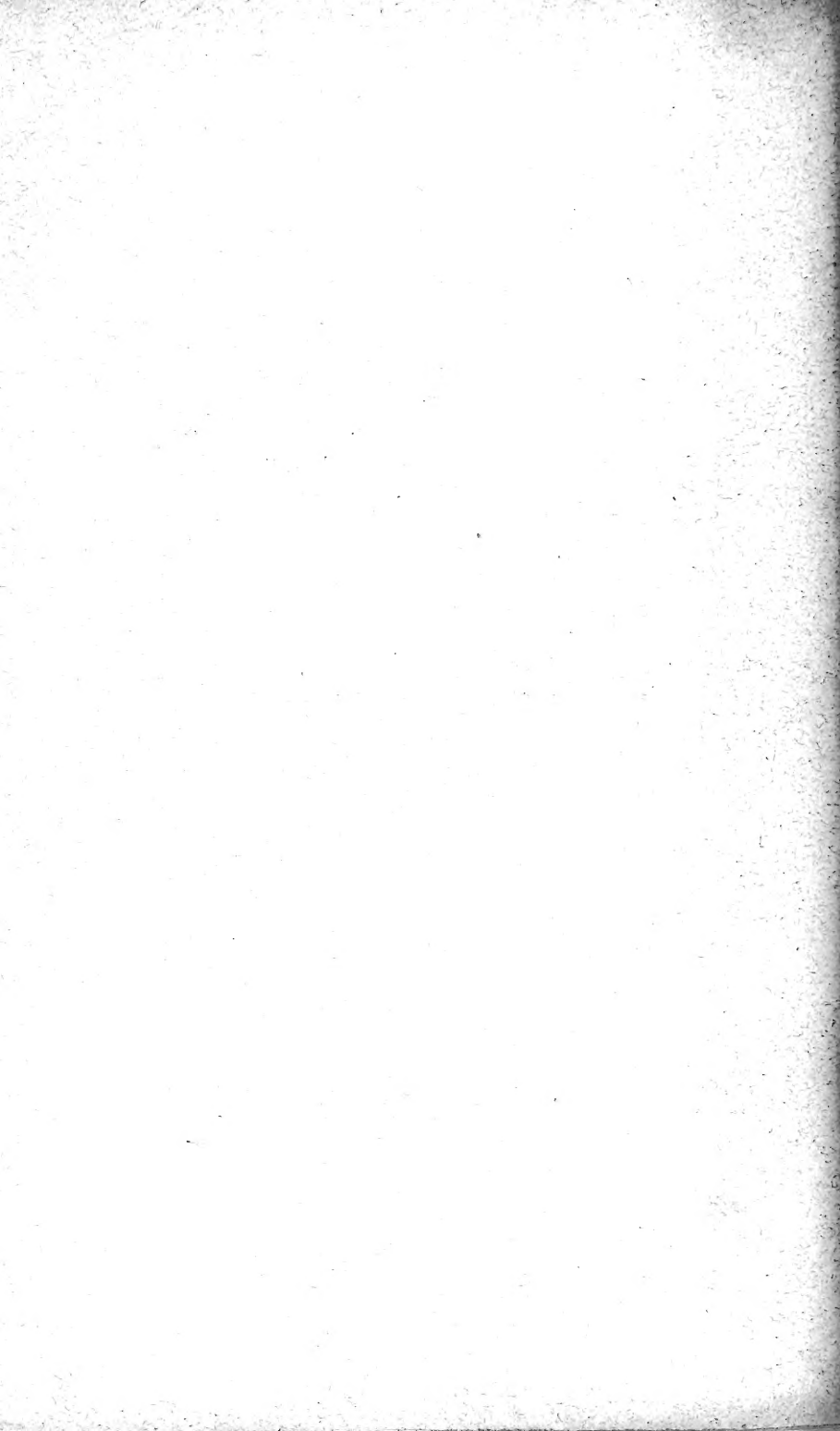
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No. XII.

REPORT FOR 1903,  
ON THE  
LANCASHIRE SEA-FISHERIES LABORATORY  
AT  
THE UNIVERSITY OF LIVERPOOL,  
AND THE  
SEA-FISH HATCHERY AT PIEL.

DRAWN UP BY

Professor W. A. HERDMAN, D.Sc., F.R.S.,

*Hon. Director of the Scientific Work,*

Assisted by Mr. ANDREW SCOTT, A.L.S., and

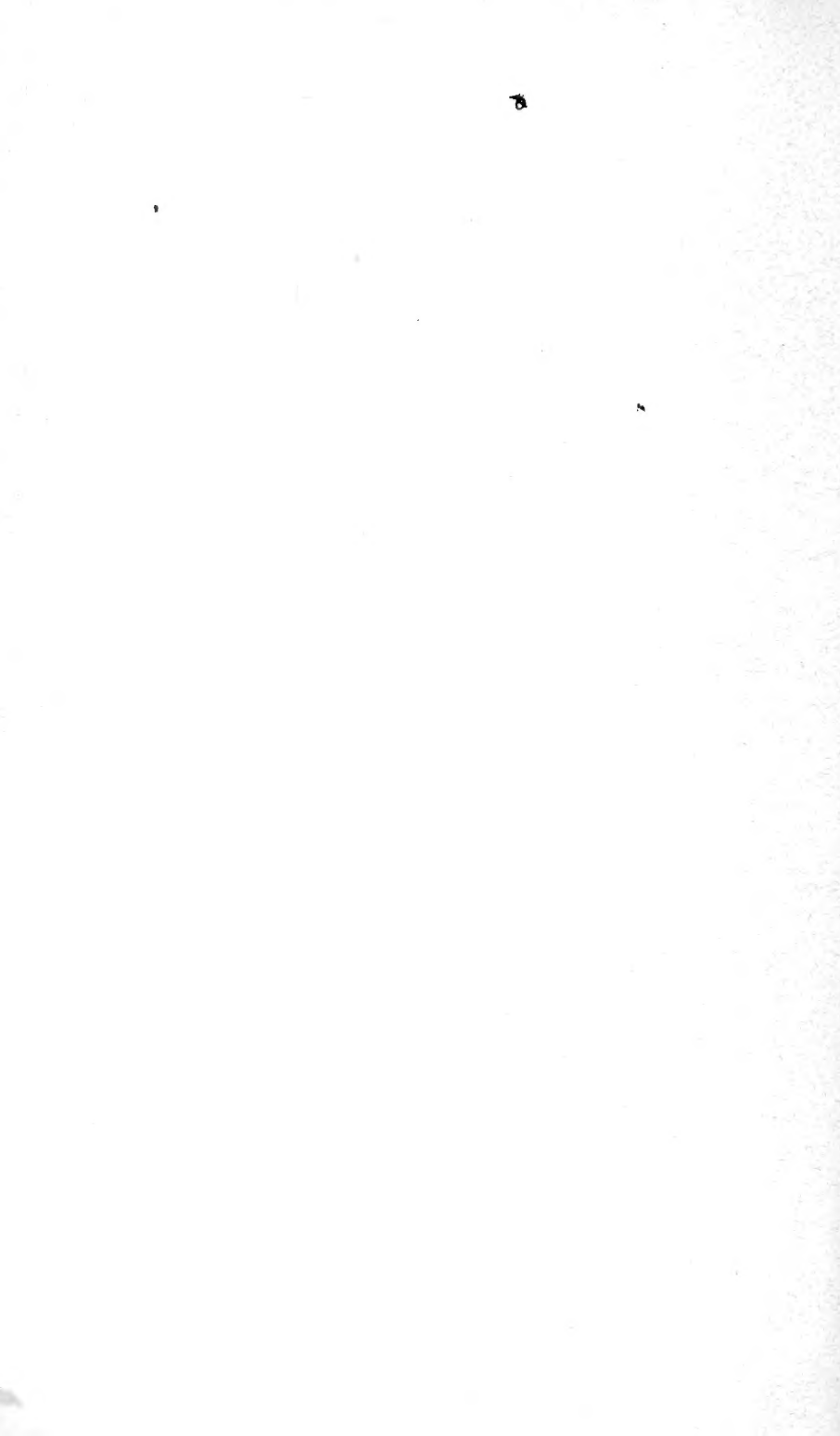
Mr. JAMES JOHNSTONE, B.Sc.

WITH ILLUSTRATIONS.

LIVERPOOL

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1904.



JUN 4 1904

REPORT on the INVESTIGATIONS carried on during 1903 in connection with the LANCASHIRE SEA-FISHERIES LABORATORY at the University of Liverpool, and the SEA-FISH HATCHERY at Piel, near Barrow.

Drawn up by Professor W. A. HERDMAN, F.R.S., Honorary Director of the Scientific Work; assisted by Mr. ANDREW SCOTT, A.L.S., Resident Fisheries Assistant at Piel; and Mr. JAMES JOHNSTONE, B.Sc., Fisheries Assistant at the Liverpool Laboratory.

*With Compliments from*

PROFESSOR W. A. HERDMAN, F.R.S.,  
UNIVERSITY COLLEGE,  
LIVERPOOL.

*who will be glad to receive your publications in exchange.*

— THE BOOKS RECEIVED :—

(1) The hatching operations and other similar work carried out at Piel by Mr. Andrew Scott;

(2) Laboratory investigations on fish and on their parasites and diseased conditions, by Mr. James Johnstone, at the University of Liverpool;

(3) Practical Laboratory Classes for Fishermen and for school teachers, conducted by Mr. Johnstone and Mr. Scott, at Piel;

(4) Observations at sea on board our Fisheries Steamer "John Fell."



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Assistant at the Liverpool Laboratory.

(With plates, charts and figures in the text.)

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INTRODUCTION AND GENERAL ACCOUNT  
OF THE WORK.

THE work of the past year has been chiefly:—

(1) The hatching operations and other similar work  
carried out at Piel by Mr. Andrew Scott;

(2) Laboratory investigations on fish and on their  
parasites and diseased conditions, by Mr. James Johnstone,  
at the University of Liverpool;

(3) Practical Laboratory Classes for Fishermen and  
for school teachers, conducted by Mr. Johnstone and Mr.  
Scott, at Piel;

(4) Observations at sea on board our Fisheries  
Steamer "John Fell."

Some matters that can be treated briefly I shall remark upon in this introductory part of the Report; the others will be discussed more fully in the special articles that follow.

I am glad to say that the very full account of the Fisherman's "Lugworm," which Dr. J. H. Ashworth (formerly of Owens College, Manchester, now at the University of Edinburgh) has been preparing for some years, is now finished, and I am able to add it as an Appendix to this Report. The expense of lithographing the beautiful plates that illustrate this Memoir has been largely met from an outside source.

### The Piel Hatchery.

Mr. Scott's account of the Sea-Fish Hatching at Piel will be found in the next section of the Report. As on former occasions, the fish dealt with were the Plaice and the Flounder, and out of close on seventeen millions of eggs obtained nearly fifteen millions were hatched and distributed in the sea as fry. The total loss from all causes during the operations was just under 11 per cent. It can scarcely be doubted that the natural mortality in the sea during the corresponding period in the life of the young fish embryo must be enormously greater than this. The benefit of protection would be, however, still further increased if we had the accommodation necessary for keeping and rearing the larvæ to still later stages. This is impossible without a fish pond; and Mr. Scott points out that he cannot, with his present small tanks deal with much larger numbers than those that passed through his hands this year. An open-air fish pond has proved a success elsewhere. An American fish-culturist, Professor Mead, of Brown University, who visited the Port Erin Biological Station last summer, expressed his satisfaction

at seeing the fish pond there, and evidently regarded it as a necessary addition to any hatchery. I would repeat again what I stated to the Committee last year, that no hatchery is complete without a spawning and rearing pond, and that the want of one at Piel seriously impedes Mr. Scott's operations.

### T r a w l i n g   R e s u l t s .

We are now preparing for the hatching work of the coming season, and once more we are indebted to the courtesy of the Fishery Board for Scotland for permission to trawl for large plaice in their closed waters of Luce Bay. As before the Fishery Board asked us to make observations and to give them a record of our results. Two trips were made to Luce Bay in October and November and our Naturalists who accompanied the steamer were able to make a series of interesting hauls on both occasions. The results are fully discussed by Mr. Johnstone in his article on "Trawling Observations," which will be found below. These results confirm those obtained in 1902, and reinforce the conclusions which I drew in last year's Report as to the remarkable differences in the catches which may result from very slight differences in the positions and conditions of the grounds trawled. The bearing of these observations upon the danger of any attempts to draw conclusions from samples taken relatively far apart, even on areas where uniform conditions are supposed to obtain, must be obvious.

Mr. Johnstone also draws attention to the very large average size of the plaice in the closed Scottish waters, and shows how the much smaller size on the Lancashire coast may be regarded as the natural result of constant and practically unrestricted fishing.

## The Classes in Biology at Piel.

I have also received from Mr. Scott a report upon the other work and the leading events of the year at our Piel establishment. It will be found below, and consists mainly of a record of the practical classes held in the laboratory, and of the visits of scientific men, and other Sea-Fisheries and Educational Authorities.

Our Fisheries Exhibition is still on view at the Piel Laboratory, and is a useful adjunct to the teaching resources. It can, however, be sent out on loan, as formerly, when required; and any public institutions within the contributing counties desiring to show the exhibit should apply to the office at Preston for a copy of the conditions upon which it may be obtained.

## Fish Parasites and Diseases.

Some time ago I suggested to Mr. Scott, who has been working for years, at odd moments as opportunity offered, upon the parasitic Crustaceans of our seas, that he should extend his observations to the parasitic worms and other lower animals that may cause diseased conditions, and give us a list of all the fish-parasites he was able to detect. A first, and very considerable instalment, appears below, consisting of four Protozoan, ten Trematode, one leech and forty-six Copepod parasites, taken, as will be seen, from a varied assortment of fishes, and from very different positions in the body. The Cestodes (tape-worms) and Nematodes (thread-worms), not included in this paper, will follow on some future occasion.

Mr. H. M. Woodcock, of University College, London, has examined for us two of the Protozoan parasites, and contributes a useful paper on "Myxosporidia in Flatfish," which enumerates all these parasites which have yet been



found, and also contains a description of a new species, *Sphaerospora platessæ*, from the Plaice. Mr. Woodcock in a second paper describes a very remarkable parasite, *Lymphocystis johnstonei*, from the Flounder.

#### O t h e r   W o r k .

Mr. R. D. Laurie, a former student of our Zoological Department at the University, has contributed a short note bearing on the question of the number of eggs that can be produced by an adult plaice. His results for plaice, of about 20 inches in length, from the Irish Sea, agree with those obtained for other seas. Of the three or four hundred thousand large eggs present on the average in such a fish only a comparatively small number are mature at a time, the plaice setting free its ova in successive small batches over an extended spawning period. In stripping a spawning plaice only a certain small proportion of the eggs in the ovary can be extruded.

The subject of pearl formation in Molluscs, such as mussels and oysters, has been brought into prominence of late years by several investigations and reports both in this country and abroad. As the matter is one of considerable public interest and importance, and as there is at present a good deal of misapprehension in connection with it, caused by sensational statements derived from some of the French papers, I have thought it well to give a brief account of the more important recent discoveries and views bearing upon the subject of pearl-formation.

The relation of sewage disposal to the pollution of our coasts, and to the possible infection of edible shell-fish with pathogenic organisms, has become a matter of national importance. Some previous work done in our laboratory 8 years ago,\* drew attention to the matter

\* See Report of Ipswich meeting of British Association, Sept., 1895, and Lancashire Sea-Fisheries Memoir, No. 1., 1899.

locally, and since then investigations have been carried on at various points round the British coasts, and for the last few years the Royal Commission on Sewage Disposal has been taking evidence and deliberating on the subject. Both Mr. Dawson and I, in our evidence before the Royal Commission, have drawn attention to the very serious state of affairs on some parts of the Lancashire and Cheshire coasts, and we have lately planned a more thorough examination into the condition of the shell-fish beds of the district. Mr. Scott has inspected for me several of the mussel and cockle producing areas in the northern part of Lancashire, and Mr. Johnstone has made a bacteriological examination of the samples of shell-fish that have been sent to the laboratory. As the Royal Commission has also just issued a Report† dealing with these same questions, and very much on the lines we have adopted, I have thought it appropriate to devote a few pages further on to a discussion of the matter.

A suggestion, made by Mr. Fell, that we should report upon the intricate question of the inter-relations between shrimps and young flatfish, has led to the preliminary statement of the subject which, with Mr. Johnstone's help, I have drawn up. It is evident that a great deal of exact information is still required. I have tried to focus attention upon what is known and what is unknown, upon what the essential problems are and what investigations are necessary in order to solve them. But it is clear that this is one of the matters upon which we cannot get wholly satisfactory evidence until we have, on the West Coast of England, a steamer devoted solely to scientific fisheries work, so that an organised scheme of investigation, combined with the collection of statistics, can be carried out. It would probably require a couple of years

† Pollution of Tidal Waters with special reference to contamination of Shell-fish. London, 1904.

of such organised work before reliable conclusions could be arrived at. Still the outline of the sub-divisions and relations of the investigation given below may be useful to the Committee as showing the complexity of such a question, and the need for a very thorough systematic investigation of all such fishery matters.

Finally, I may refer to the most important event of the past year in connection with Sea-Fisheries Organisation and Administration in this Country, viz., the transference of the Official Government department dealing with these matters from the Board of Trade to what is now the Board of Agriculture and Fisheries. This must be a matter of congratulation in so far as it gives to our subject more of the high relative position and the improved status to which its importance entitles it, and which it possesses in most civilised countries. Although not yet an independent department of State, with a Minister for Fisheries, it is now conjoined on an equality with the allied subject, Agriculture, under a President, Lord Onslow, who recognises fully its claims to his attention, and will assuredly give sympathetic and adequate treatment to the new division of his department. The union of Fisheries with Agriculture is a natural one, which we see working well in Ireland, in several of the Colonies and elsewhere. Aquiculture and Agriculture have similar disciplines, and similar methods should give similar results. The operations that result in the harvest of the sea being now placed under the same guiding hand as those that affect the harvest of the land, we may hope that in the former case as in the latter the returns from nature will be increased as the result of scientific cultivation.

From the scientific point of view the present division of the territorial waters of England and Wales into Sea-

Fisheries Districts is not satisfactory. The Districts are too numerous and unequal, the boundary lines are arbitrary and unnatural, the methods of the Authorities are too diverse; and, as a result, the fisheries are very unequally treated both as to administration and investigation. A glance at the details of expenditure of the different Committees is enough to show how perfunctory and inadequate the examination and protection of the fisheries must be on some parts of the coast.

For these, and other reasons, the Ichthyological Committee in their Report last year recommended for purposes of fisheries investigation a consolidation of the Districts and Authorities on each of the three great coast lines of England and Wales—East, South and West. It may be worth while to reproduce once more the accompanying rough sketch plan in order to bring this idea clearly before the mind and to show how the three suggested Districts form natural coast areas. Mr. Fell has since shown\* that there would be advantages in the amalgamation of the present Committees on each coast, not only for purposes of investigation but also for administration. He has gone into the question of probable income and expenditure and finds that each such large area would have a rateable value of about thirty millions sterling, yielding, on the rate of one-sixteenth of a penny in the pound, a sum which, according to our Lancashire scale of expenditure, ought to be ample for the proposed work. In these calculations London is left out of account. Its contribution, from a similar rate, if obtainable, might be applied to general central expenses or to special work applying to the whole country.

Looking at the West Coast as now administered it is obviously unnatural and inconvenient that the large

\* I quote from a letter and from a speech by Mr. Fell. So far as I am aware his figures have not yet been published.

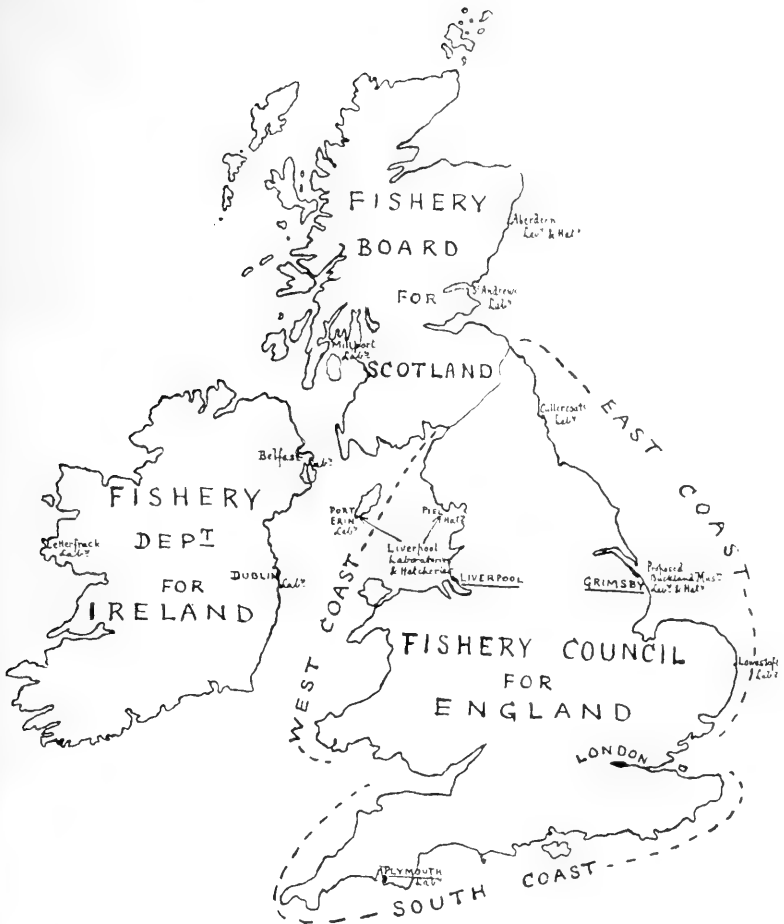


FIG. 1.—Sketch map of the British Islands for the purpose of indicating the positions of the chief marine laboratories and sea-fish hatcheries, and the proposed division of the coast of England into three great fisheries districts—the East coast, the South and the West—as recommended by the Ichthyological Committee.

Lancashire and Western area should be cut off from the Solway Firth to the north and from the Bristol Channel to the south. In studying the distribution and movements of plaice throughout the seasons of the year, and the life of the fish, it is evident, even from our few observations during the last couple of years, that the great shallow water areas of the Solway are of considerable interest. This is recognised by the Fishery Board for Scotland as well as by ourselves, and on more than one occasion now they have asked us to make observations for them from our steamer, and to aid them in obtaining young fishes for their transplantation experiments. We are also at the present time preparing to co-operate with the Fishery Board in certain drift-bottle experiments for the purpose of determining the movements of the surface waters of the Irish Sea and the Clyde Sea-area, such as may affect the drift of fish eggs and larvæ. This is as it ought to be, and if the whole west coast from the Solway to the Severn were in the control of one Authority, efficiently equipped with boats and observers, an organised co-operation with the Fishery Board for Scotland would be natural, and a joint investigation of the Solway and of any other problems of common interest would undoubtedly be effected.

The Isle of Man should also join this proposed western amalgamation. The seas round that Island are closely related to the Lancashire waters, and whatever the fishermen may be required to do, the fish respect no territorial boundaries. The Isle of Man has everything to gain by coming into line with the English counties. To administer the insular fisheries alone would be an extravagant process. Joined with Lancashire and the other counties in a western combination, in return for a very moderate rate the Manx fisheries would be adequately

exploited and policed the regulation and administration would be the same as over the adjoining English and Welsh coasts. We should then have in this northern part of the Irish Sea a natural sea-fisheries district administered by one Authority, having one steamer for police regulation and another for scientific investigation, having central laboratories in the University of Liverpool, with Marine Stations and Hatcheries at several distant points, probably in the north of Lancashire (Piel), in the Isle of Man (Port Erin), and somewhere on the coast of Wales. The co-relation between such laboratories and those on the other coasts of England, and again between England, as a whole, and Scotland, Ireland and, it may be, other Countries, might well be as was outlined in the Report of the Ichthyological Committee. If such a national scheme of fisheries co-ordination were carried out it would lead, in addition to increased efficiency of administration, to a marked increase in the scientific knowledge of our fishing grounds, the absence of which successive Select Committees and Conferences have had to deplore.

W. A. HERDMAN.

THE UNIVERSITY, LIVERPOOL,

*January, 1904.*

## SEA FISH HATCHING AT PIEL.

By ANDREW SCOTT, A.L.S.

In the operations carried on during the fish hatching season of 1903, the eggs of plaice (*Pleuronectes platessa*) and flounder (*Pl. flesus*) were again dealt with. At the beginning of the year there were 100 mature plaice and 200 flounders in the tanks. The plaice, as in former years, were brought from the closed waters of Luce Bay by the fisheries steamer, and the flounders were collected in Barrow Channel by Mr. Wright. The fish, on the whole, were not so large as those used in the hatchery work of 1902, but from the increase of numbers, and by maintaining a good circulation of water, we were able to improve upon the results then obtained.

Under the present system we have probably nearly reached the maximum number of fish that can be accommodated with safety. Further development, as we pointed out in last year's Report, can only be secured by an addition to our present resources. The fish hatchery at Bay of Nigg, and the hatchery at Port Erin have both open air ponds which are of immense use in all hatching work, and must be considered as an essential to further progress.

The first fertilised eggs were collected and placed in the hatching boxes on March 9th, and the last on May 7th, so that the spawning season with us extended over a period of two months, or about two weeks less than in the previous year. During the spawning season nearly seventeen millions of eggs were collected and incubated and those eggs produced close on fifteen millions of fry which were set free as before near the centre of Morecambe Bay. The periods of incubation of the plaice and flounder eggs were practically the same as last year, from



ten to seven days for flounder, and from seventeen to fifteen days for plaice. The total loss during incubation from all causes was just under 11 per cent.

The following tables show the numbers of eggs collected and of the fry set free on the dates specified.

## PLAICE.

		Eggs Collected.	Fry Set Free.			
March	9	... 40,000	35,000	...	March	26
"	12	.. 50,000	44,000	...	April	8
"	17	... 50,000	44,000	...	"	"
"	21	... 50,000	44,000	...	"	"
"	23	... 50,000	43,500	...	"	"
"	26	... 60,000	53,000	...	"	16
"	30	... 70,000	62,000	...	"	"
April	2	... 60,000	53,000	...	"	27
"	4	... 70,000	62,000	...	"	"
"	7	... 60,000	53,000	...	"	"
"	9	... 70,000	62,000	...	"	"
"	11	... 60,000	53,000	...	"	"
"	13	... 60,000	53,000	...	May	5
"	15	... 60,000	53,500	...	"	"
"	17	... 70,000	62,000	...	"	"
"	20	... 70,000	62,000	...	"	"
"	22	... 60,000	53,000	...	"	13
"	25	... 60,000	53,000	...	"	"
"	27	... 70,000	62,000	...	"	"
"	30	... 70,000	62,000	...	"	22
May	2	... 60,000	53,000	...	"	"
"	4	... 70,000	62,000	...	"	"
"	7	... 60,000	53,000	...	"	"
Total Eggs		<u>1,400,000</u>	<u>1,237,000</u>	Total Fry		

## FLOUNDER.

		Eggs Collected.	Fry Set Free.	
March	9	... 200,000	178,000	... March 26
"	12	... 470,000	428,000	... April 8
"	17	... 470,000	428,000	... " "
"	21	... 480,000	427,000	... " "
"	23	... 500,000	445,000	... " "
"	26	... 480,000	428,000	... " 16
"	30	... 600,000	532,000	... " "
April	2	... 600,000	532,000	... " 27
"	4	... 800,000	713,000	... " "
"	7	... 700,000	623,000	... " "
"	9	... 900,000	800,000	... " "
"	11	... 1,000,000	890,000	... " "
"	13	... 900,000	800,000	... May 5
"	15	... 700,000	623,000	... " "
"	17	... 800,000	712,500	... " "
"	20	... 800,000	712,000	... " "
"	22	... 800,000	713,000	... " 13
"	25	... 800,000	712,500	... " "
"	27	... 1,000,000	890,000	... " "
"	30	... 700,000	623,000	... " 22
May	2	... 600,000	532,000	... " "
"	4	... 600,000	532,000	... " "
"	7	... 400,000	356,000	... " "
Total Eggs		<u>15,300,000</u>	<u>13,630,000</u>	Total Fry.

Total Number of Eggs ... .. 16,700,000

Total Number of Fry ... .. 14,867,000

## CLASSES, VISITORS, &amp;c., AT PIEL LABORATORY.

BY ANDREW SCOTT.

The Fishermen's Classes, conducted at Piel under the auspices of the Lancashire County Council and the Sea-Fisheries Committee, have now become an established institution. Each year they are held the competition for places gets keener. The grant given by the County Council in 1903 enabled us to take forty-five men, fifteen more than in the previous year. Applications for places were to be sent to Mr. Dawson, and by the beginning of March no less than sixty-two names had been received, all of fishermen from between Southport and Roosebeck. Three classes of fifteen men to each were formed, and the selected men were notified regarding the date and times of attendance. They all duly presented themselves at Piel and went through the course of instruction as in previous years. At the end of each class the usual votes of thanks to the Sea-Fisheries Committee and the County Council for the privileges afforded the men for acquiring a better knowledge of the life histories and habits of the economic marine animals were duly proposed and carried. Some of the men expressed the hope that in the near future a second and more advanced course would be possible for those who had already gone through the first one. The suggestion has also been made by some Members of the Committee that one or two prizes, such as a small microscope and some pocket lenses, might be offered for competition in each class. This would have the additional advantage of encouraging some of the men to continue the work after returning to their homes.

Mr. Johnstone, from the Liverpool Laboratory, again had charge of the classes, and the following are the names of the men who attended:—

Class held March 9th to 20th.—J. J. Peet, Lytham; Thomas Newsham, Lytham; John Leadbetter, Fleetwood; Ernest C. Leadbetter, Fleetwood; David Moss, Fleetwood; John Colley, Fleetwood; James Carter, Morecambe, James Johnstone, Morecambe; Wilfred Woodhouse, Morecambe; J. G. Gardner (Secretary Fishermen's Association), Morecambe; Hugh Rimmer, St. Anne's; James Robinson, Southport; William Jackson, Southport; Thomas Wright, Southport; John Wright, Southport.

Class held March 23rd to April 3rd.—Lawrence Abram (Ned's), Banks; John Wareing (Stephen's), Banks; Richard Sharples, Banks; Richard Wright, Marshside; Nicholas Wright (Selby), Marshside; John Harrison, St. Anne's; Harry Melling, St. Anne's; John C. Pegler, Fleetwood; T. P. Ball, Jr., Fleetwood; William Wilson, Fleetwood; Thomas Wilson, Fleetwood; James Bond, Morecambe; George Bond, Morecambe; William Brown, Morecambe; John Birkett, Morecambe.

Class held April 20th to May 1st.—Robert Wright, Marshside; Jeffrey Ball, Marshside; Robert Harrison, St. Anne's; Nicholas Parkinson, Lytham; Thomas Whiteside, Lytham; Robert Clarkson, Lytham; Fred. H. Pegler, Fleetwood; Richard Bond, Morecambe; Richard Bond, Morecambe; Walter Bell, Morecambe; James Cocking, Morecambe; William Johnson, Ulverston; J. Bouskill, Flookburgh; James Butler, Flookburgh; David Wilkinson, Baicliff.

In addition to these Classes for Fishermen, two courses in Nature Study for School Teachers were arranged, and proved a successful experiment. The first one was attended by seventeen head masters, head mistresses, and assistants from the schools under the Barrow School Board. It was held on two evenings and

the Saturday afternoon of each week during the last of the series of Fishermen's Classes. The second class was held by desire of some of the Morecambe teachers, and was taken advantage of by the head master and first assistant from three of the schools. The Barrow teachers travelled to and from Piel by train each day. The six Morecambe teachers came during the Whitsuntide vacation and lived in the establishment during that week. Four hours' instruction were given on the Tuesday, Wednesday, Thursday and Friday, and the men spent the remainder of the time investigating the shores and neighbourhood. The first class was conducted by Mr. Johnstone and myself: the second one, in the unavoidable absence of Mr. Johnstone, I carried on alone. The course in each case was a practical one, and instruction was given in the structure, life history and habits of common marine animals, such as the cod, the shore crab and its allies, the cockle, the mussel, the oyster, microscopic life in the sea water collected by the tow-net, the various animals living on the shore between tidemarks, and material washed up from the sea bottom.

Much interest was shown by the teachers in the work, and from the remarks made at the conclusion of the course, it was evident that a continuation of these Nature Study Classes at Piel would receive much support from school teachers in Lancashire. For teachers resident in Barrow, evening and Saturday meetings are most convenient; for others, vacation courses alone would be possible, unless other arrangements, involving leave of absence, could be secured.

To the school teachers of Lancashire, Piel offers many advantages for the study of common marine animals and plants. From a distance the shore at low water may seem very uninteresting and barren, but a more careful

examination shows that it is far from being so. Many species of Molluses occur, including the ordinary mussel, some of which here produce pearls in abundance; the sand is teeming with the ordinary "lug-worm," with its beautiful external branchial plumes; various forms of crustacea abound, and there is a varied assortment of fishes. The rough scars provide many species of seaweeds, and in the spring and early summer the water contains a good general floating fauna and flora which can be collected by tow-net. Even the debris washed up by storms and strewn along high water mark, as we have found from experience, proves a mine of interest. To the teacher, where time is a consideration, it may be pointed out that it is possible to leave Manchester or Liverpool by 5-45 p.m. train and arrive at the Piel Laboratory at 8-45 p.m. the same evening. Other trains from Barrow to Piel are also run. There are also convenient trains for returning.

On January 28th a meeting of representatives from the various Technical Instruction Committees in the county was held at Piel, for the purpose of hearing an address from Professor Herdman, on "Technical Instruction in Sea-Fisheries Science" (see last year's Report), and also to inspect the equipment of the Laboratory with a view to teaching fishermen and others.

The Special Subjects Committee of the Barrow School Board visited the establishment during the course of the second fishermen's class in order to see the men at work. They seemed much impressed by the keen interest displayed by the fishermen in the instruction given and in their practical work.

The Chairman and Members of a number of the Lancashire local Technical Instruction Committees under

the leadership of Mr. James Fletcher and Mr. Dawson, paid an inspection visit during the course of the third fishermen's class. Several members of the party delivered short addresses to the men.

Mr. Thomas Baxter and party from Morecambe visited the establishment while the Morecambe school teachers were at work.

Another meeting of Representatives from the Technical Instruction Committees of the Lancashire County Boroughs was held in the Laboratory at the end of July. The meeting was for the purpose of further developing the facilities for the teaching of Marine Natural History to fishermen, school teachers and others. Mr. Fell, Mr. Ragdale, and other members of the Committee addressed the Representatives. They pointed out the advantages that the Piel establishment presents, and the ease of access to it from all parts of the county. Reference was made to the satisfactory reports on the teaching work already accomplished under more or less temporary conditions.

The St. James' Rambling Club and the Barrow Naturalists' Field Club also visited the establishment during the summer.

Many visitors have been shown through the Laboratory and tank-house. We have also had an inspection visit from Mr. Fryer, of the Board of Agriculture and Fisheries. Professor Herdman and the late Mr. I. C. Thompson, F.L.S., from Liverpool, stayed over a week end in order to make some investigations, in addition to shorter visits at other times during the year.

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## TRAWLING OBSERVATIONS AND RESULTS.

By JAS. JOHNSTONE.

(1) Six Hauls in Luce Bay, on  
October 26th, 1903.

By permission of the Fishery Board for Scotland the Lancashire and Western Sea Fisheries Committee's steamer was enabled to make a series of hauls with a trawl-net in the preserved waters of Luce Bay, on October 26th, 1903. The primary object of these trawling operations was to obtain a stock of mature living plaice for the Committee's Sea-Fish Hatchery at Piel. Advantage was taken, however, of the opportunity thus afforded to compare the results usually obtained by fishing on the plaice grounds within the Lancashire and Western Sea Fisheries area, where trawling is permitted under certain restrictions, with those obtainable by fishing in waters strictly preserved against all forms of trawling. In order to obtain the fish in as healthy a condition as possible a series of short hauls were made with a rather small trawl-net of 30 feet beam, and with 7 inch meshes throughout. By using such a net a very limited number of invertebrates were captured, and few small fish were obtained. The smallest plaice obtained in any of the hauls was  $8\frac{1}{2}$  inches in extreme length. The results of these hauls are given in the table on following page.

*Physical observations during the hauls.*

Wind, S.W., light;

Sea, smooth;

Weather, unsettled;

Barometer, 29·4 to 29·6 inches;

Air temperature, 10°·5 C. to 11°·6 C.;



RESULTS OF 6 HAULS IN LUCE BAY ON OCTOBER 26TH, 1903.

FISH CAUGHT.	1st HAUL. 1½ hours duration, 2½ miles long.	2nd HAUL. 1½ hours duration, 2½ miles long.	3rd HAUL. 1½ hours duration, 2½ miles long.	4th HAUL. ¾ hour duration, 1½ miles long.	5th HAUL. 1½ hours duration, 2½ miles long.	6th HAUL. 1 hour duration, 2 miles long.
Plaice under 14 in. in total length.	53	406	42	44	333	168
Plaice over 14 in. in total length...	15	32	9	20	32	29
Dabs .....	10	68	68	15	64	12
Ray ( <i>Raja clavata</i> ) .....	32	40	26	20	12	68
Ray ( <i>Raja circularis</i> ) .....	1	...	...	...	...	...
Soles .....	1	4	1	...	1	3
Flounder .....	...	1	...	...	...	...
Codling .....	...	...	...	...	...	1
Ling .....	1	...	...	...	...	...
Grey Gurnards .....	2	...	...	...	...	...
Total Fishes caught.....	115	551	146	99	442	281

Sea temperature at surface,  $10^{\circ}4$  C. to  $11^{\circ}5$  C.;

„ „ bottom,  $10^{\circ}6$  C. to  $11^{\circ}2$  C.;

Specific gravity of sea at surface, 1.025 to 1.027;

„ „ bottom, 1.025 to 1.027.

The positions of the hauls are shown on the sketch chart below.

The results of these hauls demonstrate strikingly the restricted distribution of fishes on an area of very limited

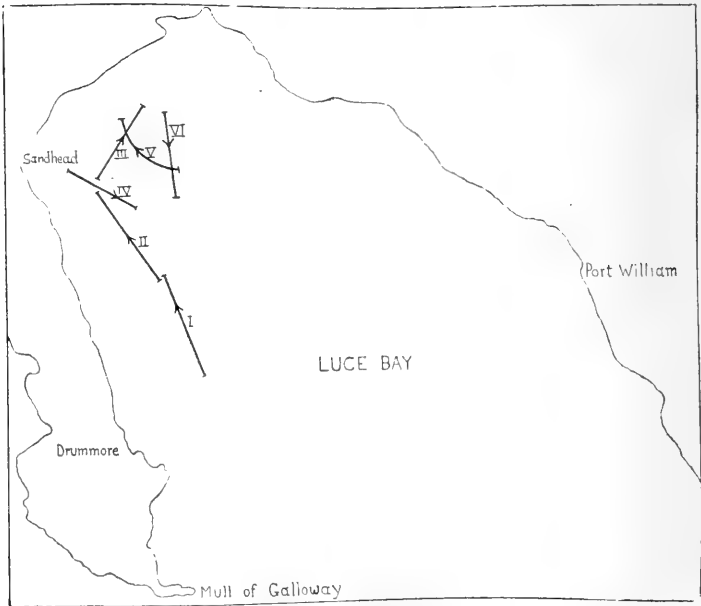


Chart of Luce Bay.

extent, and over which the physical conditions, which might be supposed to influence the abundance and nature of the bottom fauna, are very similar. The chart shows that fishing was restricted to the west and north-west margins of the bay. The bottom was uniformly sandy, and the depth varied from 9 to 5 fathoms. Hauls 1 to 3 are comparable in all respects; the depth varied from

7 to 9 fathoms, the duration and length of the drags were identical, and all three were made dragging with the stream. Nevertheless, the number of plaice taken varied from 51 to 438, the dabs from 10 to 68, and the ray from 26 to 40. The fifth haul was made dragging in towards shallow water. The fourth and sixth hauls were shorter than the others, being of one and three-quarters and two miles length respectively. They differed, however, to an extent which is not accounted for by their difference in length, as the fourth yielded only 64 plaice, while 197 were obtained in the sixth. The sixth haul, too, was made in the dark when, according to general experience, plaice are more difficult to catch in the trawl net.

There is a general similarity between the results of the first three hauls obtained on this occasion and those obtained on October 21st, 1902 (see last year's Report, p. 85). The positions, states of the tide and lengths of drag are very approximately the same in each series. Both the first hauls, dragging north from Drummore yielded poor catches (14 and 68 plaice, respectively), the second hauls gave much better results (438 and 157 plaice), while the third were again poor (17 and 51).

The results obtained on the present occasion confirm then those obtained in 1902, that is, that a trifling difference in position of the ground dragged over may correspond with very remarkable differences in the volume of the catches. This, indeed, is well known to fishermen, but it is a factor which has not been sufficiently recognised in statistical trawling operations, where it has been generally assumed that the distribution of fish on a large area is fairly constant.

*Sizes of the plaice obtained.*

The main object of the trawling operations was to obtain mature plaice, and in each haul all the fish of over

14 inches in extreme length were at once separated out and brought back alive. The number of these obtained in each haul is given in the Table, and is about 11·5 per cent. of the total number caught. The sizes of the fish caught were, so far as could be judged by a mere inspection, very similar in each haul, and it was considered sufficiently exact to measure the individual fish in one catch only, and assume that the average so obtained applied to all the others. This was done for the first haul and an average length of about 11·1 inches was obtained. As the net used had a uniform mesh of 7 inches ( $1\frac{3}{4}$  inches along each side), and the drags were short ones, no plaice of less than 8 inches in length were obtained.

The size of the plaice obtained in the preserved waters of Luce Bay is in striking contrast to that of the plaice which inhabit the corresponding areas in the Lancashire and Western Fisheries District. We have no reason to suppose that Luce Bay is a better feeding ground, or differs in any essential respect from many plaice grounds in the former area. The nature of the bottom, the depth and physical condition of the water, the bottom and pelagic faunas resemble closely the corresponding conditions on many extensive areas on the North-west Coast of England. Still, there is no inshore area known to us where 10 per cent. of the plaice caught are over 14 inches, and a fair proportion are 18 or 20 inches in length. The only difference is that Luce Bay has been closed against trawling of every kind for about 16 years, while every plaice ground off the Lancashire and Cheshire coast is the scene of an active trawl fishery, with only very limited restrictions in force. There appears to be no doubt that the comparatively large size of the plaice on this portion of the Scottish coast is directly due to the prohibition of trawling, and conversely that the much smaller average

size of the plaice further south results from the practically unrestricted exercise of this method of fishing.

The size of the plaice captured also suggests some doubts as to the accuracy of the generally accepted theory of the life history and migrations of that fish. It is usually stated that the mature plaice inhabits and spawns in comparatively deep water (10 to 20 fathoms), that the fertilised eggs and larvæ drift inwards towards shallow water, and that as the fish grows it migrates outwards towards the 20 fathom line. The observations made in Luce Bay, where plaice of 20 inches in length were found close in towards the shore in water of six fathoms and less in depth, show, however, that the above distribution is not universal, and that there is no necessary relationship between the depth of water and the size of the fish on the bottom.\* Large plaice are found in Luce Bay because trawling is prohibited there and they are not interfered with. It is, however, reasonable to suppose that the absence of mature plaice on the inshore grounds of such an area as the Lancashire and Cheshire District is due to the great extent to which these grounds have been fished over. The amount of fishing on the inshore grounds is much greater than in deeper water, and the fish on the latter area are much less disturbed. The effect of this extensive fishing inshore has been to reduce greatly the average size of the plaice present, and it is to be noted that investigations into the life history of this fish have been made in comparatively recent times—since the exploitation of the inshore grounds by the trawlers on the modern scale has taken place. This effect of trawling in reducing the average size of the fishes present on a ground is well known, and was first demonstrated by

\* Our observations, however, have been confined to the months of October and November. Possibly the large plaice may migrate from the Bay in winter and during the spawning season.

McIntosh in regard to certain fisheries on the east coast of England and Scotland.\*

*Invertebrates taken in the trawl net.*

No special attention was paid to the bottom invertebrate fauna, but the following forms were identified among the contents of the net:—

*Mytilus modiola*, *Acyonidium*, various compound and simple ascidians, *Flustra*, *Pecten*, *Asterias*, *Solaster*, *Astropecten*, *Echinus*, *Cucumaria frondosa*, *Ophioglypha*, *Fusus*, *Dentalium*, *Hyas*, *Stenorhyncus*, *Pagurus*, *Portunus*, *Porcellana*, *Aphrodite*, *Sabella* and other Polychaetes, and *Actinoloba*. *Pontobdella* (from a ray) was also taken. *Zostera* was present in the North Western portion of the bay. A female lobster of  $8\frac{3}{4}$  inches in length was caught in the first haul, and two females, one recently berried, 10 inches in length, were obtained in the third haul.

*Food of the fishes taken.*

The majority of the fishes were examined for food contents of the stomach. Ray were feeding on fishes (too much decomposed for identification); plaice and dabs on *Scrobicularia* and *Nucula* (especially the latter), and the soles on annelids with a few *Scrobicularia*.

*Plankton.*

Surface tow-nettings were taken during every haul except the last. The pelagic organisms present were, however, remarkably scarce (not more than 10 cc. in all the five hauls), and the only animals present were *Pleurobrachia* (relatively abundant), *Caligus rapax* (male), a young *Cyclopterus lumpus*, some larvæ decapods, and the Copepods *Paracalanus parvus*, and *Oithona similis*. This is in marked contrast with the fishing operations in October, 1902, when an enormous catch of Copepods (over

\* See Report of the Trawling Commission, Appdx. A., p. 378, and Report, p. xvi.; 1885.

250 cc.) was obtained in the second drag. These consisted chiefly of *Acartia discaudata*, with a few *Acartia clausi* and *Temora longicornis*.

*Average catches.*

These were greater in 1903 than in 1902, as the following table shows:—

Average catch of fishes, 1902	...	...	179.			
"	"	"	1903	...	...	272.
"	"	plaice, 1902	...	...	132.	
"	"	"	1903	...	...	197.

(2.) Two hauls in Wighton Bay on  
November 9th, 1903.

I. BEAM TRAWL of 30 ft. beam and with 4-inch meshed pockets and tails.

Dragging N.N.E. for  $1\frac{1}{4}$  hours, and for a distance of  $2\frac{1}{2}$  miles towards Innerwell Point. 8-45 a.m. to 10-0 a.m.,  $1\frac{1}{2}$  hours' ebb.

Plaice	...	...	...	47	...	$4\frac{1}{2}$ in. to 12in.
						(avge. size about 9in.).
Sole	...	...	...	1	...	9in.
Dabs	...	...	...	320	...	4in. to 10in.
Ray	...	...	...	22	...	3in. to 26in.
Skate	...	...	...	1	...	12in.
Whiting	...	...	...	122	...	6in. to 10in.
Codling	...	...	...	3	...	6in. to 13in.
Yellow gurnard	...	...	...	1	...	$4\frac{1}{2}$ in.
Herring	...	...	...	2	...	7in. to 8in.
Poor-cod ( <i>Gadus minutus</i> )	...	...	...	12	...	4in. to 8in.
Total	...	...	...	531		

II. SHANK NET, 12ft. wide,  $\frac{1}{2}$ -inch mesh; one haul off Innerwell Point for  $\frac{1}{4}$  hour and for  $\frac{1}{2}$  mile; 10-15 to 10-30 a. m.

Shrimps	...	...	...	1	pint.
Dabs	...	...	...	12	„ ... $1\frac{1}{2}$ in. long.
Plaice	...	...	...	1	„ ... 8in. „
Poor-cod	...	...	...	1	„ ... 4in. „
Gobies	...	...	...	6	„
Pipe-fish	...	...	...	3	„

During these hauls the following observations were made:—

Wind, fresh W.S.W.

Weather, fine; sea smooth;

Barometer, 30.5;

Air temperature, 80.6 C.;

Sea temperature at surface, 90.8 C.;

„ „ bottom, 100.0 C.;

Specific gravity at surface, 1.0242;

„ „ bottom, 1.024;

Depth, 4 fathoms; transparency, 5 feet.

The common invertebrates taken in the nets during these hauls were:—Oysters (about 12), *Turritella* (very numerous), *Pagurus*, *Buccinum*, *Portunus*, *Polynoe*, *Ophiura*, *Serpula* (on the oysters), small actinians (on the oysters), *Scrtularia*, *Asterias*, *Solaster*, and one male lobster  $11\frac{1}{2}$  inches long. Only one tow-netting was taken, but very few organisms were obtained, the commonest being *Pleurobrachia*.

The object of these hauls was to ascertain whether small plaice (3 to 6 inches long) were present to any great extent in Wigton Bay. It was possible to find time for only two short hauls on the west side of the Bay, and these yielded very few small fish of the size in question.



(3.) Five hauls in Luce Bay on  
November 9th, 1903.

One haul was made with a trawl net with 4-inch meshes, the object being to ascertain as before whether small plaice were present in the Bay in any abundance. The following fishes were taken:—

Plaice	...	...	7	...	14	—18	inches	long.
„	...	...	127	...	6	—13	„	„
Dabs	...	...	285	...	5	—9	„	„
Ray	...	...	105	...	4 $\frac{1}{2}$	—16	„	„
Yellow gurnards	6	...		...	6	—7	„	„
Whiting	...	6	...	...	6 $\frac{1}{2}$	—8	„	„
Herrings	...	3	...	...	7	—7 $\frac{1}{2}$	„	„
Soles	...	3	...	...	8	—11	„	„

Total ... 542

Very few small plaice (6—8 inches long) were captured.

Four hauls were then made with a net of 7-inch mesh on the north and west sides of the Bay, the results of which are as follows:—

	I. 2 $\frac{3}{4}$ miles.	II. 2 miles.	III. 2 miles.	IV. 1 $\frac{3}{4}$ miles
Plaice .....	15, 14-20 in. long.....	31	9	12
Plaice .....	120, less than 14 in. long.	545	92	136
Dabs .....	79 .....	155	77	40
Ray .....	50 .....	40	4	30
Angler Fish	... ..	1	...	...
Total .....	264 .....	772	182	218

Three of the ray captured in haul III. are the largest we have taken in the Bay; they measured 34, 36, and 48 inches respectively. Large mature plaice were, however, not so abundant as they were on October 26th, and as the trawling was interrupted by bad weather, only about 80 such fish were obtained.

*Physical observations.*

Wind, strong from W.N.W. and N.W.;

Weather, unsettled, some rain;

Sea, choppy;

Barometer, 30.5 in.

Air temperature, 9°·4 C. to 11°·7 C.;

Sea temperature at surface, 9°·4 C. to 10°·5 C.;

Specific gravity at surface, 1.0258 to 1.026;

Transparency of sea, about 5 feet.

*Invertebrates taken.*

These were similar to those taken on October 26th. Townettings only were taken on two hauls, but the organisms captured were very few, principally *Pleurobrachia*.

(4.) F o u r h a u l s o f f t h e I s l e o f M a n .

Several hauls with a 7-inch meshed trawl net were made off the North end of the Isle of Man between Jurby Point and Point of Ayre, the object being to obtain some large mature plaice for the Port Erin Hatchery.

The hauls were made in water of from 9 to 5 fathoms in depth and about half-mile from the shore, the direction of the drags being towards Point of Ayre. The ground was very rough, and the net caught three times, and had to be hauled. Five plaice were caught in these first three hauls. Two were large females, one being 19 $\frac{3}{4}$  inches and the other 23 inches long. The latter weighed 4 lbs. 11 ozs. The others were smaller fish, and

one shewed a remarkable malformation, which was evidently the result of an accident. The fourth haul was about two miles long; the net was, however, torn, and only 2 plaice (15 inches and 8 inches long) and 1 lemon sole (8 inches long) were taken.

*Physical observations.*

Wind, W. light breeze;

Weather, fine;

Sea, smooth;

Barometer, 30·85;

Air temperature, 10°·5 C.;

Sea temperature at surface, 10°·85 C.;

„ „ bottom, 11°·25 C.;

Specific gravity at surface, 1·0258;

Transparency, 20 feet in hauls 1-3, and 12 feet in haul 4.

No tow-nettings were taken during these hauls, and, in consequence of the catching and tearing of the net, very few invertebrates were taken. A dog-fish purse, with a well-advanced embryo, was picked up during the last haul.

The size of the plaice taken in these hauls is very noticeable, when it is remembered that they were made in territorial waters and in comparatively shallow water—the 23in. plaice was caught in 7 fathoms. It is apparent that large plaice are relatively abundant at the North end of the Isle of Man, and, as clean trawling ground is to be found there, there should be no difficulty in supplying the Port Erin Hatchery from its own area in a very convenient manner.

The malformed plaice (Plate I.) caught in the third haul presents a very interesting peculiarity, in that a notch of about 2 inches long has been taken out of the

anal fin and the adjacent ventral portion of the body. It is, of course, impossible to say how this injury occurred, but the shape of the notch suggests a bite by some carnivorous fish. The injury had probably occurred when the fish was much younger. Healing has been perfect, for there is no obvious cicatrix, and the pigmentation of the ocular side extends round the ventral margin on to the blind side. That this perfect healing has occurred is remarkable, for this part of the body is fairly well supplied with blood-vessels, and bleeding must have been copious. Dr. Fulton describes a somewhat similar case.\*

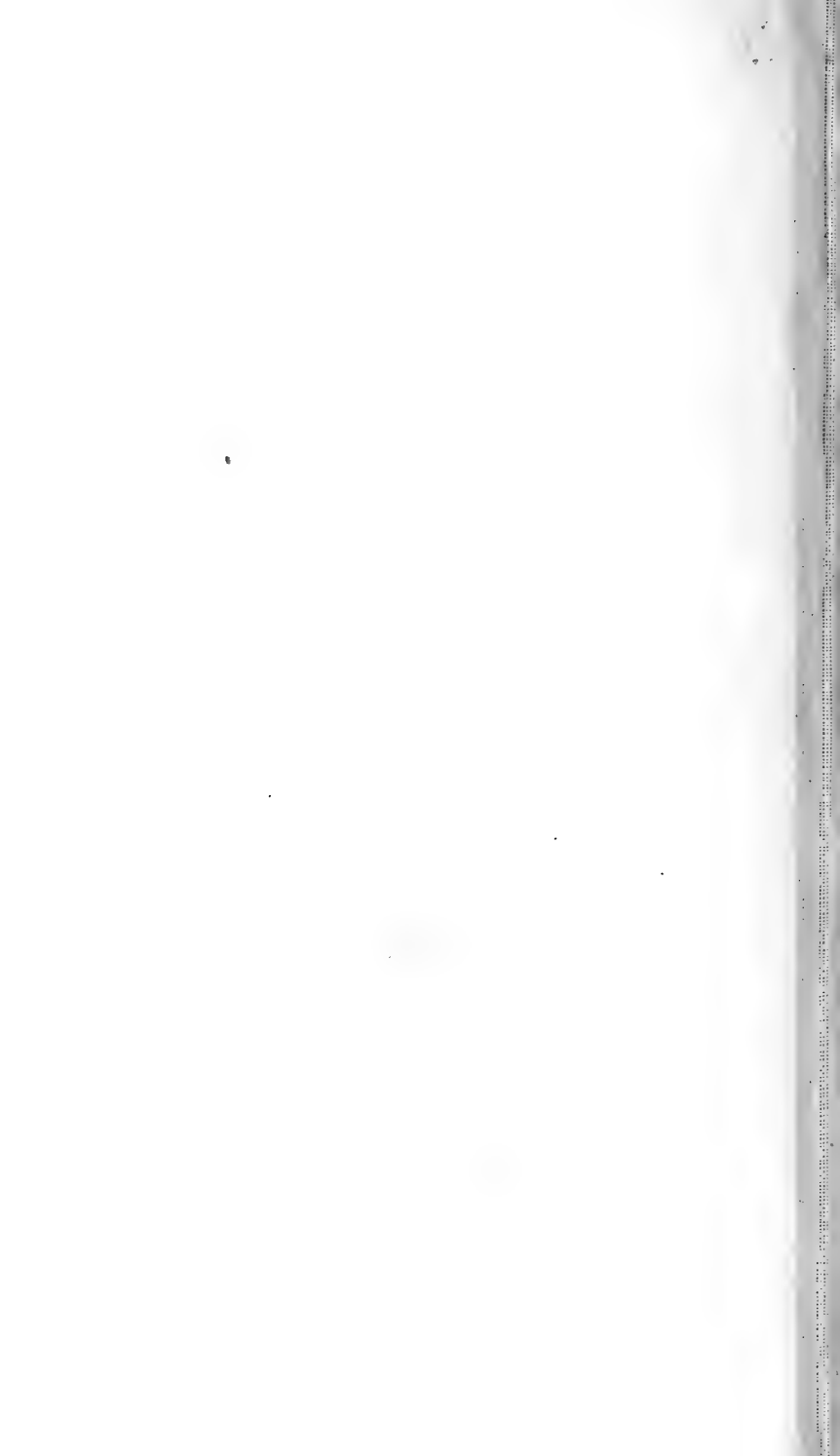
The fish is a female, of  $15\frac{1}{2}$  inches in extreme length. It ought to be mature, but the ovaries are small and thin, that of the blind side being only  $1\frac{3}{4}$  inches long. Probably the injury had inhibited the maturation of these organs. It is interesting that the white haloes round the ocelli are not clearly evident in this fish, and this lends support to Petersen's contention† that these white-edged ocelli in the plaice are indicative of the condition of sexual maturity.

\* Rept. Fish. Bd., Scotland, No. 21.

† Publications de circonstance, No. I. Cons. Perm. Internat. Explor. de la Mer. Copenhagen, 1903.



MALFORMED PLAICE.



## SOME PARASITES FOUND ON FISHES IN THE IRISH SEA.

By ANDREW SCOTT.

No subject offers a more enticing attraction to the Zoological student than the one that deals with those members of the animal kingdom which through some acquired habit have become partly or altogether dependent on other animals for their food. This branch of research has been very prominently brought to the front within the last two years by the further investigations relating to the formation of pearls in the common mussel of our sea shores, by Dr. H. L. Jamieson, and of the extremely valuable oriental pearl from the Ceylon pearl oyster, by Professor Herdman and Mr. James Hornell.

It has long been known that the completion of the life cycle in some parasites, such as the "liver fluke" in sheep, is wholly dependent on part of the development being carried on in another animal; and it is becoming more and more evident that the life-cycle of these parasites can only be completed when the creature has successfully passed from one stage to another, and reached its final host where reproduction takes place.

The difficulties that await the student who contemplates investigating the life histories of parasites are great, but by no means insurmountable. Extreme care and much patience are necessary, but the work is well worth the labour expended and there is still much to do.

In the following notes a list of the parasites that have been recorded from the fishes caught in the Irish Sea is given, with remarks on some of the more important points connected with each. It is probably far from exhaustive, but will form a foundation for future additions. Up to

the present we have four species of Protozoa, ten species of Vermes (Trematodes) and forty-six species of Crustacea (Copepoda).

The Protozoan parasites are found on and in various parts of the fish, such as the brain, the intestines, and the skin. In a few instances the presence of these parasites is easily recognised, but in the majority, careful dissection and examination of the tissues is required before they can be detected. The most noteworthy appearance of the presence of Protozoan parasites that has come under our notice is the one on the skin of the ordinary "white fluke," *Pleuronectes flesus*. It usually takes the form of small white globular bodies attached to the skin of the body and fins. In a specimen recently secured, the upper and lower lips of the mouth were thickly covered with the cysts, which doubtless interfered with the fish securing food. The gills were also much infested. Occasionally larger masses, sometimes the size of a marble, may be present. This disease in the white fluke has attracted considerable attention for many years, and is, no doubt, the origin of the theory long held by fishermen that white flukes carried their eggs attached to their body. We have a species of *Glugea* recorded from the plaice, a Sporozoan from the flounder, which is probably a new genus, also *Glugea lophii* and another new form, *Sphaerospora platessa*, which infests the auditory capsule of the plaice. The two new species will be discussed in another section of this Report, in special papers by Mr. Woodcock.

The Trematode parasites may be found attached to the gills, or skin, and, in the case of one species, even in the Cloaca. Occasionally more than one species may be found attached to the gills of a single fish. These parasites are of various sizes, and usually colourless, except as regards their excretory and reproductive systems. The



posterior sucker may be entire and circular in outline, or divided into pairs of separate suckers. One genus has a small secondary or "adhesive" disc attached to the large one. These parasites live for some time after the death of the fish and adhere very closely to the gills or skin, in fact it is sometimes quite impossible to detach them without injury. A very convenient way to remove them is to leave the gills, or portions of the skin to which they are attached, in fresh water for some hours; this treatment eventually kills the parasite and at the same time causes it to relax its hold.

I.—Species with posterior sucker entire:—

*Callicotyle kroyeri*, Diesing.

From the cloaca of *Raia clavata*. It is also found on other rays, but always in the cloaca. The posterior sucker has seven strong bands radiating from an inner circle, the spaces between the rays are conical in outline with the apices towards the centre.

*Phyllonella soleae*, van Beneden and Hesse.

Attached to the scales on the "white side" of the common sole. This trematode is of moderate size but easily overlooked owing to the colour resembling that of the fish.

? *Placunella pini*, van Beneden and Hesse.

This trematode which we identify as *P. pini* has only been found on the gills of the yellow gurnard, *Trigla hirundo*. It has eight distinct and two indistinct rays in the posterior sucker.

*Deplectanum æquans*, Diesing.

Found on the gills of the "bass." A very small species and sometimes abundant.

II.—Species with no distinct posterior sucker, having

instead, a number of cupules which may be either stalked or sessile.

*Phyllocotyle gurnardi*, van Beneden and Hesse.

On the gills of the common gurnard, *Trigla gurnardus*, and yellow gurnard. A small species and easily overlooked. Three pairs of sessile cupules.

*Microcotyle labracis*, van Beneden and Hesse.

On the gills of the "bass," *Labrax lupus*. A moderately large species with a number of sessile cupules, but not common on any of the fishes examined by us.

*Octobothrium scombri* (Kuhn).

On the gills of the common mackerel. A very slender species and easily overlooked. Four pairs of sessile cupules.

*Octobothrium merlangi* (Kuhn).

On the gills of the whiting. A large species of a dark colour. Four pairs of cupules on short stalks.

*Dactylocotyle pollachii*, van Beneden and Hesse.

On the gills of the pollack. A large species of a dark colour. Four pairs of cupules on moderately long stalks.

III.—Species with cupules, and a bifurcated median appendage:—

*Onchocotyle appendiculata* (Kuhn).

On the gills of various Elasmobranch fishes (rays and dogfishes), sometimes very common. Three pairs of large cupules and a slender appendage, bifurcate at the apex.

In addition to these Trematoda one representative of the Hirudinea is known from the Irish Sea fishes. The large skate leech, *Pontobdella muricata*, Leach, with its corrugated, warty skin, is often found on rays captured by the fisheries steamer.

There are many different kinds of worm parasites infesting local fishes, and as this group has only recently been seriously investigated in our district, we expect that the present list will soon be greatly extended. The Cestodes and Nematodes have still to be worked out. In addition to the fishes, many marine invertebrata are also more or less infested with parasites, some containing immature stages of the fish parasites.

The Copepod parasites are almost entirely confined to places in direct communication with the exterior. They may be found on the skin, the fins, in the mouth and branchial chamber, in the lateral line canal, attached to the gills and operculum, in the nostrils, in the eye and sometimes even burrowing into the abdominal cavity. In many cases their life history and anatomical structure are only partly known. Some are almost free, but the majority are true parasites, depending entirely upon their host for food, and having no means of locomotion. The males of the attached forms are extremely small and easily overlooked. All hatch from the egg as Nauplii and undergo very considerable metamorphoses before arriving at maturity. In some genera the development is wholly progressive. In others, such as *Lernæa*, when a particular stage is reached it becomes retrogressive. At one end of the group the parasites are practically free swimmers, and at the other they are mere inert sacs whose relationship with such forms as *Caligus* appears very remote.

*Bomolochus soleæ*, Claus.

In the nostrils of cod, greater fork-beard, plaice, &c., also on the skin of the common sole. A number of specimens of this copepod can usually be secured by forcing out the mucus from the inner part of the nostrils of medium sized cod. On placing the mucus in a watch

glass with water, the copepods, if present, can easily be seen.

*Caligus curtus*, Müller.

A very common species, and may be found on various kinds of fishes, cod, ling, hake, plaice, &c.

*Caligus rapax*, M. Edwards.

Also a very common species and as widely distributed amongst fishes as the former. The males and immature females are often taken in tow-net collections.

*Caligus diaphanus*, Nordmann.

In the gill chamber of the common gurnard and yellow gurnard, occasionally moderately frequent.

*Caligus gurnardi*, Kroyer.

The only record of this species from the district is the one by my late colleague and fellow-worker, I. C. Thompson, whose reports have been of much service in the preparation of the present list. The specimen was obtained from dredged material and had evidently been detached from its host.

*Caligus brevicaudatus*, A. Scott.

This is a moderately large species, 5.3 mm., and has only so far been found in the mouth of the common gurnards caught at Piel.

*Caligus labracis*, T. Scott.

Frequently found attached to the gills of the wrasse.

*Caligus scomberi*, Bassett Smith.

Occasionally found in the gill chamber of the mackerel.

*Caligus minimus*, Otto.

This well-marked species is sometimes very common in the mouth of the bass, *Labrax lupus*.

*Pseudocaligus brevipedes* (Bassett Smith).

Found attached to the inner side of the operculum of the three bearded rockling, *Onus tricirratus*. This genus is distinguished from *Caligus*, its nearest ally, by the structure of the fourth pair of feet, which have no exopodite.

*Lepeophtheirus pectoralis* (Müller).

Our experience of this species is that it occurs more frequently on the flounder than on any other member of the Pleuronectidæ. The pectoral and pelvic fins often have their under surfaces covered with this parasite; on one occasion thirty-two adult females were counted adhering to a pectoral fin. Males and immature females appear to be more common on the general surface of the fish.

*Lepeophtheirus nordmanni* (M. Edwards).

This is one of the larger species of *Lepeophtheirus*, and sometimes measures half an inch in length. It is found on the short sun-fish *Orthogoriscus mola*.

*Lepeophtheirus hippoglossi* (Kroyer).

Found generally distributed on the surface of halibut, occasionally in considerable numbers. On the "white" side of one specimen we have counted 100 individuals, but that was on a fish in the Aberdeen fish market.

*Lepeophtheirus stromi* (Baird).

Frequently found on the salmon captured in the estuaries. Its black metallic lustre makes it a conspicuous object on the bright scales of the fish.

*Lepeophtheirus obscurus*, Baird.

From the gills and inner surface of the operculum, and under the pectoral fins of the brill. It is probable that this species may only be a form of *L. thompsoni*, Baird, which is found on the gills of the turbot.

*Lepeophtheirus pollachii*, Bassett Smith.

Attached to the inside of the mouth of the pollack.

*Trebius caudatus*, Kroyer.

On the skin and inside the mouth of various species of Raia. The pink colour of the living animal renders its presence easily visible to the naked eye.

*Echthrogaleus coleoptratus* (Guerin).

On the skin of the picked dog-fish, *Squalus acanthias*, but not of very frequent occurrence. The ovisacs are very slender and of great length.

*Pandarus bicolor*, Leach.

Also occasionally found on the skin of the picked dog-fish. It has been recorded by other observers from *Galeus canis* (tope), *Scyllium catulus* (greater spotted dog-fish), and *Carcharias glaucus* (the blue shark).

*Dichelestium sturionis*, Kroyer.

A large white cylindrical species only found in the gill chamber of the sturgeon. The fish is not often caught in the district, and this makes the parasite rather rare.

*Clavella labracis*, van Beneden.

This is a very small parasite, egg-bearing females being scarcely one millimetre in length. It usually occurs in abundance, however, and is found on the gills of the wrasse, *Labrus mixtus* and *L. maculatus*.

*Cygnus pallidus* (van Beneden).

A small slender species, sometimes very common, on the gills of conger.

*Eudactylina acuta*, van Beneden.

Attached to the gills of the angel-fish, *Rhina squatina*. A number of specimens are found on each moderately-sized fish.

*Eudactylina acanthii*, A. Scott.

Of frequent occurrence on the gills of the picked dog-fish. Van Beneden records *Eudactylina acuta* from the angel-fish and the picked dog-fish, but we have never seen it on the latter. The present species is very distinct from *E. acuta*. Another species, *E. similis*, T. Scott, from the gills of *Raia radiata*, and a fourth from the gills of *Trygon* are known, but, so far, have not been met with in the Irish Sea.

*Lernæenicus sprattæ* (Sowerby).

Anchored in the eyes of the sprat, *Clupea sprattus*; not very common, but one or two specimens are frequently found in each catch of sprats. Usually only one specimen is found on a fish; occasionally two, one on each eye. We once met with a sprat that had two parasites in one eye and one in the other. The parasite measures 18 millimetres in length without the long egg sacs, but when these are added the length may reach to one and three-fourth inches.

*Lernæenicus musteli*, van Beneden.

On the gill rakers of the smooth-hound, *Squalus mustelus*. It is a large species, but does not appear to be a common one.

*Lernæa branchialis*, Linn.

This well-known and easily recognised parasite is moderately common on the gills of various members of the Gadidæ. The cyclops stage is often abundant on the extremities of the gills of plaice and flounder, and the next stage is occasionally taken in tow-net collections. An account of this curious form, and the changes that take place before it reaches maturity, have already been given in the Fisheries' Laboratory Report for 1900.

*Lernæa minuta*. T. Scott.

On the gills of the common goby, *Gobius minutus*. When present, this parasite is easily noticed, as it causes a considerable expansion of the operculum, and projects from under it.

*Haemobaphes cyclopterus* (Fabr.).

This appears to be a rare species and is not often met with; only one specimen has been observed by us. It was found on the gill rakers of *Cottus scorpius*.

*Oralien asellinus* (Linn.)

Occurs frequently on the gills of the common gurnard. Other workers have also recorded it from the gills of plaice and halibut.

*Chondracanthus cornutus* (Müller).

Occasionally found on the gills of plaice. Size, 6 mm. to 7 mm.

*Chondracanthus clavatus*, Bassett Smith.

On the gills of the lemon sole, *Pleuronectes microcephalus*. Size, 6·5 mm.

*Chondracanthus soleæ*, Kroyer.

A well-marked species occasionally found on the gills of the common sole. Size, 7·5 mm. to 8·5 mm.

*Chondracanthus lophii* (Johnstone).

This appears to be one of the more common members of the Chondracanthidæ, and is frequently found in the gill chamber of the Angler fish.

*Chondracanthus merluccii*, Holten.

Very common sometimes in the hake, and may be found on the roof and sides of the mouth, and also on the under side of the tongue, and attached to the inner surface of the operculum. Size 12·5 mm.

In all the *Chondracanthus* the males are very small,



being little over half a millimetre in length, and usually found on the abdomen of the female.

*Thysanote impudica* (Nordmann).

A moderately rare species and only appears to be found on the gill rakers of *Trigla hirundo*.

*Lernanthropus kroyeri*, van Beneden.

At first examination this species might be mistaken for *Chondracanthus merluccii*, but can easily be distinguished from that parasite by the structure of the appendages. Only found on the gills of the bass, *Labrax lupus*. Its deep brown colour makes it a conspicuous object amongst the bright red coloured gill filaments.

*Charopinus dalmanni* (Retz.)

This species appears to be confined to the spiracles of the grey skate, *R. batis*, where it is anchored by the large second maxillipedes. It is of large size and sometimes measures nearly  $2\frac{1}{4}$  inches from the apices of the second maxillipedes to the end of the ovisacs; usually only one specimen is present in a spiracle, but occasionally two and three specimens are found in one spiracle.

*Charopinus ramosus*, Kroyer.

A much smaller species than the last, being only about one-fourth the size. It has quite a different habitat from that of *C. dalmanni*, and is found attached to the gills and gill rakers of *Raia clavata* and *R. maculata*. A third species, *C. dubius*, T. Scott, is found on the gill rakers of *R. circularis* and *R. fullonica*. It has not been recorded from the Irish Sea but we have taken it from *R. fullonica* captured off Dubh Artach and landed at Fleetwood by one of the trawlers.

*Lernaeopoda galci*, Kroyer.

On the claspers and fins of the lesser spotted dog-

fish, *Seyllium canicula*, and occasionally on the fins of the tope.

*Lernaeopoda bidiscalis*, W. V. de V. Kane.

A remarkably distinct species, and always found on the ends of the claspers of the male tope, *Galeus canis*. These claspers, where the parasites are adhering, are almost invariably found to be torn and bleeding. A third species, *Lernaeopoda cluthæ*, T. Scott, is found on the gills of *Raia fullonica*; a fourth *L. salmonea* (Gisler), on the gills of the salmon; and a fifth *L. elongata* (Grant), attached to the eye of the Porbeagle shark and the Greenland shark, but none of these have, so far, been recorded from the Irish Sea.

*Brachiella ovalis* (Kroyer).

Attached to the gill rakers of the common gurnard, more common on half-grown fish than adults.

*Brachiella insidiosa*, Heller.

A moderately-large species, and found only on the gill of the hake. A considerable number are sometimes found on one fish. Another species, *Brachiella merlucci*, Bassett Smith, attaches itself to the gill rakers of the hake, but so far has not been seen by us.

*Anchorella uncinata* (Müller).

Very frequent in the mouth, &c., of species of *Gadus*, such as whiting.

*Anchorella appendiculata*, Kroyer.

From the hake, but the position in which they were found is not stated.

*Nicothoë astaci*, Aud. and M. Edw.

This peculiar copepod is sometimes found in considerable numbers on the gills of the common lobster. The wing-like projections of the fourth thoracic segment

give it an unusual appearance. Several times we have attempted to ascertain the fate of this copepod when the lobster casts its shell, but so far have been unsuccessful. None of the lobsters that moulted in our tanks happened to have parasites on the gills.

The Branchiurid, *Argulus foliaceus* (Linn), has been found on trout sent to us for examination from the Ribble.

## ON MYXOSPORIDIA IN FLAT-FISH.

By H. M. WOODCOCK, B.Sc. (Lond.).

Myxosporidia are Sporozoan parasites frequently found in fishes generally, and often the cause of severe disease, *e.g.*, the "Pockenkrankheit" of carp, and the "Barbenseuche" of the barbel, which have at times ravaged these fish in continental rivers.

The group is characterized (*a*) by the fact that reproduction by spores goes on throughout the growing or "trophic" period, and (*b*) by the complicated process of spore-formation and the nature of the spores. These contain, besides the germ or "sporozoite," one or more "polar-capsules" (very similar to hydrozoan nematocysts), from which filaments serving as organs of attachment to the epithelium of the new host—can be extruded.

Until recently the Pleuronectidæ were thought to be immune to the attacks of these parasites, and, in fact, to-day, there is, so far as I am aware, only one paper which specifically describes their occurrence, and at most two or three which record cases of disease in flat-fish either certainly or probably to be ascribed to infection by Myxosporidia.

In 1899 Hagenmüller (2) gave a short account (unfortunately without any figures) of a Myxosporidian infecting *Flesus passer*, Moreau (= *Pleuronectes flesus*, the flounder), a common member of the fauna of brackish, littoral pools in the neighbourhood of Endoume. At least fifty per cent. of the specimens captured harboured the parasites, which turned out, on investigation, to be a new species of *Nosema* (*Glugea*), to which Hagenmüller gave the name *N. stephani*. This genus belongs to the sub-order Cryptocystes (Microsporidia), comprising forms which

are always (at first) cell-parasites, with very minute spores possessing only one polar-capsule, invisible in the fresh condition. In all cases the parasites were found in the gut-wall, ranging, indeed, from the œsophagus to the rectum. The cysts were lodged according to the author, either in the muscular or connective tissue (sub-mucosa), or else projecting into the cœlomic cavity, and only covered by the peritoneal epithelium. They were also found under the peritoneum investing the liver, and in the mesenterial folds in which the blood-vessels run, but were completely absent from the other organs, spleen, kidney, &c. With the exception of certain species of *Myxobolus*, this invasion of gut is quite unique among Myxosporidia of fishes, the Myxobolida causing the above-mentioned devastating epidemics, usually occurring in the tissue of the liver, spleen, kidneys, bladder, &c. Hagenmüller goes on to describe briefly the minute anatomy of the parasites and their relation to the host's tissues as seen under a high power, and reference to one or two points in his account will be found below.

The next record of intestinal cysts in a flat-fish is that by Johnstone (3), who describes and figures Sporozoan cysts from two specimens of the plaice (*Pleuronectes platessa*). On examination of sections like that from which the author's fig. 2 was drawn, prepared from material which he was kind enough to forward me, I had no hesitation in deciding that these were also cases of a *Glugea*\* infection. Hagenmüller did not give the size and shape of the spores of *G. stephani*—the principal, though rather arbitrary, criterion of specificity among the

\* There is some doubt as to which of the two generic names, *Glugea* or *Nosema*, should be employed, so I have retained the former, which is well known and established.

*Glugea* - but contented himself with remarking that the cysts scarcely ever exceeded 1 mm. in diameter. Taking into consideration, however, the agreement in size and the very similar habitat and appearance of the parasites in the two cases, there is every probability that those in the plaice also belonged to this species.

About the same time Linton (4) published his systematic researches on fish parasites. Under *Pseudopleuronectes americanus*, the winter flounder, is a note (p. 485) concerning two small specimens infected with cysts. The gut-walls of one throughout almost the entire length, and of the other for a short distance, were entirely covered with "sporocysts" (*i.e.* seen from the outer or cœlomic side). The cysts were irregular in shape where crowded together, and where not—which was in but few places—they were elliptical or spherical. Their size varied, but none much exceeded 1 mm. in diameter. Fig. 1 is reproduced from Linton's fig. 4, Pl. 1, and shews the general appearance of a portion of the intestine wall. Compared with Johnstone's fig. 1, Pl. D., the similarity of the two is readily apparent, allowance being made for the difference in magnification. Linton's figure, however, does not shew the areas or regions which are observable in Johnstone's specimen. The spores were oblong-ovate, about .003 mm. ( $3\mu$ ) in length by .0015 mm. ( $1\frac{1}{2}\mu$ ) in width, and these agree very well with the dimensions I have found, both for those from Johnstone's plaice and from my own. In short, I am quite confident that this specimen of *Pseudopleuronectes* was also infected with *Glugea stephani*.

From another Pleuronectid, *Rhombus (Stromateus) triacanthus*, the butter-fish, Linton further describes (p. 455) a "sporocyst" occurring in the liver, white and globular, and about 1.5 mm. in diameter. When com-

pressed, it liberated immense numbers of spores, which were in great part aggregated into globular or oblong clusters, the larger being as much as .02 mm. ( $20\mu$ ) in diameter. The spores themselves were short and thick, with bluntly-rounded ends, their length being about .0025 mm. ( $2\frac{1}{2}\mu$ ), and a little less in breadth. Here also there is little doubt that we have to deal with a Cryptocyst (Microsporidian), but in the absence of figures it is rather uncertain in what genus this parasite should be placed. From the mention of distinct clusters or clumps of spores, and the shape of these latter, I am somewhat inclined to regard this case as one of infection by *Pleistophora*, rather than by *Glugea*—the reason will be referred to below.

My own acquaintance with these parasites began in the summer of 1901, when Mr. Todd, then Assistant to the Director of the Plymouth Marine Biological Laboratory, brought to my notice the intestine of a plaice which, although, unlike Johnstone's specimen, it was quite healthy in appearance, presented certain abnormal features.

The gut had been fixed in Corrosive and Acetic, and preserved in spirit, and shewed at intervals little oval patches, usually projecting slightly, on the outer cœlomic side. Besides these, there were little out-growths of the tissue of the wall, often in the form of pear-shaped appendages, attached by the narrow end to the gut. These patches and projections indicated the site of the infection, and were readily to be distinguished by their rather different colour, having a faint reddish tinge added to the pale nondescript shade of the preserved gut. I gathered from Mr. Todd that, when fresh, the spots were of the usual Sporozoan opaque-white. A point worth noting in the position of the parasites is that they were

all on that side of the gut to which the mesentery was attached, and in which the blood-vessels ran; indeed, often they were quite close to these latter, although never actually in their walls. The infection was, in this instance, a comparatively limited one, and the functional activity of the intestine would be in no way interfered with—a very different state of affairs from that described above. There was no enlargement of the folds or pleats (consequently no occlusion of the lumen), and on opening, the internal surface (the mucosa) appeared quite normal.

In size, the patches averaged about 1 mm., while the out-growths were sometimes  $1\frac{1}{2}$ —2 mm. in length. Figs. 2 and 3 shew some examples drawn natural size. In fig. 2 there are three appendages (par) visible on one of the pyloric caeca, and close to the pyloric branch of the mesenteric artery (art). In fig. 3 some more are seen, also mostly near the attachment of the mesenteric blood-vessels.

Johnstone's specimen and my own aptly illustrated, respectively, the two chief *modi vivendi* of the *Glugeæ*, viz., (a) cyst-formation, and (b) in the condition of "diffuse infiltration," Thèlohan's (7) term for signifying an infected area in which the parasites and the tissue of the host completely intermingle. Fig. 4 represents a section of a portion of the wall of the intestine and an appendage under a very slight magnification, where the darker shaded part (par) shews the diffuse infiltration, and the lighter region is normal uninfected tissue. The out-growth, it will be seen, is practically entirely a parasitic development, with the exception of a delicate covering of connective tissue and peritoneal epithelium, which is, in places, broken down (ep.). But, here also, at (cy.) an attempt at cyst-formation has taken place, really a pseudo-cyst, since in origin and nature these are very



different from the true cysts in Johnstone's plaice, which I will call in future specimen A. Before describing the two varieties minutely, it will, perhaps, help to understand their structure, to give briefly the course that the development of the parasites has followed in the two cases.

In both, the infection is a ripe, well-matured one, but whereas in A it is very strong, and must have proceeded from a great number of centres (in other words, the host swallowed very many spores), in my plaice (B) it is only a slight one. Perhaps this supplies, at any rate partly, the reason for the fact that here the individual parasites have tended to spread the infection further, invading the neighbouring tissue of the host largely by the process of endogenous multiplication ("multiplicative reproduction" of Doflein (1)) in the young forms. Indeed in the case of *Glugea*, where cell-infection prevails, this condition of diffuse infiltration is, to a very considerable extent, the result of endogenous reproduction, and, therefore, apparently differs rather in origin from the condition as it is generally understood to occur in the larger Myxosporidia; although I think it is by no means unlikely that, in these also, it will be found that endogenous reproduction has a larger share in the result that is at present ascribed to it, for Doflein (l.c.) describes in one instance (*Myxobolus cyprini*, the cause of "Pockenkrankheit") a similar multiplicative reproduction of young forms while still intracellular. There is, therefore, no need to restrict, as he would, the term diffuse infiltration to such larger forms, which, when adult, are intercellular. Thèlohan, in first describing the condition, was not aware of this endogenous reproductive capacity, and looked upon it as a more or less continuous ramifying infiltration of the parasitic body, with displacement and disturbance of the surrounding tissue. Whereas, as we have just seen, it is far more

likely that it is caused by a multiplication and separation of young individuals, which break down and leave the host's cells, the intercellular nature of the adults being the only point in which the diffuse infiltration here differs from its occurrence in the *Glugeæ*.

Anyhow, in these, to return to (B), the final result is that the infected area consists of a confused mass of tissue-cells (many broken down), spore-containing cells, and clusters of free spores. In A on the other hand, this process is not evident, and I greatly doubt whether it has been at work. Probably owing to the strong infection, with its attendant effects on the host's metabolism, the parasites do not seem to have attempted to spread further (except, as it were, automatically by growth), but to have concentrated their energies on becoming large spore-forming individuals. The cysts are well-defined, and sharply limited, and there is no sign of "diffuse Ausläufer." Each is, I consider, the result of growth of a single individual.

Dofflein (l.c.) is of the opinion that the cysts of *G. lophii*, which he describes (p. 334 *et seq.*), result from the fusion of 3-4, and this may well be, for he is evidently dealing with "pseudocysts" and an infiltration condition, more or less similar to my specimen B. In A, however, the minute structure shews clearly that each cyst is a single unit. Here the parasites are never, as sometimes in Hagenmüller's case, in the muscles, but entirely in the areolar tissue of the sub-mucosa. The intestinal epithelium is also free from infection, and though often broken round the much-enlarged internal end of the folds or rugæ, it is still quite evident and normal up the furrows. For a general idea of the size and arrangement of the cysts, the reader is referred to Johnstone's fig. 2, Pl. D.

Figure 5 is a section through a portion of a cyst. The outer, cœlomic, side of the gut-wall lies to the right, and the muscle-layers, though a little thin, perhaps, are free from infection, and present no feature worth drawing. At the left is a fold (endoth.) of the mucosa, also quite normal, at the head of a furrow. At ect., ect. is seen the thick, practically structureless external layer (ectoplasm) of two adjacent cysts, which are separated by a few delicate layers of connective-tissue (areol. tiss.). Hagenmüller, who maintained that the cyst membrane is formed of elements belonging to the host, was certainly referring to pseudocysts, which, however, he did not recognise were intimately connected with diffuse infiltration. He does not seem to have seen true cysts at all. In these, there is no transition whatever visible between the surrounding tissue and the firm "ectorind" (as one may term the modified ectoplasm) of the parasite, nor any signs in this latter of flattened-out nuclei or cells. I entirely agree with Thèlohan in thinking that the cyst membrane is a modified ectoplasm, and for this reason propose the term ectorind, to distinguish it from an ectocyst. Against its being an ectoplasmic secretion—comparable to the cyst-envelopes of Gregarines—I would say there is no other layer which can be regarded as the ectoplasm. Immediately internal to the ectorind is a delicate layer without any sign of spore-formation, but which is structurally identical with the endoplasm into which, indeed, it passes, and, therefore, I regard it also as such. (In some sections through a *Glugea anomala*, from a stickleback, which I possess, there is an equally distinct, here finely-striated, ectorind, thus confirming Thèlohan's figs. 138 and 139, pl. 9). Internal to the ectorind, and extending all round, we see the typical Myxosporidian endoplasm (end.). It is a comparatively narrow layer, as by

far the greater part of the cyst consists of an immense number of spores. The endoplasm is the seat of spore-formation. In the Microsporidia, this starts and proceeds either from one centre or from several. In the latter case, each centre comprises (at first) a single nucleus and a small portion of the cytoplasm in the immediate neighbourhood, the whole becoming segregated to form an "organella" of reproduction. Each such centre is termed a pansporoblast. When, as in the one family, including *Thelohania*, *Pleistophora*, &c., there is only one, it signifies that the whole individual becomes a reproductive organella and commences to sub-divide up into sporoblasts and spores (just as, for instance, a Gregarine does). So that, in *Pleistophora*, a ripe infection consists of a number of relatively small individuals, each being a single cluster of spores. This fact, together with the blunter and thicker shape of the spores in this genus, leads me to surmise that Linton's sporocyst from the liver of *Rhombus triacanthus* (see above) was probably a *Pleistophora*.

To return to our *Glugea* specimen. The endoplasm contains a great number of spore-forming organellæ in various stages of development, from little uninuclear ones to large ones which are practically clumps of sporoblasts (spbl.). The actual transition from sporoblast to spore is extremely difficult to follow in the case of these minute spores; for a full account of the development in the large forms, Thèlohan's paper should be consulted. I may add, in passing, that anyone who desires complete information on the group, cannot do better than read Minchin's up-to-date and concise résumé (5). Scattered about in the endoplasm, and also occasionally met with in the central mass, are large, sometimes drawn-out nuclei (n.) with fragmented chromatin and a distinct nucleolus (?).

They stain more deeply and brightly than the sporoblasts, and cannot be mistaken for these. Thèlohan does not describe them, and since mine are all ripe cysts I have no means of ascertaining their origin and significance. Passing inwards we come to clusters of ripe spores, and these soon almost entirely replace the endoplasm, and run together to form the central mass of spores, the organellæ themselves having broken down and disappeared. Often, however, I noticed, as it were, tongues of endoplasm projecting internally towards the centre, and in some sections appearing as isolated patches, some of which were fertile, but others apparently sterile. These are not to be confounded with the islands of residual-tissue described below. Although I have examined sections through many cysts, I have not seen any instances of the degeneration of the spores such as is described in the central part of "over-ripe" cysts. In all mine, the central spores are as healthy in appearance and as deeply-staining as those of the periphery. A noteworthy difference in the central mass of an A cyst from that of a B one, is the absence in the former of the areas or patches which are the more or less colloidal residua of the degeneration of tissue-cells, &c., of the host. Moreover, the spores themselves in the former case are quite free, and not embedded in any matrix. These facts, together with those already set forth, emphasize the distinction which I wish to bring out between the two kinds of cyst. While the one (A) is practically all parasitic tissue, and represents one individual, the other (B) the pseudocyst—is the consequence of the massing together and circumscribing of (a portion of) an infiltrated area, originally comprising many tiny, daughter, individuals (of which, in an old pseudocyst, nothing is left save spores) diffused in and among the host's tissue (also

broken down). There is not the slightest possibility that an A cyst could ever become a B cyst, or *vice versá*. And now to describe the latter.

Fig. 6 is part of a section through the periphery of the lower cyst in the appendage of fig. 4. All around (though the upper cyst happens to be close to it on one side) is the infiltration, a condition easier to describe verbally than to draw, as it would be most difficult to reproduce except very diagrammatically.

These pseudocysts arise as the result of an endeavour on the part of the host to limit the infiltrated area by the arrangement of the hypertrophied connective-tissue in a series of concentric layers round the centre of infection. (The marked hypertrophy here is in strong contrast to its complete absence in the A cysts). This re-action of the host, not always so well-defined, seems to be without much success, for the parasitic invasion usually spreads further, often leading to another attempt at restriction; in fig. 4 for example, the upper pseudocyst is of later formation than the lower. Young ones, therefore, are simply areas of diffuse infiltration with the layers concentrically arranged; but, with age, and probably, to a certain extent, the pressure of the surrounding parts on the enclosed tissue, the cells of this latter disintegrate and degenerate, leaving a mass of spores embedded in their remains as a ground-substance, with frequently patches or islands free from spores and staining only with the plasma stain (deg.). The spores are in a much more closely packed condition than in the comparatively loose, infiltrated tissue of the appendage (not shewn in the fig.). It need hardly be added that there is no trace of endoplasm, nor of sporoblast-formation round the margin, nor, of course any ectorind, and, in fact, no sharply-marked external limit—a great contrast to fig. 5. The periphery

is formed by loose clusters of spores in artificial chambers bounded by delicate strands of connective-tissue (cham.). Outside, again, there are several concentric and partially imbricate layers of connective-tissue (con. tis.). The comparative looseness of the peripheral layers is partly due to the shrinking together of the central residual mass on fixation, and also to the fact that, once the "cyst" is in the appendage, any further layer-formation, and, of course, the proliferating infiltration, is free to expand. From Hagenmüller's account it is quite evident that his cysts were nothing more or less than a similar modification of the infiltrated condition.

Before passing on to consider the spores, there is an interesting point which a comparison of the two types of infection, as set forth above, leads me to regard as being very probable, namely, the potential independence of the pansporoblasts in *Glugea* (i.e. their capability of existence as separate individuals in certain circumstances). Stempell (6), p. 263, has already suggested that *Thelohania*, *Pleistophora*, &c. (those forms where the whole individual becomes one reproductive organella), are examples of a phylogenetic individualization of the pansporoblasts. Now, I am inclined to think this may take place normally—for instance, in the condition of diffuse infiltration—as a stage in the life cycle. Quite probably "multiplicative reproduction" is, here, simply a separation of the pansporoblast rudiments, as daughter-individuals. Indeed, the whole nature of the diffuse infiltration in *Glugea* seems to me to support this idea. There is no question of the individual parasites attaining size, still less of any continuity of a protoplasmic mass ramifying in and between the host's tissue-cells. It is far rather a cell-infection, visible, when ripe, as separate clumps of spores, each formed from, and

representing, one pansporoblast, and either still surrounded by a hypertrophied host-cell, or else free, but only owing to the latter's break-down. Such an origin and nature of the daughter-individuals would obviate any necessity for Doflein's hypothetical "swarm-spores," the occurrence of which would be without analogy in the Sporozoa.

The spores themselves are seen in fig. 7 (*a*) and (*b*). They are oblong-ovate in shape, and average  $3\mu$  by  $1\frac{1}{2}$ — $1\frac{3}{4}\mu$  in size. They are the same in the two cases, those drawn in (*a*) coming from specimen A, and those in (*b*) from B. *Glugea* spores are usually pear-shaped, but, after very careful examination, I cannot, with certainty, distinguish any difference in this respect between the two ends of those of *G. stephani*. Linton (l.c.) also says nothing about the spores from his specimen being pear-shaped. Not having had any fresh material to work with, I have been unable to observe the expulsion of the polar filament in this species, but in fig. 7 (*c*) are seen a few fresh spores of *G. anomala* from a stickleback, one of which shews the filament extruded. The clearer space at the opposite end (which is nearly always evident in fresh *Glugea* spores) represents a vacuole—also generally apparent in the stained spores of *G. stephani*. In one or two instances I saw a faint longitudinal suture (*s*) marking the junction of the two valves of the spore. The contents are most difficult to interpret correctly, since, owing to the absence of a pear-shaped end, one cannot say positively where the polar-capsule is situated. There are usually two unstained clear areas, with the sporoplasm (*i.e.*, the germ) lying between. One of these is well marked, and invariably contains a small, rounded, deeply-staining granule; whilst the other seems to vary in size, and is not always very obvious. The former is,



probably, the polar-capsule, in which case the dot would be the capsulogenous-cell nucleus. The granule itself is certainly not the capsule, for, in other spores, I have never found the capsule to stain up at all, and besides, it is very rarely terminal in position. Moreover, in one instance (at x) it is distinctly double, as if the nucleus had divided—as, in fact, Stempell (l.c.) maintains it does in the spores of *Thelephania*. On this view, the other clear area (v.) would represent the vacuole seen in the fresh condition. I am somewhat inclined to think this tends to increase in size with the ripening of the spore (as indicated by the number of nuclei), and I suggest, tentatively, that it may have some such function as the oval body in the spores of *Coccidium*, to assist in separating the valves and liberating the germ. In the sporoplasm itself, there is not much to be made out save the nucleus (N). In the earlier stages this is single and round, but in its most general condition in my sections it has the form of a horseshoe, being drawn out prior to division. Sometimes, however, as in the two upper examples, it has distinctly divided into two. Stempell is of the opinion that these two nuclei (representing two germs, although the sporoplasm has not divided) again fuse, this action constituting a “conjugation,” which is, as yet, unknown for the order. This requires confirmation before it can be accepted, as it is, *a priori*, most unlikely that two germs, so closely related and, indeed, barely separated, would conjugate; such a proceeding would be entirely without precedent. Once or twice the sporoplasm possessed three nuclei, the reason for which I have not made out.

While revising these notes, I have received from Mr. Johnstone a slide, a smear preparation, made, he writes, from cysts in the otic capsule of a plaice. The car-

tilage was much hypertrophied, and the cysts were little opaque masses about 1 mm. in diameter. The smear consists of a mass of spores, two of which are seen in fig. 7 (*d*); each is spherical in shape, and possesses two polar-capsules (p.c.). The spores are 8-9 $\mu$  in diameter, and the polar-filaments, many of which are expelled, have a length of about 70 $\mu$ . There are several refractile oil granules (g) visible, but the stain has not penetrated sufficiently to shew up the nuclei. I could not make out any vacuole in the sporoplasm which would correspond to the "iodinophilous vacuole" of a *Myxobolus* spore. Moreover, the length of the filament is much longer than is usual for this genus. Hence, although there are one or two *Myxoboli* with spherical spores, I am more inclined to place this new species in the genus *Sphaerospora*, as *S. platessæ*, presumably polysporous, and with unornamented, spherical spores.

We may, therefore, summarize the Myxosporidian parasites of flat-fish as follows:—

Host.	Organ or Tissue.	Parasite.
<i>Pleuronectes flesus</i> (flounder).	Gut-wall and mesentery.	<i>Glugea stephani</i> , Hagenm.
<i>P. platessa</i> (plaice).	Gut-wall.	Do. do.
Do. do.	Otic-capsule.	<i>Sphaerospora</i> <i>platessæ</i> , n. sp.
<i>Pseudopleuronectes</i> <i>americanus</i> (winter flounder).	Gut-wall.	<i>G. stephani</i> , Hagenm.
<i>Rhombus</i> (= <i>Stromateus</i> ) <i>triacanthus</i> (the butter-fish).	Liver.	<i>Pleistophora</i> sp. (probably n. sp.).

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## EXPLANATION OF PLATE.

All figures except 7 (*c* and *d*) refer to *Glugea stephani*. Figures 4-7 drawn with the aid of the camera lucida.

Iron Haematoxylin followed by Orange, Thionin followed by Eosin, and Kleinenberg's Haematoxylin, were the most successful stains.

Fig. 1. Linton's "sporocysts" from *Pseudopleuronectes americanus* reproduced. A portion of the gut shewing the infection.  $\times 2$ . sp. = the cysts.

Fig. 2. Part of the stomach and the pyloric caeca of my plaice (specimen B), shewing the little parasitic appendages (par.). art. is the pyloric branch of the mesenteric artery.  $\times 1$ .

Fig. 3. A portion of the intestine shewing the same. The centrally placed parasite is still a swelling, not having yet become an appendage.  $\times 1$ .

Fig. 4. A section through a piece of the gut wall and an appendage. The dotted line divides the two. The darker shaded part (par.) is the "diffuse infiltration," the lighter part representing uninfected tissue. ep. is a thin layer of connective-tissue and peritoneal epithelium covering the appendage. cy. is a "pseudocyst."  $\times 20$ .

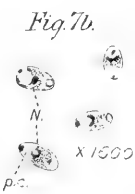
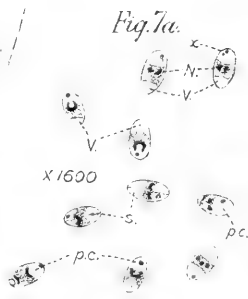
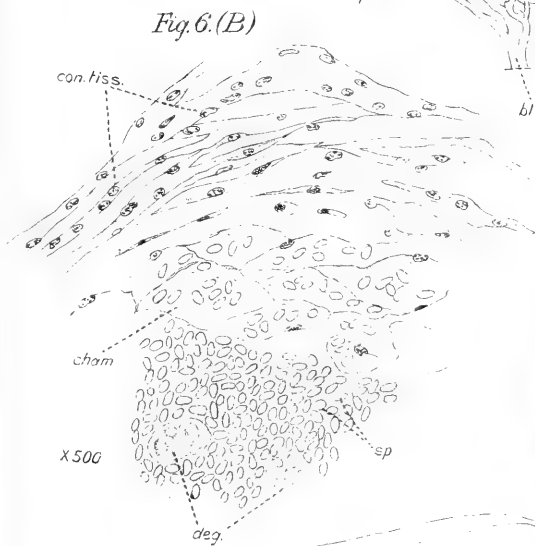
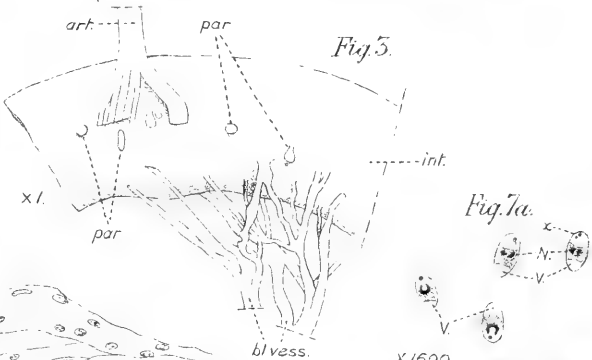
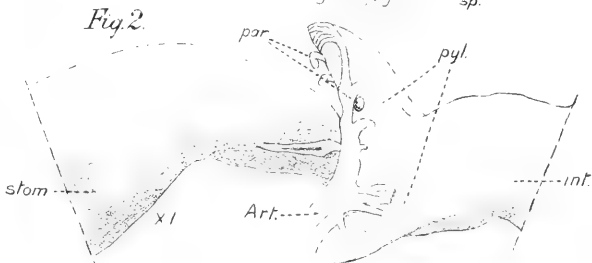
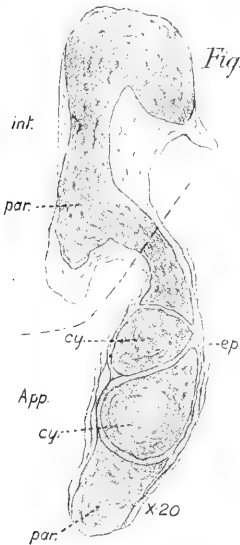
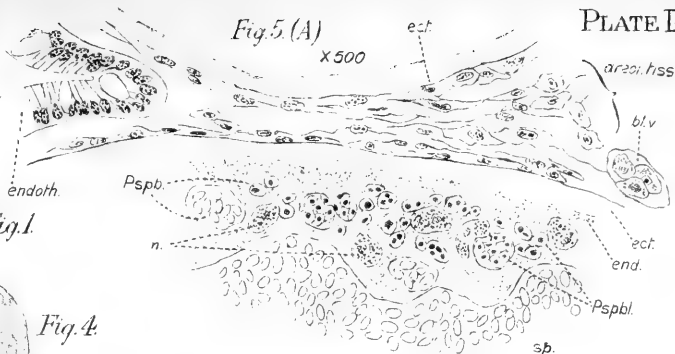
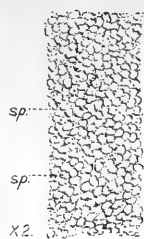
Fig. 5 is part of one of Johnstone's cysts (specimen A), and Fig. 6 is part of one of mine (B). For description, see the text.

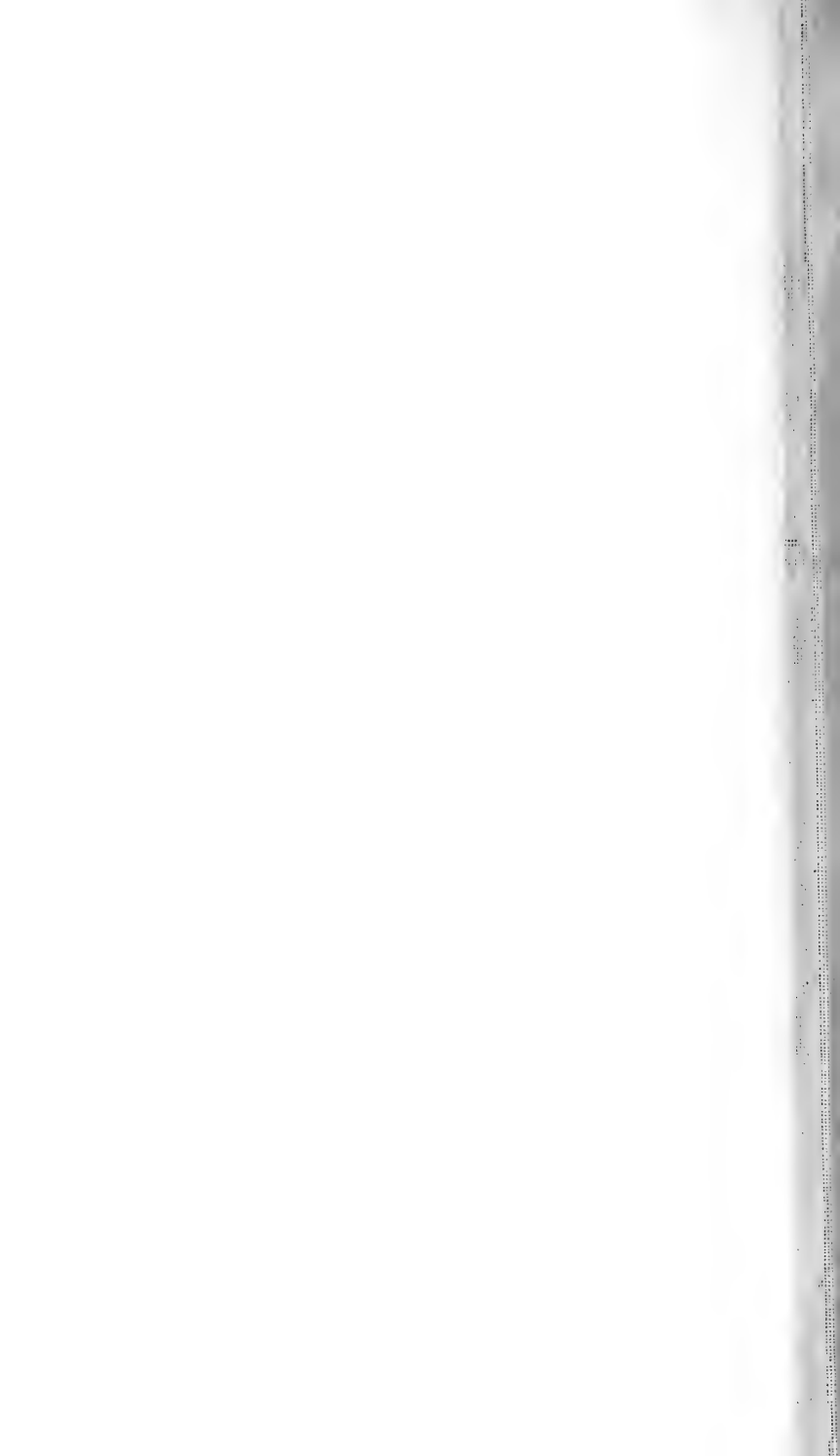
ect. = ectorind (ectoplasm), end. = endoplasm.  
 endothel. = a fold of the mucosa, shewing its normal appearance, n, = large nuclei in endoplasm. Pspbl. = various stages in the development of spores (sp.) from the reproductive organellæ.  $\times 500$ , Zs. 3mm. Apochr. 1.40 O.I., C.E. 4.

Fig. 7 (a). Spores from (A). 7 (b). Spores from (B). N. = nucleus of sporoplasm, p.c. = polar capsule, x = nucleus of latter. s = suture, v = vacuole.  $\times 1,600$ . Zs. 2mm. Apochr. 1.40 O.I. C.E. 8.

Fig. 7c. Fresh spores of *Glugea anomala*, treated with iodine sol. in 50% alc., one of which shews the polar filament extruded.  $\times 1,200$ .

Fig. 7d. Spores of *Sphaerospora platessæ*, one shewing the 2 polar-filaments expelled. g = refractile granules.  $\times 900$ . Z. 3mm. Apo. 1.40 O.I., C.E. 8.





NOTE ON A REMARKABLE PARASITE OF  
PLAICE AND FLOUNDERS.

By H. M. WOODCOCK, B.Sc. (Lond.).

In corresponding with Mr. Johnstone about the *Glugea*-infection of the plaice, above described, he informed me of what seemed to be another Myxosporidian infection of a flounder (*Pleuronectes fesus*), instances of which, he says, are not unfrequently met with. In looking up the literature on parasites of flat-fish, I came across the papers by Lowe (2), McIntosh (3), and Sandeman (4), which would appear to refer to the same thing. As Sandeman practically gives the substance of McIntosh's two papers, accompanied by figures, a brief abstract of his note "On the Multiple Tumours in Plaice and Flounders" will suffice.

The tumours were prevalent more or less all the year round, occurring principally from May to November, and giving the fish an emaciated appearance. The situation of these cyst-like swellings was in the skin and dermal tissue of the fins, the operculum, and the tail, usually projecting externally. Sandeman does not say whether the gut or other internal organs were affected. The little swellings are described as pearly-white spheres, not firmly attached, but loose in the connective-tissue, covered over by the pigmented epithelium, and exuding on pressure a creamy-white, structureless substance. The larger masses form tumours up to nearly an inch in size, composed of many spheres from 1 to  $1\frac{1}{2}$  mm. in diameter, often, however cuboidal or polygonal owing to mutual pressure, and each limited by a distinct membrane. The author remarks on their great resemblance to a mass of eggs, but admits

that it would be impossible for any animal to force such a quantity under the skin, as to give rise to a pedunculated tumour. (Since, as I shall presently shew, they also occur in the mesentery, this possibility is entirely negatived). In none, though he noticed them all the year round, could he discern any development.

Sandeman apparently only examined the cysts under a low power, and judging from his description and figures, I thought it very probable that this was another case of a *Glugea*-infection. There is considerable resemblance between his fig. 3 (a section through a tumour of many cysts), and Johnstone's fig. 2 (l.c., above), for example. In order to identify it, Johnstone kindly forwarded me a well-infected flounder from the Fisheries Museum collection in the University of Liverpool, together with a drawing of the head-region, shewn in fig. 1. On sectioning the parasites, I soon realised they were very different from what I expected to find them.

The specimen was taken in the Barrow Channel in January, 1901. Both sides of the head are plentifully covered with opaque white cysts—averaging  $1\frac{1}{4}$ - $1\frac{1}{2}$  mm. in diameter—some spherical, others more ovoid. On the dorsal side, just in front of the fin, are two or three contiguous tumour-like masses—not, strictly speaking, tumours, however, for there is, practically speaking, no proliferated tissue, the whole thing being a mass of cysts with, of course, a little vascular connective-tissue between and around them. Solitary ones are also scattered about on the operculum, ventral fin, and tail, and they shew a tendency to aggregate along the lateral line, especially posteriorly. Under the pectoral fins an Entomostracean ectoparasite (*Lepeophtheirus*) is fairly abundant, but there is no connection between the two kinds of parasite. The cysts lie beneath the skin (on the upper side



the pigmented epithelium over them is distinctly discernible) in the dermal lymph-spaces, held in place by the surrounding tissue, but not embedded in it, and they can easily be removed with only a few lymphocytes, &c., attached. They are quite absent from the somatic musculature. On opening the body-cavity, numerous parasites are seen in the gut-mesentery, usually close to, but not actually in the wall of, the blood-vessels. Fig. 2 gives an idea of their appearance in the mesentery of one loop of the intestine, from which it will be seen that, when internal, the cysts are uniformly slightly smaller than when beneath the skin. They are generally oval or elliptical, and never exceed 1 mm. in diameter. Unfortunately, I have, so far, not found any younger, or different, stages, but it is most likely that the parasites, when smaller, pass into a blood-capillary or lymph-channel from the gut, and there grow and encyst (?), since in section (fig. 3) they are surrounded by a space (spa.). All the internal organs are quite free and normal, and for this reason I should not say the hosts are harmed to any dangerous extent. The two or three afflicted specimens which I have so far seen certainly cannot be described as "emaciated."

Minute structure. Notwithstanding the size the things grow to (up to  $1\frac{1}{2}$  mm. in diameter) each is, undoubtedly, a single cell; as to that I have not the least doubt. There is no trace of cell-division, nor of cell-nuclei in the ordinary sense, in it; whatever a cyst represents, it is, as a whole, unicellular. Fig. 3 is a section, slightly magnified, through one in the mesentery, the space around representing an enlarged capillary or a lymph-channel, as already mentioned. This happened to be more spherical than the internal ones usually are; it is surrounded by a layer of amoebocytes, &c. (lym), rather

closely aggregated. (A portion of the same cyst more strongly magnified is seen in fig. 6.) The most external layer belonging to the parasite is a thick, faintly-staining, structureless membrane, which I have denoted by (ect.). Next comes a thick zone (end.), more deeply-staining, of a finely granular nature, the greater part of which presents a most unusual appearance, and gives the organism its remarkable character. Centrally is what can only be a nucleus (N), although of relatively huge size as in fig. 4, which is a section through another, larger, cyst, from underneath the skin. In each nucleus are several nucleoli (n), or rather karyosomes, since they retain the chromatic stain. Fig. 5 is part of the nucleus drawn under a high-power, and shews a faintly-staining, irregular reticulum, which traverses a finely-granular ground-substance, with karyosomes of all sizes, the larger being vacuolated and the smallest little more than granules. The nuclear-membrane is very thin and extremely irregular, and sometimes appears only as a boundary between the nucleus and the inner limit of the cortical region (shewn on the left in the fig.).

In fig. 6 I have attempted to indicate the appearance of a portion of the cortex, as seen under a high power. It consists of a finely-granular matrix, staining with the plasma stain, in which are innumerable, usually separate, reticula or net work (ret.), in every variety of shape and size. These stain up deeply with chromatic stains, and, so far as I can make out, are made up of threads of rodlets or granules, not easy to resolve. Each network is developed round a centre (which stains sometimes less, sometimes more, than the general ground-substance) apparently at its periphery. These structures do not commence quite at the external limit of the cortex and they cease some distance before its inner limit. On the whole,

the smaller ones are more peripherally situated, though there is no regular increase in size as one passes inwards. The only other point to note is a series of tiny spherules (sph.), each with one, or sometimes two granules, at the outer margin of the cortex, almost abutting on the membrane (ect.), but I have seen no transition between these and the deeply-staining reticula, nor are they obvious in all my sections. I should add that in fig. 3 these latter are more closely packed and rather more strongly-stained than in fig. 4.

In endeavouring to arrive at some idea of the nature and affinities of these remarkable cysts it will be most convenient to commence by the process of elimination. In the first place we have, certainly, not to deal with a Trematode or other Metazoan parasite. Further, Dr. Nabarro and Professor Oliver, who have kindly examined it, are of the opinion that the cysts are the result of neither a bacterial nor a fungal infection, and, indeed, it is almost inconceivable that an ordinary cell could be so enormously hypertrophied by bacterial or hyphal invasion, and retain as much of its structure as these bodies do (compare the amoebocytes and connective-tissue cells around). Moreover, Mr. Pollard, of the Bacteriological Laboratory at University College, has stained sections for me by the usual methods adopted for Bacilli, &c., without result. The localized and restricted nature of the infection is also against this view, the cells around being quite normal. So that we may dismiss the idea of the cysts being caused by a bacterial or hyphomycetic parasite. Each cyst-like body is one organic unit.

I can only think of two remaining hypotheses, namely, that the bodies must represent either eggs or parasites. Now, although, as stated above, it was absolutely impossible that they were the eggs of some

other animal, for the simple reason that they are also internal, yet it was conceivable that they represented enormously modified and hypertrophied ova, which had become detached from the genital stroma (germinal epithelium) when very young, had been carried about—absorbing a great quantity of nutriment which had formed the remarkable chromatic development—and thus finally grown into these huge cysts.\* Unlikely as this hypothesis might at first sight appear there were two or three points in favour of it, and I have, therefore, carefully considered it. For one thing, the resemblance between the nucleus of one of these bodies and that of a flounder's egg is quite striking. Though differing greatly in size (and I may here say that the diameter of a cyst averages about four times that of a normal egg, and the nucleus is relatively larger), their structure is practically identical. Indeed, the nucleus reminds me more of a germinal vesicle than anything else. It is not like a Protozoan nucleus, that of a Gregarine being the only one which can be compared with it, from which this differs chiefly in relative (to say nothing of absolute) size, and in the ill-defined membrane, lacking any marked affinity for the chromatin stain. The outermost layer (ect.) would also serve for a thick egg-membrane, but in none of my sections is any radial striation visible, corresponding to the "zona radiata" of the eggs, although I should add that Sandeman mentions and figures something of the kind in his description. The chief difference is in the cortical zone. Whereas, in an egg, the cytoplasm is filled with large, spherical, refringent, oil or fat globules, there is not the least sign of such in the cortex of these bodies

\*I should prefer to think of their origin thus (*i.e.*, from differentiated ova, however small), than to suppose they had originated from wandering ("vagrant") indifferent germ cells,—because of their single and markedly ovarian nature. In the latter case, there would more probably have resulted (by proliferation) "cell-nests" of ordinary indifferent cells.

Whether, owing to differences in nutrition and chemical metabolism, the remarkable chromatic reticular-centres could have arisen instead, it is now scarcely necessary to discuss, for while this note was being prepared for the printers I received from Mr. A. Scott, at Piel, another equally infected specimen, which turned out to be a male. This, of course, left me with only the parasitic alternative, as one cannot imagine such an abnormal ovarian development occurring in a male.

Perhaps the chief reason why I gave so much consideration to the above hypothesis was because the bodies are so utterly unlike any known Protozoan. The parasite, for which I propose the name *Lymphocystis johnstonei*, is, in truth, the strangest Sporozoan (this being the only class in which it can possibly be placed) that I am aware of, and until I obtain further stages in its life-history, I can only interpret the above-described features in very general terms. Ect. may well represent the ectoplasm, now modified into an ectorind, while end. corresponds to endoplasm. Presumably the large nucleus is the vegetative or "trophic" nucleus, although in this respect *Lymphocystis* differs from any known Sporozoan, in that while sporulation is proceeding (for what else can the chromatic centres represent?), the vegetative nucleus persists undivided. In a Myxosporidian, spore-formation certainly goes on during the trophic phase of the life-cycle, but here there are many nuclei, some only of which originate reproductive-organelle, the others continuing vegetative in function. This single nucleus in our parasite recalls more the condition in Gregarines, but there the original nucleus breaks up altogether at the close of the trophic period to form reproductive nuclei. Nor in the Sporozoan "lumber-room" (already well filled) is there anything similar. The sole form with which *Lymphocystis* seems to have any point of

agreement is *Lymphosporidium trutta*, the cause of a brook-trout epidemic in America. In most respects this parasite, described by Calkins (1), is very different from *Lymphocystis*, the adults being amoeboid, relatively minute, and with no well-defined nucleus—the chromatin being in the “distributed” form. The two parasites are, however, not without certain points of resemblance. *Lymphosporidium* is, especially in the adult sporulating stage, chiefly met with in the lymph-spaces surrounding the various organs and of the dermis. Moreover, its manner of reproduction rather recalls that of *Lymphocystis*. According to Calkins, deeply-staining granules collect in masses to form many spores. The chromatin next forms a layer around the periphery of each such centre, and breaks up into rounded granules, eight in number, which separate (?) to form the sporozoites. From Calkin's figures, it does not seem to me unlikely that the complicated reticular areas in *Lymphocystis* (i.e. the threads of rodlets or granules above described) may represent a modification of this process, although at present one cannot say so with certainty. In that case the ultimate germs must be extremely minute (only about 1 or  $1\frac{1}{2}$   $\mu$ ) and numerous. Whether the little spherules at the margin of the endoplasm have any connection with spore-formation and are in any way comparable to the reproductive organellæ or “pansporoblasts” of Myxosporidia has also yet to be ascertained.

In conclusion, *Lymphocystis* would appear to combine, to a certain extent, Gregarine and Microsporidian characters—with remarkable results. I do not feel inclined to place it in the Serosporidia (the order to which *Lymphosporidium* belongs), as these forms, though of similar habitat, are all very small. I have, unfortunately no alternative but to leave it for the present, to swell the ranks of the “unattached” Sporozoa.

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## EXPLANATION OF PLATE.

All figures refer to *Lymphocystis johnstonei*.

Figs. 3-6 were drawn with the aid of the camera.

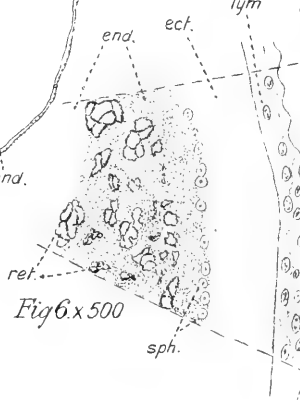
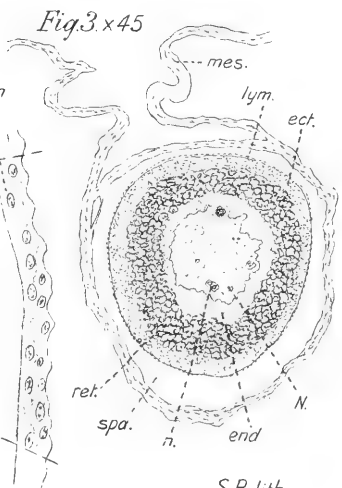
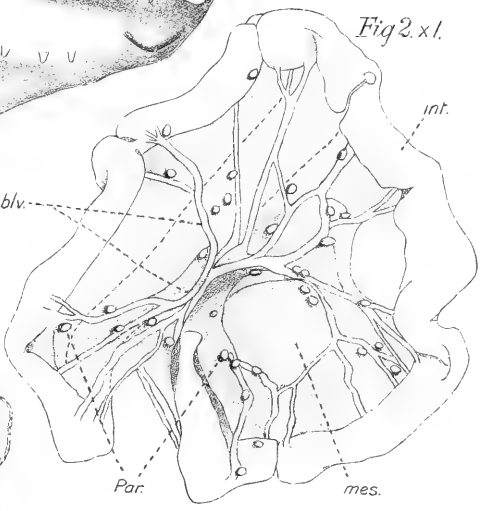
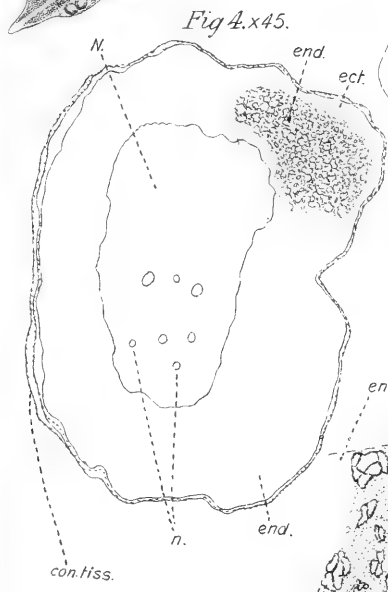
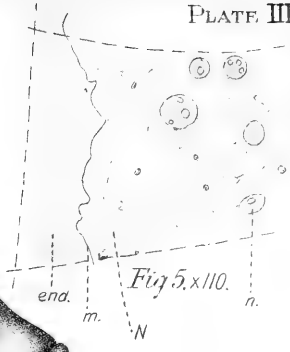
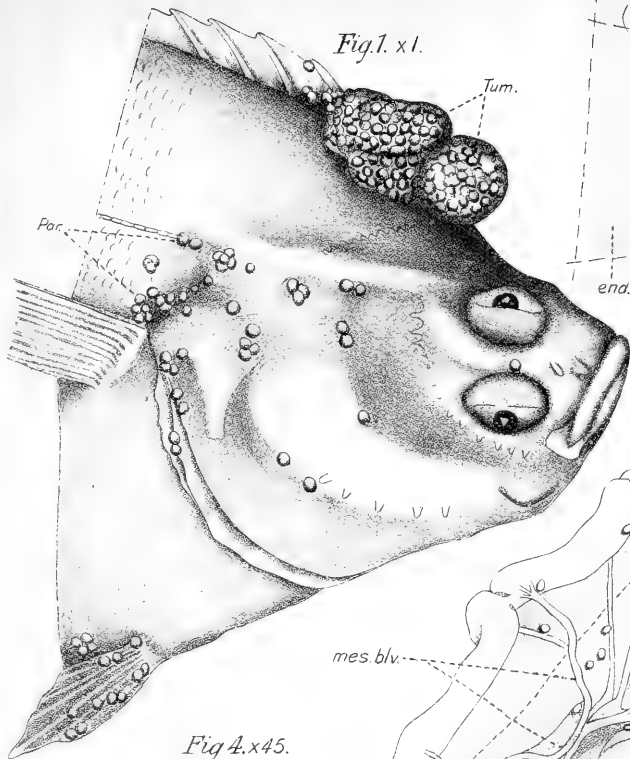
The stains mostly used were Iron Haematoxylin followed by Orange or Eosin, Kleinenberg's Haematoxylin and Fuchsin, and various bacteriological methods.

Fig 1. Head of the flounder, shewing the parasites (*par.*) beneath the skin, but projecting externally. At *tum.* are three huge aggregations of cysts (not proliferations, see text).  $\times 1$ .  
Drawn by J. Johnstone.

Fig. 2. View of one of the coils of the intestine, with attaching mesentery (*mes.*) and blood-vessels (*blv.*). *Par.* are the numerous parasites.  $\times 1$ .

- Fig. 3. Section through one of the parasites in the mesentery. It is lying in an enlarged lymph-space (*spha.*) in the latter. *Lym.*, a layer of lymphocytes around it, *ect.*, = ectoplasm, *end.* = endoplasm, *ret.* = chromatic reticula, *N* = nucleus, *n* = Karyosomes.  $\times 45$ .
- Fig. 4. A section through another, of different shape and larger, beneath the skin (only partly drawn in). *Conn.-tiss.* = a layer of connective-tissue.  $\times 45$ .
- Fig. 5. Part of a section through the nucleus, shewing the faint, irregular reticulum, and karyosomes (*n.*). *m* = the nuclear membrane (see text).  $\times 110$  (drawn in under a high power).
- Fig. 6. Part of a section through the periphery of the parasite. *sph* = the clear spherules at the margin of the endoplasm. Other letters as in fig. 3.  $\times 500$ . Zs. 3 m. Apochr. 1.4 O.I. C.E. 4.





H.M.W.del. Fig.1. J.J.del.

Lymphocystis Johnstonei.

S.B.lith.



## THE FECUNDITY OF THE PLAICE.

By R. D. LAURIE.

It may be useful to put on record two estimations of the number of ova produced by a plaice, which I made at Prof. Herdman's suggestion while working, during last Easter vacation, at the Port Erin Biological Station. The determinations were made by Dr. Fulton's method,\* in which a fractional portion of the roe (or ovary) of the fish is weighed and the ova are counted, and from the numbers so obtained, and the total weight of the two ovaries, the total number of ova present is calculated. No determinations of this kind have hitherto been made from the fish in the Irish Sea.

The fishes dealt with were two ripe mature females, which died in the tanks at Port Erin during April, 1903. The data are as follows:—

	Total length (in centimetres).	Total weight (in grammes).	Total weight of both ovaries.
1	49 ( $19\frac{3}{4}$ inches)	1,346	315·5
2	52 ( $20\frac{1}{2}$ inches)	1,318	181·5

Small portions of the ovarian substance, weighing 10 grains each, were taken from various parts of the organ, and the ova were counted. The results of these counts are given in the following table:—

\* 9th An. Report Fish. Bd. for Scotland. Pt. III., pp. 243-268.

(1.) Anterior region, 10 grains contained	...	744	ova.
"    "    "    "    "	...	762	"
Middle region,    "    "    "	...	721	"
"    "    "    "    "	...	730	"
Posterior region,    "    "    "	...	701	"
"    "    "    "    "	...	735	"
<b>Average for 10 grains</b>		<b>=</b>	<b>732 ova.</b>

(2.) Anterior ventral region, 10 grains contained	804	ova.
"    "    "    "    "	789	"
Anterior ventral lobe,    "    "    "	853	"
"    "    "    "    "	870	"
Middle region,    ...	810	"
"    "    ..."	857	"
Posterior region,    ..."	886	"
<b>Average for 10 grains</b>		<b>= 838 ova.</b>

The differences in number of the ova in the various samples were due chiefly to the mixture in various proportions of eggs of slightly different size, and also to the varying admixture of water and connective tissue—both sources of error which are unavoidable in this method of investigation. Two kinds of ova are present in the roe of the plaice, (1) large ova lightly attached or lying freely in the lumen of the ovary, and smaller immature ova still adherent to the epithelium. In fish (2) which had not begun to spawn, I counted 492 large mature ova in one ovary; their diameter was 1.32 mm. Nearly all the remaining ova were about 1 mm. in diameter, and there were few intermediate in size. This is in accord with the general belief that the process of ripening, or the absorption of fluid by the ova, is very rapid. Fish (1) had just begun to spawn, but a similar contrast between mature and immature ova was evident.

The following values were calculated from the figures so obtained:—

	Total length. Cms.	Total weight. Grms.	Total number of ova in the fish.
1	49	1,346	356,278
2	52	1,318	234,640

It may be useful to compare these results with those obtained by other investigators. Fulton, in the paper quoted, gives the following estimations:—

	Total length. Cms.	Total weight. Grms.	Total number of ova in the fish.
1	44·5	1,368	223,497
2	44·5	1,191	148,470
3	52·0	1,715	323,166
4	56·0	1,914	487,087
5	56·5	2,140	324,749

These figures present much the same appearance as those I have obtained.

Reibisch,\* in 1899, made a lengthy investigation of this nature. His method differed in principle from that devised by Fulton. The ovaries were put into cold water on being removed from the fish and the water was gradually heated to the boiling point and kept at this for  $\frac{1}{4}$  hour. This facilitates the removal of the ova from the ovarian epithelium. The ova, after being separated in this way, are counted by the method employed by Hensen in the quantitative determination of plankton. This is a more accurate method than that of weighing, for in the

\*Wiss. Meeresunt. Kiel u. Helgoland. N.F. Bd. 4. Abth. Kiel, pp. 233-248, Taf. 1. 1899.

latter method a certain amount of ovarian tissue is estimated as ova, and a number of very small ova which will not be spawned at the next spawning period are also included. In two of Reibisch's estimations the weight of the ovaries after complete spawning was 7·8 grms. and 18·3 grms. By employing this method Reibisch made a large number of estimations, some of which are quoted in the following table:—

	Total length in Cms.	Total weight in Gms.	Total number of ova in the fish.
1	54	1,770	223,250
2	45	969	109,500
3	42	1,100	558,500
4	40·5	693	736,250
5	38·5	585	250,750

Reibisch did not count the very small eggs which are always present in the ovary. If these had been included values of  $2\frac{1}{2}$  millions of ova would have been obtained in some cases, and this represents a degree of fecundity certainly not attained by the plaice. Such very small eggs he contends cannot ripen for the next spawning period.

All these results, and my own have the same tendency, show, as Reibisch has observed, that there is no recognisable relationship between the size or weight of the fish and the number of eggs produced by it at the spawning period. The age of the fish has in all cases to be considered. We know that the rate of growth is very variable. Probably most plaice attain sexual maturity at the same age, but the size and weight at this age may be very different in a number of specimens. Thus a mature female 13 inches long, and an immature female of 19 inches, have been taken from the Irish Sea.

## AN OUTLINE OF THE SHRIMP QUESTION.

By W. A. HERDMAN.

In the course of last summer Mr. Fell, the Chairman of the Lancashire and Western Committee, sent me a letter in which he suggested that it might be useful to give in this Report a detailed statement in regard to the natural conditions under which shrimping is carried on in Lancashire waters, and as to the relations between the shrimps and the young flat-fish. It is very desirable that such a statement should be drawn up, but the time has not yet come when we are in a position to do so in any detail or with any finality. Periodic investigations carried on over a couple of years, such as cannot be undertaken until we have a scientific steamer at our disposal, are necessary to clear up certain points in life-history and bionomics. Still it may be useful to give now an outline of what is known and what has still to be determined in connection with the subject, and to take what steps are possible to us during the coming year to obtain statistics which may aid us in tackling some of the unsolved problems.

The subject is a very diverse and complicated one, which leads us into economic as well as scientific questions, and although one might desire that any proposed regulations of the shrimping upon grounds frequented by young fish should be considered and settled on the scientific evidence, still it can scarcely be doubted that administrators will take cognisance of the economic questions even if they do not adjudicate wholly upon them. Consequently in any discussion of the subject we must be prepared to take fully into account the important interests involved in the shrimping industry, and not to sacrifice unduly any pre-

sent material advantages to what may be considered somewhat problematical benefits in the future. We must be prepared to give full and accurate information as to the economic effects of any suggested restrictions—the effects, that is, upon the fishermen and others engaged in the industry, and upon the markets and supply to the public, as well as upon the shrimp and fish populations in the sea.

We desire to know, amongst other things :

(1) The number of boats and men employed in shrimping on each of the grounds.

(2) The produce of the fishery throughout the year, and especially during certain periods—March to June, July to September and October to February.

(3) The approximate amount of destruction of young fishes under various circumstances.

(4) The subsidiary interests involved, *e.g.*, potting and selling the shrimps.

(5) The probable effect upon employment which would be produced by the imposition of a close season.

(6) The extent to which foreign preserved shrimps are imported, and the probable effect of any change in restrictions upon such importation.

In regard to some of these matters we already have a good deal of information, and can readily obtain more. For example:—

Under (1).—There are now 70 boats fishing on our coasts hailing from Southport and Marshside alone, all engaged in shrimping at some time of the year. These are all half-decked boats, and most of them have been built during the last six years. The catching power of this fleet is said to be now ten times as great as was the case 25 years ago. Each boat is worked by two men, and the takings are divided into 5 shares, of which each man takes two and the boat the fifth share. Each



fisherman finds one net and the boat has to find two nets when engaged in shrimping. We can readily give similar information in regard to other parts of our coast.

Under (2).—The average take at Southport during the twelve months ending December 31st, 1902, was 30 quarts per boat for each fishing day. The statistics for other boats and periods can readily be ascertained.

Under (4).—In addition to about 200 fishermen in Southport and Marshside all more or less engaged in shrimping, the potting of the shrimps is an important local industry, and provides a fair amount of work in boiling, picking and potting the catch. There are in all about 30 shrimp-potters in Southport, and they utilise nearly all of the shrimps that are caught on our coast, and distribute them to nearly every town in Great Britain. The fishermen's wives and children boil and pick the shrimps, and make them ready for the potters to prepare for market. During the last 15 years the Southport shrimp-potting industry has increased tenfold. It must also be remembered that the boat-builders, net-makers, butter merchants, printers, pot manufacturers, and railway companies all share, more or less, in the profits derived from the local shrimping industry.

So far we have been dealing with fairly easily ascertainable facts, but in (5) and (6) we come upon contentious matters which are not strictly scientific, and in regard to which it might be difficult to get agreement. In (3) we also meet with difficulties, but of a different nature. This is a scientific question, and the answer is to be obtained as the result of a large number of reliable statistics. Precautions must be taken to see that the conditions under which statistics may be taken are normal and such as hold good in the course of the fishery. Moreover, the practice may vary from time to time, or with

different boats, and so affect the result. At the best, it can only be an approximation that will be obtained; but, still, it can scarcely be doubted that the annual destruction is enormous, and that the young flat-fish so lost are potentially very valuable. It seems probable that the total annual destruction of young fish by shrimping in our district is to be measured by the hundred million. We know, moreover, that the Lancashire shrimping grounds constitute our most valuable fish nurseries, which bear a definite and important relation to the fishing ground off-shore, and we may assume that if young fishes are allowed to grow undisturbed on the in-shore nurseries, they will later on become marketable fishes on the off-shore fishing grounds. We may state also that as all the common flat-fish pass through a stage in which they inhabit a shallow-water area, the number of marketable fishes on the off-shore grounds will vary as does that of the small fishes in the nurseries. If, then, we are able to protect our young fishes in the coastal waters, under ordinary circumstances they should turn up a year or two later on the grounds outside. The preservation of immature fishes ought to be very beneficial to the off-shore fisheries—more beneficial even than the preservation of spawn, because the mortality during the period between hatching and the stage when the young fishes make their appearance in the nursery is so very great that a given number of young fishes represents many hundred or thousand times that number of eggs or embryos.

Now, this *a priori* argument is probably quite a good one, and if we had no other means of investigating the matter, we should probably be justified in relying upon it. But the points enumerated in the last paragraph in relation to the life-history of the fish, are eminently suitable for biological and statistical investigation. And it

would be manifestly unfair to the shrimping industry to impose restrictions, and possibly interfere with the livelihood of so many fishermen, before making those further investigations that are practicable, and are most likely to throw much light upon the matter.

We may take the probable effect of such restrictions upon shrimping as have been suggested as an instance that will show the problems that confront us, and the kind of information we want. It is interesting to speculate upon what would be the resulting effect upon the fish and shrimp populations if shrimping were either stopped or restricted to certain months on particular grounds. The number of immature fishes would probably increase, at least for a time—possibly permanently—and this might be expected to lead to an increase in the marketable fishes on the off-shore grounds a year or two later. The great numbers of young fish at present destroyed would be preserved, and, no doubt, the number of shrimps would also increase considerably. Interesting questions would then arise as to whether the fish and shrimps would be competitors for the same food, and whether there would be enough for both in their increased numbers. Taking the plaice as an example of the young fish, we know that when very young it feeds mainly on Copepoda—we have found their stomachs crowded with *Jonesiella hyæna* and other allied forms. But, after the metamorphosis, the young fishes from, say,  $1\frac{1}{2}$  to 4 inches in length, feed largely upon worms such as *Nereis* and *Pectinaria*, upon small Crustaceans such as *Mysis* and the Amphipoda, and even upon small shrimps. Later on, the fish adopts its proper adult food, which is Mollusca (mainly small cockles, mussels and allied bivalves).

Shrimps, we know, are general feeders (using small Molluscs and other animals, and also Algæ) and

scavengers, and will subsist largely on dead material. Consequently, if there should be any scarcity of food, I do not doubt that they might, to some extent, compete with the little fish by eating the worms and smaller crustacea, but it is improbable that there would be any such scarcity. We may put great trust in the recuperative powers of the invertebrate fauna of the seabottom. Even in the spots where fishes are most crowded, we bring up plenty of invertebrate food material in the dredge and trawl; and, moreover, if there were any scarcity of food the star-fishes and crabs, which are so abundant on these grounds, would move away.

A greater danger might be brought about by the increased numbers of shrimps and young fishes attracting many skates, rays and other larger predaceous fishes to the ground. In fact, the disturbance of the fauna might be very wide spread. Some forms of invertebrata might be either favoured or the reverse by the changed conditions, and then that change in the food might re-act upon the fish population. For example, if the smaller crabs which are usually present in enormous profusion on the shrimping grounds, found conditions uncongenial and migrated to other banks and channels, the Gadoid fishes, which feed largely upon such crabs, might in their turn be affected.

The chief enemies of shrimps in our district (see our Report for 1894) are skates and rays, whiting, gurnard, and the larger Gadoid fishes. The latter are not abundant on the shrimping grounds, but skates and rays seem to have increased on the Blackpool closed ground, possibly as a result of the more abundant feeding upon that sanctuary. A careful detailed comparison of this closed area with the open grounds on both sides of it might give information as to effects to be expected by closing other

parts of our fish nurseries to shrimping. We desire to know not merely the statistics of the "catch," *i.e.*, the fish and shrimps in each haul, but also the relative abundance of invertebrate (food) animals on closed and open grounds, and full details as to their kinds and abundance, their life-histories and changes throughout the year, and their food, for comparison with that of the shrimps and fishes.

Furthermore, we obviously require to have full information in regard to the structure and habits of both the shrimp and the fishes throughout their life-histories before we can be sure that any alteration in relative numbers will be permanent. We know a good deal about such matters already, either in our own seas or in other parts of the world. Professor J. S. Kingsley,\* in America, has made us acquainted with the development of the early stages of the shrimp (*Crangon vulgaris*), and Dr. Ehrenbaum has given us much information as to the structure and life-history of the shrimp at Heligoland.† All that is a help, but still there are many local details that must be worked up on our own ground. We must know exactly where and when the various stages occur, and in what abundance, and in what association.

Then turning to the little fishes, if we take the plaice again as an example, Mr. Johnstone has given, in the appendix to our own Report for 1901 (No. X., p. 211), a summary of what is known as to the development on our coasts, and the habits and food at the various stages. If, however, we take the young sole—a very valuable, and frequently abundant, constituent of the fauna on the shrimping grounds—much less is known, and the exact

\* Bulletin of the Essex Institute, 1887 and 1889.

† Naturgeschichte von *Crangon vulgaris*, 1890.

history of the young stages in our district has still to be written.

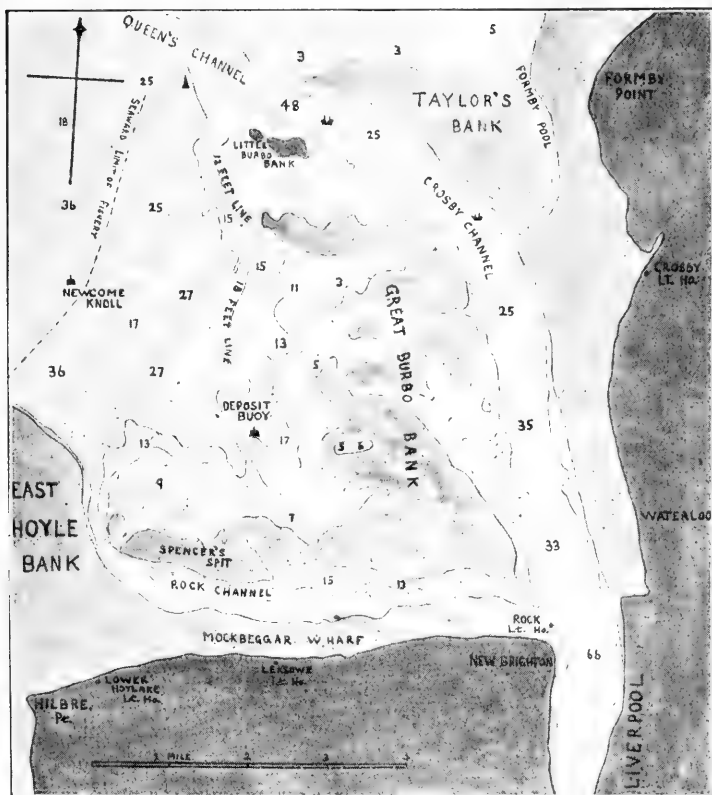
In our Report for 1900 (No. IX., p. 39), we had an article by Mr. Johnstone and Dr. Jenkins on the shrimp trawling statistics of the Mersey grounds, in which they give a description of this remarkable region, samples of various kinds of hauls and such conclusions as they were able to draw from an examination of our statistics (over 1,000 forms) for the period 1893-99.

The accompanying figure shows the great banks surrounding the mouth of the Mersey, and the channels between them. Shrimping is carried on over almost the whole of this ground, the extent of which is roughly 16 to 18 square miles. This is only a sample of the remarkable Lancashire and Cheshire shrimping and nursery grounds. Similar areas exist in Morecambe Bay, off the Ribble, and in the estuary of the Dee, and on these grounds we find associated with the shrimp an abundant fauna of both fishes and invertebrates.\* The chief fish are the plaice, dab, flounder, sole and solenette, with occasional whiting, haddock, cod, young brill and turbot, sprats, sand-eels and sting-fish. The commonest invertebrates are crabs, starfish and shrimps. Examples of hauls on different nursery grounds, and at different times of year, are given in our ninth and tenth Reports, and in the Memoir, "Fishes and Fisheries," by Mr. Dawson and myself, and need not be repeated here. Mr. Johnstone has summarised his previous work, from our statistics, as follows:—

(1.) The great abundance of young flat-fishes on the shrimping grounds. They are, in order of frequency, dabs, plaice, and soles; solenettes are also very abundant, in some areas nearly as abundant as the soles.

\* See Herdman and Dawson, Lancashire Sea Fisheries Memoirs, No. II. Fishes and Fisheries of the Irish Sea, 1902.

(2.) These fishes are generally most abundant during June, July, August and September. But on certain of the grounds in winter (December and January) plaice may be more abundant than in summer.



Mersey Shrimping Grounds.

(3.) The number of fish caught by the shrimp trawl varies greatly from time to time. On the Mersey grounds the average for September, 1893, was nearly 3,000 per haul, and the average for September, 1899, was nearly

95 per haul; the average for the seven years (1893-99) was nearly 567 per haul.

(4.) The number of fish caught on closely-adjacent grounds is sometimes very different, even when the depth and bottom are very similar. Caution is, therefore, necessary in comparing neighbouring grounds, even when these are similar physically, *unless we have previously ascertained that they are almost similar biologically.*

(5.) Our statistics for the 10 years 1893-1902 show that:—

1. Young plaice on the shrimping grounds have diminished.
2. Young soles on the shrimping grounds have increased.

The distribution and relative abundance of these young food fishes throughout the year on the various banks and other shrimping grounds will have to be still further studied from more abundant statistics before these variations can be considered as established and understood.

In regard to the shrimp, also, we want more information as to the relative abundance of the sexes, the spawning periods, the time of hatching, the rate of growth, the proportional numbers throughout the year, and the distribution on and about the shrimping grounds. It must be remembered that work done in other seas cannot be utilised in our own district except as a general guide to investigation. An intensive study of the fish and shrimp populations on the areas we propose to deal with will have to be undertaken.

Such investigations as we contemplate would require one whole day's trawling per week on each of the principal grounds to be compared, say in Morecambe Bay, on



the Blackpool ground, and on the Mersey banks. It seems unlikely that we shall gain much further, or more exact, information than we now possess with regard to the distribution and inter-relations of the shrimps and fishes with any less expenditure of time. Three hauls on different parts of the grounds should be taken during each day's work. It need scarcely be pointed out, however, that these hauls would also give us statistics in regard to other fishes, and generally add to our knowledge in various other useful directions. It would, probably, be impossible to report satisfactorily upon all the matters referred to in the above outline in less than two years, and a consideration of the work and statistics required to solve the problems shows how necessary it is that we should have a second steamer in this district, devoted solely to scientific and statistical investigation.

## RECENT INVESTIGATIONS ON PEARLS IN SHELLFISH.

By W. A. HERDMAN.

In last year's report I referred to the work done by Dr. H. L. Jameson upon pearl-formation in the Common Mussel at Piel. Since then Mr. Hornell and I have published a preliminary notice\* of our results obtained with the Ceylon Pearl Oyster: and more recently several French investigators, Seurat, Giard, Dubois and Boutan have written short notes dealing with the same important matter.

As is so often the case, the first statement of the correct view was made long ago and afterwards forgotten or contradicted, and the more modern work is largely a resuscitation, extension and demonstration of an older view. Filippi, in 1852, showed that the parasitic Trematode worm *Distomum* was the cause of pearl-formation in the freshwater mussel of rivers and lakes, and other naturalists soon after extended the discovery to other pearl-producing molluscs and to other worm parasites. To Dr. Kelaart belongs the honour of having first connected the formation of pearls in the Ceylon oyster with the presence of vermean parasites. He and the Swiss Zoologist Humbert, who was with him at a pearl fishery off Aripu, found various parasitic worms infesting the viscera and other parts of the pearl oyster, and they agreed that these worms played an important part in the formation of pearls. Kelaart moreover, in 1859, made the remarkable suggestion, in the case of the Ceylon pearl oyster, that it might be possible to increase the quantity of pearls by infecting the oysters in

\* Southport British Association Report, Sept., 1903; and Report on Ceylon Pearl Fisheries, Part I., Royal Society, Nov., 1903.

other beds with the larvæ of the pearl-producing parasites. This is exactly the idea that has lately been revived by a French professor.

Turning now to European shell-fish we find that our countryman Robert Garner in 1871 associated the production of pearls in our common English mussel (*Mytilus edulis*) with the presence of Distomid parasites.

Professor Giard, in 1897, and other French biologists since have made similar observations in the case of *Donax* and other Lamellibranchs—Giard describing\* the Distomid worm which he found as a species of *Brachycoelium*. We now come to quite recent years, during which there has been great activity. Prof. Raphael Dubois in 1901 ascribed the production of pearls in mussels on the French coast to the presence of the larva of *Distomum margaritarum*. The next year (1902) Dr. H. L. Jameson† followed with a more detailed account of the relations between the pearls in *Mytilus edulis* and the Distomid larvæ, which he identified as belonging to *Leucithodendrium (Brachycoelium) somaterie*—the same sub-genus as Giard had found some years previously. Jameson's observations were made partly at Billiers (Morbihan), the same locality at which Dubois had also worked, and partly at our Lancashire Laboratory at Piel. Dubois published a further note‡ in January, 1903, in which he stated that Jameson had come to Billiers after his departure and had confirmed the discovery made previously, first by Garner and then by himself. But Jameson had really done more than that. He had shown that it is probable that the parasite causing the pearl formation in our common mussel (not however in the Ceylon Pearl Oyster) is the larva of

\* Comptes rendus, Soc. de Biol., 13th Nov., 1897, p. 956.

† Proc. Zool. Soc., Lond., 1902, p. 140.

‡ Comptes rendus, Acad. Sci., 19 Jan., 1903.

*Distomum somaterice*, a Trematode worm, the adult of which lives in the intestines of the Eider duck and the Scoter duck. He also stated that the larva inhabits Tapes or the cockle as a first host before getting into the mussel, and gave figures of the parasite in various conditions.

Two very important matters are, however, left in a somewhat unsatisfactory condition by Jameson's paper. The first of these is the mode of origin of the epithelial sac which encloses the larval parasite, and which secretes from its cellular walls layer after layer of nacreous material so as to form a pearl. The presence of this sac was known before (Von Hessling, 1858, and Diguët, 1899), but no one has yet satisfactorily traced its origin. Jameson several times compares it with the epithelium on the outer surface of the mantle, using such terms as "similar to" and "indistinguishable from" but he evidently considers that it has nothing to do with that epithelium, although it produces an identical pearly secretion. He describes the sac round the parasite as formed by the proliferation of a few cells which "are basally continuous with fibres of connective tissue." He also says of it, "This epithelium appears to arise quite independently of the outer epidermis." Now such a mode of origin as this is very unlikely, and although I have not had the opportunity of re-examining pearl-bearing mussels on this point since Jameson's paper appeared, I think there can be little or no doubt that the cells of the pearl sac are directly and genetically connected with the exactly similar cells on the outside of the mantle. It is almost certain that the parasite in burrowing into the mantle carries in with it one or more epidermal cells which proliferate to form the sac. As the Distomid larvæ are found moving on the inner surface of the shell before coming to rest in the mantle they must traverse the epidermis, and it is natural to

suppose that in their migration they may push some epidermal cells in before them. At least this is not such a violent assumption as that the connective tissue in the centre of the mantle can produce an epithelial sac the cells of which are indistinguishable both in structure and in functions from the epidermis outside.

In giving a preliminary account of pearl formation in the Ceylon Pearl-Oyster to section D of the British Association last September I took up the position that the sacs enclosing the pearls were in all cases of ectodermal (epidermal) origin: and I am glad to find that Prof. A. Giard, in a recent note\* on the subject, takes the same view, and considers that in the case of Jameson's mussel there is a "passive immigration" of the epithelial cells caused by the migrating parasite.

The second point which I feel is not yet satisfactorily settled, is the supposed infection of the mussel with parasites by the Tapes in France and the cockle in the Barrow Channel. So far as regards the latter case, Jameson's conclusion is based upon the experiment of placing some mussels, which he supposed to be free from parasites, in a tank with French Tapes which were infected, and examining the mussels from time to time until he found they contained the parasites (*Cercaria*). Now in such an experiment it is necessary to be quite sure of the material used, to deal with sufficiently large numbers and to have control experiments. Jameson does not seem to have taken these precautions. He says of the material:—"These mussels, of which I examined a number, were practically without parasites. About one in every five of the largest examples contained a *Cercaria*, one had two *Cercariæ*, and one contained a small pearl." This can

\*Comptes rendus, Soc. de Biol., Paris, 19 Dec., '03, lv., p. 1618.

scarcely be described as free from parasites! He used 70 mussels: if we take his own figures, one in five, as correct, then about 14 of the mussels were infected at the beginning of the experiment. We find from his records that he only examined 13 of these mussels (2 after 11 days, 6 after 2 months and 5 after  $6\frac{1}{2}$  months), and found 12 of them infected. But it is obvious that that number might possibly have been infected from the beginning or may have become infected at any time from neighbouring mussels. The theory of transference of the parasite from one mollusc (such as cockle) to another (the mussel) may be true, but it is not proved by these experiments. It was not shown that the mussels were free from parasites at the start, the numbers in the recorded experiments are too small to yield definite conclusions, and the observations should clearly be repeated using hundreds of cockles and of mussels with well-devised control experiments. In order to show the necessity for large numbers in this kind of work I may add that Mr. Andrew Scott having informed me of Dr. Jameson's observations at Piel, I had some samples of these same mussels and cockles sent to the Liverpool laboratory where, along with Mr. Walter Tattersall, B.Sc., and Mr. J. Pearson, B.Sc., I made (in October, 1902), an independent examination of them with results that do not altogether agree with Dr. Jameson's.

We may distinguish between 4 kinds of mussels, examined by Jameson and myself, as follows:—

- (A.) From the beds opposite the Piel Hatchery—"where every specimen is abundantly infected . . . and almost every specimen contains pearls" (Jameson).
- (B.) From the piles of the old pier at Piel—"practically without parasites" (Jameson).

(C.) From Roosebeck Scar, outside Barrow Channel—  
“not infected” (Jameson).

(D.) Roosebeck Scar mussels transplanted to foreshore at Piel two years ago—“all were infested”—“each contained several small pearls” (Jameson).

Of (A.) I examined a sample of 25 mussels, which contained in all 151 pearls and 11 parasites, but 4 of the specimens had neither pearls nor parasites, and no less than 18 out of 25 had no parasites. I cannot therefore agree that “every specimen is abundantly infected.”

Of (B.) I examined also 25 mussels, which showed in all 21 pearls and 22 parasites, 7 had neither pearls nor parasites, and 13 had no parasites. These then showed far fewer pearls than (A), but twice as many parasites, and fewer of them were free from infection. They can scarcely be called “practically without parasites.”

Of (C.) I examined 28 mussels, which contained 73 pearls and 37 parasites, 4 had neither pearls nor parasites, and only 9 (out of 28) had no parasites. These then are evidently just as much infected as the mussels on the Piel foreshore. (A.)

Of (D.) I examined 24 mussels and they contained 65 pearls and 26 parasites, 3 had neither pearls nor parasites, and 12 out of 24 had no parasites. So in place of these transplanted “Roosebecks” having become more infected on the Piel shore, they on the whole showed rather less infection than the mussels taken direct from the parent bed.

Finally, I examined a sample of 25 cockles from Piel, and found in them eight pearls, but no parasites at all of the right kind. This does not support the view that the

cockle contains the earlier stage of the parasite, and passes it on to the mussel.

A couple of weeks later, at the end of October, 1902, Mr. Scott and Mr. Johnstone being together at Piel, examined some further samples with the following results :—

(A.) Examined 61, got 390 pearls and 191 parasites.

(B.) Examined 103, got 100 pearls and 61 parasites.\*

(D.) Examined 53, got 161 pearls and 66 parasites.

(Roosebeck Scar mussels could not be got at the time).

The most noteworthy difference between these results and mine are in the case of the parasites in (A.), where Mr. Scott found about seven times as many as we did. The sample of (B.), in this case also, it will be noticed, is by no means free from infection. Since then Mr. Scott has examined a few more samples with slightly different results : but I do not wish to attach too much weight to any of these figures. The point I desire to make is that in working with these comparatively small samples each examination gives a different result, and that consequently it is necessary that some one like Mr. Scott, living on the spot, with abundance of material at hand, and with tanks for experiments under constant observation, should make a comprehensive investigation of some hundreds of each kind of mussel and cockle, in order to clear up the distribution of pearls and parasites, and settle the question of infection.

Prof. McIntosh † describes the examination of 700 mussels from near St. Andrews, and finds that 300 in all, or nearly 43 per cent., were pearl-bearers—a small proportion compared with ours at Piel.

† Ann. and Mag. Nat. Hist., June, 1903.

\*As this was going to press Mr. Johnstone informed me that before he made the examination referred to a gale had washed away some of the piles of the old pier, and that his sample of (B) was obtained from a lower level than Jameson's, and so may have contained more parasites.



Prof. R. Dubois has recently turned his attention to the Mediterranean Coast. He found that the Southern French mussel (*Mytilus gallo-provincialis*) forms pearls caused by another Distomid, distinct from that of Brittany. He then worked at the acclimatisation of a true Oriental Pearl-Oyster ("Pintadine") in French waters, and the artificial production of pearls.\* He brought the pearl-oysters from the Gulf of Gabes, in South Tunis, to the marine laboratory at Sfax, and caused them to multiply and increase in size. The pearls produced in Tunis are small and very rare—it is necessary to open 1,200 to 1,500 oysters to find one pearl; but Dubois tells us† that by placing them on ground where *Mytilus gallo-provincialis* becomes infested with pearls and parasites, he very easily provoked the production of fine pearls in the "pintadine" to such an extent that three successive individuals opened contained each two little pearls.

This, if corroborated, is a remarkable circumstance from several points of view. First it will, if it proves a success, be a striking verification of what Kelaart in Ceylon, fifty years ago, declared might be done. Secondly, if the "pintadine" in question is really the same species as the Ceylon Pearl-Oyster (Giard considers that it is not), it is curious that a Distomid parasite should prove to be so efficacious in setting up pearl-formation, since Mr. Hornell and I found in the Gulf of Manaar that the pearl-parasite is a Cestode of the genus *Tetrarhynchus*. Thirdly it is remarkable that the parasite of the *Mytilus* should transfer itself so readily to a new host belonging to a distinct family.

\* Comba had, however, in 1898, introduced the same mollusc on the south coast of Italy, and experimented in artificial pearl formation.

† Comptes rendus, Acad. Sci., 19th October, 1903, p. 611.

It is this last paper by Dubois that has given rise to various more or less exaggerated or even erroneous statements in the public press, such as that the Pearl-Oyster must be infected with a microscopic germ in order to render it pearl-producing: or even that inoculation with a serum causes the oyster to produce artificial pearls. The parasite that causes the irritation is, as has been known for many years, not a "germ," and still less a "serum," but a worm which is visible to the eye—a worm which in *Mytilus* seems to be usually a Trematode, and in the Ceylon Pearl-Oyster (*Margaritifera vulgaris*), according to Mr. Hornell's and my observations, is certainly a Cestode.

According to an interesting note by Prof. Giard\* the discovery of Cestode larvæ as nuclei of pearls, which we made upon the Ceylon Pearl-Oyster in 1902, has been corroborated by M. G. Seurat, working independently in his laboratory at Rikitea in the island of Mangareva (Gambier Archipelago). The oyster on which Seurat worked was a *Meleagrina*, and the Cestode parasite found is, according to Giard, an *Acrobothrium*, or some allied form. It is possible that some of our Ceylon Pearl-Oyster parasites may also belong to the genus *Acrobothrium*, although others of them are certainly Tetrarhynchids.

Giard, in a further note in the same Journal (p. 1225), discusses the statements that have been made in regard to "margarose artificielle," and evidently considers that Dubois' claim to have established the artificial production of pearls is not yet justified by the facts. Last of all M. L. Boutan† shows that fine pearls do not really differ from nacre-pearls, since both are secreted from open or closed epithelial sacs, derived from the epidermis; and Giard

\* Comptes rendus, Soc. Biol., Paris, 6th Nov., 1903, lv., p. 1222.

† Comptes rendus, Acad. Sci., 14th Dec., 1903, p. 1073.

very properly replies a few days later\* that this fact is quite in accord with general principles, and was previously known. M. Boutan in a letter (20th Jan., 1904) informs me that he is on the point of departure for the East in order to investigate the matter further.

Notwithstanding this recent activity, especially amongst our French neighbours, there is still a good deal of work to be done before the whole process of pearl formation can be considered as cleared up. We want to know especially:— 1st, The exact details of formation of the pearl-producing epithelial sac when deeply placed in the tissues, and 2nd, the complete life-history of the parasite inside the sac.

\* Comptes rendus, Soc. Biol., Paris, 19th Dec., 1903, p. 1618.

## SEWAGE AND SHELL-FISH.

By W. A. HERDMAN.

The first "Lancashire Sea-Fisheries Memoir" (Oysters and Disease), by Prof. Boyce and myself in 1899, drew attention to the serious amount of sewage contamination in certain samples of shell-fish. Since then much additional evidence has been obtained, both in our own district and also from other parts of the coast, that, as the result of the discharge of unpurified sewage into tidal waters, extensive pollution of the shell-fish beds and of the waters generally of our coastal fisheries takes place—causing serious injury to health, and affecting the prosperity of the fishing industries. It has now been established, and is generally admitted, that shell-fish contaminated by sewage may produce enteric fever and other illness in human beings.

Mr. Dawson and I, in the evidence we gave before the Royal Commission on Sewage Disposal, both discussed cases of serious contamination of shell-fish beds in our district. Since then, however, Mr. Dawson has made a special survey of the mussel beds, and has drawn up a report which, with his permission, I insert here in order that it may be placed on permanent record. This report was submitted to the Lancashire and Western Sea-Fisheries Committee last November, and has since been made public.

Report by Mr. R. A. Dawson, Superintendent Lancashire and Western Sea Fisheries, on the Mussel Beds of the District with regard to danger of pollution by sewage.

“At the last meeting of the General Purposes Committee, held on the 14th August, I was instructed to lay before the Committee, at the next meeting, a list of Mussel beds, in my opinion, contaminated by sewage. Together with Dr. Sergeant (the Medical Officer of Health for the County of Lancaster), Mr. Halliwell (Chief Inspector of the Ribble Watershed), Mr. Scott (the Resident Scientist at Piel), and others, I visited and inspected the sewage outfalls in Barrow Channel, Ulverston, Morecambe, and Heysham, and the Mussel Beds in the Lune. I have also inspected the different sewer outlets in other parts of the District likely to cause contamination to Mussels.

The sewage from Barrow and Dalton is discharged into Barrow Channel in great volume, and has the appearance of being untreated. This sewage joins the stream in the Barrow Channel, and together they flow over the different grounds where Mussels are found. On the day we were there, the sewage was being discharged as late as low water. I may remark that, although some Mussels are taken from here for human food, the bulk are only used for bait: Periwinkles, however, are taken in large numbers, and London seems to be the chief Market for them. The sewage from Piel also flows into the Barrow Channel.

At Ulverston, although some treatment is attempted, the effluent appeared to me to be very dirty. This discharge joins the main stream at the west end of the Slag Bank, about a mile above a Mussel scar; they flow on together round the edge of the Mussel scar at low water

and down Channel over other scars; earlier on the ebb tide the stream will flow over the first scar.

At Morecambe and Heysham there are in all eight sewer outlets, all of which discharge some considerable distance away from the *principal* Heysham Mussel Beds. At the same time, three of them empty close to where sizeable Mussels can be taken in payable quantities. I understand, however, it is the intention of the Authorities to turn all the Morecambe sewage out through a new pipe, which is now laid down and in use, and to do away with the old sewers referred to. With regard to the new sewer outlet, it does not, to my mind, project far enough into the channel, and should be carried about 75 yards farther out. It discharges on a scar which, although not a main bed, is one where sizeable Mussels can be obtained in fair quantities, and this scar might easily become well stocked with fish. The fishermen agree that the sewer outlet should be carried farther out. I may also draw attention to the fact that, at the time I refer to, the sewage was being discharged at low water, and, although some of it is supposed to be treated, what we saw appeared to me to be untreated.

In the Lune the sewers discharge a considerable distance up the river, above the Mussel Beds; and although they join the main stream which flows alongside and, in some states of the tide, over Mussel Beds, there was nothing we saw that, in my opinion, exposed the fish to serious danger of contamination. In the Wyre, with the exception of a sewer outlet at Knott End, the sewage is discharged on the Fleetwood side of the river, and some distance from the Mussel Beds. In the Ribble the Preston sewage is dealt with at the sewage farm; but at Lytham the sewage joins the main stream and together they flow on over the Mussels attached to the training walls, and

over the Church Scar Mussel Bed about a mile below Lytham. At St. Annes the sewage is discharged not far from the Mussel Bed, and flows with the ebb tide over it.

In the Mersey five sewers discharge about the Egremont Mussel Bed, one on to the bed, and the other four—although the pipes have been carried down to low water—discharge close to the bed, and in fact over that portion which does not come adry. A few Mussels are at times also taken near the sewer mouth at Rock Ferry. The Wallasey Bed lies some distance below the sewer outlet at New Brighton.

At Rhyl very few Mussels are taken. In the Conway river there are valuable Mussel Beds. The principal beds lie a considerable distance below the sewer outlets; but Mussels are found and taken near the Deganwy sewer outlet, and also not far from the Conway sewer outlet. Opposite Deganwy, and in the Channel, a large strike of Mussels has taken place which extends along the opposite side of the river for a considerable distance, both up and down stream.

At Portmadoc the main sewer empties into an open ditch or stream about 1,000 yards above the harbour, and then flows on through the harbour down channel seawards. There are also two smaller sewers, one which empties into the harbour and the other just above it. Mussels are taken in the harbour and in different parts of the channel over which the sewage and stream together flow. Mussels are also taken from the scars which lie to the south of the harbour. At Barmouth the sewage is discharged a considerable distance below two of the beds. The third bed is situated opposite the sewer outlet, but on the south side of the river—the sewage being discharged on the north side. In the Dovey, the Aberdovey sewage outfall enters the main stream about 220 yards below a Mussel Bed, and 650

yards further down stream there is a second bed. The sewage is mixed with a large quantity of water on joining the main stream, and is carried seawards over the bar—a short distance away—on the ebb tide.

I have thought it better to describe the different sewage outfalls at length, in order that the Committee may know their position in relation to the Mussel Beds.

As regards the list of Mussel Beds which, in my opinion, are contaminated with sewage, I think there can be little doubt that grave danger to health exists in eating Mussels gathered from the Egremont Bed. Here we have direct evidence that death has resulted from eating Mussels gathered near the Egremont Ferry slip. At the moment there are not many sizeable Mussels left, but on tides which cause big ebbs a number of persons may be seen gathering them. A strike of young Mussels has, however, recently taken place, and the fish are now growing fast. Considering the very large quantity of sewage discharged into the Mersey in this neighbourhood, I am of opinion that Mussels should not be removed for human food between Rock Ferry and New Brighton. To this there can be no reasonable objection. The fishermen are not dependent on Mussels from this place; the beds are of little value; and the danger to health from eating them is, I think, a serious matter. With regard to other Mussel Beds, I hesitate to report on them, pending Dr. Sergeant's report to the Public Health Committee for Lancashire, and the report of the result of the analysis of the samples of the water taken by Mr. Halliwell from the vicinity of the beds.

The whole question of pollution of Shell-fish by sewage is both a difficult and complicated one. In the case of the Egremont Bed, which is practically situated in the centre of sewage deposit, there can be little doubt as to



the advisability of prohibiting Mussels being removed for human food: but there are other places where—although sewage outfalls are nearer the Mussel Beds than one could wish—the deposit is mixed with a large quantity of water, and there is no direct evidence that the fish are in anyway polluted. To my mind it would be of assistance in safeguarding the public if samples of Mussels were regularly sent to the Laboratories, examined by experts, and when found polluted the Sea Fishery Committees should have power to temporarily close the bed pending further action by the different County Authorities, Local Government Board, or Board of Trade.”

It will be seen that in this report, made in October, 1903, Mr. Dawson recommends that when samples of mussels were found to be polluted the Sea Fisheries Committees should have power to temporarily close beds pending further action by County and Central authorities.

As a result of Mr. Dawson's report, samples of mussels from the Mersey were sent to the Liverpool Sea Fisheries Laboratory for inspection, and I give below the report which I sent in as the result of Mr. Johnstone's bacteriological examination. This report has also been laid before the Committee and made public, and has been submitted to Lord Onslow at the Board of Agriculture and Fisheries.

Bacteriological Report on Mussels collected at Rock Ferry,  
on 4th November, 1903.

“The mussels brought to the laboratory were rather small, and the fish were very thin and soft. The mantles, or parts of the body where the spawn should be developing rapidly at this time of the year, were very thin. The byssus or ‘weed’ was very easily torn away. Quite apart from the question of sewage contamination, the fish were in very poor condition as articles of food.

Sufficient time has not yet elapsed to make a complete bacteriological analysis of the mussels. We have only attempted to demonstrate whether or not the mussel bed was grossly contaminated with sewage matter, and whether the fish had been feeding on the sewage. The stomachs of a dozen fish were examined, as well as the water in the shells. All the stomachs, with the exception of two (which were sterile), contained sewage bacteria in quantity.

In the examination of the stomach contents, cultures were made on phenolised agar, on Dr. Grünbaum's neutral-red, bile-salt, agar medium, and in milk tubes. The object of the first two cultivations was to isolate bacteria of intestinal origin, and of the latter, to isolate *Bacillus enteritidis sporogenes*. Hardly any bacteria except those belonging to the 'colon-group' and the 'typhoid-Gaertner' groups grow on these media. The presence of colon bacteria in any quantity in water or in an article of food is regarded by most bacteriologists as undoubted evidence of faecal contamination. *Bacillus enteritidis sporogenes* is a virulent bacterium which is regarded by Dr. Klein as the cause of certain outbreaks of epidemic diarrhoea. The presence of these bacilli together is regarded as certain evidence of sewage pollution.

Both of these groups of bacteria were found in nearly all of the mussels examined. Colon bacteria were abundant in all the cultures made. *Bacillus enteritidis sporogenes* was not so abundant, but was present in all the special cultures made to determine its presence, and gave all the characteristic reactions. There is no doubt then, that this mussel bed is grossly contaminated with sewage matter, on which the shell-fish have been feeding, and that, consequently, if used as food these mussels may be, in certain circumstances, the source of grave danger."

Since that date other samples of mussels from various beds have been examined bacteriologically in our laboratory, and in these also extensive pollution by sewage organisms has been demonstrated. I wish to state that in all cases in which sewage contamination has been reported we have not relied upon the bacteriological evidence alone. We have used it as the corroboration, not as the sole proof.

It is evident to anyone who has watched recent investigations that we must have still further knowledge in regard to the distribution of the organisms of the "coli" group in nature, and of the bacteriology of the absolutely clean shellfish before we can place implicit trust in bacteriological results taken alone. Professor C. A. Fuller, of Brown University in the United States, read a paper on "The bacterial flora of the Oyster's intestine," before the Society of American Bacteriologists, in March, 1903, in which he gave the results of extensive studies of the oysters of Narragansett Bay. He found that the "coli" organisms do not occur in oysters from clean sea water far from shore, but are present in those nearer land and in less pure water. He concludes "From the results of these experiments it appears that the colon bacillus is not normally present in the intestine of oysters, and when present always indicates contamination."

Also Miss Chick shows (Thompson-Yates Reports, vol. 3, pt. 1, pp. 1-29, and pt. 2, pp. 117-129) *B. coli* is not found at all in any unpolluted soils and fresh waters. When it was found there was always other evidence of sewage pollution. It was not found in *dry* road dust. It does not withstand even a short exposure to drying. (If it is found in the sea it apparently always comes from polluted land drainage).

On the other hand, in the recently issued Fourth Report of the Royal Commission on Sewage (1904) we

find that Dr. Houston examined for the Commission over one thousand oysters, from "some of the purest waters," and from others "obviously liable to pollution," with the result that nearly all the oysters, "from whatever laying they were taken, contained *Bacillus coli communis*, or other *B. coli* closely allied to it." Dr. Houston found, however, a very much smaller number of these organisms in the oysters stored in pure waters than in those from polluted waters. Consequently the Commissioners state that they should not be justified in recommending that the closing of a bed or laying should depend as a matter of routine on the results of a bacteriological examination—which is very much the conclusion at which we had arrived, and the view that I gave in my evidence.

In taking samples of suspected shell-fish I would attach great importance to personal supervision by a scientific or fisheries expert. The samples should obviously not be taken by the parties interested, and they should not be taken by disinterested, but untrained collectors who may miss seeing some qualifying factor or some important piece of evidence. A knowledge of the local conditions, of the influence of tidal and other currents, and of prevalent winds, may be of great value in judging of the presence and extent of pollution, and of the parts liable to be affected at a particular time of day or month. Consequently a personal examination of the locality by a scientific man is always important. Samples from various parts of the same bed may have to be taken at different states of the tide, and these should be chosen with knowledge and discrimination.

Any additional evidence that can be obtained from an inspection of the physical and biological conditions on the bed is all the more important because of our want of exact knowledge as to the meaning and value of some

bacteriological results. The topographical observations and the laboratory work ought always to be considered together, and must be regarded as parts of the same investigation conducted by the one authority. The bacteriological examination may at once confirm the field work in such a manner as to leave no doubt as to the purity or pollution of the locality, or it may give useful indications which suggest the necessity for further observation of the local conditions. It may also give a measure of the amount of pollution. The question has been raised as to whether it is possible to fix a standard of pollution which should be regarded as dangerous to health. Can we say that all samples yielding say 10, or say 20 *B. coli* per c. c. must be condemned, but that those showing less than say, 5 per c. c. may be tolerated? Before answering such a question we must have further investigations. There are still too many of the points involved which are left in doubt. For example, we cannot be certain that all samples yielding 10 *B. coli* per c. c. are equally dangerous. Even if we assume (as we probably may safely do) that pure oceanic sea-water is free from *B. coli* and allied organisms, and that these are to be taken as an indication of some sewage contamination, we do not know how remote in time the pollution may have been and how comparatively harmless from a pathogenic point of view it may have become. It is possible, or even probable, that *B. coli* may be distributed to considerable distances in the excreta of fish and sea-birds, possibly with some modification. Then again the bacteriology of the shrimp's alimentary canal requires examination, and we may add the fishes that feed upon the shrimp. There are also other sewage feeding invertebrates that may conceivably pass on some organisms and not others, and may favour the distribution of *B. coli* under

circumstances that deprive its presence of any special significance.

I am not arguing against the value of bacteriology, but against a possible abuse of the method, and in favour of a much wider investigation in which the laboratory work will in all cases be supplemented, guided and inspired by the marine biologists' work in the field. The case of each estuary, bed, or laying must be regarded as a separate problem to be solved with a full knowledge of all the local conditions.

## SYLLABUS OF THE LESSONS GIVEN IN THE CLASSES FOR FISHERMEN.

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*Two lessons are given each day, each lesson lasting for about two hours.*

### FIRST DAY.

The work of the first lesson is introductory, and will deal with the use of the microscope and dissecting tools.

Before going on to the study of fishes and other marine animals, it is necessary to learn something about water and air, so as to understand the way in which animals breathe.

**The Atmosphere.**—Air consists chiefly of three gases, *nitrogen, oxygen and carbonic acid gas*. In every 10,000 parts, by bulk, of air there are a little less than 2,100 parts of oxygen, about 7,900 parts of nitrogen, and about four parts of carbonic acid gas.

The oxygen of the air is necessary for the breathing of animals and plants, and for combustion. Without it animals or plants would not live, and fuel would not burn. When an animal breathes, oxygen is taken into the body, and combines with certain substances in the muscles and elsewhere, producing mechanical energy and heat. When a piece of charcoal or coke burns in the air, it combines with oxygen.

Nitrogen is not really necessary for breathing or for combustion. It merely serves to dilute or weaken the oxygen.

Carbonic acid gas in the air comes principally from the breathing of animals and from combustion of fuels.

**Water** is made up of oxygen and another gas called *hydrogen*. But water also contains air dissolved in it. When water is boiled, it loses its air, and until it dissolves more air marine animals, like fishes, can not live in it.

*Experiments will be made in order to prove these statements.*

### **The Breathing of Animals.**

Most large animals that live on the land breathe by means of lungs. Oxygen is taken into the substance of the body, and carbonic acid gas is given out. Plants also breathe in oxygen and give out carbonic acid gas. Marine animals obtain their oxygen from the sea water by means of gills instead of lungs.

## SECOND DAY.

### **The Structure of a Fish.**

The stomach, the liver and the digestion of the food.

The blood, heart and gills; the circulation of the blood through the body.

The red blood of a fish consists of a clear colourless liquid, in which there are a great number of small reddish, oval particles, about  $\frac{1}{20000}$  part of an inch in diameter. These are the corpuscles, and their use is to carry the oxygen to the tissues of the animal.

The heart is a force-pump, which propels the blood all through the body. From the heart the blood goes to the gills, where it takes in oxygen from the water, and gets rid of its carbonic acid gas. It then flows in the blood vessels all through the body.

The brain, the nerves and the senses.

**The Breeding of a Fish.**—Fishes are either male or female, but with the exception of the skates, rays,

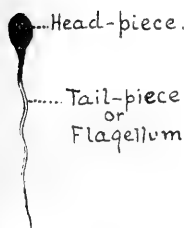


sharks and dogfishes, there is no direct connection between the male and female at the breeding season.

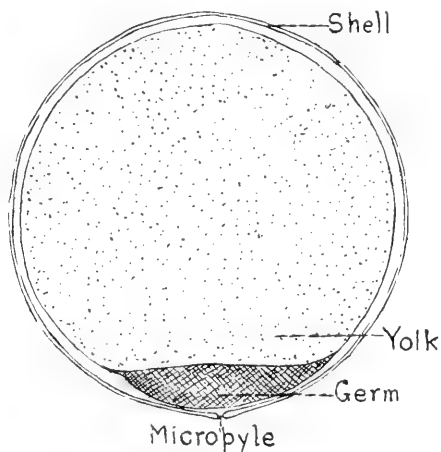
The generative organs are the parts of the body in which the spawn is formed.

THE MALE FISH.—The generative organ is the *testis* or soft roe; the *milt* is formed in this. The milt consists of *sperms*.

THE FEMALE FISH.—The generative organ is the *ovary* or hard roe; the spawn proper is formed in this. The spawn consists of eggs.



The sperm of a fluke magnified 1,500 dia.



The egg of a fluke magnified 30 dia.

Fertilisation.—The way in which the eggs of a fish, such as the flounder or cod, are fertilised will be shewn in the hatchery. If the eggs are simply taken from the female fish, and allowed to remain in clear sea-water, no development takes place. But if the milt from the male is mixed with the water in which the eggs are floating fertilisation takes place. One sperm enters each egg through the micropyle, which then closes. The development of the egg then begins.

At the spawning season the eggs and milt are shed, and drift about in the sea. Fertilisation then takes place as described above.

The eggs of all the flat fishes, and of most round fishes (the cod, haddock, whiting, gurnard, sprat, &c.) float near the surface of the sea. The eggs of the skate, rays and dogfishes, herring, and some other fishes (which are not used as food) sink to the bottom, and pass through their development there.

The average number of eggs spawned by a single female fish in the course of one season are:—

In the Turbot ... ..	8,600,000
„ Cod ... ..	4,500,000
„ Sole ... ..	570,000
„ Haddock ... ..	450,000
„ Plaice ... ..	300,000
„ Whiting ... ..	120,000
„ Herring... ..	31,000
„ Skates, rays and dogfishes, about a dozen or less in the season.	

### THIRD DAY.

#### The Mussel.

The structure of the mussel.—The mouth and stomach; the gills; the mantle; the foot or “tongue”; the byssus or “weed.”

The feeding of the mussel.—The mussel feeds “by suction.” Its food consists of exceedingly small animals and plants that float about in the water. The water containing this food is sucked into the cavity of the shell and the food is then taken into the mouth.

The mussel breathes by means of its gills. When the gill is examined under the microscope it is seen to consist

of very fine threads or *filaments*. These are hollow tubes and blood from the heart flows down inside them. Each filament is covered by an immense number of very small hairs or *cilia*, and as long as the mussel is alive these are waving backwards and forwards and causing a current of water to be sucked into the shell. This current passes over the gills towards the mouth. The food is strained by the gills, caught up by the lips, and taken into the mouth. At the same time the oxygen in the water passes through the filaments of the gills into the blood and the carbonic acid of the blood passes out into the water.

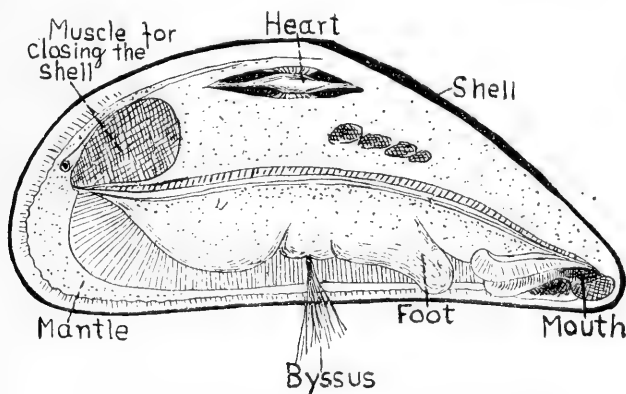
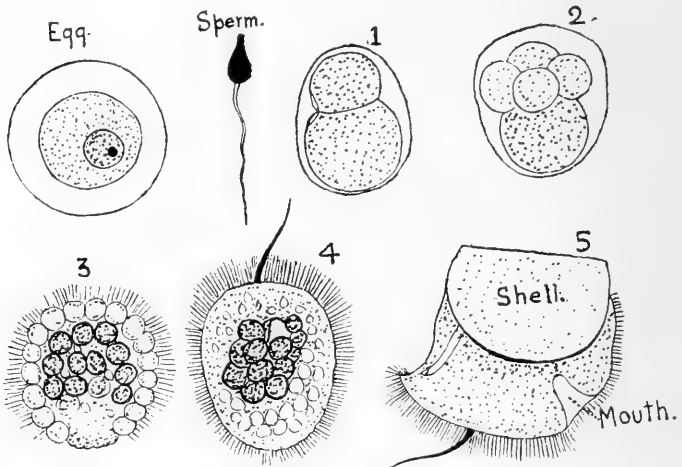


Diagram of the structure of a mussel. Natural size.

**Breeding of the mussel.**—Mussels are male and female. It is impossible to tell the sex except by examining the animal microscopically. The eggs and milt are contained in the mantle. Spawning takes place in the spring, and immediately on spawning the fish become thin and in poor condition because the eggs and milt have been expelled from the body.

The little animal which hatches out from the egg is not at first like a mussel, and it swims about in the water.

After a time it grows a shell and settles down as a fully formed little mussel.



Various stages in the growth of the mussel. (All magnified).

**The Development of the Flounder.**—Eggs of the flounder will be examined with the microscope every afternoon. On the second day of the class a number of eggs will be taken from several female fish, and these will be fertilised by milt from male fish. A sample of this batch of eggs will be examined every afternoon, and the little fishes studied as they form inside the egg-shell. These fish will hatch out in from seven to ten days, according to the temperature of the sea-water.

#### FOURTH DAY.

#### **The Tow-net and Plankton.**

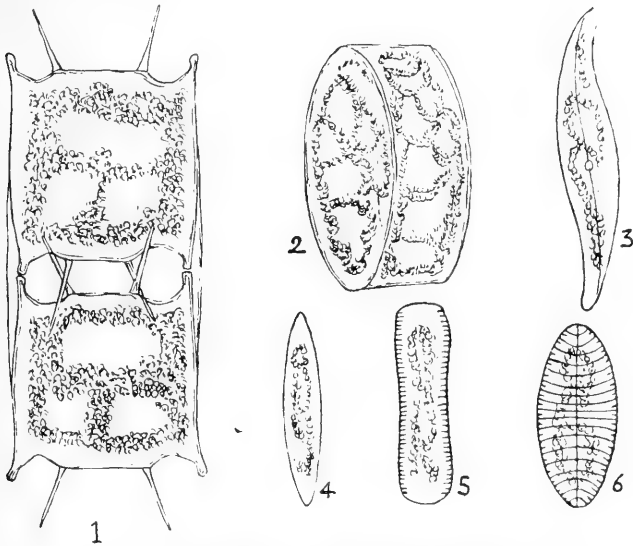
Although the sea-water may look perfectly clear, yet it nearly always contains great quantities of very small animals and plants. These can be caught by means of a tow-net, which is a small net made of fine silk, or muslin,

with 100 or more meshes to the inch. It is towed slowly behind a boat, and when it is fished the catch is shaken out into a bottle of clean water.

Examination of a tow-netting taken in Barrow Channel. (*Such a catch will be examined in the class.*)

The commoner things present in the tow-netting.

**Diatoms.**—These are very small plants, some of which live in the sand and mud at the sea bottom, but most of which float about in the sea. They are yellow-green in colour. Their bodies are enclosed in hard, glass-like, flinty shells.



Diatoms.—1 and 2 are floating diatoms, but 3 to 6 live at the bottom. All highly magnified.

**Diatom Ooze.**—In some parts of the sea far away from land and in very deep water (about 2,000 fathoms) the mud at the bottom is soft and white, and when it is examined with the microscope it is seen to consist of nothing but diatom shells. These little plants

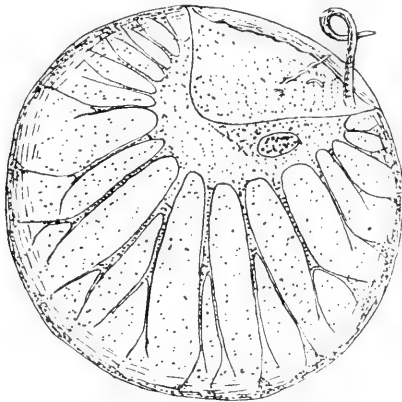
are so extremely abundant in the sea, that when they die their dead shells fall to the bottom, and form a layer of mud or ooze.

The difference between an animal and a plant.

Most animals have a very different appearance from plants, but in the case of diatoms, and other small creatures, it is quite impossible to tell by their appearance whether they are animals or plants. The real difference lies in the methods of feeding.

An animal must live on food which is either alive, or has been alive. So animals feed on the living or dead bodies of other animals or plants.

A plant lives on material which has never been alive—on the carbonic acid and the substances containing nitrogen, which are in the air or are dissolved in the sea-water.



Noctiluca.—Magnified about 70 dia.

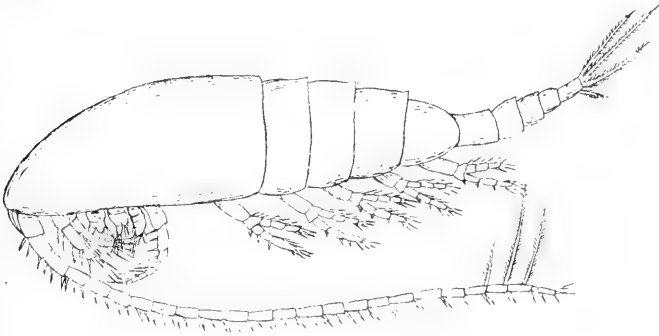
*Noctiluca*.—This is a small animal which is always present in some parts of the Irish Sea during the summer. It is sometimes so abundant as to colour the water brown. It is simply a little round blob of jelly-like material. It feeds on diatoms, and these will usually be

seen inside its body. It is one of the things which cause the phosphorescence, or "fox-fire," of the sea.

*Ceratium* is another small animal which is sometimes extremely abundant in the sea. It forms a large portion of the food of some small fishes.

#### FIFTH DAY.

**Copepods.**—These are the most abundant animals in the sea. They are very small animals belonging to the same class as the shrimp. Many young fishes live on them, and they are the principal food of many full-grown fishes like the herring and mackerel.



A Copepod.—Magnified about 45 dia.

**The arrow worm** is a little worm about an inch long. It is nearly always found in the sea. It forms part of the food of some fishes and it is said to eat fish eggs.

Other common animals found in the tow-nettings.

**Food of marine animals.**—Everything in the sea lives on something smaller than itself. But this cannot go on without coming to a stop, and in the long

run we find that the smallest animals in the sea live on the Diatoms.

Thus—

The **Cod** feeds largely on **Crabs**,  
 The **Crab** „ „ „ **Worms**,  
 The **Worm** „ „ „ **Diatoms**.

The **Plaice** feeds largely on **Shellfish** (mussels and  
 cockles),  
**Shellfish** „ „ „ **Diatoms**.

The **Herring** and **Mackerel** feed largely on **Copepods**,  
**Copepods** feed on **Diatoms**.

So everything in the sea depends for its food on the Diatoms or on other plants living in the sea. The Diatoms and other plants feed on the carbonic acid in the sea water and on substances containing nitrogen which are washed down from the land into the sea:

*A number of pictures will be shown by the lantern on the last afternoon of the first week of the class. These pictures will be explained, and they will illustrate the work of the first week.*

#### SIXTH DAY.

##### **The structure of Fishes.**

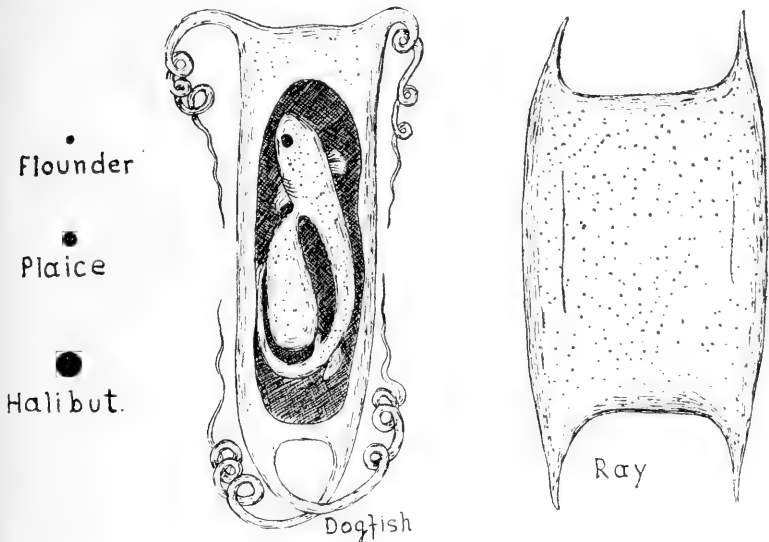
**Skates and Rays and Dogfishes.**—These fishes differ in many ways from the round fishes like the cod and haddock. Their bones are gristly, they breed in a different manner, and their gills are different.

**The structure of a skate.**—Its brain and nerves; the ear; the heart, stomach and intestine; the gills.



The electric apparatus of a skate; other fishes with electric apparatus.

**Breeding.**—Skates, rays, dogfishes and sharks breed quite differently from all other fishes. In the case of most round fishes, flukes and soles, the eggs and milt are shed into the sea and fertilization takes place there so that there is no actual connection between the male and female. But in the case of the skates, rays, dogfishes and sharks the male makes connection with the female at the breeding



Eggs of various Fishes.—All natural size.

season and the milt is passed into the body of the female and fertilizes the eggs before they are shed.

The eggs of these fishes are quite different from those of the other fishes. They are very much larger and they are enclosed in a hard horny case called the purse.

Then most other fishes lay a great number of eggs at one time (30,000 to 9,000,000), while a skate or dogfish

only lays two. But the other fishes only spawn during one or two months during the year, while the skate probably spawns during most of the year.

**The plaice and dab.**—These fishes (and also the other flukes and the sole) are very like the round fishes as far as their structure and method of breeding is concerned. But they are flat instead of round. The coloured side of a plaice is not its back, but is really its right side. Its head has been twisted so that both its eyes appear to be on the right side of the body.

**The sole and solenette.**—The solenette is often mistaken for a young sole. It never grows to more than about five inches in length. It differs in appearance from a young sole in being redder in colour and in having bigger scales. If a solenette of 4-5 inches long is opened about March, spawn will be found in it—either eggs or milt. This shews that it is a full-grown fish. Spawn will never be found in a sole of the same size.

Male and female soles.

**Sizes at which fishes spawn.**—A fish is called *mature* when it spawns for the first time. The sizes at which various fishes come to maturity differ.

The **Plaice** becomes mature at about 15 inches long.

<b>Dab</b>	..	..	..	5	..	..
<b>Flounder</b>	..	..	..	11	..	..
<b>Sole</b>	..	..	..	12	..	..
<b>Cod</b>	..	..	..	30	..	..
<b>Haddock</b>	..	..	..	12	..	..
<b>Whiting</b>	..	..	..	11	..	..

These are the average sizes for the females. The males are usually smaller than the females, and also produce milt for the first time at a smaller size.

**The sprat and herring.**—This is another

case like the solenette and sole. The sprat differs from the herring of its own size in having a row of prickles along the edge of the belly. It is a full-grown fish, and produces ripe spawn when it is about three inches long, whereas the herring does not become mature till it is over eight inches long. The eggs of the herring sink to the bottom when they are spawned. The eggs of the sprat float on the surface.

#### SEVENTH DAY.

##### **The Shrimp and Crab.**

**The Shrimp.**—The stomach, liver, ovaries and testes.

**Breeding of the Shrimp.**—Difference between the male and female shrimp.

All shrimps carrying "berries," or eggs, are females. The males never carry berries. All shrimps without eggs are not males however, for some may be females which have hatched out their eggs. The males are always smaller than the females, and not so numerous; but the only certain way to tell the sex is to examine their legs. The fifth pair of legs, counting from the tail, are different in male and female.

Also, if the shrimp is opened up, and the generative organs examined, it will be seen that the male produces sperms, while the female produces eggs.

**Spawning.**—The shrimp spawns twice in the year—in the late spring and in the autumn. When it spawns the eggs pass out of the ovaries and become fastened on to the legs. The mother then carries them till they hatch out, which takes place some months after spawning. When newly spawned the eggs are white, but the nearer they are to hatching the darker they become.

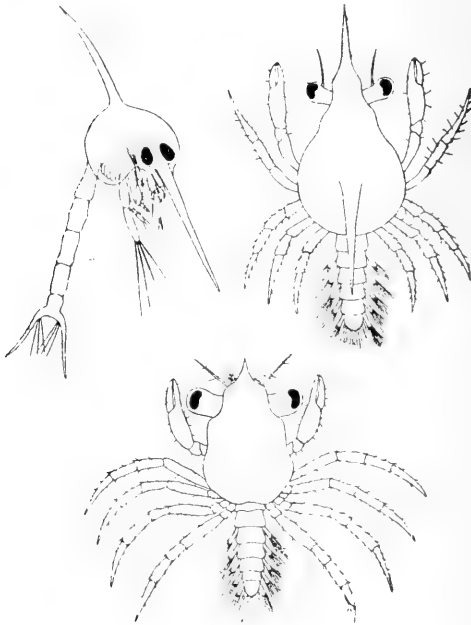
A large berried shrimp carries about 5,000 eggs.

*Examination of the eggs under the microscope. Also the berries will be hatched in the tanks and the newly-hatched little shrimps examined.*

The Lobster breeds in just the same way as the shrimp, except that it spawns only once in the year.

Male and female lobster.

The Crab.—The stomach, liver, ovaries and testes.



Three stages in the growth of the Crab. (All magnified).

The "Casting" of the Crab.—The shell of the crab is "cast," or thrown off, many times during the first year of its life. Afterwards it casts its shell about once in each year. Growth only takes place while the crab is "soft," and before it grows a fresh shell.

A female edible crab becomes mature for the first time, and produces spawn, when it is about five inches broad.

Rate at which the Shrimp Breeds.—The shrimp produces on the average about 5,000 eggs. If there were no destruction in the sea, and all these eggs developed into young shrimps, then starting with a single pair of shrimps—

one male shrimp }  
one female shrimp } 1st generation.

The female spawns 5,000 eggs, which develop into

4000 females }  
and 1000 males } 2nd generation.

At the next spawning each of these 4,000 females will spawn 5,000 eggs, that is  $4000 \times 5000 = 20,000,000$ , and they will develop into

16,000,000 females }  
and 4,000,000 males } 3rd generation.

At the next spawning again each of these 16,000,000 females will spawn 5,000 eggs, that is  $16,000,000 \times 5000 = 80,000,000,000$ , and these will develop into

64,000,000,000 females }  
and 16,000,000,000 males } 4th generation.

Thus if we start with two shrimps, and imagine that they only breed once in their lifetime, and that all the eggs come to maturity, then in the fourth generation we shall have **80,000 millions of shrimps**, and at the fifth generation there would be spawned **320 billions of eggs**.

## EIGHTH DAY.

**The Cockle.**

**Structure.**—Mouth and stomach, foot, siphons, gills, manner of feeding and breathing; male and female cockles.

**Food of cockle.**—The cockle like the mussel and oyster feeds by "suction." It gets its food from the diatoms and other microscopic life which are present in the water sucked into the shell by the action of the gills. These diatoms, &c., are to be seen in the intestine of the cockle, mixed with a great quantity of sand and mud.

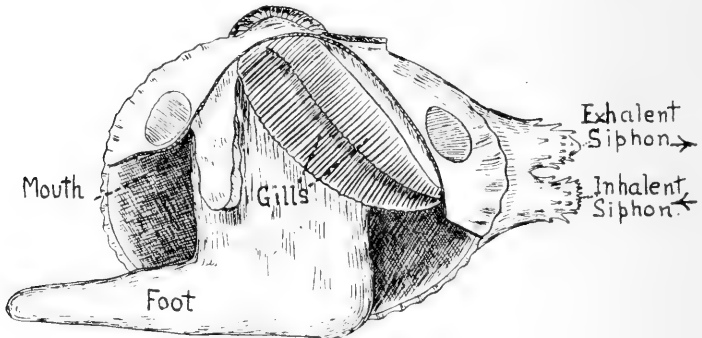


Diagram of the structure of a Cockle.

**The Oyster.**

Structure of oyster—mouth, palps, gills, stomach, heart.

Manner of feeding and breathing.

Male, female and hermaphrodite oysters.

Different kinds of oysters—Deep-sea oysters, American oysters, Portuguese oysters.

Tropical oysters, pearl oysters, pearls and pearl shells.

Pearls in the common mussel. The parasite which is the cause of the pearls.

Artificial cultivation of oysters in France.

## NINTH DAY.

**Fish Parasites.**

Nearly all animals harbour parasites. When one animal lives attached to another, either clinging to the skin, or living inside the body, and feeding on the blood or juices of the animal to which it is attached, it is called a parasite.

Thus the fish-louse found on the skin of the flounder is a parasite of this fish, and the flounder is said to be the *host* of the fish-louse.

But the pea-crab which lives inside the shell of the mussel is not a parasite, for it does not live on the blood or juices of the mussel. It gets its food from the small animals and plants in the water which enters the shell of the mussel.

The little white warts which are occasionally found on the skin of the flounder are also parasites, and not the eggs of the fish as many fishermen imagine.

**Barnacles.**—Three different kinds of barnacles are frequently found by fishermen.

1. The common barnacle, or “scab,” which grows on the bottoms of boats during the spring.

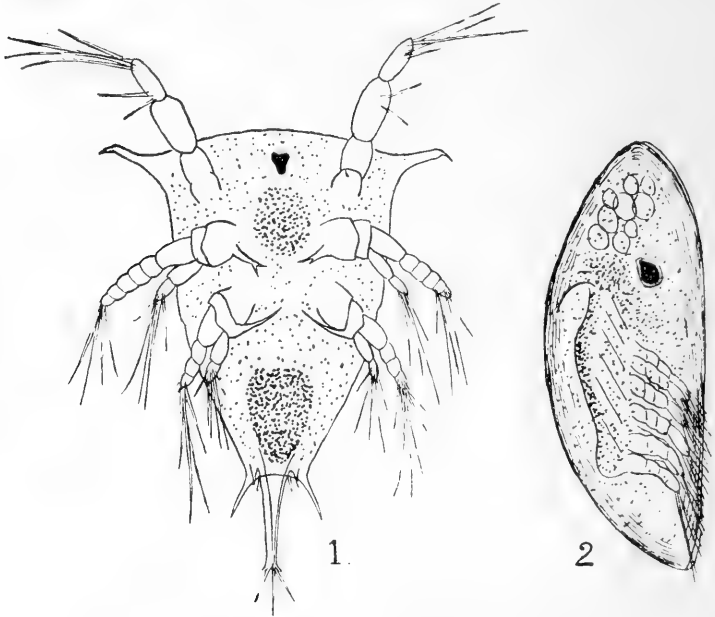
2. The ship barnacle. This does not grow in British Seas, but occasionally comes here attached to floating wreckage.

3. The crab barnacle. This is a round soft animal which is often found attached to the under side of the tail of different kinds of crabs. It is not the spawn of the crab as many fishermen imagine; if it is opened its own spawn will be found inside the body.

The common barnacle and the ship barnacle are not parasites, but the crab barnacle is. It is always a female which is parasitic on the crab, and the male barnacle is parasitic on the female, and lives attached to it.

These three barnacles are very like each other when they are just hatched.

In these two stages the young barnacle drifts about in the sea. After passing through stage 2 they fix themselves—the common barnacle to stones, piles, the bottoms of boats, &c.; the ship barnacle to the bottoms of ships, &c.; and the crab barnacle to various kinds of crabs.



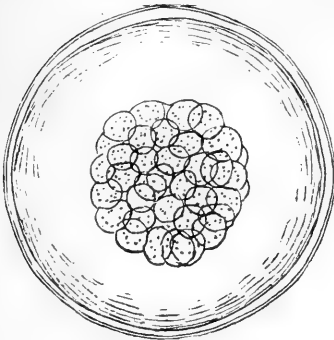
1—Barnacle just hatched. 2—Later stage of the same.  
Magnified about 60 dia.

These young barnacles are always found in the tow-nettings during the early spring, and will be studied.

Development of the Flounder.—The egg of the flounder just before hatching. The newly-hatched fish. When it issues from the egg, the little flounder is a round fish, like the newly-hatched cod or haddock. When it is about six weeks old the body flattens out and the eyes twist round until they both appear to be on the same side of the fish. Up to this time the little fish swims about in the sea, but when it has become a “flatfish” it sinks to the bottom.



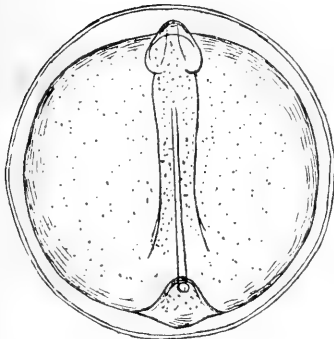
The Eggs of the Plaice and Newly-Hatched Plaice.



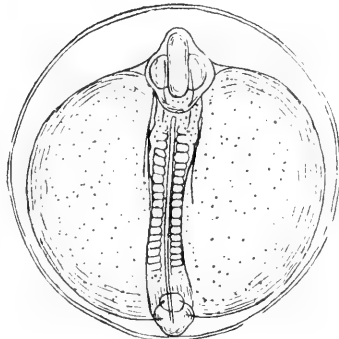
1 Day.



About 3 days.



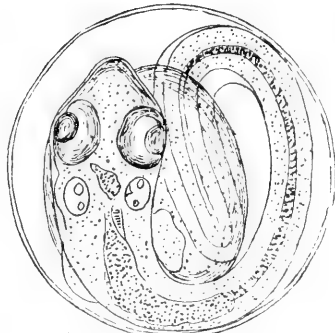
4 days.



5 days.



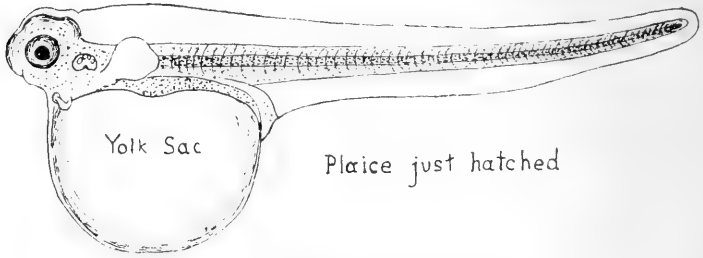
9 days.



17 days: ready to hatch.

Various stages in the development of the Plaice.—Magnified 25 dia.

When the plaice hatches from the egg it is unable to feed, and for about a fortnight it obtains its nutriment from the food-yolk contained in the yolk-sac which is attached to its belly.



Magnified 14 dia.

After about a fortnight the young plaice begins to feed. The yolk has then been used up. The food of the little fish is at first diatoms and then copepods. Afterwards it begins to feed on small shellfish—little cockles, &c., and then as it becomes older it feeds on larger animals such as “hen-pens.”

#### TENTH DAY.

*The work of this day will be a general review of what has been done during the fortnight.*

## APPENDIX.

## MEMOIR ON ARENICOLA.

The Fisherman's Lugworm.

By J. H. ASHWORTH, D.Sc.

*(Lecturer in Invertebrate Zoology in the University of Edinburgh.)*

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## INTRODUCTION.

*Arenicola*\* is a genus of abundant and widely-ranging Polychæt worms, represented in the British fauna by three species,† all of which may be found between tide marks in the neighbourhood of Port Erin (Isle of Man) and Plymouth.

The common lugworm, *Arenicola marina*, forms a convenient type of the genus. It is abundant on most of the sandy shores around the British Isles. Its presence is indicated by the small heaps or coiled castings of sand, which are familiar objects between tide marks. The animal may be found on digging to a depth of one to three feet near such castings, and it is commonly so obtained by fishermen, by whom it is extensively used as bait.‡ Its general appearance may be seen in Fig. 1. It is almost invariably slightly inflated in the anterior region, being widest at a point about one-sixth of its length from the anterior end.

## HABITS, VARIETIES.

There are on the Lancashire coast and in the Firth of Forth, two varieties of *A. marina* which differ in habits and in two structural points.

\* While the following account owes much to the memoir published in 1898 by Dr. F. W. Gamble and myself (Quarterly Journal of Microscopical Science, Vol. 41) it should be said that a large portion of it is new, consisting of observations on the development and post-larval stages and of new points relating to some of the systems of organs. All the statements from the earlier memoir used in the present one have been carefully revised and some of them corrected in the light of further investigations I have made on the genus during the last six years.

† *A. marina*, Linnæus; *A. grubii*, Claparède; *A. ecaudata*, Johnston.

‡ I have added a section (see p. 219) dealing with some points of economic importance, such as its abundance in various sands, its efficacy as compared with that of other bait, etc.

Typical specimens of *A. marina* are found in the littoral zone. Their burrows are usually U-shaped (see below). One end of the burrow is marked by a casting and the other by a funnel-shaped hole through which the head was probably withdrawn, and through which it may be again protruded when the burrow is covered by the rise of the tide. The worm is almost invariably found head downwards in such a position that the tail and posterior part of the body lie in the limb of the burrow connected with the casting, while the head and anterior part of the body lie either in this or in the horizontal limb of the U. The worms average about seven to nine inches in length,\* and their gills, which are best developed in old, deeply-pigmented specimens, are composed of nine to eleven stems each provided with three to five pairs of short lateral branches (fig. 22). These littoral forms breed in the spring, usually from the end of February onwards for about a month but specimens containing ripe ova may be occasionally met with up to the end of April,† and on the Lancashire coast even later. The smallest specimens found in the sand are 17 mm. in length (see below), possibly smaller ones are present but have escaped observation. Young specimens are pinkish in colour, due to the fact that numerous blood-vessels are seen through the translucent body-wall, and their gills are usually bright

\* The largest specimens of the littoral variety I have seen, are several found near Musselburgh, on the Firth of Forth, measuring 340 mm. (about 13½ inches) in length.

† I am inclined to believe that, in some localities at any rate, *Arenicola marina* also breeds about August or September. Large specimens were taken near Musselburgh in July, 1903, which contained great numbers of ova and sperms in an almost ripe condition. In other full-grown specimens from the same area, taken near the end of October, not a single large egg or mass of spermatids was to be found. Among the specimens examined on both occasions were both littoral and Laminarian forms. In others, taken early in January, 1904, genital products are small and scarce in the coelomic fluid. A month later, in males, spermatogonia and spermatids, and in females young ova (.06 mm. in diam.) are present in quantity.

red. Older specimens, especially if taken from sand in which the amount of organic matter is considerable, are nearly black in colour and their gills, which are also pigmented, are better developed than in young specimens.

The second variety of *A. marina* is found in the upper part of the Laminarian zone, and can only be readily obtained at very low tides. It breeds in the spring, from the beginning of March onwards.† When fully grown it is one of the largest Polychaets of our shores, measuring as much as 400 mm. (16 inches) in length and nearly three inches in girth at its widest point. It is dark brown or almost black in colour. Its burrow, in which the worm is found head downwards, appears to be simply vertical and not U-shaped. The most distinctive character of this variety is, however, the gill, which is a highly developed pinnate structure (fig. 19) consisting of about twelve stems united by a connecting membrane at their bases and bearing ten or more branches on each side of the axis. The Laminarian differs from the littoral form also in the sub-division of the interval between the second and third chætigerous annuli; in the former this region is divided into two rings and in the latter into three (see figs. 1 and 2). There do not appear to be any other structural points of difference. The Laminarian variety is abundant on certain parts of the Lancashire coast, *e.g.*, near Blackpool, where specimens are obtainable at low tides on digging to a depth of about three feet in the sand near extreme low-water mark. I have recently found four specimens\* of this variety in a collection of *Arenicola* from the sand between Portobello and Musselburgh on the Firth of Forth. The Laminarian variety has also been found in

† See footnote (†) on p. 131.

\* The gills of two of these were not as obviously pinnate as those of the Laminarian forms of the Lancashire coast.

Jersey and has been figured, probably from the coast of Normandy, by Milne Edwards in the plates of Cuvier's "Règne Animal" (Edition de Disciples, Plate VIII., Fig. 1), where it may be recognised by its characteristic gills. From the records of *A. marina*, made by Scandinavian and other naturalists who have examined specimens from northern seas, it would appear that the littoral variety is the only one with which they are acquainted.

#### DISTRIBUTION.

*Arenicola marina* has been recorded from both sides of the Atlantic, *e.g.*, from the shores of Western Europe, Norway, Spitzbergen, North Siberia, Iceland, Greenland, and along the northern coast of America from the Bay of Fundy to Long Island.\* Latitude 40° N. marks approximately the southern limit of *A. marina* on the Atlantic shores. South of this it is replaced on the American coast by *A. cristata*, and in the Mediterranean by *A. claparedii* and *A. cristata*.

#### EXTERNAL CHARACTERS.

The description of the external characters of *Arenicola* will be better understood after a brief reference to those of some other Polychæt, such as *Nereis*, in which the segmentation is more clearly shown.

At the anterior end of *Nereis* is the distinct "head," at the posterior end the "tail," while the intervening portion is sub-divided by a series of shallow grooves into a number of segments which are practically identical in their external characters except in size. Each segment bears laterally a pair of muscular out-growths provided with bundles of bristles, or setæ, and with certain sensory

\* It is also reputed to occur on the shores of Vancouver Island and in Angra Pequena Bay (West Africa).

organs termed cirri (fig. 9). These out-growths, or parapodia, are locomotor organs, and, by their active movement in the living animal, enable it to creep or swim as occasion demands. Each parapodium consists of a basal piece from which extend outwards two almost equal, usually bilobed, processes, a dorsal one (the notopodium), and a ventral one (the neuropodium). In each of these two rami of the parapodium there is a large setal sac, formed by invagination of the epidermis, the lips of which are well seen in *Nereis*. From the inner end or bottom of this sac the setæ arise, each being formed by products from a single cell. The bundle of setæ contained in each sac may be protruded or retracted or moved in various directions by means of a number of slender muscle bands inserted into the bottom of the sac. In addition to these setæ, which may project a considerable distance beyond the lips of the setal sac, there is in each notopodium and neuropodium a stouter, dark-coloured, needle-shaped aciculum of a similar constitution to the setæ. Its base extends further inwards than the bases of the ordinary setæ, but its pointed tip projects only a short distance beyond the surface of the parapodium. The two acicula serve as an internal skeleton to the parapodium, and to them the muscles which move the bundles of setæ are attached. On the dorsal side of the notopodium, and on the ventral side of the neuropodium, is a finger-shaped, or filamentous sensory process, or cirrus.

The head or prostomium of *Nereis* (figs. 7 and 8) is pre-oral. It is followed by the peristomium on the antero-ventral surface of which the mouth is situated. The prostomium bears on its dorsal surface two pairs of eyes, in front a pair of short sensory tentacles and ventro-laterally a pair of much stouter sensory and muscular palps. The peristomium, which bears some resemblance



in shape to a body segment, though frequently larger and always without parapodia, bears on each side at its anterior edge four long slender filiform sensory cirri.

On the last or anal segment of the body, the "tail," the anus opens posteriorly, and though without parapodia or setæ this segment bears a pair of long anal cirri, which probably correspond to the ventral cirri of the trunk segments.

The prostomium and parapodia of *Arenicola* are reduced compared with those of *Nereis*, and the peristomium and the segmentation of the body are much less distinct in the former than in the latter.

The prostomium of *Arenicola* may be seen on examining the anterior end of the animal in the mid-dorsal line. It is a trilobate structure which, in fully-grown specimens, is about 1 mm. in length by nearly 2 mm. in breadth. It is liable to be retracted within the crescentic nuchal groove which lies immediately behind it, and in that case is difficult to see, but if well-expanded the prostomium assumes the form shown in fig. 5. It does not bear any sensory processes, and eyes, though present, are minute and sub-epidermal, and are not visible in the adult except in sections.

There is good reason for believing that the peristomium is represented by the first two annuli, and that the third and fourth annuli represent a segment which has lost its setæ, but the limits of the peristomium and the composition of the region between the prostomium and the first chætigerous annulus will be discussed below.

#### Segmentation, Annulation, Parapodia.

Leaving out of account the prostomium, the peristomium, and the following achætous segment, *Arenicola* may be, for descriptive purposes, divided into three por-

tions:—(1) an anterior bearing parapodia, but no gills, (2) a middle region bearing both parapodia and gills, and (3) a posterior portion or tail, which has neither setæ nor gills. Each of these parts is composed of a number of segments, but the segmentation is somewhat obscured by the sub-division of each segment into a number (generally five) of rings or annuli.

One of the annuli of each segment is larger than the rest and bears the parapodia. The latter may be best seen in the middle branchiferous segments. They are obviously much reduced compared with those of *Nereis*, and their two rami are different in shape and more dissociated than they are in *Nereis*. The notopodium is a small conical elevation situated dorso-laterally, on the rounded apex of which is the mouth of the setal sac from which the tips of a small pencil of hair-like setæ project to a greater or lesser extent (fig. 19). The neuropodium is a muscular ridge traversed dorso-ventrally by a narrow slit, the mouth of the setal sac, in which are situated in a linear series the numerous chætæ or crotchets. The first few neuropodia are small, but in the posterior part of the branchial region they are well developed and extend ventrally until those of the right and left sides almost meet in the mid-ventral line. Each has the appearance of a pair of closely applied tumid lips between which only the tips of the curved chætæ project. There are no cirri or acicula in the parapodia of *Arenicola*. For a detailed description of the setæ, see below.

There are thirteen branchiferous segments, and in the region in front of the first gill six chætigerous segments may be recognised. The interval between two chætigerous annuli is sub-divided into four rings, except between the first and second chætigerous annuli, where there are only two rings, and between the second and

third, where there are either two rings (as in the Laminarian variety), or three (as in the littoral variety).

The exact limits of the segments are not obvious, but from a consideration of the internal anatomy, it can be shown that the second groove behind each chætigerous annulus marks the posterior limit of that segment. Internal septa, where present, are inserted at this level, not only in *A. marina*, but in all other species examined (see below). Each of the segments posterior to the third consists of the annulus which bears the parapodia, together with the three annuli in front of and the one behind it. The parapodia are therefore situated slightly behind the middle of the segment to which they belong. The third chætigerous segment consists, in the littoral variety, of four annuli, viz., the chætigerous one, two in front of it, and one behind it; while in the Laminarian variety this segment contains only three annuli, the middle one of which bears the setæ. The extent of this third segment is easily determined, as it is delimited both anteriorly and posteriorly by internal septa. The second segment consists of three annuli, the middle one bearing the parapodia. The first segment is composed of two rings, the anterior of which is chætigerous. This segment is bounded in front by the first diaphragm (fig. 2.)

Between the first chætigerous annulus and the prostomium there is a region sub-divided into four rings, each of the first three of which may be again sub-divided into two; but usually there is little difficulty in recognising the four primary annuli (figs. 5, 6). This region is composed of the peristomium and a body segment, the setæ of which are minute, and disappear early. The evidence in support of this statement is derived from (1) an examination of post-larval stages,

(2) a comparison of this region in the adult and in post-larval stages, and (3) a consideration of the position of certain giant nerve cells present in the adult.

In all post-larval stages the region between the first chætigerous segment and the prostomium is clearly divided into two parts by a groove (figs. 57, 58). The posterior of these is a body segment, which in all the seventeen post-larval stages I have examined is achætous, but in which a small seta was observed by Benham. This seta, though it disappears early, indicates that there is a distinct trunk segment in front of the first adult chætigerous segment. The anterior part of the region under discussion is undoubtedly the peristomium, and never bears setæ. The otocysts, which in most other Polychæta in which they are present are peristomial, may be seen near its anterior margin (figs. 56, 57).

In post-larval stages in which the annulation is making its appearance, the peristomium and the achætous body segment are each divided by a shallow groove into two annuli, together therefore consisting of four annuli. In two or three of these specimens the metastomial grooves, which mark the track of the œsophageal connectives, may be clearly seen uniting ventrally near the middle of the third annulus (fig. 58). The annulation of these post-larvæ corresponds, therefore, to that of the adult (cf. figs. 6, 58). It is evident from the above facts that of the four annuli in the adult between the prostomium and the first chætigerous annulus, the first two belong to the peristomium and the third and fourth to a segment from which the setæ have disappeared in early life. Confirmatory evidence is afforded by the giant nerve cells present in the nerve cord. These cells occur singly or in couples near the hinder limit of each segment, *i.e.*, just behind the level

of the parapodia (see below). Just behind the meeting point of the œsophageal connectives there is a single giant cell, which is therefore situated either in the posterior part of the third or, more usually, in the fourth annulus (fig. 52). This cell, whose position near the hinder border of the somite corresponds with that of the giant cells of other somites, must therefore belong to the segment from which the setæ have disappeared. The first chætigerous segment of an adult *Arenicola* is, therefore, really the third segment, as it is preceded by a segment bearing a giant cell and vestigial seta, and this again by the peristomium.

The tail, which is without parapodia and gills, varies considerably in different specimens. It is usually marked by a number of slight constrictions, best seen when the tail is stretched, which indicate the boundaries of the somites and correspond in position to the internal septa (fig. 1). There may be as many as seventy segments, but in most specimens there are fewer as the worm has a tendency to throw off the last few segments when irritated. New segments are apparently formed at the anterior end of the tail region. Here the segments are short from before backwards, but further back they are longer and sub-divided into a number of annuli, as many as ten in some of the last segments. Near the posterior limit of each segment in the mid-tail region there is an annulus slightly larger and often more deeply pigmented than the rest, and upon which the epidermal papillæ are distinctly larger than on the other annuli. Each of these larger annuli occupies a position in the segment roughly corresponding to that of the chætigerous annulus in the pre-caudal segments of the worm. These larger annuli and their papillæ are best seen near the middle of the tail, behind that point the distinguishing characters above

described become gradually less obvious, and near the anus practically disappear.

Traversing the whole length of the mid-ventral line of the animal is a shallow groove which marks the position of the ventral nerve cord (fig. 6). Just behind the middle of the third annulus (*i.e.*, a short distance in front of the first chætigerous annulus) this groove unites with two others which pass round the sides of the peristomium in an antero-dorsal direction to the nuchal organ. These are termed the metastomial grooves; they indicate the course of the œsophageal connectives.

#### External Apertures.

The mouth, when the proboscis is withdrawn, is a crescentic transverse slit on the antero-ventral aspect of the peristomium. It is overhung by a small upper lip, and bordered all round by a series of papillæ (fig. 2).

The anus is terminal, opening on the end of the last tail segment. Through it the terminal part of the intestine occasionally protrudes for a short distance.

The nuchal organ is a transverse crescentic, or V-shaped, ciliated sensory groove formed by invagination of the epidermis of the sides and hinder end of the prostomium (fig. 5). The prostomium may be withdrawn into this groove so as to be almost hidden from view.

The openings of the otocysts are minute and difficult to see. They may, however, be found in large specimens. They are situated dorso-laterally in the peristomium close to the point where the metastomial groove crosses the first inter-annular groove (fig. 5).

The openings of the six pairs of nephridia are small oval slits situated immediately dorsal and slightly posterior to the upper ends of the fourth to the ninth neuropodia (figs. 1 and 19).

## Gills.

There are thirteen pairs of gills borne on the seventh to the nineteenth chætigerous segments inclusive (fig. 1). They are well supplied with blood, and, therefore, generally red in colour, but in old specimens they usually become pigmented, and then have a dark brown colour. The first pair is nearly always considerably smaller than any of the others, and may be much reduced or even suppressed (fig. 10). The largest gills are usually found about the middle of the branchial region.

The form of the gills varies considerably in the two varieties. In the Laminarian variety the gill consists of about eleven to fourteen main stems, 5 to 6 mm. long in full-grown worms, which radiate from a point situated slightly dorsal and posterior to the notopodium (fig. 19). These stems are connected at their bases by a web-like membrane. The ventral stems are the smallest and apparently the last formed. Each of the main stems bears from ten to twelve pairs of branches, which are, however, not strictly opposite, but in some cases almost alternating. Each branch divides dichotomously a number of times, and may give rise to as many as twenty or twenty-five gill filaments (fig. 21). This type of gill, with its well-developed and numerous lateral branches, is known as the pinnate type. In some specimens the gills have lost some of their branches, either owing to friction against the sand, or to the attacks of enemies.\* The gill of the littoral variety is not so well developed (fig. 22). It forms a more bushy structure, consisting of eight to twelve stems, about two to three

\* See, for example, the description of the attacks of the Amphipod *Corophium longicorne* upon *Arenicola*. M. C. d'Orbigny. *Journal de Physique*, tome 93, p. 198, 1821.

millimetres long, each of which bears only about three to six pairs of lateral twigs, which are not so richly branched as in the pinnate gill described above. This may be called the dendritic type.

Each gill is a hollow out-growth of the body wall enclosing an extension of the cœlom (fig. 36). The wall of the gill is thin; it is formed of an external cubical or flattened epithelium, below which is a thin layer of muscle fibres. The cavity of the gill is lined by the cœlomic epithelium, between which and the muscle layer the branchial vessels are situated. The cœlomic cavity of the gill is crossed at intervals by muscle strands. The gill is a contractile structure, and, as Milne Edwards pointed out, the successive contraction of the branchiæ, which often proceeds regularly from behind forwards, must exert a considerable influence in forcing the blood into the efferent branchial vessels and into the vessels of the body wall.

The gills arise in the post-larval stage. One specimen, 3.9 mm. long, has already the full complement of gills, but in other specimens examined the gills are not formed until the animal is considerably longer. In the fifteenth to the eighteenth chaetigerous segments of a post-larval specimen, 4.6 mm. long, the blood-vessels immediately behind the notopodium form a loop preparatory to the production of a gill, and there is a very slight elevation of the body wall in this region. These elevations soon become conical outgrowths, then digitiform and commence to branch. As Benham has pointed out, the gills of *Arenicola* are from the first special respiratory structures, and not as in *Eunice*, and some other Polychæta, modifications of dorsal cirri. There is no trace of sense-hairs upon the epidermis of the developing gill, while such hairs are present in cirri.



The afferent branchial vessels arise from the ventral vessel. They enter the gills, and at once divide to supply a trunk to each of the main stems. These trunks in turn give off branches into each pinna, and these again to each gill filament. There is a corresponding series of efferent vessels. The afferent and efferent vessels merge into one another in each gill filament, where one limb of the vascular loop is derived from the afferent, while the other is in connection with the efferent vessel. The first six gills return blood to the sub-intestinal vessels, while the efferent vessels of the remaining gills open into the dorsal vessel (fig. 23).

#### S e t æ .

Each notopodial seta is a slender capillary chitinous structure inserted at its proximal end, along with many other similar setæ, in a setal sac, which is moved by special retractor and protractor muscles (figs. 24 and 36). The seta has an almost uniform diameter for a considerable portion of its length, but it tapers at its distal end to a fine point (fig. 12). On the distal fourth or fifth of its length the seta bears numerous minute, regularly arranged, pointed processes, which are usually present on both sides of the seta. They are moderately obvious on one side, but on the other they are very minute and borne on the edge of a thin border or lamina (fig. 14). This is well marked in the large setæ of the Laminarian variety, in which the lamina may be traced for about a millimetre along the seta, and attains a width of  $20\mu$  (fig. 13). In some of the setæ the lamina is not denticulate at its margin, and in others is only very faintly so; but it is crossed by fine oblique lines the intervals between which correspond roughly to the size of the teeth on dentigerous laminae. It seems probable that the lamina at first

possesses an entire margin, but later this tends to break up from the edge inwards, thus giving rise to the minute teeth or processes which are usually seen on full-grown setæ. The notopodial setæ may attain a length of 7.5 mm. (in a specimen 250 mm. long).

Each neuropodial chæta or crotchet consists of a shaft, generally somewhat curved, bearing at its distal end a beak-like rostrum placed at an angle to the shaft varying from about  $90^{\circ}$  to about  $130^{\circ}$  (figs. 15-18). There is generally a slight dilatation of the shaft near the middle of its length. Near the end of most chætæ from young specimens, immediately behind the rostrum there are visible two or more minute pointed teeth, the tips of which are directed towards the tip of the rostrum, while below the rostrum at its junction with the shaft there is often a minute process—the subrostral process. On careful focussing slightly above the level of the teeth and subrostral process there comes into view a number of other fine teeth situated on the sides of the rostrum, so that the latter projects from the centre of a series of teeth arranged round its base. The small subrostral process marks the position of the base of the lowest (and often the smallest) tooth of the series. Only unworn chætæ show these lateral teeth. By isolating the entire band of neuropodial chætæ the number in each neuropodium and the different stages in their growth may be seen. New chætæ are formed at the ventral end of the series, the rostrum being first formed, then the teeth, and finally the shaft which is at first comparatively short (fig. 18).

The neuropodia of the anterior segments are short and contain few setæ, but those further back extend by the addition of crotchets ventrally so as to almost reach the mid-ventral line. The neuropodia of young specimens contain very few crotchets, but in old specimens there is

a large number in the neuropodia of the branchial region. One of the neuropodia of a specimen only about six inches long contained 110 fully formed chætæ and eight or nine others in course of formation at the ventral end of the series.

The crotchets show considerable differences according to the age of the specimen from which they have been taken. Those of post-larval specimens (5·1 mm. long) are ·03 to ·04 mm. long, and bear two, or occasionally three, sharply marked teeth behind the rostrum and also a well-marked process, ending in a fine point, under the rostrum (fig. 15). In older specimens 17 mm. long, which have assumed the adult characters and mode of life the crotchets are ·1 to ·12 mm. long, and bear two well-marked teeth. Specimens about 100 mm. long have chætæ which are ·4 to ·5 mm. long, each of which bears two or three very small teeth and also a very small subrostral process. The crotchets of large specimens, especially those of the Laminarian variety, may attain a length of ·85 mm. From the time of their formation they are devoid of teeth, but bear a rather blunt subrostral process. It is interesting to note that as the animal grows in size the rostrum of the crotchet, which in the post-larval stages is at right-angles to the shaft, in later formed chætæ makes a considerably greater angle with the shaft, the angle increasing with the age of the worm from which the chætæ are obtained, so that in a chæta from a large worm, *e.g.*, a Laminarian specimen about 250 mm. long, the angle between the rostrum and shaft is almost  $130^{\circ}$ . Concurrently there is a reduction in the size of the teeth of successive generations of chætæ, so that although in post-larval stages the teeth are large and comparable to the rostrum in size, they are entirely absent from the chætæ of very old specimens. (For method of isolation and preparation of setæ, see under practical work).

## E p i d e r m i s : P i g m e n t .

There is a well-developed cuticle formed by the mucus cells of the epidermis.

In adult specimens the skin of the anterior and middle portions of the animal is sub-divided by grooves into a series of squarish, oval or polygonal areas (fig. 5). These are composed chiefly of large club-shaped mucus cells and of elongate columnar cells, many of which are almost filled with pigment granules, especially in old specimens. These granules, which are dark yellow or brown when viewed singly, but nearly black in the aggregate, are more abundant in the distal part of the cells, and give to many of them a clavate appearance. Pigment cells are present all over the epidermis, extending on to the prostomium, but are less numerous in the nuchal organ than elsewhere. The epidermis in the grooves between the raised areas is composed of shorter cells, in which the pigment is present in less quantity and mucus cells are wanting.

In the tail the epidermis is raised into rounded papillæ (see above) the structure of which corresponds to that of the raised areas described above.

Sensory cells are also present in the epidermis, being specially abundant in many of the grooves.

When specimens of *Arenicola* are handled a yellowish or greenish pigment exudes from the skin and stains the hands. This, which is readily soluble in alcohol, and to some extent in sea-water, is probably a lipochrome. There is also a brown or black pigment, the fine granules of which are insoluble in alcohol, and are readily seen in sections, especially of old specimens. According to McMunn (Q.J.M.S., Vol. 30, p. 74), this pigment resembles melanin in its resistance to solution, and he

suggests that it is possibly derived from the yellow lipochrome. Fauvel (C.R. Acad. Sciences, Paris, Tome 129, p. 1273) has recently adduced evidence in support of this suggestion, showing that the formation of melanin granules may be due to a chemical modification of the lipochrome taking place in the interior of the cells under the influence of some acid.

#### GENERAL ANATOMY OF THE INTERNAL ORGANS.

(See Plate III.)

The body cavity is best opened by an incision through the body wall along the mid-dorsal line. It often happens that in freshly killed specimens the middle part of the alimentary canal is forced out through the incision owing to the fact that the animal has died somewhat contracted, and the contents of the cœlom are under considerable pressure. At the same time a quantity of the cœlomic fluid will escape through the aperture.

The cœlom is spacious and continuous from one end of the animal to the other. In front it is sub-divided transversely by three fenestrated septa, or diaphragms. The first of these is placed at the anterior boundary of the first chætigerous segment, and is inserted into the body wall at the level of the anterior edge of the first chætigerous annulus. It is perforated by some of the retractor muscles of the pharynx by which, in some specimens, the septum is pulled back ventrally, so that at first sight its ventral edge appears to be inserted near the posterior margin of the first chætigerous annulus. Further examination shows that the diaphragm is *not* obliquely placed, but is situated dorso-ventrally at the anterior edge of the annulus. The second and third diaphragms mark the posterior limit of the second and third chætigerous

segments. They are placed at the posterior margin of the ring which succeeds the second and third chætigerous annuli. Between the first and second diaphragms there are dorsal and ventral mesenteries\* supporting the corresponding blood-vessels; the dorsal mesentery terminates in front about the level of the posterior margin of the first chætigerous annulus. The funnels of the first pair of nephridia perforate the third diaphragm, and lie on its anterior face. Behind this diaphragm the body is uninterrupted by septa almost to the base of the tail, but rudimentary septa may be recognised as strands of connective tissue accompanying some of the afferent and efferent vessels connected with the nephridia and gills. Towards the posterior end of the gill region this tissue increases in amount so as to form in the last, or last two, gill segments an almost complete septum.

It will be seen that all vessels connected with the stomach are attached to its ventral face. This arrangement, together with the absence of mesenteries from this region, allows considerable freedom of motion to this part of the gut without endangering the blood-vessels which, by their length and flexibility, appear to readily permit this motion. During digestion the stomach is swung backwards and forwards by movements of the body, and the arrangement of the blood-vessels indicates a considerable amplitude of swing. There is, however, a structure which probably acts as a sort of safety cord to prevent the backward motion becoming so great as to rupture the vessels. This is a solid pinkish cord of connective tissue, which may easily be mistaken for a vessel; it is attached ventrally to the anterior wall of the stomach, lies alongside the afferent vessel of the fourth nephridium,

\* The ventral mesentery also runs backwards through a considerable part of the third chætigerous segment.

and is inserted into the body wall just behind the level of the sixth chætigerous annulus. This cord and the other rudimentary septa are all placed at the boundary of the segment to which they belong.

Between the intestine and the body wall in the tail there is a comparatively small cœlomic cavity crossed by septa, which are much closer in the anterior than in the posterior caudal region, especially in young specimens, in which the anterior tail segments are very short.

(For a description of the alimentary canal, the vascular system and the nephridia, see below.)

#### MUSCULATURE.

There is a small amount of connective tissue between the epidermis and the well-developed underlying musculature of the body wall, which consists of a layer of circular muscles below which are seen the bands of longitudinal muscle fibres. The latter, which are covered by the thin cœlomic epithelium, abut upon the cœlomic cavity (see figs. 24, 35, 36 and 54). In the anterior region of the body there are usually a few circular muscle bands which are stronger and more obvious than the rest (fig. 23).

The thin bands of muscle seen in dissections arising at the sides of the nerve cord and inserted right and left into the body wall at the level of the notopodial sacs are the oblique muscles which are very characteristic of Polychæta. They commence behind the third diaphragm and extend to the base of the tail. They divide this region of the cœlom into three longitudinal compartments, a dorsal median portion containing the alimentary canal, and two ventro-lateral portions containing the nephridia (fig. 36). The oblique muscles partially cover the nephridia and one of the bands is usually attached to the

nephrostome and binds the latter to the body wall (figs. 23, 24 and 25).

Each bundle of notopodial setæ is enclosed in a sac to the inner end of which are attached (1) a single retractor muscle strand, which is inserted into the body wall at the side of the nerve cord, and (2) six to ten protractor muscles, which are inserted into the body wall at the level of the setal sac. Contraction of the latter causes protrusion of the setæ beyond the lips of the setal sac, while by shortening of the retractor muscle they may be almost entirely withdrawn into the setal sac (fig. 24).

There is a strong sheath of muscle fibres attached to the pharynx and to the neighbouring body wall (fig. 23). Contraction of these muscles produces withdrawal of the pharynx. It is probably by the help of the cœlomic fluid, which can be collected here and subjected to pressure, that the pharynx is protruded.

The prostomium is provided with a small sheet of retractor muscle arising from the musculature around the œsophageal connectives and inserted into the ventral surface of the brain and hinder edge of the nuchal organ (fig. 34).

The position of the three anterior diaphragms and the dorsal and ventral mesenteries in the second segment has already been noticed (see above). Each of these diaphragms is perforated by numerous rounded, usually oval, apertures which are best seen in the third diaphragm where they are moderately close together and about  $\cdot 02$  to  $\cdot 03$  mm. in diameter (fig. 45). These openings permit the passage of the cœlomic fluid and its cells but prevent all but the smallest ova and spermatogonia from passing into the anterior segments. Each diaphragm is covered on both its faces by an endothelium composed of flattened cells between which is a thin layer of connective tissue



and intercrossing muscle fibres. The first diaphragm is perforated by some of the retractors of the proboscis, and it bears two backwardly projecting pear-shaped or finger-shaped out-growths which lie to the sides of, and ventral to, the œsophagus (fig. 23). These are *not* œsophageal glands. They open anteriorly into the cœlomic space in front of the first septum. Their walls are muscular and very vascular and they contract at frequent intervals during life. Their function is unknown, they may possibly be of use in aiding the eversion of the proboscis.

The occurrence of rudimentary septa accompanying some of the afferent and efferent vessels of the nephridia and gills, and the probable function of the two strands (septa) attached to the efferent vessel of the fourth nephridium have been described above.

The caudal septa are incomplete ventrally and ventrolaterally, *i.e.*, above and at the sides of the nerve cord. Each is composed of two thin layers of cœlomic epithelium, forming the anterior and posterior faces between which is a small quantity of connective tissue and muscle fibres. These septa are not fenestrated like the three anterior diaphragms. There are in the tail, in addition to these septa, dorsal and ventral mesenteries, by means of which the intestine is attached above and below to the body wall and in which lie the dorsal and ventral blood-vessels. Owing to their small size these mesenteries are with difficulty seen in dissections, but they are readily distinguished in transverse sections of the tail.

#### CÆLOM AND CÆLOMIC FLUID.

The cœlom, which is lined by a layer of flattened cells, is spacious and continuous from end to end of the animal. It is partly sub-divided by septa in the anterior

region and in the tail, but in the middle portion of the animal it is uninterrupted by septa, although, as mentioned above, these are represented by certain thin bands which accompany the segmental blood-vessels across the cœlom (fig. 23).

There are numerous canaliform prolongations of the cœlom, best seen in sections of the anterior end of the animal, which penetrate into the musculature, or insinuate themselves between the brain and the prostomial epithelium, and often accompany the blood-vessels which supply the body wall. In most of the canals the thin lining of cœlomic epithelium may be recognised, and cœlomic corpuscles may be found in many of them. They probably act as nutritive, and possibly also as excretory and respiratory, channels.

The cœlomic fluid is a mixture of sea-water and globulins (Krukenberg). Its specific gravity is on the average 1·0288, but it varies somewhat according to circumstances. It was found to be greater (1·0311) in worms which had been kept for thirty-six hours in moist sand and seaweed than in others which had been transferred from the moist sand and seaweed to sea-water for three hours (1·0285), or over-night (1·0270). The specific gravity of the sea-water used was 1·0264.

The fluid contains cœlomic cells and, during a considerable portion of the year, reproductive products. The cœlomic corpuscles (fig. 44) are abundant, and of two chief types—(1) fusiform cells about  $\cdot 04$ — $\cdot 05$  mm. long, which are very numerous, and (2) smaller amœboid, or subspherical cells, many of which contain yellow or brown refringent granules. On exposure to the air a delicate fibrous network is formed, with which the fusiform cells (and to a less extent the amœboid cells) become united to form a clot.

The cells of the cœlomic epithelium and the cœlomic corpuscles perform excretory functions; carmine granules injected into the cœlom are taken up by these cells and by the nephridia (see G. Schneider, *Zeitschrift für Wiss. Zoologie*, Band 66, pp. 505-507, 1899). Chlorogogen granules are present in many of the cells of the cœlomic epithelium.

A great proportion of the reproductive cells present in the cœlomic fluid, instead of floating freely, accumulates in the space between the oblique muscles and the ventral body wall. In females oocytes in various stages of growth and varying in diameter from  $\cdot 02$ — $\cdot 16$  mm. may be seen. In males various stages in the development of spermatozoa from groups of young spermatogonia, consisting of about eight cells to the masses of spermatids seen in Fig. 65, may be found.

The cœlomic fluid is kept in motion by contractions of the body wall. In a specimen freshly taken from the sand one usually observes a series of peristaltic waves arising in the posterior part of the gill region, and running forwards to the anterior end, producing a progressive swelling of the body, due to the carrying forward of the cœlomic fluid, which, as soon as the wave is past, flows backwards again. This motion is of considerable importance in promoting the efficient circulation of the cœlomic fluid, in inflating the anterior portion of the animal, thus aiding in burrowing, in assisting the comparatively weak gut muscles to cause the backward motion of the sand in the alimentary canal, and, when the animal is in its burrow, in providing frequent changes, practically a current, of sea-water to bathe the external surface and the gills.

## ALIMENTARY CANAL: BURROWING.

The alimentary canal consists of (1) an eversible buccal mass and pharynx or "proboscis" generally of a pinkish colour in young or middle aged specimens, due to the contained blood-vessels, but in old specimens liable to become darkly pigmented; (2) a cylindrical, pinkish or greenish brown œsophagus, often transversely wrinkled, which pierces the three diaphragms, and, just behind the level of the last of these, bears a pair of glands; (3) the stomach, which has yellow walls on which are numerous blood streams, extends from the level of the heart to that of the eleventh or twelfth setæ, and gradually merges into (4) the intestine, which is yellowish brown or dark olive green in colour, and extends to the posterior end opening at the anus (fig. 23).

During life the "proboscis" is being constantly everted and withdrawn, carrying sand into the œsophagus. During eversion the buccal mass is first extruded, this is armed with several rows of curved, bluntly pointed, vascular papillæ, which in old specimens are capped with chitin (figs. 1, 3 and 4). Then the more globular pharynx covered with minute rounded processes is protruded. These papillæ, which are covered by a thin cuticle, have an axis containing muscle fibres and connective and nervous tissue covered by a columnar epithelium, in which numerous mucus-forming cells are present. Among the columnar cells there are here and there fine fusiform sense-cells, the drawn-out tips of which project into or through the cuticle covering the papillæ.

The œsophagus is a thin-walled, distensible tube, which is lined by elongate, columnar cells, among which are swollen mucus-forming cells and occasional gland cells of other kinds, the latter being more numerous in the

posterior part of the tube. The anterior part of the œsophagus is non-ciliated, but the portion extending from about the level of the third diaphragm to the stomach is lined by cells which bear short cilia.

The œsophageal glands or pouches are somewhat flask-shaped and open into the posterior part of the œsophagus by a hollow stalk (fig. 23 and 35). They are usually greenish in colour, due to the contained secretion, but they may also have a pinkish tinge owing to the large amount of blood present in the sinuses in their walls. These blood sinuses are connected with the lateral œsophageal and dorsal vessels. The cavity of each gland is sub-divided by twenty to thirty incomplete partitions, which, in young specimens or in strongly dilated glands, are mere ridges upon the inner wall of the gland, but in old ones they are, as a rule, lamellæ projecting well towards the centre of the pouch (fig. 35). Each partition is produced by infolding of the wall, and is, therefore, covered on each face by the epithelial lining of the gland. These infoldings of the wall enormously increase the secreting surface of the organ. Between these two epithelial layers is a blood sinus which is slightly enlarged near the free edge of each partition. The pouch is lined by cubical cells among which are numerous gland cells. The secretion of these glands forms a mucous fluid with a neutral re-action.

The stomach is covered with patches of yellow cells—the chlorogenous tissue—which in front are arranged in symmetrical oval areas right and left of the dorsal blood-vessel, while more ventrally they are placed in two or three less regular series (fig. 23). They are separated from one another by blood sinuses. About the level of the tenth setæ these areas become sub-equal and are arranged in a spiral manner, and behind the level of the

fourteenth setæ they are either indistinct or absent. The epithelial lining of the stomach is strongly folded. It consists of columnar cells among which are numerous goblet-like cells, some of which produce a secretion which is probably digestive, while others form mucus (fig. 51). Commencing near the middle of the stomach, *i.e.*, about the level of the ninth setæ, there is a well-marked ventral groove, the cells lining which are provided with long cilia which produce a current from before backwards (fig. 36). There are numerous smaller ciliated grooves on the lateral walls of the stomach and intestine in which the flow is downwards and backwards into the ventral groove. The ventral groove extends to the anus. The intestine is lined by columnar cells, ciliated in the above mentioned grooves, among which only a few gland cells are present.

Circular and longitudinal muscle fibres are present in the walls of the alimentary canal, but they are well marked only in the œsophagus; in the stomach and intestine they are so feebly developed that these parts of the alimentary canal can have only slight powers of peristalsis.

The process of digestion has not been fully investigated, but the series of events appears to be somewhat as follows:—During life the buccal mass and pharynx are constantly being everted and withdrawn, carrying sand into the œsophagus. As the sand passes along the œsophagus it is mixed with the mucus from the cells lining this part of the gut, and further back the secretion of the œsophageal glands is poured upon it. The mixture then passes into the stomach, where the secretion from the mucus-forming and digestive cells is added to the mass. The swinging backwards and forwards of this part of the alimentary canal, brought about by the muscles of the body wall and by the protrusion and retraction of the

pharynx, tends to produce a thorough mixing of the sand and the digestive substances, and in this way the food, which consists of organic substances of various kinds in the sand, is brought into contact with the digestive secretions. The ciliary action of the lateral and ventral grooves probably separates the digested substances from the sand, and carries them slowly backwards. The ciliated grooves are in close association with the blood sinuses in which the flow of blood is probably slowly forwards. The ventral groove is in especially close relation to the sub-intestinal vessels (fig. 36). It seems probable, therefore, that the blood in the gastric plexus absorbs the nutrient materials, conveys them to the hearts which pump the blood along the ventral vessel to various parts of the body.

A thin cord of mucus from the ventral groove may often be seen in freshly-formed castings.

The great abundance of *Arenicola* wherever the sand or mud contains a large proportion of decomposing matter or sewage, and its absence from or scarcity in long stretches of coast where the beach is formed of clean sand, indicate that these animals feed on decaying animal or vegetable matter. They remove some of the decomposing organic matter in the sand, and as they burrow to a depth of two feet or more, they cleanse the sand to about this depth, and discharge it on the surface in a purer condition. (See also under Economic section.)

#### BURROWING.

The burrowing of *Arenicola* is performed by the combined action of the proboscis, the swollen anterior region of the body, and the waves of muscular contraction which pass along the body from behind forwards. The

proboscis is protruded, pressed into the sand and withdrawn full of sand and again everted. The body is thrust forward partly by the action of the longitudinal muscles of the body wall, and partly by the peristaltic waves produced by the circular muscles, by means of which the anterior end is also rendered swollen and tense, and is thus enabled to enlarge the burrow. By these means a passage is eaten and forced through the sand, smoothed by contact with the skin, and may be lined with mucus secreted by the epidermis. The gill region, being narrower than that which precedes it, is to some extent protected from friction, and the notopodial setæ are also directed so as to protect the gills. After burrowing vertically downwards to a depth of from one to two feet, the littoral forms may make a horizontal or oblique gallery, and then a second vertical one, which opens on the surface of the sand in a small funnel-shaped aperture.\* The burrows of Laminarian worms, and those of some littoral specimens, are simple vertical excavations (not U-shaped), in which the animal is almost invariably found head downwards.†

The amount of work done by *Arenicola* has been estimated by Davison‡ on the Holy Island Sands. As the result of observations upon the number and weight of

\* G. Bohn ("Observations Biologiques sur les Arénicoles," Bulletin du Muséum d'histoire naturelle, 1903, No. 2, p. 62) states that the burrow of the littoral form is not U-shaped, and that it does not open at the funnel-shaped aperture. He believes that the latter is due to subsidence of the surface sand brought about by the subjacent sand being removed during feeding by the proboscis of the worm. All other observers agree that the burrows are, in the majority of cases, U-shaped.

† For an account of curious seasonal changes in *Arenicola* and other burrowing marine animals see G. Bohn, "Les intoxications marines et la vie fouisseuse," Comptes Rendus de l'Académie des Sciences, Paris, tome 133, pp. 593-596; and "L'histolyse saisonnière," *ibid.* pp. 646-648.

‡ Geological Magazine, vol. viii., 1891, p. 489.



the castings in measured areas, he estimates that the amount of sand brought up by the worms in a year is 1,911 tons per acre, which, if evenly spread, would form a layer thirteen inches in thickness. Taking two feet as the average depth to which the worms descend, the layer of sand in which they burrow passes through their bodies once in every twenty-two months.

#### VASCULAR SYSTEM (See Plate III.).

The dorsal blood-vessel arises near the anus, and runs on the alimentary canal to the anterior end, when it breaks up into small vessels and capillaries. It contracts moderately regularly from behind forwards. Connected with it, in the tail, are intestinal vessels (a pair in each segment), each of which runs round the intestine and opens into the ventral vessel. The last seven pairs of gills send efferent vessels to the dorsal vessel, and between consecutive efferent branchial vessels there are two or three pairs of intestinal vessels. After receiving the most anterior of these efferent branchial vessels at the level of the thirteenth seta the dorsal vessel receives no segmental vessels until it reaches the œsophageal pouches, but it has numerous connections with the gastric plexus or sinus.

The dorsal vessel has no direct connection with the heart. In front of the heart the dorsal vessel receives on each side (1) vessels from the first three nephridia and neighbouring body wall, (2) from the œsophageal pouches, (3) from the second diaphragm and the body wall near the second setal sacs, and (4) from the body wall near the first setal sacs. It then runs forward, pierces the first diaphragm, and breaks up into capillaries supplying the buccal muscles, prostomium, otocysts, &c.

Small vessels collect the blood from these parts and unite to form the ventral vessel which, soon after its origin, gives off a median vessel to the first diaphragm and its pouches, then a pair of vessels to the nerve cord and the body wall in the neighbourhood of the second setal sacs. A little further back it gives off a median vessel running on the second diaphragm to the nerve cord, and a similar vessel on the third diaphragm to the nerve cord and first nephridium. From this point the ventral vessel, as it proceeds backwards, supplies the chætigerous sacs, body wall, nephridia and gills (in those segments in which the two latter are present) by segmentally arranged branches. As it runs along the intestine the ventral vessel is connected with the dorsal vessel by the intestinal vessels (see above), and it finally breaks up into capillaries near the anus.

On the walls of the stomach and intestine are numerous intersecting blood-streams. In young specimens the blood is contained in a plexus of small vessels, but as the animal grows these vessels become converted into a system of sinuses, and in old specimens the stomach practically lies in a gastric blood sinus, which is situated between the gut epithelium and the cœlomic epithelium which covers it. The dorsal vessel is, however, more or less distinct.

It is convenient to refer here to two portions of the plexus or sinus which are somewhat differentiated. (1) There are two vessels (or sinuses) ventrally situated, and known as the sub-intestinal vessels (figs. 23, 36). They commence just behind the heart, and may be traced backwards to the level of the twelfth setæ, behind which point they gradually disappear. They receive efferent vessels from the first six pairs of gills. (2) On each side of the anterior part of the stomach there is a lateral gastric

vessel (figs. 23, 36). This is usually first distinguishable posteriorly about the middle region of the stomach, and becomes more clearly differentiated as it proceeds forwards. Each receives blood from the dorsal and sub-intestinal vessels through the gastric plexus, and opens into the auricle, which is a thin walled expansion, probably of the gastric vessel.

After giving off the lateral œsophageal vessel, the auricle opens into the ventricle, the walls of which are muscular, and drive the blood into the ventral vessel. The lateral œsophageal vessel on each side gives off a branch to the œsophageal pouch, and then runs forward, supplying the lateral walls of the œsophagus, and breaks up into capillaries between the first and second diaphragms.

On each side of the nerve cord there is a small vessel which accompanies the cord along the whole length of the body. These neural vessels arise in front in the triangular area between the œsophageal connectives by union of capillaries from that region. Branches of the ventral vessel enter into connection with them in the five segments (second to sixth inclusive).

The vessels of the body wall are well developed (figs. 24, 36). There are on each side two longitudinal vessels distinguishable from the other vessels of the body wall by their somewhat greater size—(1) the nephridial longitudinal vessel which runs on the inner face of the body wall just ventral to the level of the nephridiopores, and (2) the more obvious and more important dorsal longitudinal vessel which runs parallel to but above the former, being slightly dorsal to the level of the notopodial setal sacs. The former (the nephridial longitudinal vessel) is only well marked in and for a short distance anterior and posterior to the nephridial region. The dorsal longitu-

dinal vessel is distinguishable anteriorly just behind the first seta, and may be traced to the posterior end of the animal. It receives blood chiefly from branches of the afferent vessels which supply the nephridia and gills. There is in each chætigerous annulus a series of connections between these vessels of the body wall, viz., (1) a short transverse vessel on each side connecting the nephridial and dorsal longitudinal vessels, (2) a circular vessel passing across the dorsal middle line, and connecting the right and left dorsal longitudinal vessels, (3) a vessel on each side connecting the nephridial and neural vessels, and (4) a short vessel passing over the nerve cord connecting the right and left neural vessels (fig. 24.) The dorsal and nephridial longitudinal vessels supply the body wall in the dorsal and lateral regions, while the neural vessels supply the nerve cord and ventral region of the body wall. All these vessels, which are best seen in transparent young specimens, are indirectly connected by a network of capillaries—the parietal vessels (fig. 36).

The ventral vessel is large and turgid in the greater part of the branchial region, and bears along its course tufts of dark brown filaments (fig. 23). The extent to which these are developed depends upon the size and age of the specimen; they are much more numerous in old than in young specimens. In old examples the brown filaments are found in other situations also, *e.g.*, along the gonidial vessel of the first nephridium (fig. 27), and upon many of the small vessels situated upon the inner face of the body wall, especially in the branchial region. Each filament (fig. 42) consists of a blindly-ending branch of the blood-vessel on which it is supported, covered with cells which contain large numbers of yellowish or brownish granules which are similar to those found in the chlorogenous

cells upon the stomach. Willem\* finds on examining the cells in the fresh state that they contain olein and acid urate of sodium (brown), the former substance being probably of a nature of a nutritive reserve and the latter an excretory product.

The blood plasma is red, due to the presence of hæmoglobin. The corpuscles are minute colourless rounded or ellipsoidal, nucleated cells  $\cdot 005$  to  $\cdot 01$  mm. ( $5$  to  $10\mu$ ) in diameter. They are comparatively few in numbers and their origin is unknown.

#### HEART AND HEART BODY.

The hearts are a pair of contractile bulbs connecting the gastric and ventral vessels. They are capable of great dilation. The thin walled auricle is merely a swelling on the gastric vessel. The ventricle has thicker muscular walls and its cavity is, in adult specimens, invaded by a "heart-body." In post-larval and young specimens the heart contains no trace of this body, but it has appeared in examples 65 mm. long (fig. 37). At this stage of growth the ventricular wall consists of an outer cubical peritoneal epithelium and an inner but indistinct endothelium between which muscular tissue is barely recognisable. The cavity of the ventricle is, however, invaded by processes which repeat the structure of the ventricular wall and are probably invaginations of it. Later on, as the muscular tissue develops in the wall, fresh invaginations occur (fig. 39). In full grown specimens each process of the heart body is seen to be composed of a very delicate endothelium, a muscular layer and a mass of cells, some granular and some glandular, either forming a fairly definite lining to the invagination or

\* *Miscellanées Biologiques dédiées au Professor Alfred Giard, Station Zoologique de Wimereux, p. 556. Paris, 1899.*

forming a mass loosely filling the process (figs. 33 and 40). The granules in the cells are in some cases united into a spherical mass lying in a vacuole; in others they are minute and scattered. They agree in appearance with the chlorogogen granules of the peritoneum. These ingrowths begin and are throughout best seen upon the posterior (and to some extent on the outer) wall of the heart. In large specimens they encroach on the cavity of the ventricle to such an extent as to sub-divide it into a large number of spaces so that the ventricle in section has a somewhat spongy appearance (fig. 39). The heart body appears to be a means of preventing regurgitation of the blood into the gastric plexus after systole, and of ensuring its passage into the ventral vessel. The presence of the chlorogogen granules suggests that it may also have an excretory function.

#### NEPHRIDIA.

There are six pairs of nephridia which open to the exterior on the fourth to the ninth chætigerous annuli, just dorsal and posterior to the neuropodia.

Each nephridium may be divided into three regions, an anterior funnel or nephrostome,\* a middle secreting portion and a posterior vesicle or bladder.

The funnel is always bright red in colour owing to its rich vascular supply. It opens into the cœlom by an elongated slit-like aperture which is generally directed forwards and inwards and is bordered by two lips which may be described as ventral and dorsal. The ventral lip is entire and almost semi-circular. The dorsal lip is slightly larger and fringed by ciliated, vascular processes placed close together on the edge of the lip (figs. 24 and

\* See also under **Post-Larval Stages**.

28). These processes are flattened spatulate or triangular structures fixed by their narrower end to the lip of the nephrostome. In young specimens (up to about 50 mm. long) these processes are represented by small ciliated tubercles on the edge of the lip (fig. 25), but later these not only increase in number but become larger and subdivided distally. Each of the processes eventually forms a flattened structure the edge of which is sub-divided by deep notches into from two to fourteen rounded lobes (fig. 29). Each of these lobes contains a blind diverticulum of the blood-vessel which traverses the edge of the dorsal lip, hence the processes are bright red in colour. The edges of both the dorsal and ventral lips are richly ciliated, and there are also numerous long cilia within the mouth of the funnel the motion of which is very obvious in fresh nephridia examined on a slide in sea water. The action of these cilia creates a current passing down the funnel to the oval aperture which leads into the middle or excretory part of the organ. This current carries into the nephridium small particles of foreign matter introduced into the cœlom and cœlomic cells burdened with excretory or with foreign particles.

The excretory part of the nephridium is a moderately thin-walled sac, usually dark brown, sometimes almost black in colour, owing to the presence of large numbers of brown excretory granules in the cells lining the sac. These excretory products are doubtless derived from two sources—from the blood flowing through the network of vessels upon the sac and from effete materials carried into the nephridium from the cœlom. This portion of the nephridium tapers posteriorly and opens into the bladder, which is usually greyish or brownish in colour. When expanded the bladder is more or less spherical, but when contracted it is rosette-like. Each bladder opens to the

exterior by a small oval aperture situated just above and behind the dorsal end of the neuropodium (fourth to ninth).

Immediately behind the posterior part of the funnel there is a small pinkish ovoid, club-shaped or cylindrical mass of cells. This is one of the reproductive organs of the worm, an ovary or testis as the case may be. There is no gonad on the first nephridium.

The funnel and anterior part of the excretory portion of the organ are attached to the body wall by a thin mesentery, and the funnel is further held in its place by one of the oblique muscles which crosses it and is fused to it.

The nephrostomes of the first pair of nephridia are situated on the anterior face of the third diaphragm. They are somewhat smaller than those of succeeding nephridia (fig. 27). The first and last pairs of nephridia are subject to considerable variation, and the former especially are often appreciably smaller than any of the succeeding nephridia. Out of about 160 specimens examined eight were found in which there is some departure from the normal condition. Six of these specimens show reduction or loss of the first nephridium. In one example the first nephridium of each side is very small, its secreting portion being only as thick as an ordinary pin; in the second example the first left nephridium is normal but the right one has no funnel; in the third case the first nephridium on each side has no funnel; in the fourth and fifth examples the first nephridium of one side has a very small funnel while the corresponding one of the other side is represented by a funnel only; and in the sixth specimen the first left nephridium is totally suppressed, the corresponding right one being present, but without funnel. In the other two cases of variation the



last pair of nephridia is affected. In a large specimen (250 mm. long) the sixth nephridium of the right side is normal, but the corresponding one of the left side consists of a funnel only. Another specimen, 100 mm. long, has only five pairs of nephridia opening on the fourth to the eighth chætigerous annuli, the sixth pair being totally absent. These cases seem to show that the funnel and the rest of the nephridium are, to some extent, independently formed, as in the examples mentioned above there are four cases in which the funnel is absent, and three in which a funnel only is present.

In worms about 17 mm. long the nephridia, which are about 5 mm. long, have already assumed the adult form. The funnel is of considerable size and has well-marked lips, the dorsal one bearing from three to five short, blunt, conical elevations which later become the large spatulate processes of the older nephridium. The secreting portion of the first nephridium is a rather wide, almost S-shaped tube; that of the following nephridia is much wider and sac-like. There is a rich vascular supply to all parts of the nephridium. In a specimen 44 mm. long the nephridia are about a millimetre in length (fig. 25). There is little change from the condition described above except that all the parts are larger; the dorsal lip of the nephrostome bears from four to seven small, blunt, conical processes. Nephridia of large specimens may attain a length of 8 mm. The funnel of such nephridia is large and its dorsal lip bears as many as thirty-two spatulate processes sub-divided distally into twelve to fifteen lobes.

The lips of the nephrostome are lined by a single layer of ciliated columnar or cubical cells, supported by a thin film of connective tissue. There is a fairly sharp line of demarcation between the cells of the funnel which have no concretions, and the cubical, columnar, or pear-

shaped cells of the excretory portion of the nephridium, which, even in young specimens, contain some excretory granules. Sections of the nephridia of young specimens show that the cells lining this part of the organ bear one or two long cilia on their inner ends, and that their somewhat vacuolated protoplasm contains comparatively few concretions (fig. 30). In older specimens the concretions in the cells are very numerous, filling up the middle and part of the distal region of the cell. The granules are either black or yellowish, and some at least consist of acid-urate of sodium (Willem). The distal fourth of the cell is almost free from granules; they appear to have been extruded into the cavity of the nephridium. Considerable collections of such yellow granules are occasionally found in the lumen of the organ. Finely powdered carmine, when injected into the cœlom, is taken up by the cells lining the excretory portion of the nephridium, and afterwards extruded in small masses. Below the excretory epithelium there is a thin, almost structureless, layer of connective tissue, while in the walls of the vesicle there is a thin layer of muscle fibres in a corresponding position. In young specimens the cells lining the vesicle contain fewer granules than those of the middle part of the nephridium; but in old specimens there is little difference in this respect between the cells of the two regions.

The network of blood-vessels with which the nephridium is provided lies between the excreting epithelium and the thin cœlomic epithelium which covers the organ.

From the dorsal vessel a branch is given off which runs on the anterior face of the first diaphragm. This divides—one part supplies the body wall in the region of the third setal sac, and is there connected with the longitudinal dorsal vessel; the other part enters the funnel of

the first nephridium, and after traversing the nephrostome and the excretory part of the organ ramifies finally on the body wall near the nephridial longitudinal vessel, or opens into the latter. The funnel of the first nephridium is also connected with the ventral vessel. Similar branches of the dorsal vessel pass to the second and third nephridia, and have a course corresponding to that connected with the first nephridium. A branch of the ventral vessel enters the funnel of each of these two nephridia a little in front of the middle of its length. Just behind the heart the ventral vessel gives off a branch which forks near the fourth nephrostome, one part passing to the dorsal longitudinal vessel, and the other taking the usual course through the funnel and the excretory part of the organ to the nephridial longitudinal vessel. Similar branches of the ventral vessel are given off to the fifth and sixth nephridia, but these send afferent branchial vessels to the first and second gills before entering the nephridia. Their course in the nephridia is as described for the corresponding vessel in the fourth nephridium.

Connected with all the nephridia there is also a small branch from the dorsal longitudinal vessel, which enters the anterior part of the excretory portion of the organ and ramifies there. This vessel is apparently sometimes missing from one or more of the nephridia.

The vessel which enters the funnel of the nephridium gives off a branch to the ventral lip, but the main trunk traverses the dorsal lip close to its edge, sending a blind vessel into each of its ciliated processes. Numerous branches are given off from the main vessel in both the dorsal and ventral lips, forming a close network; some of the vessels on the nephrostome have blind dilated terminations (fig. 28). After traversing the dorsal lip of the nephrostome, the large vessel leaves it at its posterior

angle, where it is usually joined by the vessel which has traversed the ventral lip. The gonidial vessel formed by their union runs over the excretory part of the organ, and sends branches to the vesicle, finally opening into the nephridial longitudinal vessel. In the upper part of its course the gonidial vessel is surrounded by the gonad, except in the first nephridium, where it usually bears filamentous blind outgrowths, covered with chlorogenous tissue (figs. 26, 27). The excretory part of the nephridium is covered with a network of vessels, which lie between the excretory cells and the cœlomic epithelium. This network is well seen in the nephridia of young specimens (fig. 25).

#### REPRODUCTIVE ORGANS.

The reproductive organs are closely associated with the nephridia. They are found immediately behind each nephrostome, except the first, as a small, pinkish, ovoid, club-shaped or cylindrical mass of cells,  $\cdot 4$  to  $1\cdot 0$  mm. long, surrounding the gonidial vessel and apparently produced by proliferation of its cell covering (fig. 24). The gonidial vessel is developed on the nephridia in very early life, it may be recognised even in post-larval stages. In a specimen 44 mm. long the gonads, though minute, are recognisable (fig. 25). The gonad is well seen in an adult stained nephridium, being distinguished by its affinity for stains (*e.g.*, carmine or hæmatoxylin). It is a closely packed mass of cells in which at the anterior end, *i.e.*, the end in contact with the nephrostome, the cells are small, almost uniform in size, and have well-marked, deeply-staining nuclei. In the middle and posterior portions of its length the cells on the surface of the gonad become differentiated, and in females young oocytes, and

in males young spermatogonia may be recognised. The anterior portion of the gonad is covered by a thin layer of cœlomic epithelium, but the posterior portion, from which oocytes or spermatogonia are being shed is not covered by an epithelium. The genital products are shed at an early stage from the gonad into the cœlomic fluid where they complete their growth. The oocytes leave the ovary when they have reached a diameter of  $\cdot 016$  to  $\cdot 02$  mm. While floating in the cœlomic fluid they increase in size and the nucleus becomes vesicular, its diameter being about half that of the oocyte. Very small yolk granules are deposited in the protoplasm, they are rather more abundant round the nucleus, the peripheral portion of the protoplasm contains less yolk. The protoplasm is surrounded by a thin vitelline membrane about  $1\mu$  in thickness. Oocytes must be produced in the gonads at a great rate, for the body cavity of large worms is filled with them almost to bursting by about the end of February. Ripe ova are not spherical but discoidal. The face of the egg is either circular (usually) and about  $\cdot 15$  mm. in diameter, or it is oval with diameters of  $\cdot 16$  mm. and about  $\cdot 14$  mm., while the third axis of the egg measures  $\cdot 08$  to  $\cdot 09$  mm. (figs. 67 and 68).

On the surface of the posterior part of the testes groups of two, four or eight cells, young spermatogonia, may be seen. They are shed into the cœlom: the youngest stage usually found in the cœlomic fluid is formed of eight spermatogonia arranged around a vesicular mass of protoplasm—the blastophore (figs. 60 and 61). The cells undergo numerous divisions, the products of which remain attached to the central blastophore. There is then a period during which the cells do not divide but increase in size becoming spermatocytes. Each of these divides probably twice successively (as in the better known

spermatogenesis of other invertebrates) to form four spermatids. Disc-shaped masses of spermatids are thus produced, the thickness of which is equal to about one-fourth the diameter of the face (fig. 65). In each mass there is a central cavity containing the remains of the blastophore, a small quantity of a slightly fibrous coagulum being present. Each spermatid undergoes no further division, but is gradually transformed into a spermatozoon. An early stage of this transformation is seen in fig. 64. The nucleus is an oval compact body at one end of which is a small conical mass of protoplasm, by which the spermatid is attached to the blastophore. This becomes the apical body or acrosome of the spermatozoon. At the opposite end of the nucleus there is a clear substance from which the "middle piece" of the spermatozoon is apparently largely derived, and following this is the rest of the protoplasm, which is being drawn out to form the tail of the sperm. When shed into the sea the ripe spermatozoa soon become free from the blastophore and move by means of the tail, which appears to be a somewhat stiff filament capable of comparatively limited movements (fig. 66). A ripe spermatozoon is about  $\cdot 058$  mm. long, it has a curiously shaped head  $\cdot 04$  mm. long, and a long slender tail ( $\cdot 054$  mm. long). The nucleus forms the greater part of the head of the sperm. At one end is the apical body forming a cap slightly sub-divided by a median groove. It is by means of this apical cap that the sperm is later attached to the egg with which it is about to unite. At the other end of the nucleus is the middle piece which is apparently notched behind to receive the basal part of the tail. Ripe sperms may usually be obtained about the end of February or the beginning of March.

The genital products, instead of floating freely in the cœlomic fluid, often accumulate in the space between the

oblique muscles and the ventral body wall. In ripe specimens there is often also a considerable mass of genital products behind the third diaphragm, and pushing it forwards into a pouch-like outgrowth (or two such outgrowths, one at each side of the alimentary canal). This is due to the fact that while the perforations in the diaphragm allow the passage forwards of the waves of cœlomic fluid (see above), they do not permit the passage of the ova, or spermatid masses, to any extent. These are, therefore, as it were filtered out of the cœlomic fluid, and collect in a more or less compact mass behind the ventral portion of the diaphragm.

The genital products escape from the animal by means of the last five pairs of nephridia, the terminal vesicles of some of which are often found to be distended with ova or spermatozoa during the breeding season (fig. 26). In a specimen 200 mm. long the vesicles of some of the nephridia were 14 mm. long and 6 mm. in width owing to distension by ova, while in another specimen the vesicles were filled with sperms, and were 5 mm. in length and 4 mm. broad. The funnel of the nephridium is usually widely open during the breeding season. During the discharge of ova from the female the eggs are caught in considerable numbers by the slimy mucus which covers the body. Nothing further is known, however, about the oviposition. Ripe females, with the cœlom filled with ova, are found during the later portion of February and during March, but by the end of the first or second week in April the ova are usually all discharged.

#### NERVOUS SYSTEM.

The central nervous system is composed of the brain, the œsophageal connectives, the stomato-gastric system and the ventral nerve cord.

**The Brain** is situated in the prostomium, and even in large specimens (250 mm. long) is only about a millimetre in length. It consists of a pair of anterior lobes placed well forward in the prostomium, a pair of posterior lobes which lie below the nuchal organ, and an intermediate region which connects the anterior and posterior lobes (figs. 46, 47).

The anterior lobes are short but broad—in fact this is the broadest part of the brain; behind these lobes the brain gradually tapers. Their shape may be seen from fig. 46. They are separated in front by a cœlomic space. Each gives off anteriorly and dorsally a series of nerves to the epithelium of the prostomium, and ventrally nerves to the upper lip and neighbouring part of the eversible buccal mass. The anterior part of these lobes consists of small clusters of cells, separated from one another by fibrous tracts and by neuroglial tissue. Further back the delicate neuropile which forms the core of the anterior lobes is well seen covered by clusters of cells and fibrous tissue. Bundles of nerve fibrils may be traced from the bases of the prostomial epithelial cells into the neuropile. Larger unipolar ganglion cells are found just outside the neuropile, and particularly on the side nearest the middle line. These cells are more numerous immediately in front of the point of union of the anterior lobes, and for a short distance behind that point. The œsophageal connectives arise from the anterior lobes at the point where the neuropile reaches its greatest development. The eyes are found on the dorsal side of this part of the brain. Passing backwards along the middle region of the brain, it is seen that the ganglion cells become more restricted to the dorsal and lateral faces, the middle and ventral parts being composed largely of neuropile, in which also neuroglial cells and fibrillæ may be recognised. The cells



situated on the dorsal aspect of this part of the brain are in close association with the epithelium of the middle part of the prostomium.

The posterior lobes are small and tapering. On tracing them backwards, the cells are seen to decrease in quantity, and each lobe is continued as a fibrous tract, accompanied by a thin covering of cells, which lies on the inner side of the nuchal organ just below its sensory epithelium (figs. 34, 47). The posterior lobes are separated by a coelomic space, containing muscles and blood-vessels.

The brain has a strong ventral neurilemma sheath, especially on the anterior lobes, into which some of the prostomial muscles are inserted. Other muscles pass between the anterior lobes, and are inserted into the connective tissue underlying the epidermis. The brain derives its blood supply from the dorsal vessel, small branches of which break up into capillaries on its ventral surface.

On examining a number of specimens of gradually increasing sizes, two series of changes are seen to take place in the brain. Firstly there is an increase in the number of elements involving a growth of the brain, and secondly a differentiation of form and a tendency to the formation of groups of cells or "centres." In a post-larval specimen, 4.5 mm. long, the brain is .11 mm. long, .07 mm. wide and .05 mm. deep. In a worm 7.5 mm. long the brain has about twice these dimensions (length .2 mm.), and in another 17.5 mm. long the length of the brain has again doubled (.4 mm.). Beyond this point the rate of growth is much slower, and in a specimen 10 inches (250 mm.) long, the brain is only .9 mm. in length. The description of the minute structure of the brain given just above is drawn from specimens about 60 mm. in length, in which the brain is a little more than half a

millimetre long. In older specimens the fibrous portion of the brain becomes proportionately larger and more complex, and the neuroglia is better developed. The nerve cells also become aggregated into groups, separated by bands of fibrous tissue.

**The Œsophageal Connectives** arise from the anterior lobes of the brain. They run beneath the epidermis and circular muscles passing round the sides of the pharynx, and uniting about the middle of the third chætigerous annulus. The course of the connectives is marked externally by the metastomial grooves (figs. 2, 6). The connective of each side gives off (1) a nerve to each of the first two inter-annular grooves, (2) a nerve to the otocyst, (3) a nerve to the buccal mass which is probably connected with the stomato-gastric system, (4) numerous nerves to the epidermis of the peristomium and following segment. Each connective is a stout fibrous cord, with numerous cells upon its outer face. At the point of origin of the nerve to the otocyst, and along the course of this nerve, there is a considerable number of ganglion cells (fig. 49). The connective is enclosed in a sheath of neurilemma, which is better developed in old specimens, and by ingrowths partially sub-divides the fibrous part of the connective into two or three.

**The Ventral Nerve Cord** is usually separated from the epidermis by the layer of circular muscles, but in some specimens the cord in the tail and in the last chætigerous segment, lies only just below the epidermis. Its blood supply is derived from the two lateral neural vessels, which are connected with a series of capillaries lying chiefly on the dorsal face of the cord (fig. 24).

The cord is non-ganglionated, ganglion cells occur moderately evenly distributed along the whole length of the ventral and lateral surfaces of the cord except at the

points where the spinal nerves are given off. The cord gives off a pair of nerves situated in each inter-annular groove, and lying below the circular muscles, and in each chætigerous annulus either a pair of stout nerves or two bundles (right and left) of two to four nerves, which run outwards towards the parapodia, supplying the circular and longitudinal muscles between which they lie (fig. 55).

In transverse section the cord is usually oval in shape, being flattened from above downwards. The cells are arranged on the ventral and ventro-lateral faces of the fibrous part of the cord. The entire cord is invested by a thin sheath of neurilemma, and the fibrous part of the cord is partially sub-divided into two by a median vertical sheet of neuroglia (fig. 54).

The ganglion cells are chiefly unipolar, and are small and sub-equal, though here and there are larger cells, generally in the neighbourhood of the giant cells.

**The Giant Cells and Giant Fibres.**—These cells are much larger nerve cells, placed at segmental intervals along the cord (fig. 52). There are no giant cells in the brain or oesophageal connectives; the first one occurs just behind the point of union of the connectives near the level of the groove between the third and fourth chætigerous annuli. This cell belongs to the achætous segment (composed of the third and fourth annuli) immediately following the peristomium. The next cell is found in the annulus behind the first chætigerous annulus, and the cells in most of the other segments are found in a corresponding position, but they may be a little anterior or posterior to this level. The cells are, therefore, situated close to the posterior limit of each segment. In many of the segments only a single giant cell is present, but in about one-third or one-half of the segments two cells are found near together, one in front of the other. They are present in

the tail, but are not usually recognisable in the first few segments, which are very small, and probably only recently formed. Typical giant cells may be seen in some of the middle and posterior caudal segments, but careful search has failed to reveal them in a considerable number of the tail segments of the two worms examined. In the mid-dorsal region of the cord there are one, two or three giant fibres seen in section (figs. 35, 36). Anteriorly there is only one, in the middle region of the body either two or three, and in the tail usually one. At first sight they appear to be tubes with distinct walls and homogeneous contents, but by suitably staining longitudinal sections the contents are found to be distinctly fibrillar, but the fibrils are so fine that they are scarcely recognisable in transverse sections. The wall of the tube is composed of nucleated cells. These giant fibres arise from the giant cells in the cord. This is not so easily demonstrated in *Arenicola marina* as in *A. grubii*, in which the cells and fibres are larger (fig. 53). In the latter species each giant cell gives off a stout process, which runs in a somewhat sinuous course through the fibrous part of one side of the cord towards the dorsal surface. Shortly after leaving the cell the process gives off one or two branches, which pass into and ramify in the fibrous part of the cord. The process then passes dorsally and enters either the median or, more usually, one of the lateral giant fibres. The single median fibre present in the anterior region of the cord arises from the first giant cell situated just behind the union of the œsophageal connectives. In those portions of the cord where two or three giant fibres are present there are connections between them, generally in each chætigerous annulus. Branches from one or other of the giant fibres may occasionally be traced into the spinal nerves.

The giant cells and giant fibres of *A. marina* (fig. 54) have similar relations to those of *A. grubii*. As a rule the giant cells are almost mid-ventral in position, and their diameter in *A. marina* is from about '04 to '08 mm. The cell is pyriform in shape, and the narrow extremity is prolonged upwards, and follows the course described above. Each cell has a fibrillar sheath. The protoplasm is clear, but in favourable preparations is seen to be traversed by delicate, darkly-staining fibrillæ, which branch; but the branches do not appear to reach the nucleus. These neuro-fibrillæ may be traced into the process of the cell, and for some distance towards the giant fibre, with which the process is connected. The nucleus is a large vesicular structure, with a diameter about one-third that of the cell to which it belongs; within the well-marked nuclear membrane is a small amount of chromatin reticulum, and usually one deeply staining nucleolus. The nucleus is nearly always excentric, being placed near the broader end of the cell.

The giant cells are not differentiated in a post-larval specimen 4·5 mm. long. In specimens 17·5 mm. long and upwards they are well marked, but they are apparently no larger in specimens 250 mm. long than in others one-fourth this length.

#### SENSE ORGANS.

The sense organs are the otocysts, the nuchal organ, the eyes and the prostomium. To this list of sensory structures should be added (1) the papillæ of the proboscis, in the epithelium of most of which sense cells may be distinguished; (2) scattered sense cells in the epidermis, and (3) the notopodial setæ, as Retzius has found nerve endings around their bases. Even when the animal is at rest the

constant movements of protraction and retraction of these setæ suggests that they may have a sensory function.

**The Otocysts** (figs. 46, 49 and 50) are the best developed sense organs of *Arenicola*. They may be seen in dissections close to the outer edge of the dorso-lateral portions of the œsophageal connectives. Each is a vesicle communicating with the exterior by a narrow tube, the external opening of which is situated in the peristomium close to the point where the metastomial groove crosses the first inter-annular groove (fig. 5). The otocyst is placed at an angle to its tube and both are lined by a very thin cuticle, best seen in old specimens. The epithelial wall of the otocyst is thicker in old specimens than in young ones, due to the elongation of the cells. In specimens 65 mm. long the epithelium is about  $25\mu$  thick, but it is twice as thick in specimens 250 mm. long. The cavity of the otocyst does not, however, increase in the same way, it is practically the same size in these two specimens, its mean internal diameter being  $\cdot 12$  to  $\cdot 13$  mm. (cf. figs. 49 and 50). The epithelium is composed of non-ciliated sense cells and supporting cells. The sense cells are not always easily recognisable, but in some preparations they may be distinguished by their fusiform shape, their more deeply staining nuclei and by the possession of delicate neurofibrillæ. In favourable sections, stained with iron hæmatoxylin, each sense cell is seen to be traversed by a delicate, deeply-staining fibril which terminates either just below or at the surface of the cuticle. Similar cells and their fibrillæ may also be seen in the wall of the adjacent part of the tube of the otocyst. The tube is lined by columnar cells among which are gland cells; the cells lining its proximal part (immediately after it leaves the otocyst) are ciliated. The epithelium of the distal part of the tube gradually merges

into the epidermis. The nerve supply to the otocyst is derived from the œsophageal connectives. Around the point of origin of this nerve and along its course are numerous large ganglion cells with vesicular nuclei. The nerve comes into contact with the otocyst at the point where the tube leads off to the exterior and is intimately related to both structures as it provides them with a sheath of nervous elements. The nervous sheath lies below the epithelium of the otocyst and tube, among the nerve fibres occur scattered fusiform or stellate cells.

The otocyst, in life, contains a fluid of a somewhat viscous nature which consists of a secretion of the walls of the otocyst and its tube mixed with sea water. It also contains numerous otoliths in the form of foreign bodies such as quartz grains, portions of spicules, frustules of diatoms, &c. In some specimens the original otoliths, which were irregular in shape, have been covered by layer upon layer of secreted substance of chitinoid nature, the resultant otoliths having rounded outlines. This condition is met with in moderately old specimens (130 to 250 mm. long) in which also the tube of the otocyst has become closed either by apposition of its walls or by the blocking of the lumen by a granular substance secreted by the gland cells in the wall of the tube (fig. 50). That this rounded character of the otoliths depends upon the closure of the tube is shown by the fact that in other specimens (about 170 mm. long), in which the lumen of the tube is a fair-sized slit, the otoliths are irregular and uncoated foreign bodies. In most young specimens the otoliths consist of irregular bodies which are almost naked, *i.e.*, they have either no secreted covering or else it is a mere film the presence of which is indicated by its staining with hæmatoxylin.

Whenever the otocyst is examined fresh under the

microscope, either *in situ* in young transparent specimens or after being rapidly dissected out, the otoliths exhibit a peculiar quivering and rotatory movement. This motion is probably due to the action of the cilia in the entrance to the tube which leads to the exterior.

The otocyst does not appear to be an organ of hearing as the animal takes no notice of even loud sounds made in its immediate vicinity. It is more probably an organ for enabling the animal to appreciate its position in the sand or in the water and thereby to direct its movements.

**The Nuchal Organ** (figs. 5, 34 and 47) is a U-shaped ciliated groove formed by an invagination of the epidermis of the posterior part of the prostomium. The epithelium of the organ is composed of columnar cells, some of which are distinguished as sense cells by the presence of neurofibrillæ. The intervening supporting cells are rather stouter, and many of them are ciliated. In old specimens pigment granules are deposited in many of the cells. Beneath the epithelium there is a layer of nervous elements in connection with the posterior brain lobes. The organ probably has an olfactory function.

**The Eyes** (figs. 47 and 48).—Two to five eyes, one of which (the oldest) is larger than the others, are present on each side of the prostomium in post-larval stages (figs. 56 and 59). Only in very young specimens from the sand are they visible externally. They may be found in sections of specimens up to 70 mm. long by which time they have sunk below the epidermis and have become imbedded in the mass of ganglion cells on the dorsal surface of the anterior cerebral lobes just in front of their point of union. They are difficult to find in older specimens owing to the increase of pigment in the prostomium, and they are apparently wanting in some old specimens.

The eyes are very simple in structure. Each is com-



posed of a cup-shaped mass, about .01 mm. in diameter, of reddish brown pigment spherules, grasping the base of a spherical or ovoid lens.

The animal is sensitive to light. If its anterior end be protruding from the burrow and light be thrown upon it, the worm at once disappears from view. In its reaction to light *Arenicola* resembles the earthworm, and, as in the latter, the re-action may be due to the general sensitiveness of the anterior end.

**The Prostomium** (figs. 5, 47 and 48).—The epithelium of the anterior and dorsal face of the prostomium consists of columnar cells among which slender fusiform sense cells may be distinguished. These are generally in small groups and their slender tips are level with or project slightly beyond the outer surface of the cuticle. The bases of the cells are in intimate relation to either the cells of the brain itself or to the fibres of the nerves which connect the epithelium to the brain. In some preparations the neurofibrillæ traversing the sense cells may be seen. Among the ordinary columnar cells of the prostomial epithelium there are, especially in the mid-dorsal region, numerous swollen gland cells.

#### DEVELOPMENT.

The early stages of development of *Arenicola marina* are unknown. A brief resumé may be given of the development of *A. claparedii*. We may presume with moderate certainty that the development will follow similar lines in the two species.\*

\* The following account is from observations made during my occupancy of a Table in the Zoological Station in Naples, in April, May and June, 1900:—From April 7th onwards specimens of *A. claparedii* were examined at frequent intervals, and on April 21st the first ripe males and females were obtained, the eggs fertilised, and the early stages of segmentation observed. The observations on the

Ripe ova of *Arenicola claparedii* were artificially fertilised by adding to the sea-water in which they were contained a small quantity of sea-water containing spermatozoa taken from the cœlomic fluid of a mature male. In one to two hours after this the two polar bodies were extruded, and in about four hours after the addition of the spermatozoa nearly all the eggs had divided into two cells (fig. 69), a larger (*C D*)† and a smaller (*A B*), each of which in less than an hour divided again.‡. Three of the cells (*A B C*) so produced were nearly equal in size; the fourth (*D*) was considerably larger than these (fig. 70). By the next division each cell was cut into an upper or anterior and a lower or posterior portion. The four upper cells (*1a, 1b, 1c, 1d*) so produced become displaced, or rotated, with respect to the axis of the egg, so that each no longer lies directly above the cell from which it arose (fig. 71). This rotation, which affects nearly all the cleavages of the first day, is characteristic of the "spiral" type of cleavage found in *Nereis*, and other Polychæta. These four upper cells form the first quartette of ectomeres. From the four lower cells (*1A, 1B, 1C, 1D*) a second (fig. 72) and a third set of four (*2a, 2b, 2c, 2d*, and *3a, 3b, 3c, 3d*) are successively

development given in the following account were made on various batches of eggs and larvæ and extended from this date until May 19th, when the last of the larvæ died. I beg to express my sincere thanks to Professor Dohrn for his generosity in placing a Table and the resources of his Station at my disposal, and to the Government Grant Committee of the Royal Society for a grant towards the expenses of this and other work done in Naples.

† I have used Dr. Child's nomenclature of the cells, which is based on that proposed by Wilson in his classical memoir on "The Cell Lineage of *Nereis*," *Journal of Morphology*, Vol. VI.

‡ The early development of *A. claparedii* appears to be very similar to that of *A. cristata*, which has been studied in great detail by Dr. C. M. Child (see *Archiv für Entwicklungs-mechanik der Organismen*, Band IX., Heft 4, May 22nd, 1900). See also E. B. Wilson, *Studies from the Biological Laboratory, Johns Hopkins University*, Baltimore, Vol. II., 1883, pp. 271-299.

Diagram showing the early cleavages of the egg. The first division of the members of the first quartette is shown, but the other quartettes are not followed. The mesoblast and endoderm cells are labelled; all the cells which are derived from the first, second and third quartettes (labelled 1, 2 or 3 followed by a small letter) are ectomeres.

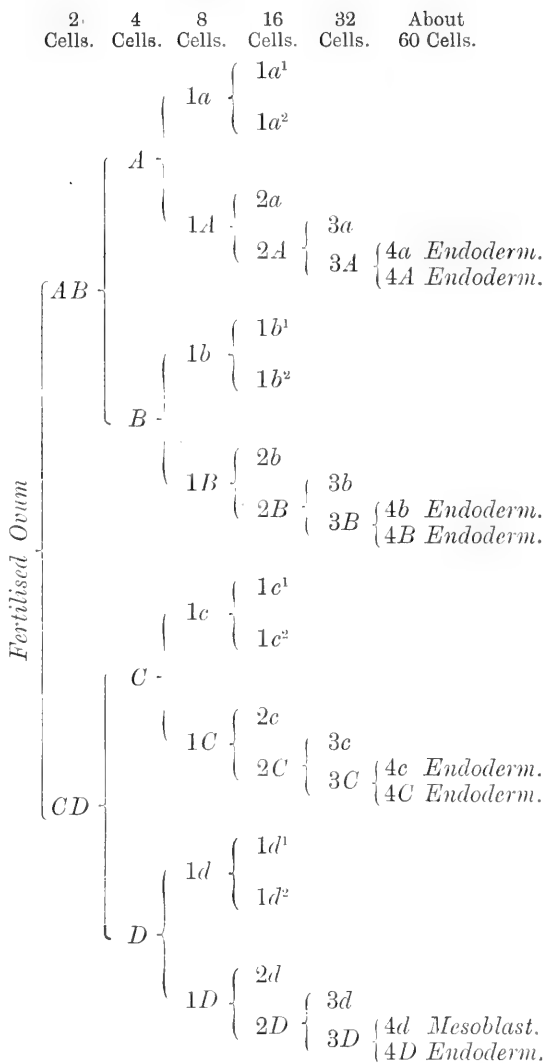


FIG. 69. FIG. 70. FIG. 71. FIG. 72. In FIG. 73 all the endo-  
 derm cells except 4c  
 2B, 2C, and  
 2D, are not  
 seen in the  
 figure.  
 derm cells except 4c  
 have again divided,  
 and the mesoblast  
 cell has sunk into  
 the segmentation  
 cavity.

cut off, all of which are ectomeres. Then a fourth set of four ( $4a$ ,  $4b$ ,  $4c$ ,  $4d$ ) is cut off from the lower cells. One of these four cells ( $4d$ ) is very large; it is the mesoderm cell, or somatoblast, and it soon sinks below the surface into the cleavage or segmentation cavity. The other three cells of the fourth quartette ( $4a$ ,  $4b$ ,  $4c$ ), and the four lower cells ( $4A$ ,  $4B$ ,  $4C$ ,  $4D$ ) are the endoderm cells, and they occupy a position at the lower or posterior end of the larva. At this time the larva consists of (1) about 50 ectomeres, products of division of the cells of the first, second and third quartettes; (2) the seven endoderm cells mentioned above, and (3) a mesoderm cell. At the end of twenty-four hours<sup>c</sup> after fertilisation, the larva, when seen from the lower pole, has the appearance shown in fig. 73. The ectoderm cells are by this time more numerous (about 70 to 80), and the endoderm cells, distinguished by the numerous yolk granules in their protoplasm, are in this case thirteen in number, each of the seven primary endoderm cells, except  $4c$ , having divided into two. Lying in the segmentation cavity, and not shown in the figure, are two mesoblast cells produced by division of the primary mesoblast cell ( $4d$ ). The larva, now a blastula, is converted into a gastrula by growth of the ectoderm cells over the yolk-laden endoderm cells. About twenty-eight hours after fertilisation the anterior ciliated band is just recognisable and the larvæ were found to be rotating slowly within the vitelline membrane. The stomodæal invagination had also made its appearance (fig. 74). Twenty-four hours later both the ciliated bands of the larva were well marked, a broad one just anterior to the mouth, and a narrow one near the posterior end of the animal, and by this time the rotatory motion was much

\* It is very probable that in shallow water, in sunlight, the development would be more rapid than it was in the cool laboratory.

more rapid. One or two eyes of orange-red colour were also present on the anterior portion of the larva. During the third day the animal elongated and contracted at intervals, and evidently both longitudinal and circular muscle fibres were present. At the end of the third day the telotroch\* larva worked its way out of the vitelline membrane through a thin area which had previously made its appearance (fig. 75).

When hatched the larva was about .25 mm. long. It either crawled on the bottom of the dish or swam actively by means of its cilia. Between the two bands of cilia there appeared on the ventral surface a broad longitudinal band of short cilia. Soon after hatching a small spade-shaped seta ( $7\mu$  long) was observed in one specimen. It could be protruded and retracted (fig. 76). The larvæ grew very slowly in the laboratory and it was not until more than four days after hatching that the setæ of the following segment appeared. In the meantime the spatulate setæ of the first segment had been reinforced by the addition of a seta with a long, drawn-out tip, and a little ventral to this the first crotchet appeared. Two days later the alimentary canal seemed to be complete from mouth to anus, its central part was rather distended with yolk. The cœlom was quite obvious post-orally, and the ventral body wall was thickened, due to the formation there of the ventral nerve tract. The two belts of cilia and the longitudinal ventral band gradually decreased in size from this point onwards. Two days later another segment acquired its setæ. Both the chætigerous segments in front of this had on each side two setæ and a crotchet, the third segment bore only the newly formed spatulate seta. The larvæ crawled about the bottom of

\* The term applied to Polychæte larvæ in which the cilia are arranged in two bands forming a preoral and a peri-anal ring.

the dish surrounded by a film of mucus. Although the mouth was open it was found impossible to induce the larvæ to feed and they died as soon as the yolk in the stomach was absorbed, *i.e.*, about a fortnight after hatching, without having advanced beyond a stage showing three or four chætigerous segments (fig. 77).

#### POST-LARVAL STAGES.

Post-larval stages of *A. marina* are found in the surface waters of the sea, and are almost invariably enclosed in a mucous or gelatinous tube, which overhangs the worm at each end. The external diameter of the tube is from two to three times that of the animal. The young worm is capable of wriggling movements, which are not seriously impeded by the enveloping tube.

The term "post-larval stage" was given by Benham to a stage in the development in which the animal possesses the full adult number of segments, and is divisible into an anterior chætigerous region and a posterior achæteous region, or tail, but in which the gills are not yet completely formed, or have not even made their appearance. By this definition all the pelagic specimens of *A. marina* which I have examined, with one possible exception, are post-larval stages. This one, a Lytham specimen, 3.9 mm. long, bears the full complement (thirteen pairs) of gills, and all of them except the first two have become branched, being formed of two or three finger-shaped filaments (fig. 56). In this specimen also the annulation is well marked, and the prostomium proportionately smaller than in any of the other specimens examined. This animal has reached the end of the post-larval stage, and would doubtless soon have settled down to its littoral habitat. It is the only recorded specimen

in which all the gills are present, and it would therefore appear that most specimens take to a littoral habit rather earlier than this one. In point of length it is not, however, a large specimen, being only 3.9 mm. long. In life it would certainly be longer; this measurement is taken from the mounted specimen, which was unfortunately preserved in a somewhat contracted condition. The longest post-larval stage I have seen measures 6.5 mm. from anterior to posterior ends, and this one has no gills. Evidently, therefore, the time of appearance of these organs varies in different specimens. Perhaps the two specimens under consideration belong to the two different varieties of *A. marina*, but there are no means of establishing this point in the larger specimens; the smaller almost certainly belongs to the littoral variety.

Nothing is known concerning the duration of the pelagic life of these post-larval stages, but it is apparently at any rate some days, judging from the varying sizes of the specimens captured within a few days of each other. For example, in a series of eight specimens taken near Plymouth during the last days of March and early days of April, 1901, there are specimens varying from 3.5 to 6.5 mm. in length. Again, the varying sizes of these specimens, and the length of the period during which they may be taken in the tow-nets, show that the spawning period of *A. marina* may occasionally extend over two or three months, *e.g.*, post-larval stages have been taken as early as March 8th, and as late as May 29th. The latter date is, however, exceptionally late; most specimens are taken either in March or in the early days of April. The smallest worms I have found in the sand were taken near the end of June, and are 17 mm. long. They possess thirteen pairs of well-branched gills, the prostomium has assumed the adult form and relations.

the nephridia are sac-like, and their dorsal lip bears from two to five papillæ—in fact these worms are miniature copies of adults. Other specimens up to 44 mm. in length were found at the same time and place which show a similar condition of the prostomium and nephridia (fig. 25), and are probably but little older than those 17 mm. long. It is probable that these specimens were produced from eggs laid in the early spring of the same year, so that the young worms are three or four months old.

I have examined seventeen post-larval stages of *A. marina* varying in length from 3·5 to 6·5 mm. Three of the specimens (4·6, 3·6 and 3·6 mm. in length) were examined living. The following description is drawn from specimens 4·6 to 5·1 mm. in length. Such examples are ·2 to ·3 mm. in diameter and the chætigerous portion of the worm is nearly three times as long as the tail.

The prostomium is a moderately large and somewhat lozenge-shaped or spatulate structure at the anterior end overhanging the mouth (figs. 57, 58). Upon it are seen the eyes to the number of two to five on each side of the middle line (figs. 56, 59). One of these, the oldest, is larger than the rest. Their structure is described above. The epithelium of the prostomium bears small scattered groups of sense hairs (fig. 59). Immediately behind the prostomium is the nuchal groove, the degree of development of which seems to vary in different specimens. In some it is scarcely recognisable, but in others it is a well-marked groove extending across the posterior border of the prostomium. When the lips of the groove are everted the rapid movement of the cilia may be observed.

Behind this is the peristomium in which the otocysts are well seen (fig. 57). The internal diameter of these organs is about ·04 to ·06 mm., and the external aperture



of the tube was at times comparatively large. The otoliths, consisting of quartz grains, were quivering and revolving rapidly all the time the living animals were under observation. This motion is probably due to the action of the cilia of the proximal part of the tube. The motion of the otoliths almost ceased when sea water which had been standing over chloroform was added. On the ventral side of the peristomium at its anterior edge is a crescentic or nearly semi-circular aperture—the mouth (fig. 58).

Between the peristomium and the first chætigerous segment there is a segment which, in all the post-larval stages I have seen is achætous. Benham\* and apparently also Ehlers† have found a small seta in this segment, but evidently this is a transitory seta which soon disappears leaving the segment achætous as it is in the adult. Both the peristomium and this achætous segment are rather smaller than the succeeding chætigerous segments and both are usually divided into two by a faint groove so that, as in the adult, the region between the prostomium and the first chætigerous segment is divided into four rings. In the third of these the œsophageal connectives unite. By comparing these post-larval stages with adults it is clearly seen that the first two rings of the latter belong to the peristomium and the next two to the first true body segment which has lost its setæ and has become fused with the peristomium (cf. figs. 5 and 57).

There are nineteen chætigerous segments, in each of which notopodial and neuropodial setæ may be seen (fig. 56). The notopodial setæ are of two kinds. Some are

\* Journal Marine Biological Association, New Series, Vol. III., p. 49, 1893.

† Zur Kenntniss von *Arenicola marina*, L., Nachrichten von der Königl. Ges. der Wissenschaften, Göttingen, 1892.

capillary setæ, about  $\cdot 2$  to  $\cdot 3$  mm. long, bearing along one edge of their distal fourth a narrow lamina, which may be slightly dentate distally (fig. 11*C*). In some setæ the whole of the lamina seems to be broken up into a series of minute pointed processes similar to, but, of course, smaller than, those of the adult setæ. Some of these setæ have a narrow but shorter lamina on the opposite side. The other notopodial setæ are quite different (fig. 11 *A, B*). They are only about  $\cdot 18$  mm. long. Each of them ends in a long fine point and beyond the middle of its length the seta bears a thin but broad lamina or wing on each side upon which faint oblique striations are occasionally visible. Each notopodial pencil contains only one seta of the latter kind accompanied by one to five of the longer capillary setæ. The shorter setæ with a broad lamina on each side soon disappear, they are not recognisable in young specimens, 17 mm. long, from the sand.

The neuropodial crotchets are about  $\cdot 04$ — $\cdot 05$  mm. long (fig. 15). Each has a thickening upon the shaft lying just below the level of the epidermis. On examining the rostral region there are seen to be two or three well-marked teeth behind the rostrum, and a small process often ending in a fine point under the rostrum. These structures are in focus along with the rostrum, and lie approximately in the same plane. On careful focussing, it may be seen that the teeth are not confined to the region behind the rostrum, but that on the sides of the latter there are small teeth, so that the rostrum projects from the centre of a series of teeth situated around its base. The sub-rostral process marks the position of the smallest of these teeth. In their natural position the crotchets lie so that the tip of the rostrum is directed dorso-laterally. In most post-larval specimens about 5 mm. long each neuropodium bears only two to six

crotchets, the smaller number being present in the first three or four neuropodia. In the branchiate Lytham specimen 3.9 mm. long, the crotchets are rather more numerous, there being eight or nine in many of the neuropodia. The crotchets of post-larval stages differ from those of adults in the relatively large size of the teeth, and in the angle which the rostrum makes with the shaft. In post-larval crotchets the rostrum is approximately at right angles to the shaft, while in older specimens it makes a much greater angle, in large specimens as much as 130 degrees (cf. figs. 15, 18).

As stated above, there are no gills in any of the seventeen post-larval stages examined, except one. This one is 3.9 mm. long, and the thirteen pairs of gills are present, and all except the first two are branched. The largest gills are behind the middle of the series, and this is probably the region where the gills are first formed. In other specimens no gills are indicated, but in the living specimen 4.6 mm. long, immediately behind the notopodia of the penultimate and the two or three preceding chaetigerous segments, the blood-vessels form a well-marked loop, as if preparatory to the formation of a gill, and there is a very slight elevation of the skin in this region. Later on, as shown in the Lytham specimen, the gill becomes successively a papilla, a digitiform, and then a branched structure. The gill arises, therefore, as a special respiratory structure, and is not a dorsal cirrus which secondarily becomes a branchia.

The tail is very similar to that of the adult. The terminal segment bears a number of circum-anal papillæ, each of which carries a tuft of four or five sense-hairs, which project posteriorly.

The skin is glandular. It contains numerous scattered cells, each filled with yellow granules. If the

animal, and especially its anterior end, be subjected to pressure, the yellow pigment seems to become diffused through the whole skin, and the yellow granules in the cells mostly disappear. Some of the pigment also exudes into the water. Later such a compressed anterior end becomes green, and this pigment resists solution in alcohol. The skin also contains numerous mucus-forming cells, by the activity of which the enveloping tube is formed. A specimen which was deprived of its tube was found twelve hours afterwards enclosed in an elongate mass of mucus formed by the epidermis.

The secondary annulation of the skin, which corresponds with that of the adult, is shown in many post-larval stages, except in the first two or three segments, where the annuli are less clearly indicated. The metastomial grooves are only faintly seen. They unite just behind the middle of the achæitous segment, and are there continuous with a shallow mid-ventral groove which marks the position of the ventral nerve cord.

The mouth is a crescentic or semi-circular aperture at the anterior ventral edge of the peristomium (fig. 58). The pharynx is protrusible, and bears small papillæ. It leads into the ciliated œsophagus, which bears the two glands, each of which is a simple finger-shaped outgrowth of the gut, .25—·45 mm. long, directed forwards, and showing in its posterior portion three or four slight internal ridges which are the precursors of the larger septa found in the glands of adults. The secretion of the glands is already present in the form of droplets, some of which are still in the glands, while others are in the posterior portion of the œsophagus. The stomach is marked by elongate oval areas, between which are blood-vessels. Chlorogogen cells are present, but are only distinguishable along the margin of the blood-vessels, and

not over the whole of each oval area as in the adult. Very fine débris is present in the intestine of one or two specimens, but in most the gut is practically empty.

The blood-vessels are well developed, and are arranged as in the adult. The blood is reddish or pink in colour. The heart is a simple tube, dilated at its upper end, and contracting frequently. The two hearts do not beat in unison. The wall of the ventral vessel contains numerous spherical yellowish (some nearly colourless) granules, probably chlorogenous. The flow of blood in the dorsal vessel is from behind forwards.

Cells are already abundant in the cœlomic fluid. They are mostly oval, or spindle-shaped, and about  $\cdot 02$  mm. long. Many of them, especially the oval cells, already contain yellow granules.

The musculature is similar to that of the adult. Longitudinal muscles are well developed, the circular ones only feebly so, and the oblique muscles are very slender.

There are six pairs of nephridia opening near the fourth to the ninth neuropodia, as in the adult. Each nephridium is a curved ciliated tube (fig. 31), the lips at the anterior end of which are slightly everted, and one is larger than the other. Attached to the larger lip and hanging over towards the smaller are (except in the first nephridium) from four to eight protoplasmic processes\* which are only visible in living specimens (fig. 32). They are generally of unequal length and in the living specimen,  $4\cdot 6$  mm. long, the longest processes measured only  $\cdot 03$  mm. Each process is slightly swollen about the middle of its length and tapers distally. It bears about ten moderately strong, long cilia directed backwards

\* See Goodrich, Quart. Journ. Micr. Science, p. 729-730, fig. 49, Vol. 43, 1900. In his specimen the processes appear to have been rather longer than in mine.

towards the aperture of the nephrostome. The length of the nephridium is about  $\cdot 3$  mm. The walls of its middle portion already contain numerous excretory granules, most of which are yellow, but others are almost colourless. The ciliary action in the tube, especially in its posterior part, is very vigorous at intervals. The first nephridium is more slender than any of the others and its lumen, especially in the anterior part, is very minute. Its anterior end (nephrostome) forms a slight swelling on the anterior face of the third diaphragm, due almost entirely to the larger lip, between which and the smaller lip there is a slit-like opening. The larger lip bears a number of well-marked cilia, but no ciliated processes like those on the other nephridia could be seen. The relation between the funnels of the nephridia of these post-larval stages and those of the adult is unknown. Goodrich holds that the adult funnel is not a nephrostome but a genital funnel which is developed from the cœlomic epithelium and is added to the anterior end of the nephridium. The gonads are not yet distinguishable.

The brain is divisible into a large anterior part (not yet sub-divided into two lobes) consisting of neuropile with a thick covering of cells, a posterior part which is bilobed, each lobe having a fibrous core with a cellular covering, and a middle region uniting the two. The ventral nerve cord is more closely united with the epidermis than it is in the adult. No giant fibres or giant cells are recognisable.

#### SYSTEMATIC POSITION AND CLASSIFICATION OF THE ARENICOLIDÆ.

Benham\* divides the Polychæta into two branches—the Phanerocephala and the Cryptocephala. The chief

\* Polychæta in Cambridge Natural History, Worms, Rotifers and Polyzoa. 1896.

characters of the former are—the prostomium is distinct and often bears, in addition to the paired eyes, sensory tentacles and palps (as, for example, in *Nereis*); the body segments are more or less alike and not divisible into two sharply-marked regions, distinguishable by the different arrangement and character of their setæ and by internal differences. In the *Cryptocephala* the prostomium is compressed or hidden by forward growth of the peristomium and thus becomes insignificant; the tentacles are reduced but the palps become greatly enlarged and subdivided, forming the crown of gills; the body is divisible into a “thorax” and “abdomen,” distinguished by the form and arrangement of their setæ and by certain internal differences. *Arenicola* belongs to the branch *Phanerocephala* which is divided into five sub-orders.

I. NEREIDIFORMIA.—Prostomial tentacles and palps well developed; peristomial cirri usually present; parapodia well developed with acicula, dorsal and ventral cirri; setæ usually jointed; muscular pharynx with chitinous jaws; septa and nephridia regularly repeated throughout the body. Chiefly predaceous and carnivorous worms, *e.g.*, *Nereis*.

II. SPIONIFORMIA.—Prostomium without tentacles and palps; peristomium with a pair of long cirri; parapodia project only to a slight extent, their dorsal cirri may be large and act as gills, setæ unjointed; no jaws; septa and nephridia regularly repeated. Chiefly tubicolous or burrowing worms.

III. TEREBELLIFORMIA.—Prostomium forms a prominent lobe (upper lip) with or without tentacles, but without palps; peristomium may bear cirri or “tentacular filaments”; parapodia feebly developed, ventral cirri absent, dorsal cirri may form gills, setæ unjointed, uncini (short, sharply-curved dentate hooks) usually present;

no jaws; septa incomplete except one strong diaphragm in front; anterior to this the nephridia are large and excretory while the posterior ones are mere funnels and act as genital ducts. Tubicolous or burrowing worms.

IV. CAPITELLIFORMIA.—No prostomial processes; parapodia do not project, setæ unjointed, capilliform in the anterior segments, hooded crotchets in the posterior segments (this division does not correspond to marked internal differences), no cirri; no jaws; nephridia small and may be more than one pair per segment; no blood-vessels, but the cœlomic corpuscles are red. Burrowers.

V. SCOLECIFORMIA.—Prostomium rarely with sensory processes; peristomium without cirri; parapodia not well-developed, setæ unjointed, no uncini present; no jaws; septa not regularly developed (some being absent); nephridia often reduced in number but all alike. Burrowers.

*Arenicola* belongs to the last sub-order, which contains six families.

(1) OPHELIDÆ.—Comparatively short worms, many of which have a pearly lustre; no prostomial processes; parapodia obscure, but dorsal cirri often present, *e.g.*, *Ophelia*.

(2) MALDANIDÆ (or CLYMENIDÆ).—Prostomium truncated; body formed of long and few segments, never provided with gills or other processes; the neuropodial setæ are hooked crotchets, each with characteristic sub-rostral tuft. Inhabit sandy tubes, *e.g.*, *Nicomache*, *Achiothea*, *Clymene*.

(3) ARENICOLIDÆ.—See below for definition.

(4) SCALIBREGMIDÆ.—Arenicoliform or maggot like; gills, if present, confined to the first five segments; prostomium small and either drawn out at its lateral angles into short processes or bluntly rounded; parapodia con-



sisting of almost identical notopodia and neuropodia each ramus bearing capillary and furcate setæ; four diaphragms at the posterior end of the first, second, third and fourth segments; heart median; a pair of slender, tubular nephridia in each segment, except in a few of the anterior ones. Burrowers, e.g., *Scalibregma*, *Eumenia*.

(5) CHLORILEMIDÆ.—Prostomium bears a pair of grooved processes and several tentacles which act as gills and are usually greenish, due to the colour (chlorocruorin) of the contained blood; capillary setæ in all segments except the peristomium; those of the anterior segments are directed forwards forming a protection for the head; limits of segments not clearly marked; only one or two internal septa present and a corresponding number of pairs of nephridia. Burrowers in mud, e.g., *Siphonostoma*.

(6) STERNASPIDÆ containing the single genus *Sternaspis*; body short, anterior region thickened and carrying on each side three rows of setæ; on the ventral surface at the posterior end there is a bilobed horny plate, round the edges of which are some fifteen or sixteen tufts of long setæ, and dorsal to these two bundles of filamentous gills; the ends of the genital ducts project freely.

The family ARENICOLIDÆ contains two genera—*Arenicola* and *Branchiomaldane*. The latter is, however, only provisionally and doubtfully placed here, and as its anatomy and affinities are so uncertainly known it may be neglected in this account. The definition of the family may, therefore, be taken as that of the genus *Arenicola*, which may be summarised as follows:—

Limnivorous Polychæta provided with numerous pairs of branched gills not present on the anterior seven segments. Prostomium small or moderately well developed, bounded posteriorly by the nuchal organ, no tentacles or palps. Parapodia each consisting of a conical

notopodium bearing capillary setæ and a transversely thickened neuropodium bearing a vertical row of crotchets. In the chætigerous region, except in the first three segments, there are four annuli between successive chætigerous annuli. Internally there are three diaphragms at the anterior end of the first, third and fourth segments. The pharynx has no armature. A pair of hearts present placing the gastric vessels in connection with the ventral vessel. There are five, six or thirteen pairs of nephridia. A pair of otocysts lies in the peristomium (absent in *A. clapedii*).

The genus is divisible into a caudate and an ecaudate section.

I.—The characters of the caudate Arenicolidæ may be briefly stated thus: A distinct tail present; the parapodia and gills do not extend to the posterior end of the animal. The body is often swollen anteriorly. Gills pinnate or derivable from the pinnate type, eleven to thirteen pairs, the first (which may be small or even absent) on the seventh or eighth chætigerous segment. Prostomium consisting of a median and two lateral lobes, brain with distinct anterior, middle and posterior regions. Funnel of nephridium with dorsal lip well provided with flattened, spatulate, ciliated, branched, vascular processes; ventral lip ciliated, entire (*i.e.*, not deeply notched as in the ecaudate Arenicolidæ). Gonads small, ova discoidal, vitelline membrane comparatively thin (1 to  $3\mu$ ).

(a) *A. marina*, Linnæus. Nineteen chætigerous segments. Thirteen pairs of gills; the first, which is on the seventh segment, may be reduced (or suppressed). Otocysts opening to the exterior. Otoliths, numerous foreign bodies (quartz grains, &c.) which may, however, be covered with a layer of secreted chitinoid substance giving them a rounded outline. Six pairs of nephridia opening on seg-

ments 4 to 9. One pair of œsophageal pouches, cylindrical, club-shaped or conical. Diaphragmatic pouches (on the first diaphragm) small, globular or flask-shaped.

Found on both sides of the Atlantic north of 40° N. latitude.

(b) *A. assimilis*, Ehlers.\* Twenty chætigerous segments. Thirteen pairs of gills, the first of which is situated on the eighth segment (the first gill is liable to be reduced or suppressed). Otocysts large, opening to the exterior. Otoliths numerous, spherical or rounded chitinoid bodies. Six pairs of nephridia opening on segments 4 to 9. Several pairs of œsophageal pouches; the anterior pair long, club-shaped or filiform, the others much smaller and pear-shaped. No pouches on the first diaphragm.

Recorded from the extreme south of the American continent.

(c) *A. assimilis*, var. *affinis*, Ashworth.† Nineteen chætigerous segments. Thirteen pairs of gills, the first (liable to reduction or suppression) on the seventh segment. Otocysts large, opening to the exterior. Otoliths numerous, and composed either of foreign bodies (quartz grains, &c.), or of spherical chitinoid bodies. Other characters as in the type of the species (see above).

Recorded from Otago Harbour, New Zealand, the Macquarie Islands, the Falkland Islands.

(d) *A. clapedii*, Levinsen.† Nineteen chætigerous segments. Thirteen pairs of gills, the first on the seventh segment (this pair of gills is liable to reduction or suppression especially in specimens from the west coast of North America). Lateral lobes of prostomium well

\* For an account of these forms see *Ashworth, J. H.*—Q.J.M.S., Vol. 46, pp. 737-785. Pls. 36-37.

† For an account of these species see *Gamble, F. W.* and *Ashworth, J. H.*—Q.J.M.S., Vol. 43, pp. 419-569. Pls. 22-29.

developed. No otocysts. Five pairs of nephridia opening on segments 5 to 9. Two or more pairs of œsophageal pouches, the anterior pair long and slender or club-shaped, the others shorter and usually pyriform. No pouches on the first diaphragm.

Recorded from the Mediterranean and from the west coast of the United States.

(e) *A. cristata*, Stimpson.\* Seventeen chaetigerous segments. Eleven pairs of gills, the first on the seventh segment. Otocysts, closed spherical sacs each containing a single large spherical chitinoid otolith. Six pairs of nephridia opening on segments 5 to 10. One pair of œsophageal pouches, cylindrical or club-shaped. Diaphragmatic pouches (on the first diaphragm) large and finger-shaped.

Found in the Mediterranean, in the West Indies and on the eastern shores of North America, south of latitude 40° N.

II.—The characters of the ecaudate Arenicolidæ are as follows: No tail, the parapodia and gills extend to the posterior end of the animal. Body generally of an almost uniform diameter. Number of gills variable (according to age and species), gills branched unilaterally. Prostomium simple, conical, non-lobate. Brain commissural. Otocysts closed spherical sacs, otoliths spherical. Diaphragmatic pouches long and finger-shaped. Œsophageal pouches, one pair, flask-shaped. Nephridia with dorsal lip bearing somewhat cylindrical digitiform, often branched, ciliated vascular processes, ventral lip deeply notched in middle, the two halves semi-circular in shape. Ova generally oval with thick vitelline membrane (5 to 6 $\mu$ ).

(a) *A. grubii*, Claparède.\*—First gill on segment 12

\* For an account of these species see Gamble, F. W. and Ashworth, J. H.—Q.J.M.S., Vol. 43, pp. 419-569. Pls. 22-29.

(may be small). Five pairs of nephridia opening on segments 5 to 9. Twelve to twenty-eight pairs of gills. Gonads small.

Recorded from the English Channel (Plymouth, the Channel Islands and the French coast), Isle of Man, Ireland (Valencia), Scotland (Loch Linnhe), the Mediterranean.

(b) *A. caudata*, Johnston.\*—First gill on segment 16. Thirteen pairs of nephridia opening on segments 5 to 17. Nineteen to forty pairs of gills. Gonads large, twelve pairs; in females each ovary bears as many as thirty digitiform vascular processes bearing ova; in males each testis is produced into one or more large, thin reniform lobes.

Recorded from Scandinavia, the English Channel (Plymouth, the Channel Islands and the French coast), Isle of Man, Ireland (near Fairhead, Antrim).

#### The Affinities of *Arenicola marina*.

*Arenicola marina* is the only caudate species found in Britain. As a comparison of the characters given above shows, its nearest ally is the southern *A. assimilis*, var. *affinis*, from which it differs in only two respects. In the latter species there are several pairs of œsophageal glands and no pouches on the first diaphragm, while in *A. marina* there is a single pair of œsophageal glands and a pair of septal pouches. The type specimens of *A. assimilis* are rather further removed from *A. marina*, additional and striking points of difference being found in the number of chætigerous segments (19 in *A. marina*, 20 in *A. assimilis*), and the position of the first gill (on the seventh segment in *A. marina*, on the eighth in *A. assimilis*).

\* For an account of these species see Gamble, F. W. and Ashworth, J. H.—Q.J.M.S., Vol. 43, pp. 419-569. Pls. 22-29.

*A. claparedii* is at once marked off from the common lugworm and from all other known species of *Arenicola* by the fact that it has no otocysts. Other points, such as its well developed lateral prostomial lobes, the presence of several pairs of œsophageal glands, the presence of only five pairs of nephridia and the absence of septal pouches, further separate this species from *A. marina*. *A. marina* is also well removed from *A. cristata* by the number of chætigerous segments (19 and 17), and gills (13 and 11 pairs), the position of the nephridia (opening on segments 4-9 and on 5-10), and the nature of the otocysts and otoliths.

Between the two ecaudate species and *A. marina* there are only the points of agreement which are common to the genus. The common lugworm differs especially from *A. ecaudata*, and these two species may be regarded as lying almost at the opposite ends of the series.

#### THE AFFINITIES OF THE ARENICOLIDÆ.

The discussion of the affinities of the Arenicolidæ is rendered somewhat difficult owing to the fact that the details of the anatomy of one or two of the neighbouring families are very imperfectly known. The three families Arenicolidæ, Scalibregmidæ, and Opheliidæ have several features in common, as they are limnivoruous and present certain of the peculiarities characteristic of such Polychæta. They have a spacious cœlom sub-divided anteriorly by diaphragms and non-septate in the middle of the animal; the alimentary canal consists of an eversible pharynx followed by an œsophagus (bearing one or more pairs of lateral glandular outgrowths), a dilated stomach and a straight intestine usually with a ventral groove, and the blood-vessels in the middle region of the

animal are so arranged as to leave the stomach considerable freedom of movement.

The Arenicolidæ agree with the Scalibregmidæ in the points named above, in the general shape of the body, the sub-division of the segments into annuli, the sculpturing of the skin, the small lobed prostomium, and the presence (in *Scalibregma* and *Eumenia*) of gills of a similar type. The brain and non-ganglionated nerve cord of the caudate Arenicolidæ is similar to that of the Scalibregmidæ. There are also points of difference between these two families which are of considerable importance. In the Scalibregmidæ the two rami of the parapodia are practically identical, but they are very different in the Arenicolidæ. In the latter the neuropodium bears crotchets only, and the notopodium bears capillary setæ, while in the Scalibregmidæ both rami bear two kinds of setæ, capillary and furcate, the latter being characteristic of the family. In some of the Scalibregmidæ the parapodia form laminate appendages bearing dorsal and ventral cirri, which are not found in *Arenicola* (cirri are occasionally seen in the posterior region of American specimens of *A. cristata*). The gills of *Scalibregma* and *Eumenia* are confined to the first five segments, on which they never occur in *Arenicola*. The heart in the Scalibregmidæ is a median structure, while in *Arenicola* there is a pair of hearts. The nephridia of the Scalibregmidæ are minute, but numerous, the simple microscopic funnel leading into a slender U-shaped tube; the nephridia of *Arenicola* are few in number and wide and sac-like, each having a large funnel fringed with ciliated vascular processes. Several of the Scalibregmidæ bear complex segmental lateral sense-organs which are not found in *Arenicola*.

The Arenicolidæ have a few characters in common with the Opheliidæ, but the accounts of the latter family

are so fragmentary as to preclude detailed comparison. Besides the points mentioned above as common to the three limnivorous families, the Opheliidæ and Arenicolidæ agree in the character of their nephridia and in their non-ganglionated nerve-cord. The main points of difference between them are (1) the shape of the body; (2) the skin, which is not sculptured and sub-divided into annuli in Opheliidæ; (3) there are no branched gills and no hearts in Opheliidæ; (4) the parapodia, on many of which, in *Ophelia*, there is a filamentous dorsal cirrus.

On comparing the Arenicolidæ and Maldanidæ the differences are again more obvious than the resemblances. They agree in the general form of their parapodia, in the small number of nephridia and gonads, and in the simple brain and ventral nerve cord with uniform covering of ganglion cells. They differ in habit, the Maldanidæ have few but long segments, they have no gills and no otocysts, and the tail segment is specialised. The alimentary canal of Maldanidæ is simple and bears no special glands, and there are no hearts. The genus *Branchiomaldane* presents features some of which are intermediate between these two families.

The anatomy of the Chlorhæmidæ is imperfectly known, but there appear to be no points in which they approach the Arenicolidæ. The former family differs from the latter in its setæ, septa, nephridia, median heart, the processes of the prostomium and the ganglionated nerve-cord. The Sternaspidæ are still further removed from the Arenicolidæ by the peculiar arrangement of their gills and setæ, the presence of ventral shields, special genital ducts, coiled alimentary canal and only a single pair of nephridia.

It may be concluded that the Arenicolidæ form a compact family clearly distinguished from the neigh-



bouring families, but having some affinities with the Scalibregmidæ, Opheliidæ and Maldanidæ.

#### PARASITES.

I.—A considerable number of specimens of *Arenicola*, from the Lancashire coast near Blackpool, were found to contain small ovoid bodies attached to, or imbedded in, the muscles of the anterior region. These proved to be Distomid cercariæ which had migrated into this position and encysted there. Each is about half a millimetre long and a third of a millimetre broad and is surrounded by a moderately thick cyst-wall. In each specimen the two suckers, the muscular pharynx, the two limbs of the intestine, about a dozen flame cells and the rudiments of two of the reproductive organs (testes ?) may be distinguished. These would remain encysted in *Arenicola* until the worm was eaten by the final host (probably a fish or a bird), in which the cercaria would be liberated from the cyst and would grow into a "fluke."

II.—In other specimens Coccidia are occasionally very abundant in the walls of the stomach, intestine and nephridia. They are much more common in *A. grubii* than in *A. marina*. They are spherical or ovoid cells, each about 0.14 mm. in diameter when fully grown, with a vesicular nucleus containing a large nucleolus. A few cysts, 0.2 to 0.25 in diameter, each containing a few thousands of rounded spores (sporozoites) have been met with on the outer surface of the gut or attached to the muscles of the body wall. Each spore is 3 to 4 $\mu$  in diameter, and consists of a thin covering of protoplasm enclosing a (comparatively) large nucleus.

As the lugworm is a common food of fishes (especially of flat fish), a knowledge of its parasites is desirable, as

these may have an important bearing on the parasitic diseases of some of our food fishes.

#### DIRECTIONS FOR PRACTICAL WORK.

*Arenicola* should be dissected as soon as possible after it is taken from the sand, and especially in warm weather, as the animal soon dies and changes rapidly take place in both the external and internal structures. Specimens at least eight or nine inches long should be obtained if possible; the large Laminarian variety is excellent for dissection.

Specimens intended for dissection may be killed by placing them in sea water in a jar and adding sea water which has been shaken up with chloroform. By this method the specimens are gradually narcotised, and they usually die in a moderately expanded condition. If the process of killing be too rapid, as, for example, if the worms were dropped into chloroform, the contraction of the muscles is sometimes so strong as to cause rupture of the body wall, in which case the cœlomic fluid is lost and a considerable portion of the alimentary canal is forced out through the opening, and some of the blood-vessels may give way. As soon as the specimens are dead they should be transferred to the dissecting dishes containing sea water. If ordinary sea water be not available make up beforehand a sufficient quantity of artificial sea water by adding about 35 grammes of sea-salt to each litre of fresh water required.

**External Characters.** Note the shape of the worm; its division into an anterior abbranchiate chætigerous portion, middle branchiate chætigerous region and posterior achatous and abbranchiate tail; the segmentation;

the annulation; the parapodia and setæ\*; the gills\*; the prostomium and nuchal organ; the peristomium and the succeeding achætous segment; the external apertures, the mouth, anus, nephridiopores, and apertures of otocysts.

### Dissection.

Extend the animal under sea water with the dorsal surface upwards, fixing it down by two pins through the sides of say the first segment, and two through the sides of one of the posterior branchial segments. Open the animal by making an incision with fine scissors along the mid-dorsal line, beginning about the middle of the chætigerous portion. Raise the flaps gently with the forceps and extend the cut anteriorly to within about an eighth of an inch of the prostomium, and posteriorly about an inch into the tail. The tail is difficult to open satisfactorily, unless great care be taken the alimentary canal will be cut open with the body wall. The flaps of the body wall should be regularly pinned out right and left so that the animal is moderately well stretched both longitudinally and transversely.

If it is desired to examine the cœlomic fluid before proceeding with the dissection the first incision should be made while holding the worm in the hand over a watch glass. The cœlomic fluid which at once escapes through the cut must be examined immediately, as on standing even a short time the corpuscles collect into clots. Examine the fluid fresh and determine the sex of the specimen. A few permanent preparations (for method see below) are also useful.

At the breeding season the ova and masses of spermatids are so abundant as to partially obscure some of the

\* The detailed examination of the setæ and gills is better deferred until later. See Sections VII. and VIII. below.

organs, *e.g.*, the nephridia. In this case the reproductive products should be carefully washed away. A change of sea water in the dish may even be necessary.

The internal organs may be examined in the following order:—

I. **Cœlom** (see above).

II. **Musculature**.—Note the longitudinal and circular muscles, the muscles of the pharynx, the setal muscles, the oblique muscles, the three anterior septa, the two pouches on the first septum, the rudimentary septa (the safety cord accompanying the afferent vessel of the fourth nephridium, and the small septa in two or three of the posterior branchial segments), the caudal septa, the mesenteries of the first and second segments.

III. **Alimentary Canal**.—Note the proboscis, pharynx, œsophagus, œsophageal glands, stomach covered with the vessels of the gastric plexus between which are the areas of chlorogogen cells, intestine, anus. Later (after section V.) the stomach and intestine may be slit open and their contents washed out so as to show the ventral ciliated groove and the subsidiary grooves. The stomach and intestine may be pushed over to one side, care being taken not to break any of the blood-vessels, so as to leave the greater part of the body wall in this region open to view.

IV. **Vascular System**.—Note the dorsal, intestinal, gastric, œsophageal, ventral, sub-intestinal, neural, nephridial longitudinal and dorsal longitudinal vessels, the afferent and efferent vessels of the nephridia and gills and the hearts. The sub-intestinal vessels are difficult to see, and are only visible on carefully drawing the ventral vessel a little further away from the wall of the stomach. Note the chlorogogenous tissue on the ventral vessel.

V. **Nephridia and Gonads**.—Look out for any

departure from the normal in the number or structure of the nephridia. After examining these organs in a general way, carefully remove the oblique muscles which cover one or two of the nephridia and examine these with a lens, noting the various parts. Note the funnel, its dorsal lip fringed with processes, its ventral simple lip, the excretory portion of the organ, the bladder, the gonad (not present on the first nephridium), the blood supply, &c. At the breeding season the vesicles may be distended with ripe ova or spermatozoa. Remove one of the nephridia entire and transfer it, with as little disturbance as possible, to a glass slide and examine under the microscope. If the worm has been freshly killed the strong action of the cilia of the funnel will be well seen. Note the vessels of the funnel, some of which have blind dilated endings. Note also the gonad traversed by the gonidial vessel. The excretory portion of the organ may be cut across; at the cut edges the action of the long cilia borne by the cells of this portion may be seen.

VI. **Nervous System.**—The greater part of the ventral nerve cord may be easily seen on pushing the alimentary canal to one side. It may be readily traced forwards as far as the first diaphragm, but the rest of the cord, the connectives, the brain and the otocysts are only exposed after the first diaphragm has been carefully cut away from the body wall. Very little further dissection is required. It is often, however, an advantage to cut away the pharynx, as shown in fig. 46. The exposure of the brain requires some care. The otocysts are usually rather yellowish bodies about the size of a small pin's head, their position is seen in fig. 46. Remove an otocyst to a slide, cover it and examine it under the microscope. The peculiar quivering motion of the otoliths will be seen if the specimen has been only recently killed.

VII. **Gills.**—Examine one of the gills about the middle of the series. Note its attachment to the body wall, its basal webbing, the main trunks and their branches. Remove the gill to a slide and examine it under a low power to see the mode of branching and the blood-vessels. Note the small size and simple character of the first gill.

VIII. **Setæ.**—Notopodial setæ may be easily obtained from a dissected specimen by taking hold with the forceps of the inner end of the setal sac and drawing it inwards. In this way the whole bundle of notopodial setæ will be removed. To clean off the tissues of the setal sac and any adhering strands of muscle the preparation may be warmed in 5 per cent. caustic soda. After washing in water the setæ may be placed in glycerine for a time and then permanently mounted in glycerine jelly. Only unworn setæ should be selected for examination.

By treatment of an excised neuropodium with warm caustic soda the muscles become gradually softened, and by the aid of a pair of needles may eventually be separated into an anterior and a posterior mass, between which the chætæ lie. With care the entire band of neuropodial chætæ may be obtained. It should then be washed in water, placed for a time in glycerine and mounted in glycerine jelly. The various stages of formation of the new chætæ at the ventral end of the series may be seen. There are usually two or three fully developed crotchets which have not yet come into use. These should be selected for observation of their characters as they are uninjured by wear.

I have found it very useful to have dissections of small specimens, 17 to 50 mm. long, for the study of young nephridia, which are too small to be easily removed from the body wall, gonads, blood-vessels, &c.

Preserved specimens were generally used and the dissection was performed under 70 per cent. spirit.\* The specimen was opened in the usual way and pinned out on a piece of weighted cork in a small glass dish, and the alimentary canal carefully removed. The specimen and the cork were then put into stain, either carmine or Mayer's acid hæmalum, for 12 to 24 hours. After the excess of stain had been removed the worm and the cork were passed into 90 per cent., and then into absolute alcohol. The body wall, which by this time was moderately certain to retain its shape, was unpinned, and after another change of absolute alcohol was placed in oil of cloves and afterwards mounted in balsam. Fig. 25 was drawn from such a preparation, in which the blood-vessels, &c., are very clear. Useful preparations may also be made by splitting the anterior portion of preserved specimens into two by a median sagittal cut. Such sections give a good idea of the position and relations of the nuchal organ, diaphragms, proboscis, &c.

Preparations of the **Cœlomic fluid** may be made thus. Immediately after the fluid is removed from the animal spread a small drop evenly over the middle of a glass slide so that it forms a very thin film. This may be treated in either of the two following methods:—

(1) Invert the slide so as to bring the film over the mouth of a bottle containing glacial acetic acid and hold it there for several seconds. The vapour of the acid will kill and "fix" the cells, *i.e.*, will coagulate their proto-

\*If a fresh worm be used, after the dissection under sea water has been completed, the specimen should be "fixed" in a saturated solution of corrosive sublimate. In this case, however, the dissection should be pinned out with cactus needles, and not with ordinary pins, as the latter would produce a deposit of mercury in the neighbouring tissues. After fixation wash in water and in 50 per cent. and 70 per cent. spirit, until all traces of sublimate are removed. Then stain as directed.

plasm. The slide may then be warmed *gently* over a flame until the film begins to dry. It may then be stained by placing a drop of safranin or dahlia over the film. After a few minutes (three or four) the excess of stain may be removed by washing with a little 50 per cent. and 70 per cent. spirit. A few drops of absolute alcohol are then run over the film, followed by oil of cloves, and finally the preparation is mounted in balsam.

(2) The film may be allowed to gradually dry (a matter of only a minute or two if it be even and thin), but before it is actually quite dry a little saturated solution of corrosive sublimate is gently run over it and the whole slide immersed for a few minutes in this solution. The slide is then washed in the usual way in water and in the alcohols (several hours), and may then be stained by any of the methods applied to films or sections, *e.g.*, iron-alum-hæmatoxylin, hæmalum, safranin, dahlia, carmine. These preparations show the various cœlomic cells and also the reproductive products.

#### S e c t i o n s .

Specimens not above six inches long are large enough for this purpose. Larger ones are more difficult to deal with on account of the hardness of the musculature after imbedding.

Specimens intended for sectioning must, of course, be treated so as to remove all the sand from the alimentary canal, and it is a matter of some little difficulty to ensure this. The worms should be kept in dishes of clean sea water, into which a small stream of water is allowed to trickle or else the water in the dishes should be changed at least twice a day. The dishes should be examined several times a day, and the sand, which has been voided by the worms, removed. After four or five days of this treat-



ment the alimentary canal will contain very little sand. This can usually be ascertained by examining the worms by strong transmitted light, when any sand present in the gut will be detected. As soon as all the sand has been voided the worms may be placed in a dish containing a smaller quantity of sea water. They may then be narcotised by dropping absolute alcohol little by little on to the sea water until the liquid contains about 5 per cent. of alcohol. After a few hours the worms will be sufficiently narcotised and may then be killed in an extended condition. Before attempting to kill them ascertain by touching them if they are thoroughly narcotised. Care should be taken not to allow the worms to become quite dead in the alcoholised sea water, as the tissues suffer.

A good killing and fixing mixture is sublimate acetic (95 parts saturated sublimate solution and 5 parts glacial acetic acid). As soon as the worms are dead transverse incisions\* should be made in the body wall at intervals of  $\frac{1}{2}$  to  $\frac{3}{4}$  inch, so as to allow the re-agents to penetrate rapidly and fix all the internal organs. If the operations above described be carefully carried out the worms will be killed straight, a great convenience when sectioning.

After a few hours in sublimate acetic the worms are washed in running (fresh) water for a few minutes, and then transferred to 50 per cent. alcohol (two or three hours), to 70 per cent. and to 90 per cent. alcohol (two or three changes). To the latter a few drops of tincture of iodine should be added to dissolve from the tissues of the worm the last traces of sublimate. Fresh iodine should be added as the liquid becomes decolourised, and this

\* This should be done as rapidly as possible, as on contact with metal a deposit of mercury is liable to be formed in the tissues. The worms should be transferred from one vessel to another (where that is necessary) by means of glass, horn, or paper lifters, and all contact with metal avoided.

treatment continued until the straw-colour of the spirit remains permanent. It may then be assumed that all the sublimate has been removed. The worms may then be transferred to fresh 90 per cent. spirit, and kept therein until required for sectioning.

The segments required for sectioning are dehydrated by passing through absolute alcohol (three changes), and are then placed in either xylol or cedar-wood oil. The writer has found the latter very satisfactory, provided that care be taken to ensure the thorough removal of the oil when imbedding in paraffin wax. That is, the specimen is transferred from cedar-wood oil to the first vessel of wax and afterwards to a second and third, before finally imbedding. The time required in the bath, of course, depends on the size of the tissue, but for a piece say half an inch long by an eighth of an inch in diameter from 2 to 3 hours would be sufficient, except in the case of the anterior end where a little longer time is desirable. Wax with a melting point of  $56^{\circ}$  to  $58^{\circ}$  C. is best for *Arenicola*.

The best results are obtained by staining the sections on the slide by means of iron-alum-hæmatoxylin. If this be too long a process the specimen may be stained in bulk (before cutting into sections) with borax carmine, or Mayer's acid-hæmalum. Some of the sections, especially those in which it is desired to show gland cells (*e.g.*, in the stomach\*) may, with advantage, be stained with Mayer's acid hæmalum or with Grenacher's or Delafield's hæmatoxylin.

Transverse sections are useful for the study of the general anatomy, alimentary canal, nephridia, &c.; sagittal sections of the anterior end are helpful in the

\* The histology of the alimentary canal of starved specimens should be checked by the study of sections of small pieces removed from a worm immediately after taking it from the sand.

study of the brain, nuchal organ, &c.; and horizontal sections are almost essential for the study of the nervous system, particularly the brain, otocysts, the ventral nerve cord, and the giant cells. If sections of certain organs only be desired, it is best to procure these from a worm chloroformed and opened as soon as it is taken from the sand. The organs are excised, separately preserved and sectioned as desired. The following parts may be suggested—the otocyst, various portions of the alimentary canal to show the condition of the gland cells, &c., when the gut is full of sand (of course, the sand must be rapidly washed out with sea water before preserving each part), a nephridium with gonad,\* and a heart (one *not* dilated with blood, as when imbedded the blood becomes very brittle and impossible to cut).

### Post-Larval Stages.

These are not readily obtained. Occasionally living specimens may be got from Plymouth in March and April. These are, of course, best for examination, as some of the structures described, such as the processes on the nephrostomes, can only be seen in living specimens. Preserved specimens should be lightly stained in borax carmine, and examined in cedar-wood oil, so that they may be turned over as occasion requires. For preparation of setæ cut out two or three of the posterior chætigerous segments† and warm gently in a watch glass in 5 per cent. caustic soda. Before the muscular tissue is dissolved transfer by means of a pipette to water, and later to glycerine. After

\* Whole mounts of the first nephridium, and one or two others with their gonads are very useful. Stain with carmine and mount in balsam.

† From a spirit specimen. If the specimen be in oil remove the latter by treatment with absolute alcohol before warming in caustic soda.

soaking in the latter, the specimen may be picked out on the point of a needle and placed in the middle of a melted drop of glycerine jelly on a glass slide. The cover glass (which may be warmed) should be gently pressed down on to the specimen so as to bring the setæ into one plane. Sections of post-larval stages should be thin (not more than 4 or 5 $\mu$ ) if they are to be of much use.

## ECONOMIC SECTION.

*Arenicola*, the "Lugworm," is of considerable importance to the fisherman, being largely used as a bait for flat-fish, codling, haddock, &c.\*

The worms are obtained, at low tide, by digging in the sand below the funnel-shaped opening and the coiled casting which mark the head and tail ends of the animal's burrow. Specimens obtained from near low-water mark are larger than those found higher up the beach, and the largest specimens are found in those parts of the beach which are exposed only at low spring tides. Where there is an abundance of organic matter in the sand the worms are plentiful and usually of good size, so that fishermen find it worth while to walk a moderate distance to such sands, where they can more readily obtain a supply of bait. Larger specimens usually bear exposure to the air better than small young ones and the former may be kept longer before use. At best however lugworms can only be kept for a limited period, and should be used as soon as possible after they are dug from the sand. In cold weather they may be kept for a day or two without serious detriment, but in hot weather they must be used within a few hours. They are best kept in a cool place in a quantity of moist sand sufficient to separate them from one another. If they are allowed to lie together in a heap they soon become soft and almost useless as bait. The presence of a few burst or broken specimens expedites this change, the ruptured worms lose their cœlomic fluid and usually some blood, which fluids seem to affect rapidly the

\* In addition to the acknowledgments made in the footnotes on three of the following pages, I wish to thank Mr. J. Johnstone, B.Sc., of Liverpool, Mr. H. C. Chadwick, of Port Erin, and Mr. Cyril Crossland, B.A., of St. Andrews, for kindly sending to me information on this subject.

worms with which they come into contact, so that in a few hours, especially on a hot day, the whole of the worms become soft and flabby, and soon die.

No attempts have been made, so far as the writer is aware, to preserve lugworms for subsequent use as bait. It seems doubtful, judging from the experiments which have been made on the preservation, by means of chemical substances such as boracic acid, of more hardy animals for use as bait, whether such experiments on *Arenicola* would be attended with any great success. Cold storage would probably be more successful than chemical means of preservation in the case of *Arenicola*. Undoubtedly the best plan is to use the worms as soon as possible after they are dug, but during the brief interval between digging and using them they should be placed in a moderate amount of sand containing just enough moisture to make it coherent, and kept cool.

Lugworms are abundant on most of our sandy beaches. In some places, however, *e.g.*, near Aberdeen, the force of the sea is so great that the worms cannot live in the constantly shifting sands. In other places, especially where organic matter is plentiful, *e.g.*, near some sewage outfalls, they may be found in large numbers and of good size. The organic matter may be almost entirely absent from the surface layer of sand, so that the beach may have the usual yellow colour, but it may be present in such quantities in the subjacent layer as to produce in the latter a dark-grey or even almost black colour. In this case the castings brought to the surface by the worms are usually of the same dark colour, and are conspicuous on the yellow beach. In thus passing through their bodies the sand laden with organic matter, in removing part of this during digestion and in discharging the partially cleansed sand on the surface of the beach,

where it may be subjected to the action of the air and water, these worms are playing an important part in the removal of products, which, if left to accumulate, would soon become objectionable.

The number of worms, and therefore of castings, varies considerably in different sands. Davison\* has given an account of observations made in August, 1891, on the Holy Island sands. He counted the castings in nineteen measured areas, and found that the smallest number in any one of these areas was 8.2 per square yard, while the largest number was 42 per square yard. In both cases the castings were fairly large. In other two of these nineteen areas, where the castings were very large (their average weight, when dried, being three ounces and two and a quarter ounces respectively), their numbers were 11 and 14.5 per square yards respectively.

At Musselburgh, on the Firth of Forth, there are extensive sands laid bare at low tide in which the subjacent layer contains a moderate amount of organic matter. Here worms and castings of large size are abundant near and for some distance above low-water mark. There are (January, 1904) about twelve to fifteen large castings per square yard, the average weight of (eight of) which when dried is three ounces. In the course of a few minutes seven fine worms, the mean length of which was thirteen inches, in addition to other smaller ones, were obtained here. Although these sands have been visited almost daily for a long period of years by fishermen in search of bait, there seems to be no scarcity of worms, although probably over a thousand per day are, on an average, obtained.

Near Portobello the beach near low water is gravelly in parts, and castings are scarce, but higher up the beach,

\* Geological Magazine, Vol. VIII., 1891, p. 189.

about midway between tide marks, they are very numerous. In one large area situated not far from a sewage outfall I marked out a rectangular portion six yards long and two yards wide, which was found to contain 404 castings, that is an average of 34 per square yard. These castings were small, and the worms which formed them probably did not exceed about five inches in length. The conditions described above for Musselburgh are closely reproduced on many parts of the Lancashire coast, for example on the extensive sands in the Ribble estuary.

At Piel (near Barrow-in-Furness) the main source of supply of *Arenicola* consists of an area fully half a mile square lying to the north of the old steamboat pier. The worms are most plentiful along the eastern side of this area, that is adjacent to the railway embankment. They do not extend down to low-water mark, even of neap tides, as they would then be in the tide-way (the channel to Barrow). The large area above described is beyond the reach of tidal currents. There are other beds in which *Arenicola* is plentiful, but they are not much visited by fishermen in search of bait. The surface layer consists of fine clean yellow sand to a depth of about six inches. Below this it is black in colour and strongly charged with organic matter. The number of castings in the large area described above varies from ten to twenty-four per square yard, on the best portions they average about twenty. The largest worms obtained are about seven inches in length.\*

Lugworms are usually found in the warmer parts of the year (late spring, summer and early autumn) on digging in the sand to a depth of one to two feet, but the large Laminarian forms seem to burrow more deeply and are found at a depth of nearly three feet. In frosty and

\* I thank Mr. Andrew Scott, A.L.S., of the Lancashire and Western Sea Fisheries Laboratory, Piel, for sending me this information.



in stormy weather the worms sink more deeply, and are, of course, more difficult to obtain.

Although lugworms have been so long used as bait, and fishermen have searched the same areas of sand day after day, there is usually no lack of them on most beaches. Professor M'Intosh\* suggests that they have "resisted the attacks of man probably because a sufficient stock of ripe examples and the very young are covered at all times by the tide." It is certain that plenty of old ones are covered by the tide, judging from the large specimens to be taken at low spring tides.† The habits of young ones are unknown, except those of the post-larval stages which are pelagic.

*Arenicola* produces enormous numbers of eggs, as is seen from the condition of ripe females in the spring.‡ In these the coelomic fluid contains many thousands of full-sized ova.||

Unfortunately nothing is known about either the oviposition or the early stages of development of the common lugworm. We can only suggest, by inference from the known facts of development of other species of *Arenicola*, that the eggs are laid, probably entangled in mucus, on or in the sand in shallow water, and the early

\* Resources of the Sea, p. 14.

† Reckoning only the specimens which are exposed at an ordinary low tide their number is so great that the number removed at any one time for bait really makes no appreciable difference. Take, for example, the area from which the Musselburgh fishermen dig their bait. For a distance of about a mile from East to West there is a zone from one to two hundred yards wide immediately above ordinary low-water mark in which the castings average twelve to fifteen per square yard. This area would, therefore, contain from three to four million worms, and the removal of a thousand per day would produce little effect upon the enormous number of worms accessible even at ordinary low tides. How far seawards the worms extend below ordinary low-water mark it is impossible to say, but no doubt there are here very substantial reserves.

‡ And also, in some localities, in late summer or autumn.

There were about 80,000 ova (a rough estimate made by the dilution method) in the coelomic fluid of one specimen examined.

stages of development are passed there. The larvæ grow until the full number of adult body segments has been produced and then enter upon the pelagic post-larval stage. This is the earliest known stage of development of the common lugworm. The young worms do not settle down to their littoral habitat and characteristic mode of life until they have attained a length of about 5 to 7 mm. Young specimens 17 mm. long have been found in the sand in June. Although it is difficult to estimate their age, it may be suggested that these specimens 17 mm. long and others up to 44 mm. long taken near the end of June were probably produced from ova laid in the preceding February or March. If that be the case, then worms five or six inches long are probably about a year old. Some of the large deeply-pigmented specimens obtainable at low spring tides are certainly much older, but on this point no information is available. It may be taken that the smaller specimens of *Arenicola* used as bait are at least a year old. Considering the abundance of *Arenicola* on many beaches, it is astonishing that we know so little of its life history, and that so few post-larval stages have been taken in the tow nets.

The present state of our knowledge does not permit one to make any suggestions with regard to the possible cultivation of lugworms in any given area in which they are wanting. The writer has made several attempts to fertilise artificially the ova of the common lugworm removed from a ripe female, by adding spermatozoa obtained from a mature male, but without success, although both ova and spermatozoa appeared, when examined microscopically, to be quite ripe and favourable to the experiment. Others have also experienced these failures when dealing with this and other species of *Arenicola*.\* Evidently, therefore, it is more difficult to

\* I eventually succeeded in fertilising artificially the eggs of *A. claparedii*. For an account of the early stages of development of this species, see under development.

bring about artificial fertilisation in *Arenicola* than, for example, in many Echinoderms, and this fact would prove a serious obstacle in the endeavour to cultivate *Arenicola* with the idea of re-stocking exhausted sands. This question is, however, not likely to become of practical importance as, judging from reports from various parts of the coast, the supply of lugworms is quite equal to, and even more than, the demand, except, of course, in certain restricted areas where conditions are unfavourable for their life and growth.

As mentioned above, the sands around Aberdeen form one of these unfavourable areas, so that lugworms are either absent or scarce. In consequence they are imported from the Moray Firth. The worms are gathered chiefly near Campbeltown, near Fort George, Inverness-shire, and sent by rail,\* a distance of about 100 miles, the journey occupying at least five hours, to Aberdeen. The worms cost ten shillings per stone, and railway charges, &c., amount to 1s. 2d. per stone.\* This quantity† would bait four small lines.\* A baiting of mussels for the same lines would cost about 3s. 6d., but as a rule the lugworms would catch three or four times as many fish.\* It is almost impossible, in warm weather, to transport lugworms over such a long distance and to deliver them in good condition at their destination. This probably accounts for the fact that they are used in Aberdeen only in the colder part of the year, from December to April. No doubt, the comparatively high price restricts their use for bait during the colder months. This is the only

\* For this information I desire to thank the Secretary of the Fishery Board for Scotland and the principal Fishery Officer of the Aberdeen District.

† Taking fourteen average lugworms, such as would be used for bait, I find that their mean weight is rather over half an ounce each. There would be about four hundred such worms in a stone (14 lbs.)

instance known to the writer, of lugworms being imported and sold in quantities in the market.

Although lugworms are fairly abundant they are apparently not as extensively used for bait as formerly. This is probably because they are troublesome to obtain, and may be kept only for a short time. These disadvantages account for the fact that in some cases mussels are used in preference to *Arenicola*, which was formerly employed, as these molluscs are readily obtainable in quantity, and may be kept for a reasonable period with very little trouble. In other cases Nereids are used, as these are also more hardy than *Arenicola*.

On the coast of Durham and Northumberland, for example, *Arenicola* is not nearly so extensively used as formerly. Nereids are, when procurable, used in preference to *Arenicola*, the order of preference being, for "hard bottom"—mussel, *Nereis*, *Arenicola*; for "soft bottom"—*Nereis*, *Arenicola*, mussel. The special fishing for soles is not a feature of the present-day fishing in this district as it was in the past. Possibly this accounts for the fact that worms are, on the whole, less used, and that the principal baits at present employed here are mussels and limpets.\*

Fullarton† has made experiments which give interesting information regarding the preference of fishes for the four baits most commonly used on the Scottish coast. The experiments were carried out in various parts of the Firth of Forth, and in various depths of water. The line used had 1,200 hooks, provided with different baits in batches of forty hooks, so that a large variety of baits could be used under identical conditions. The fish

\* My thanks are due to Mr. Alexander Meek, B.Sc., of the Durham College of Science, Newcastle-on-Tyne, for this information.

† Seventh Annual Report of the Fishery Board for Scotland, p. 352.

obtained by each kind of bait were carefully noted. The following table is an abstract of the results obtained at fourteen stations where lugworms, clams (*Pecten opercularis*), mussels and limpets were used together. The experiments extended from August 30th to December 26th, 1888.

BAIT.	No. OF HOOKS.	FISH CAUGHT.									
		HADDOCK		COD.		WHITING		DAB.		TOTAL.	
		No.	%	No.	%	No.	%	No.	%	No.	%
Lugworm.....	3500	130	3·7	39	1·1	67	1·9	54*	1·6	290	8·3
Clam.....	3960	182	4·6	44	1·1	50	1·3	53	1·3	329	8·3
Mussel.....	4540	266	5·8	75	1·6	94	2·1	42	·9	477	10·5
Limpet.....	3320	92	2·7	15	·5	8	·2	28	·8	143	4·3

\* Including four plaice.

From the table it is seen that *Arenicola* is the most successful bait for flat-fish, that it is about equal to mussel for attracting whiting, but mussel is superior for cod and haddock. Comparing *Arenicola* and *Pecten* it is seen that the former is rather more effective as a bait for flat-fish and whiting, but the latter for haddocks, while they are equal in their catch of cod. These two baits are practically equal in value. Limpets are inferior to each of the other three baits. It may be stated here that flat-fish are not plentiful in the Firth of Forth. In the above described experiments they form only one-seventh of the catch while haddocks form more than half.

Haddocks form the majority of fish taken by the line fishermen of Musselburgh, and other places on the Firth of Forth. These men, who work over the same area as that in which the above-named experiments were con-

ducted, prefer lugworms as a bait for haddocks, in fact, the results shown in the above table are not in agreement with the general opinions of fishermen at several points of the Scottish coast. Both in Aberdeen district and in the Firth of Forth the order of preference seems to be lugworms, clams, mussels. On the Lancashire coast and in other parts of England, lugworms are preferred to mussels.

Lugworms are used for baiting both long and short lines, the custom varying in different localities. On the Cheshire coast at New Brighton, at Piel, Lancashire, and on the coast of Northumberland, short lines are usually employed, those of the New Brighton fishermen bearing only about twenty hooks. In the Aberdeen district the lines carry about three hundred hooks, and the Musselburgh fishermen employ long lines bearing as many as twelve to sixteen hundred hooks baited with lugworms.

The lugworm has a faint odour and an ethereal extract proved to be attractive to certain fish, *e.g.*, turbot and rockling.\* It is doubtful whether this scent is sufficiently strong to play any part in attracting the fish which are usually caught with lugworm bait, as most of these fish—cod, whiting, plaice, flounder and dab—appear to find the bait by the sense of sight.†

\* F. HUGHES.—Journal Marine Biological Association, Vol. 2, p. 92.

† W. BATESON.—Journal Marine Biological Association, Vol. 1, p. 241.

## DESCRIPTION OF PLATES.

## Reference Letters.

- A.B.S.* = Achætopus body segment.  
*Ac.* = Aciculum.  
*Aer.* = Aerosome or apical body of spermatozoon.  
*An.* = Anus.  
*Ant.L.* = Anterior lobe of brain.  
*Au.* = "Auricle."  
*B.* = Blastophore (residual mass of protoplasm).  
*Bl.V.* = Blood vessel.  
*Br.* = Gill.  
*Br.Aff.* } = Afferent and efferent  
*Br.Eff.* }      branchial vessels.  
*Br. and N.Aff.* = Afferent vessel to gill and nephridium.  
*B.Sh.* = Sheath formed of buccal muscles.  
*Bucc.M.* = Buccal mass.  
*Bucc.Pap.* = Papillæ of buccal mass.  
*Caud.Sep.* = Caudal septum.  
*Ch.Seg.* = Chætigerous segment.  
*Chl.Gr.* = Chlorogogen granules.  
*Chl.Tiss.* = Chlorogogenous tissue.  
*Cel.* = Cœlom.  
*Cel.Epith.* = Cœlomic epithelium.  
*Conn.Tiss.* = Connective tissue.  
*Cut.* = Cuticle.  
*Diat.* = Diatom (as otolith).  
*D.L.V.* = Dorsal longitudinal vessel.  
*D.Mes.* = Dorsal mesentery.  
*Dphm.* = Diaphragm.  
*Dphm.P.* = Pouch of first diaphragm.  
*D.V.* = Dorsal vessel.  
*Endth.N.* = Nucleus of endothelium.  
*Ep.* = Epidermis.  
*Exc.Gr.* = Excretory granules.  
*Ext.Op.Ot.* = External opening of oocyst.  
*Gang.C.* = Ganglion cell.  
*Gast.Lat.* = Lateral gastric vessel.  
*G.C.* = Giant nerve cell.  
*G.F.* = Giant nerve fibre.  
*Gl.C.* = Gland cell.  
*Gon.* = Gonad.  
*Gon.V.* = Gonidial vessel.  
*Ht.B.* = Heart body.  
*Int.V.* = Intestinal vessel.  
*M.Circ.* = Circular muscles.  
*Met.Gr.* = Metastomial groove.  
*Mid.Com.* = Middle commissure of brain.  
*M.Long.* = Longitudinal muscles.  
*Mo.* = Mouth.  
*M.Obl.* = Oblique muscles.  
*M.P.* = Middle piece of spermatozoon.  
*Musc.* = Muscles.  
*N.* = Nucleus.  
*N.Aff.* = Nephridial afferent vessel.  
*N.C.* = Nerve cord.  
*N.Ep.* = Nerve to epidermis.  
*Nfb.* = Neurofibrillæ.  
*Ngl.* = Neuroglia.  
*Nlm.* = Neurilemma.  
*N.L.V.* = Nephridial longitudinal vessel.  
*Nm.* = Neuropodium.

- Nm.Ch.* = Neuropodial chætæ.  
*Nm.Cirr.* = Neuropodial cirrus.  
*N.O.* = External opening of nephridium.  
*Not.* = Notopodium.  
*Not.Cirr.* = Notopodial cirrus.  
*Not.Pr.* = Protractors of *Not.S.*  
*Not.Retr.* = Retractors of *Not.S.*  
*Not.S.* = Notopodial setæ.  
*Nph.F.* = Funnel of nephridium.  
*Nph.F.D.* = Dorsal lip of do.  
*Nph.F.V.* = Ventral lip of do.  
*Npile.* = Neuropile.  
*N.S.* = Nervous sheath of otocyst.  
*Nuc.Gr.* = Nuchal groove.  
*Nuc.Retr.* = Retractors of nuchal organ (and of prostomium).  
*N.V.* = Neural vessel.  
*Æ.* = Œsophagus.  
*Æ.Conn.* = Œsophageal connective.  
*Æ.Gl.* = Œsophageal gland.  
*Æ.Gl.V.* = Blood vessel of do.  
*Æ.Lat.* = Lateral Œsophageal vessel.  
*Ot.* = Otocyst.  
*Otl.* = Otolith.  
*Ot.N.* = Nerve to otocyst.  
*Ot.T.* = Otocyst tube to exterior.
- Par.V.* = Parietal vessel.  
*P.B.* = Polar bodies.  
*Per.* = Peristomium.  
*Per.Cirr.* = Peristomial cirrus.  
*Per.C.* = Peritoneal cell.  
*Ph.* = Pharynx.  
*Pigm.* = Pigment.  
*Post.L.* = Posterior lobe of brain.  
*Proc.* = Process of giant cell.  
*Pr.Pr.* = Protoplasmic processes of nephrostome.  
*Prost.* = Prostomium.  
*Prost.Epith.* = Prostomial epithelium.  
*S.C.* = Sense cell.  
*S.H.* = Sense hairs.  
*Sp.* = Spicule (as otolith).  
*Sp.N.* = Spinal nerve.  
*Stom.* = Stomodæum.  
*S.V.* = Subintestinal vessel.  
*T.* = Tail of spermatozoon.  
*Tent.* = Tentacle (prostomial).  
*U.L.* = "Upper lip" (part of peristomium).  
*V.* = Ventricle of heart.  
*Ves.* = Vesicle of nephridium.  
*V.Gr.* = Ventral ciliated groove.  
*V.M.* = Vitelline membrane.  
*V.V.* = Ventral vessel.

All the figures are drawn from *Arenicola marina*, except figs. 7, 8, 9, which are of *Nereis pelagica*, fig. 53 of *A. grubii*, and figs. 69-79 of *A. claparedii*.

## PLATE I.

Fig. 1. A Laminarian specimen, 190 mm. long, from right side. Proboscis fully protruded; the buccal portion first protruded during eversion is provided with numerous backwardly directed papillæ, the following pharyngeal



portion (smooth in the figure) is covered with closely set rounded papillæ too small to be shown. Prostomium, segmentation, annulation, parapodia, gills and nephridiopores also shown. Natural size.

Fig. 2. Anterior end of a littoral specimen about 60 mm. long. Ventral aspect. In front the mouth is seen surrounded by a series of papillæ. Behind the "upper lip" the three lobes of the prostomium are seen lying retracted in the nuchal organ. Note the metastomial grooves uniting in third annulus at anterior end of groove over ventral nerve cord. Segmentation and annulation in relation to the internal septa also shown. Compare from second to third chætigerous annuli with fig. 1.  $\times 6$ .

Fig. 3. Portion of "proboscis" from fig. 1, to show the buccal papillæ, which gradually merge into the epidermal papillæ of the peristomium.  $\times 9$ .

Fig. 4. Chitinoid caps from the tips of the buccal papillæ.  $\times 24$ .

Figs. 5, 6. Dorsal and ventral views of the anterior end of a large littoral specimen, 340 mm. long. The "proboscis" is half extruded. cf. figs. 1, 2.

Fig. 5. Dorsal aspect, showing the trilobed prostomium fully extended, the origin of the two metastomial grooves, the apertures of the otocysts, the sculpturing of the skin, &c. Each of the first four annuli is subdivided into two. The first two annuli form the peristomium, the third and fourth represent a body segment from which the setæ have disappeared. The first chætigerous annulus and the following annulus represent the first chætigerous segment.  $\times 3$ .

Fig. 6. Ventral aspect, showing metastomial grooves, groove over ventral nerve cord, and very small neuropodia of first chætigerous segment.  $\times 3$ .

Figs. 7, 8. *Nereis pelagica*. Dorsal and ventral

aspects of the anterior end for comparison with the two preceding figures of *Arenicola*.

Fig. 7. Dorsal aspect. Prostomium with pair of tentacles, pair of much stouter palps and four eyes, the peristomium bearing on each side four cirri (the basal portions only are shown on the left), the first and second body segments with their parapodia.  $\times 6$ .

Fig. 8. Ventral aspect. Note prostomium with tentacles and palps, peristomium bearing mouth on anterior margin, the first and second body segments with their parapodia. The structure at the base of the palps is part of the peristomium forming the upper lip.  $\times 6$ .

Fig. 9. *Nereis pelagica*. A parapodium, from the 74th chaetigerous segment, divided distally into bilobed notopodium and neuropodium, each of which bears a sensory cirrus, an aciculum and a bundle of setae (jointed) situated in a setal sac, the lips of which are well seen in the neuropodium.  $\times 20$ .

Fig. 10. The seventh notopodium and the small and simple first right gill of the Laminarian specimen shown in fig. 1. Compare with fig. 19.  $\times 6$ .

## PLATE II.

Fig. 11. Three notopodial setae from a post-larval specimen 4.35 mm. long. Most of the setae are of the type shown in fig. 11C ( $\times 800$ ), but in each notopodium there is one seta of the kind shown in figs. 11A, 11B. ( $\times 1,000$ .)

Fig. 12. Distal fourth of a seta (7.5 mm. long) from a Laminarian specimen 250 mm. long.  $\times 80$ .

Fig. 13. A portion of same seta (from the region marked +) more highly magnified. On the left the broad lamina, with its finely dentate margin, is shown.  $\times 800$ .

Fig. 14. A portion (0.2 mm. from the tip) of a seta (2.5 mm. long) from a littoral specimen 125 mm. long. On the left the lamina, with its dentate margin, is shown.  $\times 800$ .

Fig. 15. Two crotchets from neuropodia of a post-larval specimen 5.1 mm. long. The teeth on the sides of the rostrum are indicated. The dotted line indicates the level of the epidermis.  $\times 1,000$ .

Fig. 16. Neuropodial crotchet from a young littoral specimen 17 mm. long.  $\times 500$ .

Fig. 17. Neuropodial crotchet from a littoral specimen 125 mm. long.  $\times 150$ .

Fig. 18. Ventral portion of neuropodium of Laminarian specimen 250 mm. long. On the left (which is ventral) the various stages of formation of the crotchets are seen. The crotchet on the right is fully formed. *Nm* shows outline of the neuropodial sac.  $\times 75$ .

On comparing the four preceding figures there is seen to be, as the worm grows, an increase in the length of the crotchets, a decrease in the size of the teeth behind the rostrum, and also a gradual change in the inclination of the rostrum to the shaft, the angle increasing with the size of the specimen.

Fig. 19. Right aspect of ninth and tenth chaetigerous segments of Laminarian specimen (190 mm. long). The fourth gill has eleven main stems united at their bases by a membrane. Five of these, and many lateral branches, have been cut away to show the remainder. The ventral stem alone, in which the lateral branches have the simplest form, is shown entire. Lateral branches somewhat simplified. Fig. 21 shows one in detail. The third gill has been almost entirely cut away. The capillary setae are seen projecting beyond the lips of the notopodial setal sac. Each neuropodium resembles a

pair of tumid lips between which is the narrow opening of the neuropodial setal sac which contains the crotchets. The external opening of the sixth (last) nephridium is just behind the dorsal end of the ninth neuropodium.  $\times 6$ .

Fig. 20. Ventral stem of gill of younger Laminarian worm. This stem is about half the length of that shown in the preceding figure, and its lateral branches are still comparatively simple.  $\times 20$ .

Fig. 21. Lateral branch, from one of the dorsal stems of the gill shown in fig. 19, bearing twenty-two gill filaments.  $\times 20$ .

Fig. 22. Fourth right gill of a littoral specimen about 110 mm. long. The gill consisted of ten main stems, eight of which have been cut away. The blood-vessels are seen traversing the basal membrane. The lateral twigs are fewer in number and not so richly branched as in Laminarian specimens.  $\times 20$ .

### PLATE III.

Fig. 23. Dissection of a large Laminarian specimen (250 mm. long) cut open along the mid-dorsal line to show the anatomy. The alimentary canal is pushed over to the left side, and most of the tail has been cut out. The proboscis is about half extruded. The dorsal portion of each left gill is seen. The gastric plexus is shown full of blood, and the heart about half expanded. The funnel only of the first left nephridium is seen, the rest of the organ being covered by the œsophagus. The funnel of the third left nephridium is hidden by one of the œsophageal glands. The funnel of the fifth right nephridium has pushed its way through the interval between two of the oblique muscles. The blood-vessels of the sixth right nephridium have been pushed forward one annulus,

so as to show the afferent branch to the nephridium. The afferent vessel of the fourth nephridium is accompanied by a solid cord of connective tissue.  $\times 2$ .

#### PLATE IV.

Fig. 24. Dissection of left side of fourth and fifth chaetigerous segments of a Laminarian specimen, showing second nephridium with blood-vessels, gonad, the muscles and the ventral nerve cord. Three of the oblique muscles are interrupted to show the nephridium.  $\times 4$ .

Fig. 25. Second right nephridium of a young littoral specimen 44 mm. long in dorsal aspect showing especially the capillary network on the funnel and excretory part, the processes on the dorsal lip, the position of the external opening (dotted), and the gonad upon the gonidial vessel. The edge of the ventral lip of the funnel is seen through the dorsal lip.  $\times 65$ .

Fig. 26. Fifth right nephridium of an adult male showing the vesicle distended with spermatozoa in dorsal aspect. The funnel of this nephridium was widely open, as is usual at the breeding season.  $\times 4$ .

Fig. 27. First left nephridium of a medium-sized worm, in dorsal aspect. This nephridium is much more slender than any of the others. The ventral lip of the funnel is shown (dotted) through the dorsal one. The gonidial vessel bears a number of blindly ending vascular processes which are covered with chlorogogen cells.  $\times 8$ .

Fig. 28. The anterior portion of the funnel of the fourth nephridium in ventral aspect. The dorsal lip bears numerous, almost semi-circular, lobed ciliated processes, each of which contains a blind diverticulum of the large blood-vessel which traverses this lip. On both lips there is a network of blood-vessels, but they are shown only on

the ventral lip. Some of these vessels have blind dilated endings. The ventral lip is simple and slightly everted.  $\times 20$ .

Fig. 29. Two of the processes of the dorsal lip, from the second nephridium of a Laminarian specimen 190 mm. long, to show the multi-lobate edge.  $\times 20$ .

Fig. 30. Section of part of the excretory portion of the nephridium, from a young specimen 65 mm. long, showing two excretory cells with reticulate protoplasm and excretory granules in the distal portion. The granules become more numerous in older specimens, and are apparently extruded from the distal portion of the cell into the cavity of the nephridium. When this has recently occurred the distal part of the cell is very clear and free from granules. Each cell bears one or two long flagella. Below the cells is a thin film of connective tissue (represented by a line) which is thicker in older specimens, and outside this the thin layer of cœlomic epithelium, which covers the blood-vessels.  $\times 650$ .

Fig. 31.—The last nephridium of a living post-larval specimen 465 mm. long, in optical section. The larger (right in the figure) lip of the nephrostome bears seven protoplasmic processes (three are seen in the figure), bearing long cilia directed towards the opening. The middle of the nephridium is slightly dilated and its walls contain excretory granules. The whole nephridium is ciliated.  $\times 150$ .

Fig. 32. The anterior end of the same nephridium drawn later. Optical section, showing the peculiar ciliated protoplasmic processes of the nephrostome, the cilia of the nephridial tube, and the concretions in the walls of its middle position.  $\times 600$ .

Fig. 33. A process from the heart body of a specimen 250 mm. long (see figs. 39 and 40), to show the

endothelium, the muscle layer and the core of loosely arranged peritoneal cells. In this and in several other processes in the same heart there is a large collection of chlorogogenous granules in many of the peritoneal cells. These are shown in red in the figure, but they are naturally of a yellow colour. The granules are often aggregated into small heaps, sometimes lying in a vacuole. The outlines of the cells which are loaded with chlorogogenous granules are difficult to distinguish. The protoplasm of many of the peritoneal cells is vacuolated and of others is granular, but the granules are very minute and of a different nature from the chlorogogen granules.  $\times 500$ .

Fig. 34. Transverse section of a young specimen, showing the nuchal organ, below which are the two posterior brain lobes, an otocyst with its contained otoliths and the tube leading to the exterior, the two œsophageal connectives, the buccal mass with its sheath of muscles and its retractors, the retractors of the prostomium and nuchal organ, the muscles of the body wall, blood-vessels, &c. The upper portion of the brain lobes and the outer portion of each œsophageal connective contains nerve cells (diagrammatically shown in the figure). The fibrous portion of these structures is dotted.  $\times 65$ .

Fig. 35. Transverse section of a young specimen at the level of the openings of the glands into the œsophagus. The partitions which partially subdivide the gland, each formed by a fold of the epithelium lining the gland and enclosing a vascular sinus, are seen. Also the muscles of the body wall and their blood-vessels; the oblique muscles; the nerve cord; the vesicle and external opening of the second right nephridium; portions of two notopodial setal sacs and the blood-vessels connected with the alimentary canal. The ventral vessel and the two branches near

it are covered with chlorogogen cells. (See fig. 43.) The nerve cord is divisible into a dorsal fibrous mass, in the mid-dorsal line of which one giant-fibre is seen, and a ventral portion composed largely of nerve cells the nuclei of which are indicated.  $\times 38$ .

Fig. 36. Transverse section of the same specimen in the region of the last nephridium. To the right of the two vertical lines was drawn from a section passing through the anterior part of the ninth chaetigerous segment, while the portion on the left is from a section passing through the tenth parapodium, and showing the notopodium with its setal sac and protractor muscles and the whole length of the neuropodium. We see also a portion of a gill, with its afferent and efferent vessels, the funnel (opening into the coelom) and excretory part of the nephridium, the stomach with its ventral groove and blood sinuses, the subintestinal and the lateral gastric, the dorsal and ventral vessels, the latter surrounded by chlorogogen cells (fig. 43), the muscles of the body wall and their blood-vessels; two oblique muscles; the nerve cord, the dorsal fibrous mass of which contains two giant-fibres, and the ventral portion with nerve cells, the nuclei of which are shown.  $\times 38$ .

#### PLATE V.

Fig. 37. Longitudinal section of the heart of a young *Arenicola* 65 mm. long to show the heart body in an early stage of its formation. On the posterior (right) side and also on the antero-external side (left of the figure), the wall of the heart is being invaginated at several points, thus giving rise to the heart body.  $\times 100$ .

Fig. 38. A portion of the wall of the ventricle from the point marked + in the preceding figure, and composed



of three layers, an endothelium internally, a muscle layer and a peritoneal covering. The muscle layer is only feebly developed in worms of this size, it becomes much more highly developed in older specimens (see figs. 33, 40). The protoplasm of the peritoneal cells is moderately homogeneous, in older stages it becomes either vacuolated or granular and in many cases contains chlorogogen granules. The wall of the heart is invaginated forming a club-shaped process which contains a central cavity. The excessively thin endothelium is partly obscured by a mass of blood which closely invests the process. (Cf. figs. 33, 40).  $\times 600$ .

Fig. 39. Longitudinal section of the much thicker, muscular ventricle of a large specimen 250 mm. long. The invaginations of the wall are very numerous and complex, filling up the greater part of the cavity and forming the well-marked heart body.  $\times 45$ .

Fig. 40. The tip of one of the invaginated processes of the heart-body, from the previous figure, showing the endothelium, the muscle fibres and the core of loosely arranged peritoneal cells, some of which are vacuolated and some granular.  $\times 260$ .

Fig. 41. Three blood corpuscles. The one on the right from an old specimen (250 mm. long), the two others from a young specimen (65 mm. long).  $\times 1000$ .

Fig. 42. The terminal portion of one of the small caecal branches of the ventral vessel covered with irregular dark brown chlorogogen cells. The latter do not extend to the tip of the vessel.  $\times 150$ .

Fig. 43. Section of the covering of the ventral vessel, the columnar cells loaded with yellowish chlorogogen granules of various sizes.  $\times 500$ .

Fig. 44. Cœlomic cells. In the upper part of the figure a typical fusiform cell, on the right four amœboid cells, three of which contain chlorogogen granules; on the

left three rounded cells, one of which is vacuolated, the other two contain chlorogogen granules. These may be amœboid cells which had temporarily withdrawn their pseudopodia.  $\times 1000$ .

Fig. 45. A portion of the third diaphragm, from a stained preparation, to show the numerous oval apertures by which the septum is perforated. The diaphragm is covered on both faces by a flattened endothelium between which is a thin layer of connective tissue and inter-crossing muscle fibres. The endothelial nuclei of one face only are shown.  $\times 300$ .

#### PLATE VI.

Fig. 46. Dissection of anterior end of large specimen, with pharynx and the first diaphragm removed, showing anterior and posterior lobes of brain; middle cerebral region of each side connected by broad band of nervous tissue; nerve to otocyst arising from œsophageal connective; muscle strands from otocyst to body wall; anterior portion of ventral nerve cord; first notopodial sacs and muscles: the longitudinal muscles and the buccal sheath.  $\times 6$ .

Fig. 47. Horizontal section of prostomium of a specimen 60 mm. long. The anterior and dorsal portions of anterior lobes of the brain are formed largely of pyriform ganglion cells shewn diagrammatically in the figure. Three cœlomic spaces, lined by cœlomic epithelium and containing blood-vessels, are seen. On the anterior left side of brain is an eye slightly exaggerated. The posterior lobes of the brain underlie the nuchal organ, most of their nerve cells are small and their nuclei only are shown. The middle region of the brain, consisting largely of neuropile (finely punctate), shows a transverse band of fibres connecting the right and left halves. Attached to

the posterior end are the retractor muscles of the prostomium and nuchal organ.  $\times 70$ .

Fig. 48. Part of a similar section showing an eye imbedded in the brain. In the upper part cuticle and prostomial epithelium are shown, on the right a gland cell and on the left a bundle of slender sense cells. Beneath the epithelium is the anterior brain lobe and an eye, which consists of a cup-shaped mass of reddish-brown pigment spherules grasping a spherical lens. In the right of the pigment mass is a nucleus and small amount of protoplasm, probably the remains of the cell in which the eye has been formed.  $\times 550$ .

Fig. 49. Section of otocyst and tube leading to exterior (from specimen 60 mm. long). The otocyst is lined by epithelium continuous with that of the tube and the epidermis. In the outer portions of the tube there are fusiform sense cells, but in the otocyst itself the sense cells are distinguished only by the presence of neurofibrillæ (not shown in the figure). The proximal (and narrowest) portion of the tube is ciliated. Below the epithelium of the otocyst is the nerve sheath, connected with the stout nerve from the œsophageal connective. This nerve, along the course of which are numerous ganglion cells, also sends off branches to the skin. The otoliths are irregular bodies, chiefly quartz grains.  $\times 210$ .

Fig. 50. Camera-drawing of the cuticle lining the otocyst of a specimen 250 mm. long. This is thicker than in younger specimens. The tube is almost blocked and the otoliths are now assuming a rounded outline, due to the deposition of layers of secretion.  $\times 210$ .

Fig. 51. A small portion of epithelium from anterior end of stomach, showing two large goblet cells. The secretion of these cells has been coagulated on preservation and now appears as a reticulum.  $\times 210$ .

## PLATE VII.

Fig. 52. Plan of the nerve cord of a young specimen about 25 mm. long to show the position of the giant cells. Transverse lines mark the posterior limits of the segment, the numbers of some of which are given at the side. The cells are drawn too large ( $\times 40$ ) in proportion to the width of the cord ( $\times 25$ ), and the latter is about twice too broad in proportion to its length ( $\times 12$ ). The first giant cell is situated in the achætopous segment which lies behind the peristomium. There was no giant cell in the sixteenth chætigerous segment of this specimen. Giant cells are not distinguishable in the small anterior tail segments, but a few are present in the more posterior segments. One is shown in the twelfth caudal segment.

Fig. 53. Transverse section of the nerve cord of *Arenicola grubii*. The section shows the dorsal fibrous part of the cord, in the mid-dorsal region of which are three giant fibres, the nuclei in the sheaths of which are shown. In the ventral portion of the section note the numerous small nerve cells, the nuclei of which are shown on the right and left; a bundle of pyriform ganglion cells whose processes are directed into the fibrous part of the cord; and a giant nerve cell. The protoplasm of the latter is reticulate and its nucleus vesicular. The cell gives off a large process which, after sending branches into the fibrous part of the cord, enters the right lateral giant fibre at the point where there is a transverse connection between the two lateral giant fibres. The neurofibrillæ of the process are shown. This drawing of the giant cell and its process was obtained by superposing camera drawings of the four consecutive sections in which these structures occur.  $\times 210$ .

Fig. 54. Transverse section of nerve cord of *Areni-*

*cola marina*, showing a giant cell and its process in which the neurofibrillæ are seen; also sheath of neurilemma enclosing the cord; fibrous part of cord partially subdivided by a median sheet of neuroglia: two giant fibres: nuclei of small nerve cells situated in ventral part of cord; root of a spinal nerve on the right; epidermis; circular muscles; longitudinal muscle bands, the fibrils and nuclei of which are seen; two oblique muscles and the cœlomic epithelium.  $\times 300$ .

Fig. 55. Horizontal section of fourth chætigerous segment of a specimen 45 mm. long passing through the ventral nerve cord, to show the giant-cell and the spinal nerves. A pair of nerves is given off from the cord opposite each inter-annular groove and in addition a nerve or a small bundle of nerves to the chætigerous annulus.  $\times 15$ .

Fig. 56. A post-larval specimen 3.9 mm. long taken in the tow-net near Lytham. The animal is seen from the left side, and shows the eyes on the prostomium, the segmentation and annulation, the parapodia, the two kinds of notopodial setæ, the gills, and the otocyst.  $\times 50$ .

Fig. 57. Dorsal view of anterior end of a post-larval specimen 5.4 long, to show the prostomium, the nuchal organ, the peristomium in which lie the otocysts (outline dotted), the achætous body segment and the first chætigerous segment. The peristomium and the achætous segment are divided into two, thus forming four annuli, corresponding to the subdivision of this region in adults. Cf. fig. 5.  $\times 100$ .

Fig. 58. Ventral view of the corresponding portion of a post-larval specimen 3.5 mm. long. The mouth is situated on the anterior edge of the peristomium and the two œsophageal connectives are seen uniting in the third annulus. Cf. fig. 6.  $\times 100$ .

Fig. 59. The prostomium and nuchal organ drawn

from a living post-larval specimen 4.6 mm. long. The prostomium bears six eyes, two of which, those first formed in the larva, are larger than the others. Scattered over the surface of the prostomium are groups of fine sense hairs. Only those on the margin are shown in the figure.  $\times 100$ .

### PLATE VIII.

Figs. 60—65. The formation of spermatozoa.

Fig. 60. One of the earliest stages found in the coelomic fluid consisting of sixteen cells (spermatogonia).  $\times 500$ .

Fig. 61. Optical section of a similar stage showing eight of the spermatogonia arranged round a vesicular residual mass of protoplasm—the blastophore.  $\times 500$ .

Figs. 62, 63. Later stages produced by continued division of the spermatogonia.  $\times 500$ .

Fig. 64. Two cells—spermatids—from a stage much later than the preceding. At one end (the left) of each is a small mass of protoplasm which later forms the apical body of the sperm, following this is the nucleus and a clear substance from which the middle piece of the sperm is derived, while on the right the protoplasm is being drawn out to form the tail.  $\times 2000$ .

Fig. 65. A discoidal mass of almost ripe spermatozoa.  $\times 500$ .

Fig. 66. A ripe spermatozoon. The head is divisible into an apical body, the nucleus and the middle piece, which is notched behind to receive the tail.  $\times 3000$ .

Fig. 67. A ripe egg from the body cavity. The flat face is shown. The nucleus is large and vesicular, the yolk granules in the protoplasm are rather more numerous around the nucleus, the vitelline membrane is thin ( $1\mu$ )  $\times 100$ .

Fig. 68. Side view of a ripe egg.  $\times 100$ .

Figs. 69—79. Segmentation of the egg and larval stages of *Arenicola clapedii*.

Fig. 69. Two-cell stage. The two polar bodies are shown. Four and a half hours after addition of the spermatozoa to the sea water containing the eggs.  $\times 210$ .

Fig. 70. Four-cell stage, seen from the anterior pole.  $\times 210$ .

Fig. 71. Eight-cell stage, seen from the anterior pole. The first four ectomeres (1*a*, 1*b*, 1*c*, 1*d*) have been cut off from the lower cells.  $\times 210$ .

Fig. 72. Sixteen-cell stage, seen from the anterior pole. Each of the first ectomeres has divided into two and the second quartette of ectomeres (2*a*, 2*b*, 2*c*, 2*d*) has been cut off from the lower cells. Of the latter only one (2*A*) is shown in the figure. 2*d* is the largest cell in the egg and forms a conspicuous landmark.  $\times 210$ .

The cleavages shown in the three preceding figures occupied about two hours

Fig. 73. The larva (blastula) at the end of 24 hours, seen from the posterior pole. The four quartettes have been formed, the first three of which are ectomeres. One of the fourth quartette (4*d*) is the mesoblast cell and it has already sunk into the segmentation cavity. The three other members of the fourth quartette (4*a*, 4*b*, 4*c*) and the four lower cells (4*A*, 4*B*, 4*C*, 4*D*) are endoderm cells. In this specimen each of these seven primary endoderm cells, except 4*c*, has again divided, so that there are thirteen endoderm cells at the posterior pole of the larva.  $\times 210$ .

Fig. 74. Larva about six hours later than the preceding. Ventral aspect. The pre-oral band of cilia is shown just in front of the stomodæal invagination. The larva is at this stage constantly rotating in the vitelline

membrane. The latter shows a thin area through which later the larva forces its way.  $\times 210$ .

Fig. 75. Larva three days (72 hours) after fertilisation of the egg. Dorsal aspect. Both ciliated bands are now present, and there are two eyes on the prostomium. Ten minutes after the drawing was made the larva forced its way through the thin area in the upper portion of the vitelline membrane.  $\times 210$ .

Fig. 76. Larva one day after hatching. Dorsal aspect. Two short spade-shaped setæ are present. The anterior and posterior ciliated bands and the eyes are also shown. The specimen was slightly compressed by the cover-glass and therefore appears a little broader than it should be.  $\times 210$ .

Fig. 77. Larva twelve days after hatching. Seen from the right side. The prostomium bears two eyes and several small groups of sense hairs. Setæ are present in three segments, in the first two of which both notopodial and neuropodial setæ may be seen. The notopodial setæ are of two kinds. The alimentary canal is complete from mouth to anus, the middle portion is dilated with yolk. The cœlom is obvious post-orally. The anterior girdle of cilia has entirely, and the posterior one almost, disappeared. The anterior portion of the ventral band of cilia still remains.  $\times 210$ .

Fig. 78. A (notopodial) seta from the larva shown in fig. 76.  $\times 1000$ .

Fig. 79. Two setæ from the larva shown in fig. 77.

Fig. 79A. One of the neuropodial crotchets. The dotted line indicates the level of the epidermis.  $\times 1000$ .

Fig. 79B. The distal portion of one of the notopodial setæ. The other seta in the same notopodium is similar to the one shown in fig. 78.  $\times 1000$ .



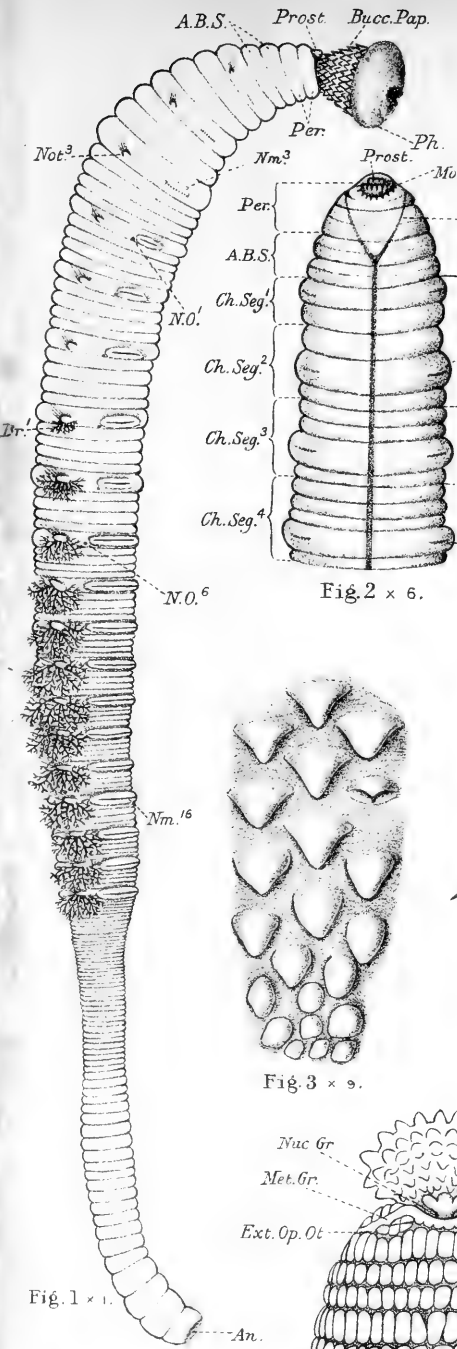


Fig. 1 x 1.

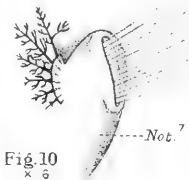


Fig. 10 x 6.

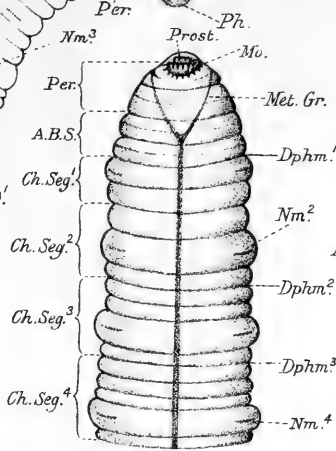


Fig. 2 x 6.

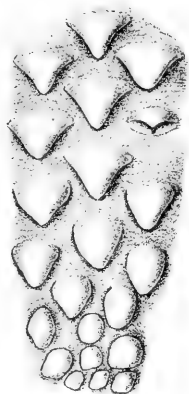


Fig. 3 x 9.

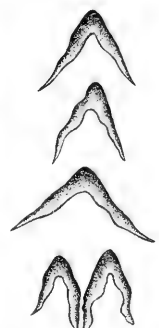


Fig. 4 x 24.

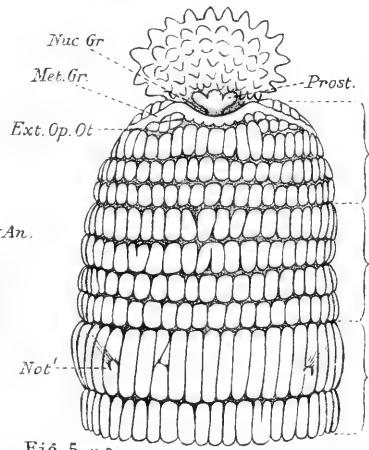


Fig. 5 x 3.

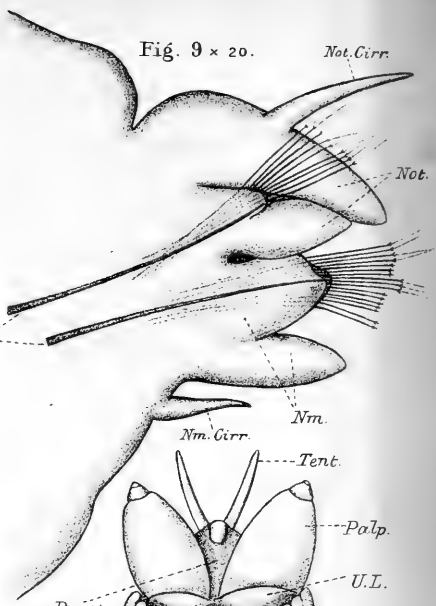


Fig. 9 x 20.

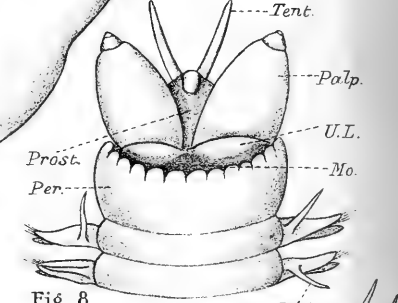


Fig. 8 x 6.

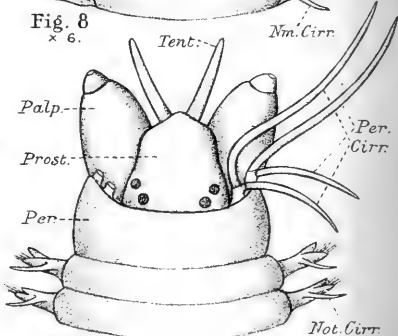


Fig. 7 x 6.

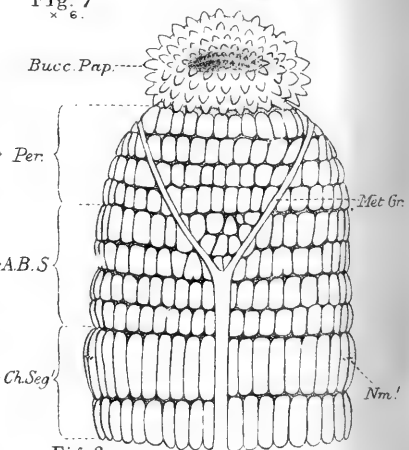
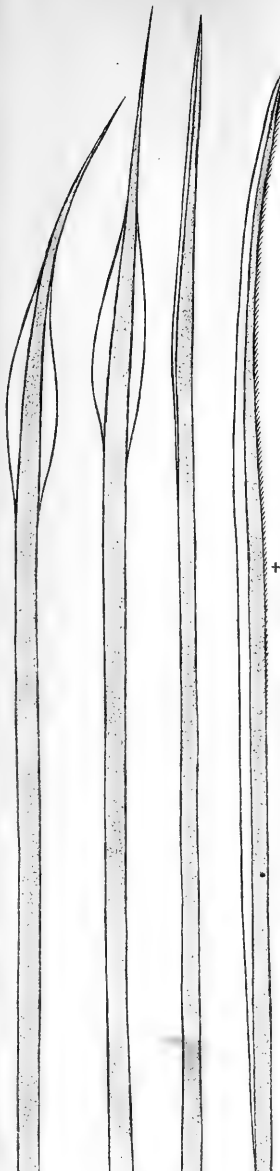


Fig. 6 x 3.





A. x 1000. B. x 1000. C. x 600.

Fig. 11.

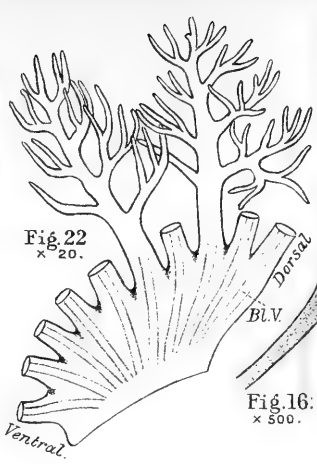


Fig. 16. x 500.

Fig. 22 x 20.



Fig. 21. x 20.

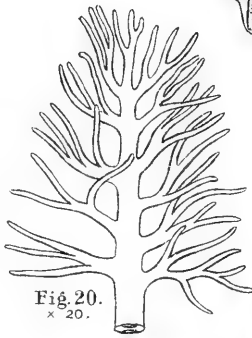


Fig. 20. x 20.

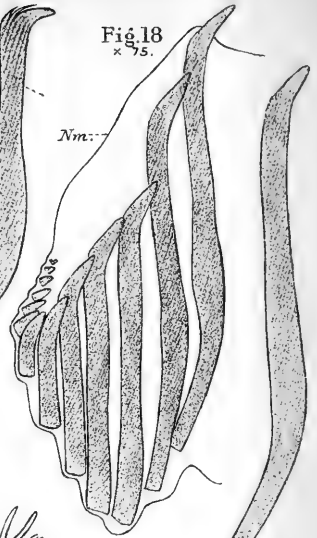


Fig. 18 x 75.

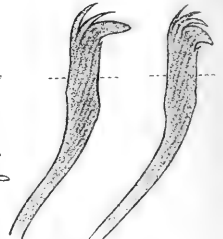


Fig. 17 x 150.

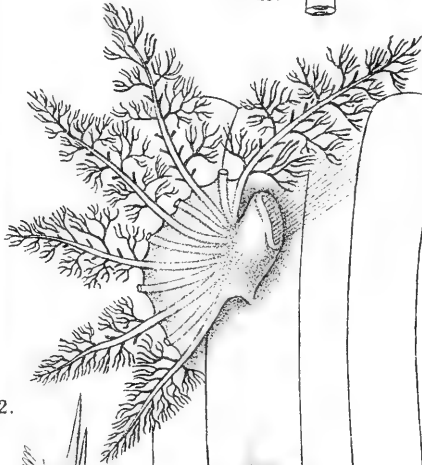


Fig. 14 x 800.

Fig. 15 x 1000.

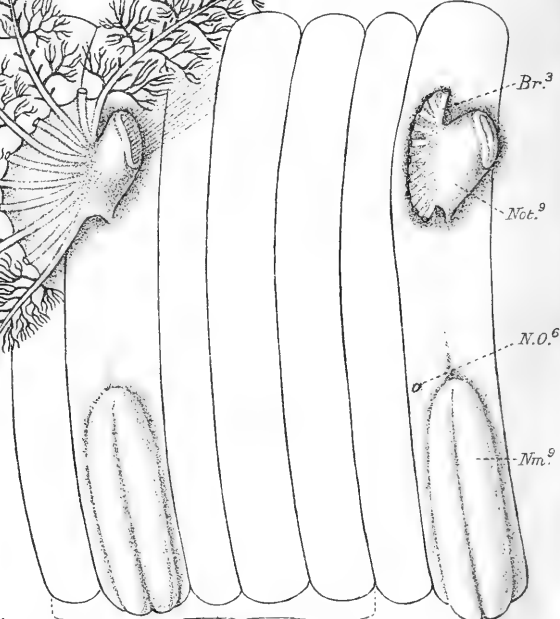
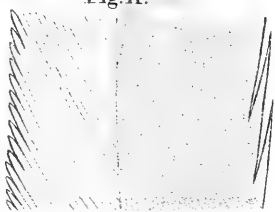


Fig. 19 x 6.

Ch. Seg. 10

Fig. 13 x 800.



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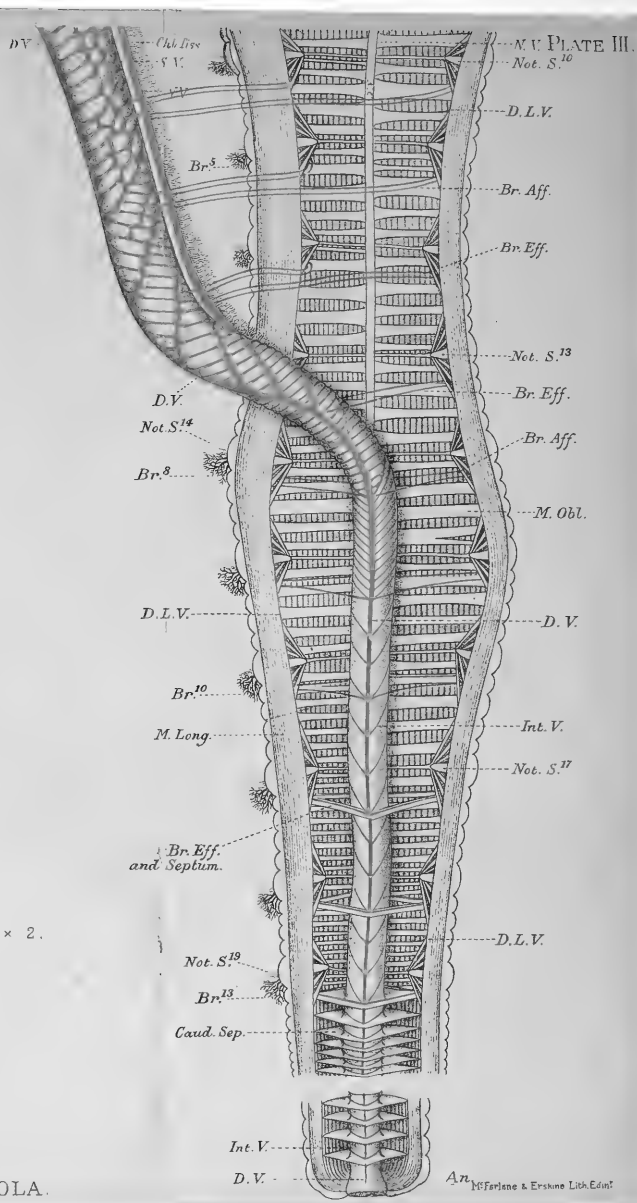
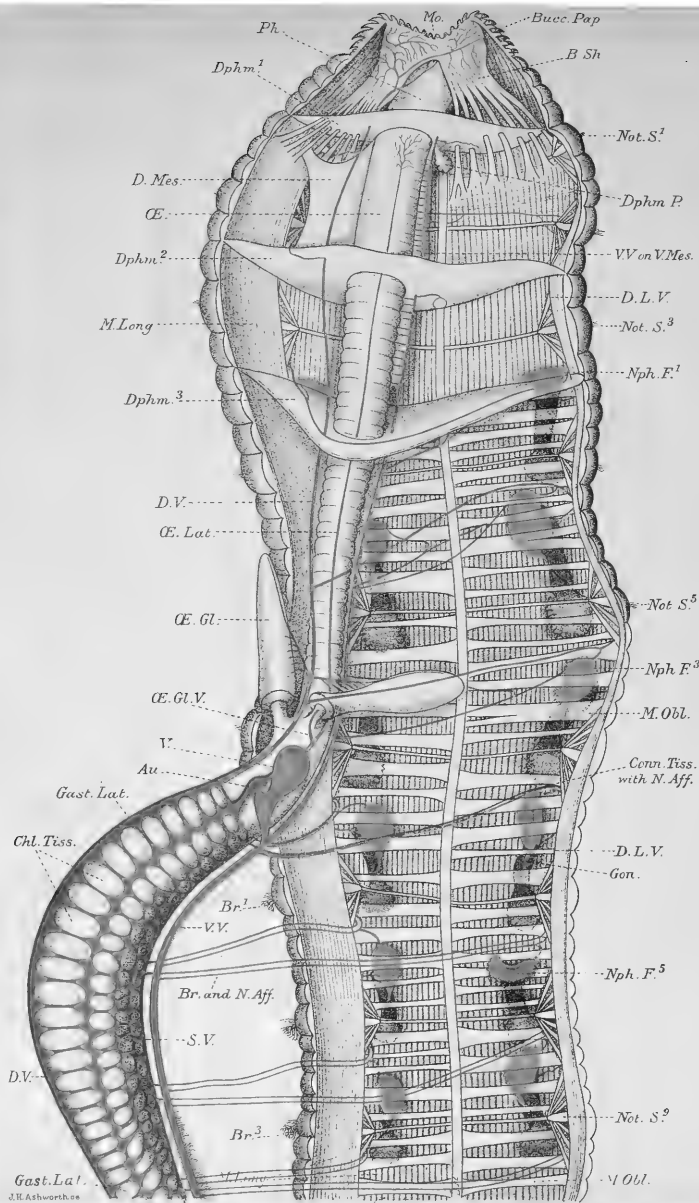


Fig 23 x 2.

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Fig. 24 x 4.

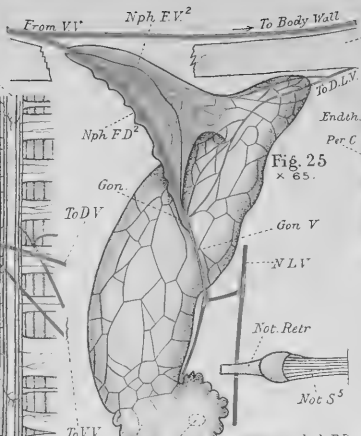
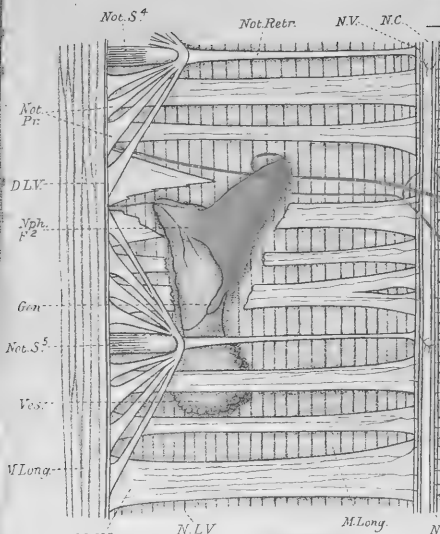


Fig. 25 x 65.

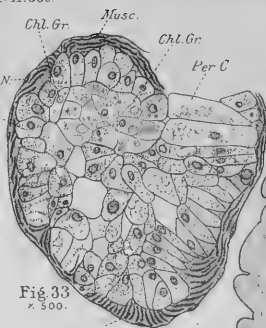


Fig. 33 x 300.

Fig. 34 x 65.

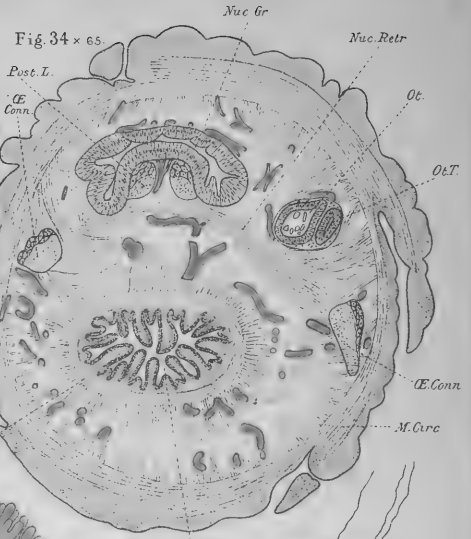


Fig. 26 x 4.

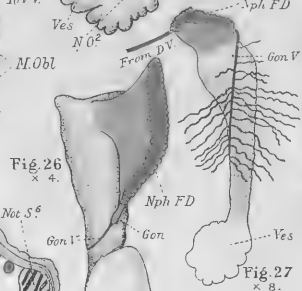


Fig. 27 x 8.

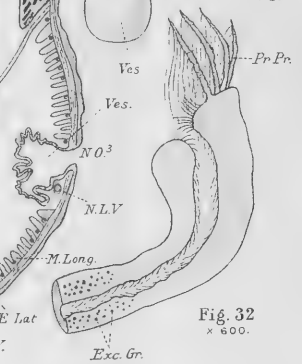


Fig. 32 x 600.



Fig. 28 x 20.



Fig. 29 x 20.

Fig. 30 x 650.

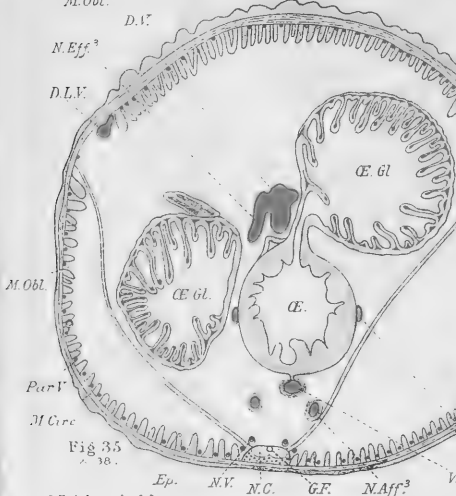


Fig. 35 x 38.



Fig. 31 x 150.

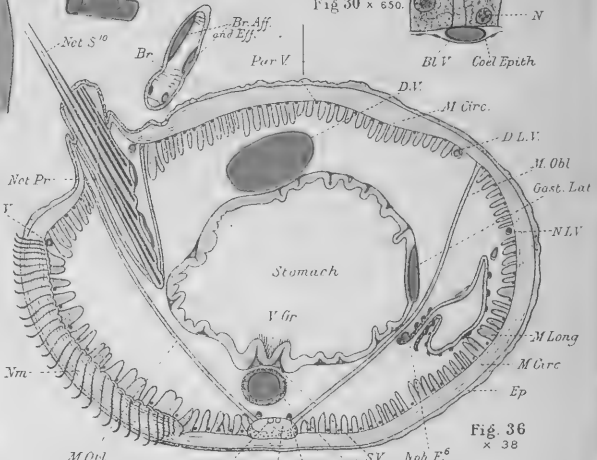


Fig. 36 x 38.





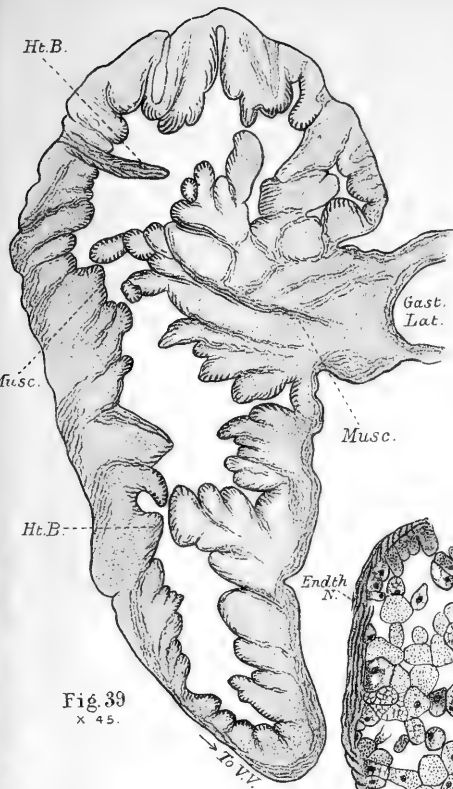


Fig. 39  
x 45.

Fig. 37  
x 100.

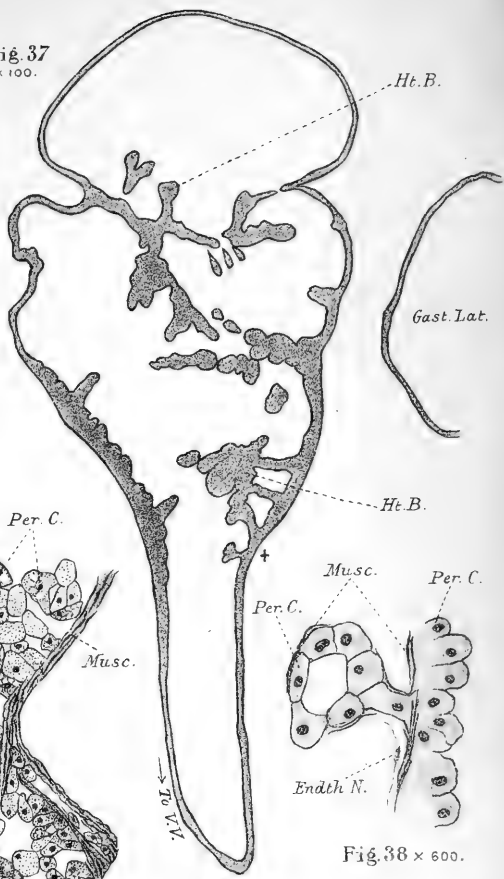


Fig. 38 x 600.

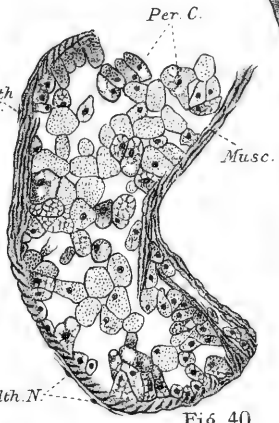


Fig. 40  
x 260.

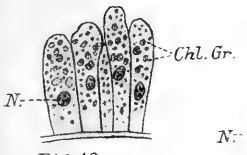


Fig. 43 x 500.

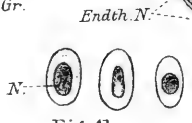


Fig. 41 x 1000.

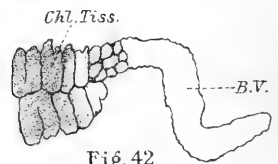


Fig. 42  
x 150.

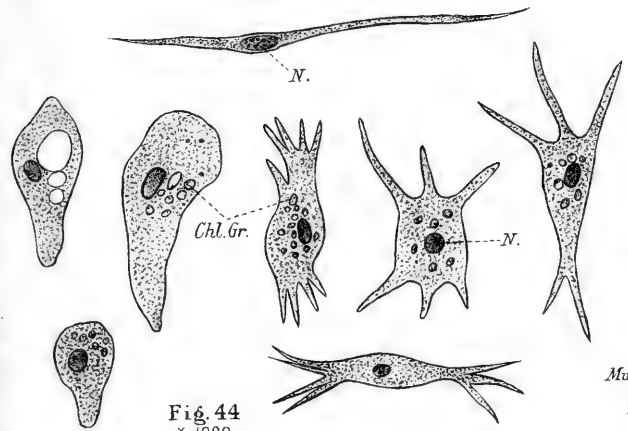


Fig. 44  
x 1000.

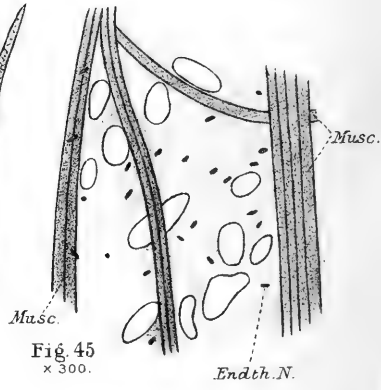
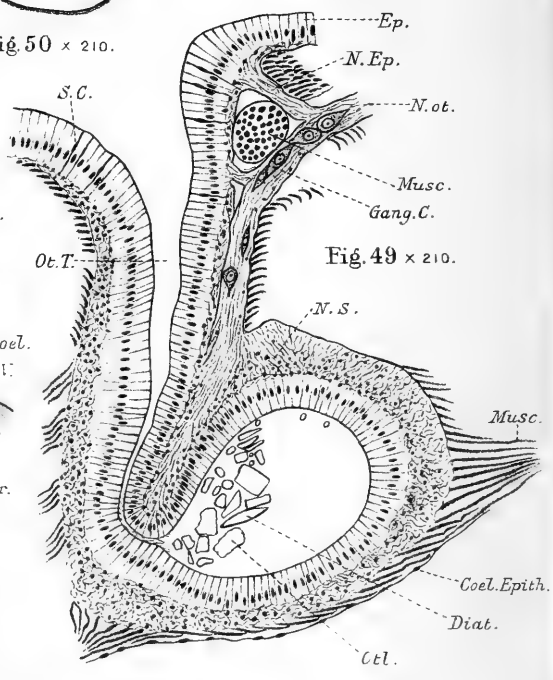
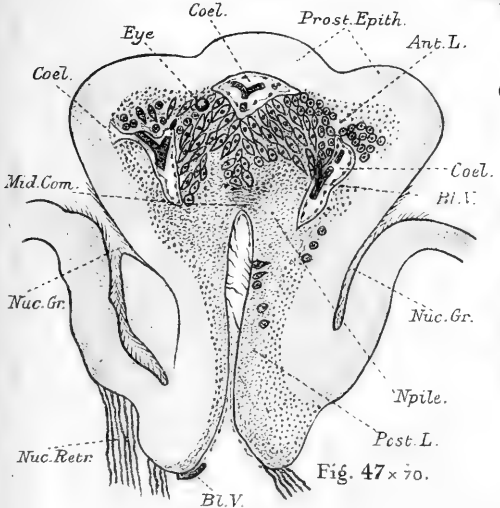
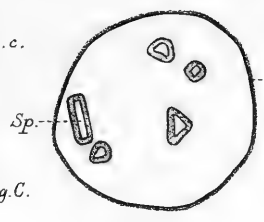
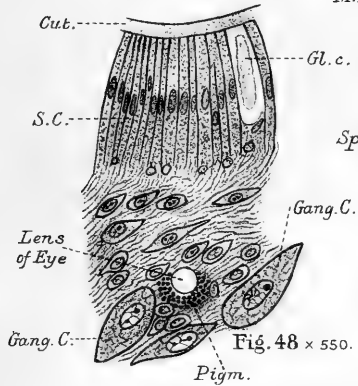
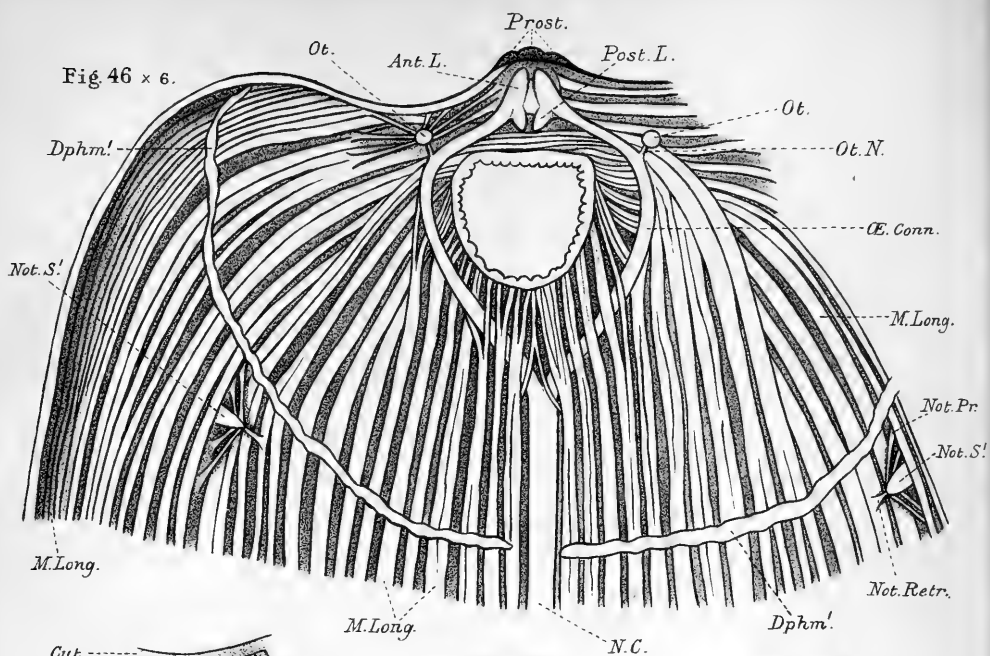


Fig. 45  
x 300.

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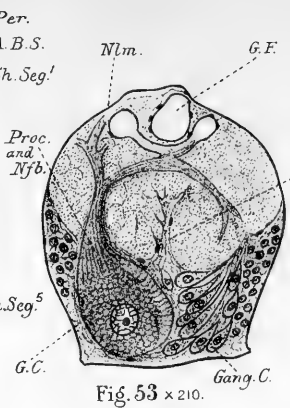
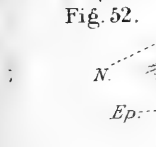
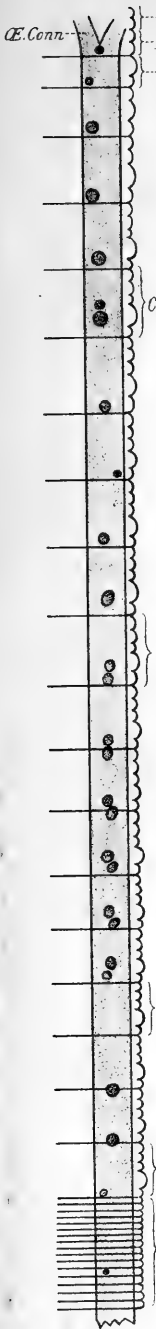


Fig. 53 x 210.

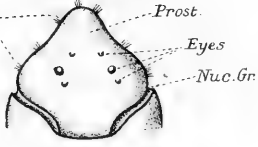


Fig. 59 x 100.

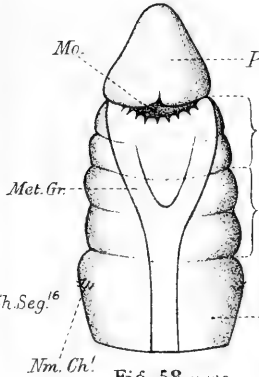


Fig. 58 x 100.

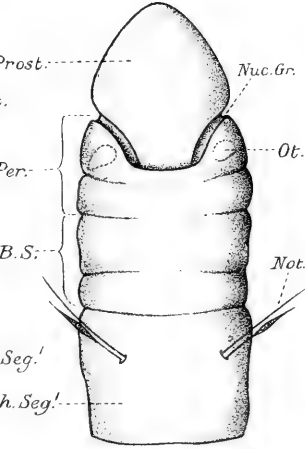


Fig. 57 x 100.

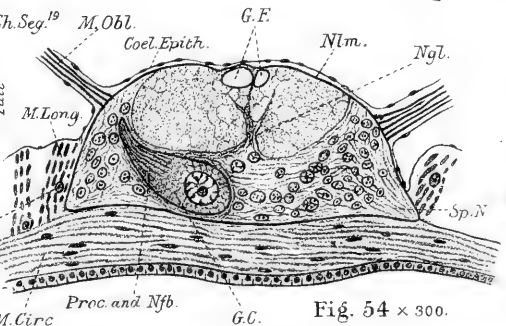


Fig. 54 x 300.

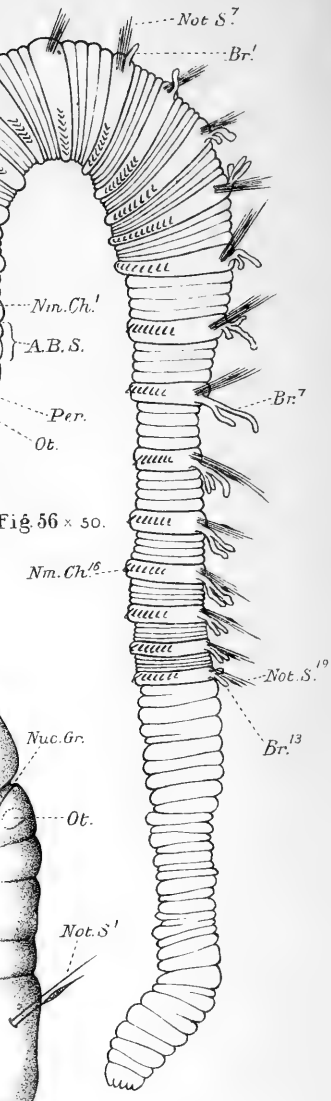


Fig. 56 x 50.

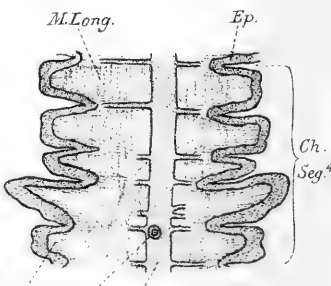


Fig. 55 x 15.

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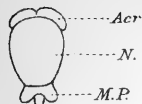


Fig. 66  
x 3000

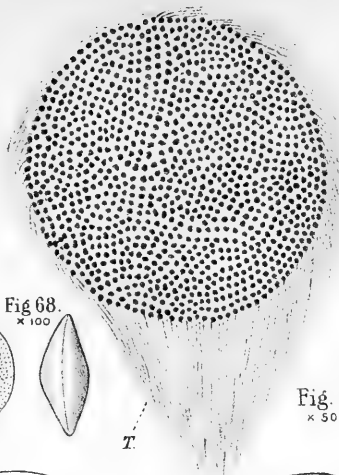


Fig. 60.  
x 500



Fig. 61.  
x 500



Fig. 62.  
x 500



Fig. 63.  
x 500

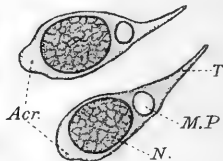


Fig. 64.  
x 2000

Fig. 65.  
x 500

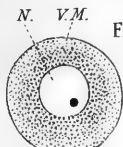


Fig. 67.  
x 100



Fig. 68.  
x 100

T.

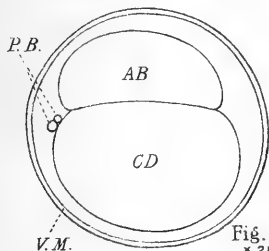


Fig. 69.  
x 210

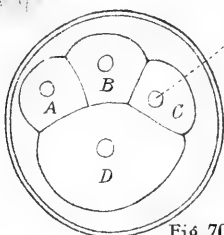


Fig. 70.  
x 210

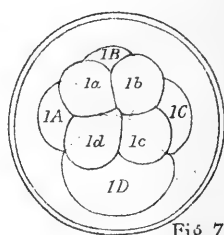


Fig. 71.  
x 210

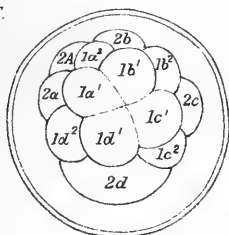


Fig. 72.  
x 210

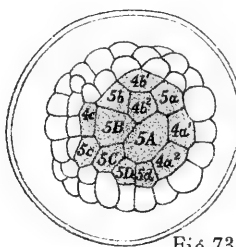


Fig. 73.  
x 210

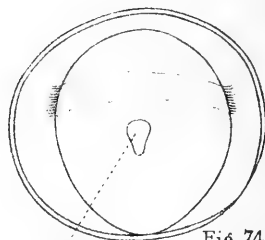


Fig. 74.  
x 210

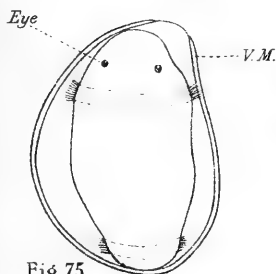


Fig. 75.  
x 210

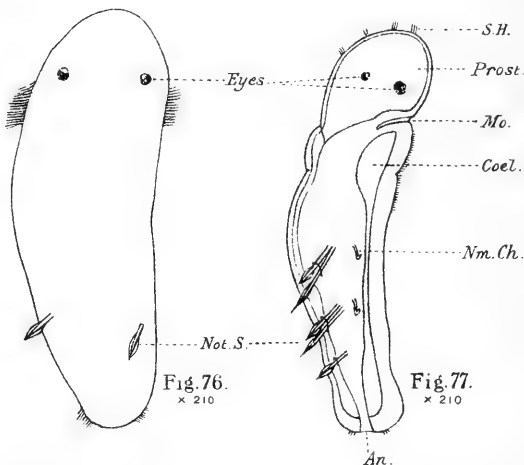


Fig. 76.  
x 210

Fig. 77.  
x 210

Fig. 78.  
x 1000



Fig. 79. x 1000

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