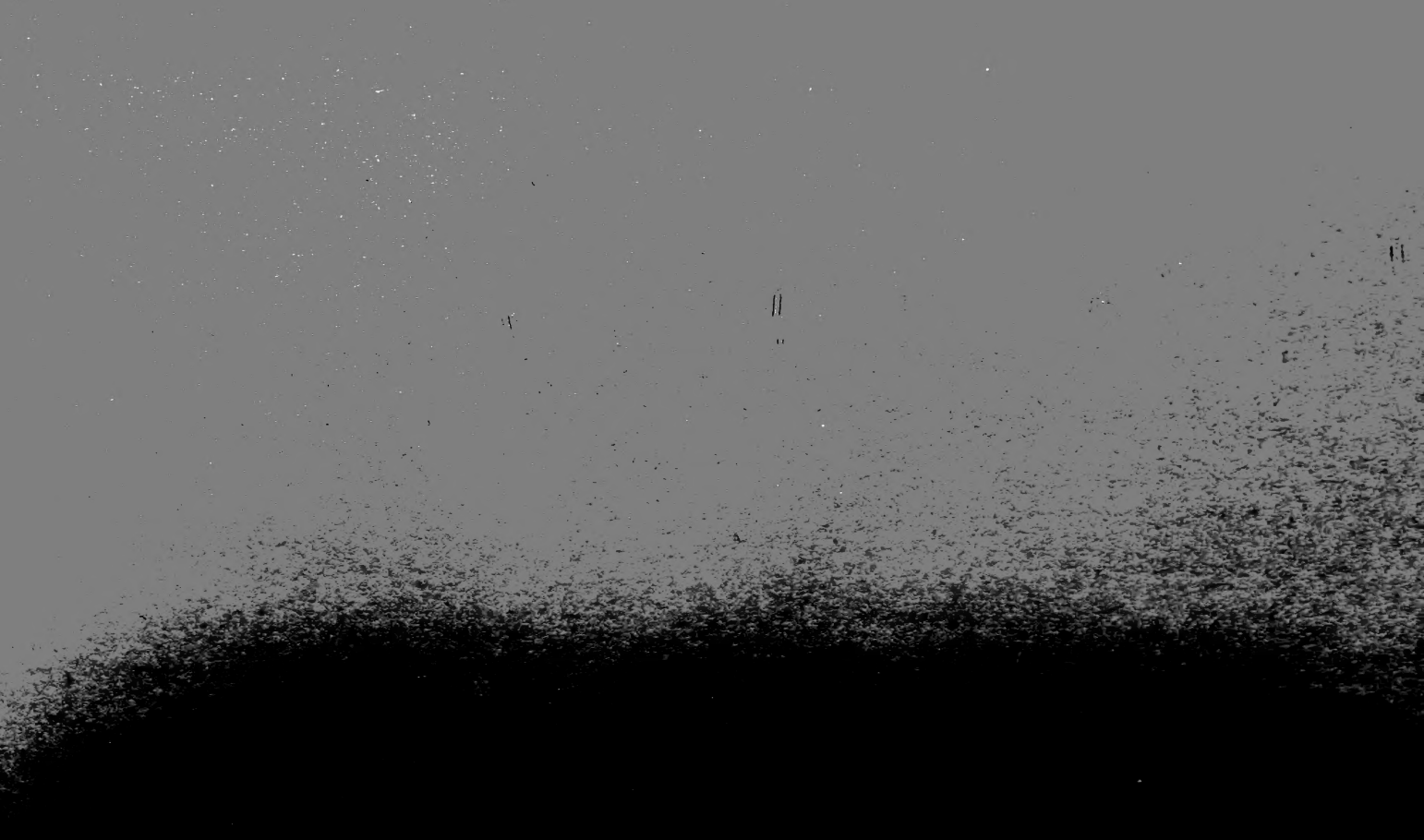

PROCEEDINGS

AND

TRANSACTIONS

OF THE

LIVERPOOL BIOLOGICAL SOCIETY.

VOL. XXIX.

SESSION 1914-1915.

LIVERPOOL:
C. TINLING & CO., LTD., PRINTERS, 53, VICTORIA STREET.

1915.

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PROCEEDINGS

OF THE

LIVERPOOL BIOLOGICAL SOCIETY

OFFICE-BEARERS AND COUNCIL.

Ex-Presidents:

1886—1887 Prof. W. MITCHELL BANKS, M.D., F.R.C.S.
1887—1888 J. J. DRYSDALE, M.D.
1888—1889 Prof. W. A. HERDMAN, D.Sc., F.R.S.E.
1889—1890 Prof. W. A. HERDMAN, D.Sc., F.R.S.E.
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1899—1900 J. WIGLESWORTH, M.D., F.R.C.P.
1900—1901 Prof. PATERSON, M.D., M.R.C.S.
1901—1902 HENRY C. BEASLEY.
1902—1903 R. CATON, M.D., F.R.C.P.
1903—1904 Rev. T. S. LEA, M.A.
1904—1905 ALFRED LEICESTER.
1905—1906 JOSEPH LOMAS, F.G.S.
1906—1907 Prof. W. A. HERDMAN, D.Sc., F.R.S.
1907—1908 W. T. HAYDON, F.L.S.
1908—1909 Prof. B. MOORE, M.A., D.Sc.
1909—1910 R. NEWSTEAD, M.Sc., F.E.S.
1910—1911 Prof. R. NEWSTEAD, M.Sc., F.R.S.
1911—1912 J. H. O'CONNELL, L.R.C.P.
1912—1913 JAMES JOHNSTONE, D.Sc.
1913—1914 C. J. MACALISTER, M.D., F.R.C.P.

SESSION XXIX., 1914-1915.

President:

Prof. J. W. W. STEPHENS, M.D., D.P.H.

Vice-Presidents:

Prof. W. A. HERDMAN, D.Sc., F.R.S.
C. J. MACALISTER, M.D., F.R.C.P.

Hon. Treasurer:

W. J. HALLS.

Hon. Librarian:

MAY ALLEN, B.A.

Hon. Secretary:

JOSEPH A. CLUBB, D.Sc.

Council:

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S. T. BURFIELD, B.A.
G. ELLISON.
H. B. FANTHAM, D.Sc., M.A.
W. T. HAYDON, F.L.S.
J. JOHNSTONE, D.Sc.
DOUGLAS LAURIE, M.A.
W. S. LAVEROCK, M.A., B.Sc.
J. H. O'CONNELL, L.R.C.P.
MAY RATHBONE, (Miss).
WM. RIDDELL, M.A.
E. THOMPSON.

Representative of Students' Section:
Miss M. BRADLEY, B.Sc.

REPORT of the COUNCIL.

During the Session 1914-15 there have been six ordinary meetings and one field meeting of the Society.

The communications made to the Society at the ordinary meetings have been representative of many branches of Biology, and the various exhibitions and demonstrations thereon have been of great interest.

By invitation of the Council, Prof. C. J. Patten, M.D., Sc.D., of Sheffield University, lectured before the Society, at the March Meeting, on "The Migration of Birds studied at Irish Light-Stations."

The Library continues to make satisfactory progress, and additional important exchanges have been arranged.

The Treasurer's statement and balance-sheet are appended.

The members at present on the roll are as follows :—

Ordinary members	52
Associate members	8
Student members, including Students' Section, about	30
Total	90

SUMMARY of PROCEEDINGS at the MEETINGS.

The first meeting of the twenty-ninth session was held at the University, on Friday, November 20th, 1914.

The President-elect (Prof. J. W. W. Stephens, M.D., D.P.H.) took the chair in the Zoology Theatre.

1. The Report of the Council on the Session 1913-1914 (see "Proceedings," Vol. XXVIII, p. viii) was submitted and adopted.
2. The Treasurer's Balance Sheet for the Session 1913-1914 (see "Proceedings," Vol. XXVIII, p. xvii) was submitted and approved.
3. The following Office-bearers and Council for the ensuing Session were elected :—Vice-Presidents, Prof. Herdman, D.Sc., F.R.S., and C. J. Macalister, M.D., F.R.C.P.; Hon. Treasurer, W. J. Halls; Hon. Librarian, May Allen, B.A.; Hon. Secretary, Joseph A. Clubb, D.Sc.; Council, H. C. Beasley, S. T. Burfield, B.A., G. Ellison, H. B. Fantham, D.Sc., B.A., W. T. Haydon, F.L.S., J. Johnstone, D.Sc., Douglas Laurie, M.A., W. S. Laverock, M.A., B.Sc., J. H. O'Connell, L.R.C.P., May Rathbone (Miss), W. Riddell, M.A., and E. Thompson.
4. Prof. J. W. W. Stephens, M.D., D.P.H., delivered the Presidential Address on "The mode of transmission of some Tropical Diseases" (see "Transactions," p. 3). A vote of thanks was carried with acclamation.

The second meeting of the twenty-ninth session was held at the University, on Friday, December 11th, 1914. The President in the chair.

1. Prof. Herdman submitted the Annual Report on the work of the Liverpool Marine Biology Committee (see " Transactions," p. 21).
2. Prof. Dakin, D.Sc., gave an interesting account of a recent visit to the Abrollos Archipelago, off the West Coast of Australia, for the purpose of investigating the Coral formations of the Islands.

The third meeting of the twenty-ninth session was held at the University, on Friday, January 22nd, 1915. The President in the chair.

1. Mr. J. W. Cutmore exhibited, with remarks, a suggested sparrow-hawk's larder, found near Mold, N. Wales.
2. Dr. Clubb exhibited, with remarks, a series of specimens of a melanistic variety of the Water Vole, found at Leasowe, living in a colony, isolated from colonies of the ordinary species living in the same pond.
3. Mr. Heron-Allen, F.L.S., gave an address, illustrated by a beautiful series of lantern slides, on a collection of Foraminifera obtained from marine deposits, collected by Prof. Herdman off the West Coast of Scotland.

The fourth meeting of the twenty-ninth session was held at the University, on Friday, February 12th, 1915. The President in the chair.

1. The President exhibited some old surgical instruments.
2. Mr. R. D. Laurie exhibited and described specimens from Lord Howes' Island, off the East Coast of Australia.
3. Dr. Johnstone submitted the Annual Report of the Investigations carried on during 1914 in connection with the Lancashire Sea-Fisheries Committee (see " Transactions," p. 63).

The fifth meeting of the twenty-ninth session was held at the University, on Friday, March 12th, 1915. The President in the chair.

1. Prof. Herdman exhibited, with remarks, the gigantic colonial Foraminifer, *Ramulina*, from the Indian Ocean.
2. Mr. R. D. Laurie gave an account of the distribution of crabs in the Indian Ocean.

The sixth meeting of the twenty-ninth session was held at the University, on Friday, May 14th, 1915. The President in the chair.

1. Prof. C. J. Patten, M.D., Sc.D., of Sheffield University, lectured before the Society on the invitation of the Council, on "The Migration of Birds studied at Irish Light-Stations." The lecture was much appreciated by a large audience, and a cordial vote of thanks was accorded to the lecturer.

The seventh meeting of the twenty-ninth session was the Annual Field Meeting, held on Saturday, June 19th. A very pleasant afternoon was spent in a Biological ramble through footpaths, via Meols, Newton, Frankby, and Grange Hill to West Kirby. At the short business meeting held after tea, on the motion of the President from the chair, Prof. E. E. Glynn, M.A., M.D., was unanimously elected President for the ensuing session.

LIST of MEMBERS of the LIVERPOOL BIOLOGICAL SOCIETY.

SESSION 1914-1915.

A. Ordinary Members.

(Life Members are marked with an asterisk.)

ELECTED.

1908 Abram, Prof. J. Hill, 74, Rodney Street, Liverpool.

1909 *Allen, May, B.A., Hon. Librarian, University, Liverpool.

1888 Beasley, Henry C., Prince Alfred Road, Wavertree.

1913 Beattie, Prof. J. M., M.A., M.D., The University, Liverpool.

1903 Booth, jun., Chas., 30, James Street, Liverpool.

1912 Burfield, S. T., B.A., Zoology Department, University, Liverpool.

1886 Caton, R., M.D., F.R.C.P., 78, Rodney Street.

1886 Clubb, J. A., D.Sc., Hon. Secretary, Free Public Museums, Liverpool.

1910 Ellison, George, 52, Serpentine Road, Wallasey.

1910 Fantham, H. B., D.Sc., M.A., School of Tropical Medicine, University, Liverpool.

1902 Glynn, Dr. Ernest, 67, Rodney Street.

1886 Halls, W. J., Hon. Treasurer, 35, Lord Street.

1910 Hamilton, Mrs. J., 96, Huskisson Street, Liverpool.

1896 Haydon, W. T., F.L.S., 55, Grey Road, Walton.

1912 Henderson, Dr. Savile, 48, Rodney Street, Liverpool.

1886 Herdman, Prof. W. A., D.Sc., F.R.S., Vice-President, University, Liverpool.

1893 Herdman, Mrs. W. A., Croxteth Lodge, Ullet Road, Liverpool.
1912 Hobhouse, J. R., 54, Ullet Road, Liverpool.
1902 Holt, A., Dowsefield, Allerton.
1903 Holt, George, Grove House, Knutsford.
1903 Holt, Richard D., M.P., 1, India Buildings, Liverpool.
1912 Jackson, H. G., M.Sc., Zoology Department, University, Birmingham.
1898 Johnstone, James, D.Sc., University, Liverpool.
1894 Lea, Rev. T. S., D.D., The Vicarage, St. Austell, Cornwall.
1896 Laverock, W. S., M.A., B.Sc., Free Public Museums, Liverpool.
1906 Laurie, R. Douglas, M.A., University, Liverpool.
1912 Lyon (Miss), Una, High School for Girls, Aigburth Vale, Liverpool.
1912 Macalister, C. J., M.D., F.R.C.P., VICE-PRESIDENT, 35, Rodney Street, Liverpool.
1915 Macdonald, Prof. J. S., B.A., The University, Liverpool.
1905 Moore, Prof. B., University, Liverpool.
1913 Mottram, V. H., Physiological Department, University, Liverpool.
1904 Newstead, Prof. R., M.Sc., F.R.S., University, Liverpool.
1904 O'Connell, Dr. J. H., 38, Heathfield Road, Liverpool.
1913 Pallis, Mark, Tätoi, Aigburth Drive, Liverpool.
1903 Petrie, Sir Charles, 7, Devonshire Road, Liverpool.
1915 Prof. W. Ramsden, University, Liverpool.
1903 Rathbone, H. R., Oakwood, Aigburth.
1890 *Rathbone, Miss May, Backwood, Neston.
1910 Riddell, Wm., M.A., Zoology Department, University, Liverpool.
1897 Robinson, H. C., Malay States.
1908 Rock, W. H., 25, Lord Street, Liverpool.
1894 Scott, Andrew, A.L.S., Piel, Barrow-in-Furness.

1908 Share-Jones, John, F.R.C.V.S., University, Liverpool.
1895 Sherrington, Prof., M.D., F.R.S., University, Liverpool.
1886 Smith, Andrew T., 21, Croxteth Road, Liverpool.
1903 Stapledon, W. C., "Annery," Caldy, West Kirby.
1913 Stephens, Prof. J. W. W., M.D., PRESIDENT, University, Liverpool.
1903 Thomas, Dr. Thelwall, 84, Rodney Street, Liverpool
1905 Thompson, Edwin, 25, Sefton Drive, Liverpool.
1889 Thornely, Miss L. R., Nunclose, Grassendale.
1888 Toll, J. M., 49, Newsham Drive, Liverpool.
1891 Wiglesworth, J., M.D., F.R.C.P., Springfield House, Winscombe, Somerset.

B. ASSOCIATE MEMBERS.

1905 Carstairs, Miss, 39, Lilley Road, Fairfield.
1914 Cutmore, J. W., Free Public Museum, Liverpool.
1913 Hamilton, Erik, M.Sc., 96, Huskisson Street, Liverpool.
1905 Harrison, Oulton, 18, Limedale Road, Mossley Hill.
1910 Kelley, Miss A. M., 10, Percy Street, Liverpool.
1912 Parkin, Miss A. B., 3, Cairns Street, Liverpool.
1913 Smith, Miss E. M. G., 39, Parkfield Road, Liverpool.
1903 Tattersall, W., D.Sc., The Museum, Manchester.
1910 Tozer, Miss E. N., Physiology Laboratory, University, Liverpool.

C. UNIVERSITY STUDENTS' SECTION.

President: Miss M. Bradley, B.Sc.

Secretary: J. Ronald Bruce, B.Sc.

(Contains about 30 members.)

D. Honorary Members.

S.A.S., Albert I., Prince de Monaco, 10, Avenue du brocadéro, Paris.

Bornet, Dr. Edouard, Quai de la Tournelle 27, Paris.

Claus, Prof. Carl, University, Vienna.

Fritsch, Prof. Anton, Museum, Prague, Bohemia.

Haeckel, Prof. Dr. E., University, Jena.

Hanitsch, R., Ph.D., Raffles Museum, Singapore.

Solms-Laubach, Prof.-Dr., Botan. Instit., Strassburg.

LIST OF SOCIETIES AND ACADEMIES WITH WHICH PUBLICATIONS ARE EXCHANGED.

(Twenty-seven additions made since 1905 are marked with an asterisk.)

Adelaide.—Royal Society of South Australia. *Memoirs ; Transactions.*

Agram.—Societas Historico-Naturalis Croatica. *Glasnik.*

Amsterdam.—K. Akad. van Wetenschappen. *Proceedings of the Section of Sciences ; Verslagen Gew. Vergaderingung ; Verhandelingen ; Jaarboeken.*
Natuurkundig Tijdschrift voor Nederlandsch-Indie.

Baltimore.—Johns Hopkins University. *Circulars.*

Basle.—Naturforschende Gesellschaft. *Verhandlungen.*

Bergen.—Bergens Museum. *Aarbog ; Meeresfauna ; Skrifter.*

*Berkeley, Calif.—University. *Publications.*

Berlin.—Deutscher Fischerei Verein. *Allgemeine Fischerei Zeitung ; Mitteilungen ; Zeitschrift für Fischerei.*
K. preussische Akademie der Wissenschaften. *Sitzungsberichte.*

Birmingham.—Birmingham andMidland Institute Scientific Society. *Records of Meteorological Observations.*
Natural History and Philosophical Society. *Proceedings ; Report.*

Bonn.—Naturhistorischer Verein der preussische Rheinlande. *Verhandlungen.*
Niederrheinische Gesellschaft für Natur- und Heilkunde. *Sitzungsbericht.*

Bordeaux.—Société Linnéenne. *Procès-Verbaux.*
*Société Scientifique et Station Biologique d'Arcachon. *Bulletin.*

*Boston, Mass.—American Academy of Arts and Sciences. *Proceedings.*
Society of Natural History. *Proceedings.*
Tufts College. *Studies.*

*Brisbane.—Queensland Museum. *Annals.*

Brooklyn.—Institute of Arts and Sciences. *Cold Spring Harbour Monographs; Science Bulletin.*

Brussels.—Academie Royale des Sciences, des Lettres, etc., *Annuaire; Bulletin (Classe des Sciences).*

*Société Royale Zoologique et Malacologique de Belgique. *Annales.*

*Bryn Mawr.—College. *Monographs.*

Buenos Ayres.—Museo Nacional. *Anales.*

*Buffalo.—American Microscopical Society. *Transactions.*

Society of Natural Sciences. *Bulletin.*

Caen.—Société Linnéenne de Normandie. *Bulletin.*

*Calcutta.—Indian Museum. *Memoirs; Miscellaneous Zoological Publications; Records.*

Cambridge, Mass.—Museum of Comparative Zoology. *Bulletin.*

Cape of Good Hope.—Department of Agriculture.—*Reports.*

Chicago.—*Botanical Gazette.*

Field Museum of Natural History. *Publications.*

Christiania.—Videnskabs Selskabet. *Forhandlinger.*

Cincinnati.—Lloyd Library. *Bulletin.*

Colombo.—*Ceylon Marine Biological Reports.*

*Ceylon Museum. *Spolia Zeylanica.*

*Concarneau.—Laboratoire de Zoologie et de Physiologie Maritimes. *Travaux Scientifiques.*

Copenhagen.—Conseil Permanent International pour l'Exploration de la Mer. *Bulletin des Resultats; Bulletin Hydrographique; Bulletin Statistique; Publications de Circonstance; Rapports et Procès-Verbaux.*

Danish Biological Station. *Report.*

Kommissionen for Havundersögelser. *Meddelelser; Skrifter.*

K. Dansk Videnskabernes Selskab. *Oversigt; Skrifter.*

Naturhistorisk Forening Videnskabelige. *Meddelelser.*

Dublin.—Royal Dublin Society. *Economic Proceedings; Scientific Proceedings; Scientific Transactions.*

Edinburgh.—Royal College of Physicians. *Laboratory Reports.*

Royal Physical Society. *Proceedings.*

Royal Society. *Proceedings.*

Fishery Board for Scotland. *Report.*

*Frankfort-on-the-Main.—Senckenbergische naturforschende Gesellschaft. *Abhandlungen; Bericht.*

Freiburg-in-the-Breisgau.—Naturforschende Gesellschaft. *Bericht.*

Geneva.—Société de Physique et d'Histoire Naturelle. *Mémoires.*

Giessen.—Oberhessische Gesellschaft für Natur und Heilkunde. *Bericht.*

Glasgow.—Natural History Society. *Glasgow Naturalist.*

Göttingen.—K. Gesellschaft der Wissenschaften. *Nachrichten.*

*Gratz.—Naturwissenschaftlicher Verein für Steiermark. *Mitteilungen.*

Güstrow.—Verein der Freunde der Naturgeschichte in Mecklenberg. *Archiv.*

HAARLEM.—Musée Teyler. *Archives.*

Société Hollandaise des Sciences. *Archives Neerlandaises des Sciences Exactes et Naturelles.*

HALIFAX, NOVA SCOTIA.—Nova Scotian Institute of Science. *Proceedings and Transactions.*

HALLE.—Academia Caesarea Naturae Curiosorum. *Nova Acta.*

*HAMBURG.—Naturhistorisches Museum. *Mitteilungen.*

*IRELAND.—Department of Agriculture and Technical Instruction. *Report.*

KIEL.—Kommission zur wiss. Untersuchungen der deut. Meere in Kiel u. d. biologischen Anstalt auf Helgoland. *Wissenschaftliche Meeresuntersuchungen.*

Naturwissenschaftlicher Verein. *Schriften.*

LA PLATA.—Museo. *Annales ; Revista.*

LAWRENCE.—Kansas University. *Experimental Station Reports ; Geological Survey Reports ; Science Bulletin.*

LEEDS.—Yorkshire Naturalists Union. *Transactions.*

LEIPZIG.—K. sächsische Gesellschaft der Wissenschaften. *Berichte über die Verhandlungen.* (*Math. Phys. Classe*).

*LINCOLN, NEBRASKA.—University. *Medical School Bulletin ; Zoological Laboratory Studies.*

LIVERPOOL.—Geological Society. *Proceedings.*

LONDON.—British Association for the Advancement of Science. *Report.*

The Naturalist.

Royal Microscopical Society. *Journal.*

*MADISON.—Wisconsin Academy of Sciences, Arts, etc. *Transactions.*

Wisconsin Geological and Natural History Survey. *Bulletin.*

MAGDEBURG.—Museum für Natur- und Heimatkunde. *Abhandlungen und Berichte.*

MANCHESTER.—Microscopical Society. *Transactions and Report.*

Victoria University. *Publications.*

MARSEILLES.—Musée d'Histoire Naturelle. *Annales* (*Zoologie*).

MELBOURNE.—Royal Society of Victoria. *Proceedings.*

*MILWAUKEE.—Public Museum. *Annual Report ; Bulletin.*

Wisconsin Natural History Society. *Bulletin.*

MONACO.—Institut Océanographique. *Bulletin ; Résultats des Campagnes Scientifiques.*

MONTEVIDEO.—Museo Nacional. *Anales.*

MONTPELLIER.—Académie des Sciences et Lettres. *Bulletin Mensuel.*

MOSCOW.—Société Impériale des Naturalistes. *Bulletin ; Nouveaux Mémoires.*

MUNICH.—Ornithologische Gesellschaft. *Verhandlungen.*

NANCY.—Société des Sciences. *Bulletin des Séances.*

NAPLES.—Reale Accademia delle Scienze, etc. *Atti ; Rendiconto.*

*University. Museo Zoologico. *Annuario.*

*NEW HAVEN.—Connecticut Academy of Arts and Sciences. *Transactions.*

*NEW YORK.—American Museum of Natural History. *Bulletin.*

OBERLIN.—Oberlin College Laboratory. *Bulletin.*
Wilson Ornithological Club. *Wilson Bulletin.*

OPORTO.—Academia Polytechnica do Porto. *Annaes Scientificos.*
Annaes de Sciencias Naturaes.

PARIS.—*Bulletin Scientifique de la France et de la Belgique.*
Museum d'Histoire Naturelle. *Bulletin.*
Société de Biologie. *Comptes Rendus.*
Société Zoologique de France. *Bulletin ; Mémoires.*

PHILADELPHIA.—Academy of Natural Sciences. *Proceedings.*

*PORTICI.—Regia Scuola Superiore di Agricoltura. *Bollettino.*

RENNES.—Société Scientifique et Medicale de l'Ouest. *Bulletin.*

ROME.—Società Romana per gli Studi Zoologia. *Bollettino.*

SAINT JOHN, NEW BRUNSWICK.—Natural History Society. *Bulletin.*

SAINT LOUIS.—Academy of Science. *Transactions.*
*Missouri Botanical Garden. *Report.*

SAINT PETERSBURG.—Academia Scientiarum Imperialis. *Bulletin.*

SAN FRANCISCO.—Californian Academy of Science. *Proceedings.*

SANTIAGO.—Société Scientifique du Chili. *Actes.*

STAVANGER.—Museum. *Aarsheft.*

STOCKHOLM.—K. Svenska Vetenskaps Academi. *Archiv för Botanik ; Archiv för Zoologi.*

SYDNEY.—Australian Museum. *Catalogues ; Memoirs ; Records.*

TIFLIS.—Kaukasisches Museum. *Mitteilungen.*

TOKYO.—Imperial University. College of Science, *Journal ;* *College of Agriculture, *Journal.*
Societas Zoologica. *Annotationes Zoologicae Japonenses.*

TORONTO.—Canadian Institute. *Proceedings.*

TURIN.—University. Museo di Zoologia ed Anatomia Comparata. *Bollettino.*

UNITED STATES OF AMERICA.—Department of Commerce and Labour. Bureau of Fisheries. *Bulletin ; Report.*

UPSALA.—Regia Societas Scientiarum. *Nova Acta.*

URBANA.—Illinois State Laboratory of Natural History. *Bulletin ; *Biological Monographs.*

*VIENNA.—K. Akademie der Wissenschaften. *Bericht der Kommission für oceanographische Forschungen ; Mitteilungen der Erdbeben-Kommission ; Sitzungsberichte (Mat.-Naturw. Classe).*
K. k. naturhistorisches Hofmuseum. *Annalen.*
K. k. zoologisch-botanische Gesellschaft. *Verhandlungen.*

*WASHINGTON.—Carnegie Institution. *Papers on Experimental Evolution.*
United States National Museum. *Annual Report ; Bulletin ; Proceedings.*
United States National Herbarium. *Contributions.*

WELLINGTON.—New Zealand Institute. *Transactions and Proceedings.*

*WOOD'S HOLL.—Marine Biological Laboratory. *Biological Bulletin.*

ZÜRICH.—Naturforschende Gesellschaft. *Vierteljahrsschrift.*

THE LIVERPOOL BIOLOGICAL SOCIETY.

Dr. IN ACCOUNT WITH W. J. HALLS, HON. TREASURER. **Cr.**

1914, Oct. 1st, to Sept. 30th, 1915.	£	s.	d.
To Teas and Attendance at Meetings	3	14	6
,, Messrs. Tinling & Co.	30	0	0
,, Hon. Secretary's Expenses	2	3	0
,, Hon. Librarian's Expenses	3	12	9
,, £125 4% Commercial Cable Co's Debenture	96	8	1
,, Brokerage and Stamps	1	2	7
,, Fire Insurance—Society's Library	1	18	6
,, Cheque Book	0	0	10
,, Balance in Bank	2	3	4
,, Cash in hand	6	5	9
	£147	9	4

1914, Oct. 1st, to Sept. 30th, 1915.	£	s.	d.
By Balance from last Account	6	2	11
,, Subscriptions	18	18	0
,, Student Society	0	15	0
,, Subscriptions in Arrear	9	9	0
,, Subscription in Advance	1	1	0
,, Sale of Volumes	10	15	0
,, Fire Insurance Claim paid	99	0	0
,, Bank Interest	0	6	6
,, Quarter's Interest on Investment	1	1	11
	£147	9	4

Audited and found correct,

W. T. HAYDON.

LIVERPOOL, *September 30th, 1915.*

TRANSACTIONS

OF THE

LIVERPOOL BIOLOGICAL SOCIETY.

PRESIDENTIAL ADDRESS

ON

THE MODE OF TRANSMISSION OF SOME TROPICAL DISEASES.

By J. W. W. STEPHENS, M.D., D.P.H.,
Sir Alfred Jones Professor of Tropical Medicine,
University of Liverpool.

[Delivered November 20th, 1914.]

In a presidential address one may, I think, expect either a consideration of the general principles underlying or on the other hand a summary of what is known about the subject. I do not feel competent to attempt the first nor perhaps is it yet time to attempt it, for the wave of discovery in tropical medicine which began about 1897 has not yet spent itself, and we are still carried along with the current of new facts. I shall endeavour, therefore, to give you simply what I am afraid must be a disconnected account of some of the more recent work on the subject. The great interest of tropical diseases lies, I think, primarily in the fact that in the most important of them one can lay one's finger definitely on the cause. This is often not the case in diseases of temperate climes, e.g., scarlet fever, measles, smallpox, numerous nervous disorders, etc., etc. And secondly, in many tropical diseases the transmission is by some insect. Again many are due to protozoa, an exceptional occurrence in temperate diseases. To this is due the fact that on the whole, perhaps, they are more amenable to attack by drugs, and moreover we have a second great object of attack, viz., the insect transmitter. Knowing thus in many cases the cause and the transmitter, there is the further great interest in studying the life histories of both, and finally the great probability of success in attacking one or other of the possible links in the chains of the life cycles.

Malaria.

As you know, this disease is transmitted by mosquitoes, but only by a certain sub-family, the Anophelinae, and further by only a limited number of these, though why some of these transmit while others do not, we do not know. The mode of transmission is as follows: At a certain stage of the fever the malarial parasites, which up to this time have multiplied in the blood and given rise to the fever, presumably by a toxin they secrete, become specialized into sexual forms male and female. When the blood containing these is sucked in by a mosquito, fertilization occurs in the mosquito's stomach. That this is so, is, I may add, rather hypothetical, as it has not been actually observed, though once or twice only, fertilization has been stated to have been observed under the microscope, in an ordinary wet blood film. One observer, indeed, believes that fertilization takes place in the blood stream, and suggests that this is the explanation why it has been observed so rarely on the slide under the microscope, and never actually in blood taken directly from the stomach of a mosquito immediately after it has fed. Whatever be the truth of this, the next stage is one that can readily be seen in a dissected-out stomach, viz., the encysted fertilized parasite—the *oocyst*. These are eventually found in numbers from one to two up to a hundred or so on the outer surface of the stomach, viz., in the muscle wall. Growth proceeds in these oocysts, and after various changes the now large oocyst is filled with large numbers of thread-like curved bodies about 12-14μ long—the *sporozoites*. These travel—how has not been followed—to the *salivary* gland of the mosquito and from there they escape, during the act of biting, into the blood, say, of a healthy person. Their actual escape has not indeed been seen, but we can safely affirm that they do escape, as healthy people bitten by infected anophelines contract malaria.

Christophers and myself made many attempts to see the sporozoites penetrating the red cells by mixing the two together in a wet film, but always unsuccessfully. Schaudinn stated that he had seen it, using sporozoites from an oocyst in the stomach (but I would remind you that many of Schaudinn's observations have not been confirmed).

This development in the mosquito takes about ten to fourteen days, so that it is only after this lapse of time that a mosquito that has bitten a malaria patient is capable of transmitting the infection. This cycle in the mosquito is known as the cycle of new infection. It is again only some ten days after being bitten that the attack of malaria develops. In this way the malaria parasite, when it has got into a mosquito develops and gets out again.

Malaria or ague has in the past existed in many parts of England, and it is still a memory in the minds of some of that somewhat rare species "the oldest inhabitant." Why malaria died out in England is not, I think, perfectly clear. The factors that are generally invoked to explain its disappearance are the use of quinine, drainage—which decreased the breeding areas of anophelines, the abolition of the window tax, etc. But there is a factor which though general in its action, is probably as great if not greater than any of these—viz., the social and hygienic improvements in the condition of the people. For it has been pointed out by Christophers in India that, *caeteris paribus*, the ravages of malaria are greatest where the social standard is lowest. Poverty, for instance, is an important determining factor. I have spoken as if malaria had disappeared from this country, but a case has recently been reported from Romney Marsh, an old haunt of the disease, in which the source of infection could not be traced, and one if not two other cases have occurred. The occurrence of malaria at the present time in this country, then, is an extremely rare phenomenon. One,

indeed, is surprised that it is so rare, for there are still in England abundance of Anophelines, e.g., in some of the ditches and marl pits in Wirral larvae abound, and it is the same elsewhere, and the return from the tropics to this country of cases of malaria is by no means rare. How the malaria parasite arose, whether from some form already existent in the gut of the mosquito, is a very interesting problem of which at present we know almost nothing.

In connection with the disappearance of malaria from England, I might note the peculiar fact that Christophers and myself found in India, in the midst of intensely malarial districts, villages which were completely free from malaria although Anophelines abounded. We could find no explanation of this very peculiar condition, but the same set of conditions is also reported in Italy, and the term *Paludismus sine malaria* is used to designate them.

Though this, baldly stated, is the malaria-mosquito cycle, yet we must consider another side to the question in order to fully understand how and why it is that Europeans contract malaria in the tropics. On examining a number of native children who appear to be in the best of health, one is surprised to find that a certain percentage, sometimes 80-90 per cent., contain parasites in their blood. These children abound in native villages, and the huts which they inhabit are infested with Anophelines, and a certain percentage of these latter are infected and infective—i.e., contain sporozoites in their salivary glands. If now Europeans live in the vicinity of native huts, as is only too commonly the case in Africa, they soon get malaria. It is the native children that constitute, so to speak, the reservoir of the disease in the tropics. They are the great danger. Prophylaxis is simple: remove the European quarter to a safe distance beyond the usual range of a mosquito's flight—quarter to half a mile. This method has been carried out in many colonies with very good results.

Of course the two fundamental methods are either (1) killing the parasites in the blood by means of quinine, a method which in Italy has yielded excellent results, or (2) destruction of mosquitoes, or rather their larvae; this in some places has also given very good results, but frequently it is difficult of performance—at least with the funds available.

Yellow Fever.

Is a disease the cause of which is unknown, but we know how it is transmitted, viz., by a particularly annoying mosquito from its persistent attempts at biting—*Stegomyia fasciata*. We know with regard to the mode of transmission that the mosquito can only transmit if it has bitten a patient not later than the third day of the disease, and that it is able to transmit the disease only twelve days later. That is practically all we know. But there is one mystery about the mode of transmission. Yellow fever is, so far as is known, only contracted at dusk or at night, not in the day time. Yet the Stegomyia bites at all times of the day and night. Only one explanation of this peculiarity has been suggested. It is that young mosquitoes bite in the day and old mosquitoes at night. This change in habits takes place when the mosquito is about three days old, i.e. when she first lays eggs. As the mosquito is only dangerous twelve days after it has bitten a yellow fever patient, no day-biting Stegomyia is ever dangerous. This explanation, however, seems to be too good to be true. The Stegomyia is a so-called domestic mosquito, i.e. it breeds in water supplies about the house, tins, barrels, water troughs, etc., etc. It is surprising what quantities of larvae and pupae a single barrel can produce. They are as thick as tadpoles in a shallow pool. It is this fact, viz., the domestic habits of this mosquito which make it comparatively vulnerable

to attack, and it is in yellow fever that prophylactic measures have obtained their greatest success. To attack the malaria mosquito—the Anopheline—breeding in marshes, rivers, streams, wells, pools, and in fact any collection of water, is a much more difficult matter, but it is largely a question of finance.

Phlebotomus Fever—Three-days' Fever—or Sand-fly Fever—Summer Influenza.

This is a very unpleasant fever while it lasts, viz., three days, but fortunately is never fatal, but presents, however, some resemblances to yellow fever. The disease exists along the Mediterranean littoral, in Egypt, India, and probably all over the world. It is a great cause of sickness among our troops in Malta, India, etc. The cause of the disease is unknown, but the blood is infective from the first two days of the fever. The infective matter, whatever it be, will pass through a fine filter candle. The disease is transmitted by certain species of sandflies. The facts of transmission are that a sandfly that bites a patient during the first day or second day of the fever becomes infected, but it is only about a week later that it can transmit the disease. Comparatively little is so far known about the life history of sandflies, the greater part of what we do know we owe to Newstead's labours in Malta. They breed in caves and dark places, and lay their eggs in cracks in the soil, in old walls, etc. The egg stage lasts about eight days, the larval stage lasts two to eight weeks, pupal stage, attached to stones, two weeks. Flies in captivity live only ten days.

The flies themselves, so far as we know at present, do not survive the winter, but do so in the larval stage. The question then arises: how does the disease survive the winter? It is

thought not to do so in man, as relapses in man are practically unknown (at least after any long period), but it is still just possible that the blood may be infective even though the patient is well, though against this is the fact that transmission experiments only succeed during the first two days of the fever. The other alternative is that the disease is transmitted through the larvae of the flies. Our knowledge is at present accordingly incomplete, and it is so, largely owing to the fact that these minute flies, which easily pass through a mosquito net, only survive captivity for about ten days, and so experimental work with them is difficult.

Sleeping Sickness.

Is due to two different species of trypanosomes, viz., *Trypanosoma gambiense* and *T. rhodesiense*. The former trypanosome is the cause of sleeping sickness in most parts of Africa where the disease exists, while *T. rhodesiense* is confined to Rhodesia, Nyasaland, and a few other adjoining territories. One peculiarity of the latter form of the disease is that the number of known cases is few, say about a hundred, but on the other hand the disease is even more deadly, or at any rate more rapidly fatal than that due to *T. gambiense*. The trypanosomes in each case exist in the blood and the disease is transmitted by tsetse flies, *T. gambiense* by *Glossina palpalis* and *T. rhodesiense* by *Glossina morsitans*. The mode of transmission is not simply mechanical, but a developmental cycle takes place in the fly, for a fly after biting a sleeping-sickness patient is only capable of transmitting the disease to a healthy person some twenty to thirty days later. The mode of transmission of *T. gambiense* is the following. The trypanosomes which are sucked into the gut disappear more or less completely in about a week, to reappear later as short stumpy crithidial forms. These eventually find their way to the

salivary glands, how is not known. I should add that the mode of development is different in the case of other trypanosomes.

A further point for consideration arises. In the case of malaria the disease is purely a man to man infection, and we have the important factor that the native children, who for the most part appear to be in abundant good health, have numerous parasites in their blood, they constitute in fact the reservoir from which, for example, the European gets infected. It is a man to man disease. In sleeping sickness this is not so, The natives do not here constitute a permanent reservoir, for the simple reason that they all die. How then does infection spread? The answer is that the same trypanosome, at least this appears to be proved in the case of *T. rhodesiense*, occurs in native game. The game enjoys an immunity to the disease. The game infects the fly and so the disease is transmitted to man. The game constitutes the reservoir. What should be done in Africa to limit this spread of the disease by the game is a question into which a special Commission enquired this year. It is proposed to try an experiment to see what will be the result of exterminating or reducing the quantity of the game from a small defined area. It is practically impossible to attack the fly.

T. tullochi and *T. grayi*. It may be further noted that tsetse flies contain in nature these trypanosomes in their gut. So far inoculation of these into animals has produced negative results, but it would appear as if they deserved re-study.

Kala-azar.

Is an extremely fatal disease prevalent in India, Sudan, parts of China, and elsewhere. The parasites that cause this disease exist in large numbers in endothelial cells in the spleen, liver, bone marrow, etc. A disease indistinguishable

clinically from this exists in the Mediterranean littoral, but there is this difference, that in these countries the disease is almost entirely confined to children, though very occasionally adults also contract it. Both these diseases are caused by parasites which are about the size of one-third of a red cell and have two nuclei. They can be cultivated on blood media, where they change into slender spindle-shaped flagellates.

Further, in the Mediterranean countries there is a wasting disease in dogs which is also due to a parasite indistinguishable from that producing the disease in children and adults in India, and it is thought that there is some connection between the disease in dogs and the disease in children.

The existing evidence is to the effect that this disease in children is transmitted from dogs by the agency of fleas, though others disbelieve this and claim that mosquitoes are the transmitting agents. There is no evidence however that in India this disease in dogs exists at all, so that we are puzzled by the apparent relationship of the dog disease in the Mediterranean and the non-relationship in India.

The matter is further complicated by the fact that in many parts of the world a disease occurs, viz., Oriental sore. This is purely local skin affection, but in the sore, parasites are found indistinguishable from those that occur in the general infection of kala-azar.

I should mention in connection with these diseases some interesting French work recently done. It is known that in the gut of many insects spindle-shaped flagellates occur. Laveran and Franchini find that the injection of these flagellates into mice gives rise to non-flagellate forms in the organs indistinguishable from kala-azar parasites, and in fact the mice die of the infection. Now in these diseases, viz., kala-azar, infantile kala-azar, dog kala-azar, and tropical sore, numerous investigators have been searching actively for the transmitting agent, e.g., fleas, bugs, mosquitoes,

sandflies, etc., and vainly experimenting to procure developmental cycles in these supposed transmitting agents on the analogy of malaria, sleeping sickness, etc. In the case of the dog disease successful transmission results have indeed, it is stated, been got through the agency of fleas. If we assume for the moment the accuracy of these transmission experiments, it does not, it seems to me, follow—keeping in view Laveran and Franchini's experiments—that they necessarily prove that the flea is the transmitting agent in the accepted sense, viz., that it transmits from dog to dog in this case. It is possible that the dog disease is due to the inoculation by fleas of their natural gut flagellates, and that each case of the dog disease arises *de novo* from the flagellates of a flea ; the disease is in fact a flea-dog disease, but not a flea-dog-flea disease. We might, indeed, call it hemi-cyclical transmission, not cyclical, and so for all the other similar infections kala-azar, infantile kala-azar, and tropical sore. But I need hardly remind you that this is sheer hypothesis. Should it prove to be true it may also apply to diseases due to helminths and possibly bacteria. For instance, we have a Nematode disease in man and cattle characterized by the presence of nodules of worms in the skin, but as no larvae apparently exist in the blood it is at present difficult to understand how the nematodes get out, i.e., the disease gets transmitted. On the hypothesis I have just put forward, the explanation would be that these nematodes were derived from forms existing in the guts of insects or possibly in water. I would suggest also that Sarcosporidia form an example of hemi-cyclical infection, i.e., they get into animals but do not get out. I must apologise for this digression into the pleasant and easily-trod path of hypothesis, but will now return to matters of fact.

Spirochaetes.

These give rise to relapsing fever in man. The term relapsing is a very appropriate one, for after a fever lasting some days and a period of apyrexia, the whole train of symptoms recurs again. There may be more than one relapse, but the succeeding relapses are not so intense as the original fever. The disease is due to the presence of spirochaetes in the blood which, however, disappear therefrom in the interval between the attacks. There are many species of spirochaete described in man. It will suffice to mention two only, viz., *Sp. duttoni*, producing the African disease, and *Sp. recurrentis*, producing the European form. The African disease is transmitted by ticks and the European probably by lice. In the African form the tick involved is known as *Ornithodorus moubata*. It is a dirty-looking, brown, wrinkled tick, and frequents rest-houses where it is found in the walls, thatch, etc., and is found in the dust around trees where caravans halt. Unlike most ticks, after feeding it crawls away. The tick is infective probably directly after biting. The life cycle of the spirochaete is, however, believed to be the following. The spirochaete in the gut of the tick breaks up into numerous ovoid granules resembling small bacilli. These pass into the cells of the malpighian tubules and into the ovary. When the tick bites, these ovoid bodies are voided in the faeces and so contaminate the coxal fluid which is secreted when the tick bites, and so presumably the wound. Those that reach the ovary pass through the egg and nymph into the adult tick, so that the disease is transmitted from mother to offspring. This is said to hold good also for a second generation. Others, however, doubt the infective nature of the ovoid granules.

Nothing much is known at present as to the mode of

transmission of the European and Indian relapsing fevers, but that lice and not bugs, as first thought, are the transmitting agent appears fairly certain.

Plague.

Is a disease by no means exclusively tropical. It is a disease of which one of the chief signs is the great swelling of the glands, and hence the term Bubonic plague. The disease is a septicaemia, that is a blood poisoning in which the plague bacilli exist in the blood. Now it has long been known that the outbreak of plague in a place or district is preceded by a mortality among the rats—in fact the rats are dying of plague. We have then to consider the rat factor. Further, it has now been proved that the disease is transmitted from rat to man by means of a particular rat flea, viz., *Pulex cheopis*. How does this come about? In India we have two rats which are important in this connection, viz., *Mus decumanus*, the ordinary brown sewer rat, and *Mus rattus*, the black or house rat. Now it is found in India that the course of plague is the following. Firstly, there is an epizootic among *Mus decumanus*, i.e., the rat mortality as shown by the dead rats collected is going rapidly up, say in July. It reaches a maximum; ten days later there is an epizootic among *Mus rattus*, and then ten to fourteen days later the plague epidemic in man starts. The relationship of plague in man is thus more close in the case of *Mus rattus* than it is in the case of *Mus decumanus*. This association of plague with *M. rattus* is shown by the following curious fact. Plague is equally distributed on all floors of a building, so is *M. rattus*, but *M. decumanus* does no go beyond the third floor. We next turn to the flea. It has been shown by numerous experiments —the experiments of Russian investigators though overlooked

were the first to prove it—that the flea transmits the disease from rat to rat, and there can be no doubt from rat to man. How does it effect this ? This has only recently been explained. When the flea sucks blood containing bacilli into its stomach, the bacilli rapidly multiply and to such an extent that they actually block the oesophagus or rather the proventriculus. The result of this block is that the flea can get no more blood into its stomach and so feels thirsty. In order to relieve its thirst it goes on sucking by means of its pharyngeal pump, but as the blood cannot pass the block some of it regurgitates, owing to the increased pressure back into the wound, but it is now contaminated by the bacilli in the blocked proventriculus and so the wound gets infected.

Filariasis.

One of the most noteworthy conditions included under this heading is Elephantiasis. This is a condition, as you know, in which there is great swelling, e.g., of the legs due to obstruction in the lymphatics caused by quite delicate worms about $1\frac{1}{2}$-4 inches long. These worms give birth to larval forms, which are about one-third mm. long, and they find their way from the lymphatics into the blood stream where they move freely about. When a mosquito sucks blood containing these larvae, the latter pass—how has not been followed—into the thoracic muscles of the mosquito and there come to rest. After a time they become active again and are now found in the proboscis of the mosquito, not in the tube which conducts the blood up, or in the tube which conducts the saliva down, but in the muscular substance of the labium, the rod in the groove of which the tubes and cutting lancets lie. They are thus in a cul-de-sac, and it is thought that they emerge by rupture of a membrane which is stretched

over the end of the rod. They now find themselves on the skin, and in all probability they burrow their own way in through the skin and so are not injected with the blood as was naturally at first thought. Here then we have an example of a nematode worm for the transmission of which an intermediate insect host is necessary. What becomes of them and what happens to them from the time they enter the skin to the time at which they are found as adults in the lymphatics, is unknown.

Ankylostomiasis.

This is a disease due to the presence of minute worms about one-third of an inch long that live in the gut of man. They bury their heads in the gut and produce their serious effect, in all probability, by a poison that they secrete into the wound that they make as they feed on the submucous layer of the gut. The ravages caused by this parasite in the Southern States of America are widespread, and the Americans are now making a great attempt to stamp out the disease. The mode of attack is two-fold.

(1) Medicinal. Luckily the worms are amenable to treatment, and in the drug Thymol, first used by Bozzolo in 1880, we have a potent weapon. (2) Improved sanitation. In order to understand how this is applied we must first understand the life-history of the worm. The worms live in the gut of man. Their eggs pass out in the excreta. Here they hatch and become larvae. These larvae are able to penetrate the skin, so that a person walking bare-footed on contaminated soil is very likely to get infected. The larvae as they pass through the skin cause a good deal of itching. They eventually, by a circuitous route through the lungs and trachea, get swallowed and so pass down the gullet into

the stomach and gut. The sanitary mode of attack then is a simple one, viz., prevention of the contamination of the soil by infected patients. Where primitive hygienic methods still exist the introduction of new methods is always difficult and expensive, but the principle involved is perfectly simple. If sanitation were at a stroke made perfect, the great scourge Ankylostomiasis would be wiped out. The medicinal treatment is necessary for the killing of the worms in the infected patients, and except for this purpose would be unnecessary if sanitation were perfect, but as this is far from being so the medicinal mode of attack is still very necessary. It is a question of breaking one link in the chain of the life-history—the thymol kills the adult worm ; improved sanitation prevents the eggs developing in the soil. The chain is in both cases broken. It should be added that the larvae can also infect if they are swallowed, e.g., in drinking water, but whether this mode of infection or that by the skin is the more important remains to be seen.

Beri-Beri.

Is a disease characterized by general oedema of the body and various paralytic symptoms. It is common in China, and was responsible for much mortality in the Japanese navy. It is seen in jails, on expeditions, and a form of it occurs on board ship, e.g., a ship put into Birkenhead in 1914 with thirty-three cases on board. It has long been suspected that there was some connection between the disease and eating of rice, but this others denied. It has now been shown that it is only particular kinds of rice that give rise to beri-beri. In fact it is the kind of rice that we consume here daily in our rice puddings, i.e., what one may call a nice clean-looking white rice. Why we do not get beri-beri will appear later. The difference between this rice and the rice in its original form,

which is called paddy, is that paddy has its layers of silver skin intact, while in polished rice there is hardly a vestige of these remaining. The difference between these two rices is shown very clearly by feeding a pigeon on polished rice. In a few days most serious nerve trouble arises, and the pigeon is quickly paralysed and dies. It is further a remarkable fact that if some rice bran, i.e., the polishings of rice, be given to the pigeon when symptoms have developed, recovery takes place in a few hours, as if in a magical way. In the case of man, the different effect of the two rices is shown by dividing the inmates of a jail into two lots, one being fed on polished rice, the other on unpolished rice, i.e., paddy. Beri-beri soon breaks out among the first lot, whereas the second is exempt. This has now been shown time after time, and it is quite clear that beri-beri in many cases is due to eating polished rice, i.e., in fact a rice which is deficient in something that native rice possesses. Beri-beri is, in fact, what is called a deficiency disease. The reason we do not acquire beri-beri is that we make up the defect which is present when we eat white rice by supplying it from various articles of the mixed diet we consume as well; but where, as is often the case in the tropics, rice is the staple diet, then if this is a white rice, beri-beri breaks out unless a mixed diet is being taken in addition and in sufficient quantity. A rice capable of producing beri-beri is, or was, in use on P. and O. steamers, but the lascars do not as a rule get beri-beri because they also receive sufficient other rations. One must, however, admit that the consumption of polished rice or other cereals which are defective from the fact that they have their skin removed, e.g., a white flour, will not explain all cases of beri-beri, but that this is very often the cause there can be no doubt.

Now the importance of this work on beri-beri not only is very great in relation to the elimination of this disease, but

it has thrown light on two other diseases not exactly tropical. The first of these is *Scurvy*, which we are wont to associate with Arctic expeditions. This disease also is a deficiency disease, for scurvy can readily be produced in guinea-pigs by feeding them exclusively on bread. It is not simply a case of starvation, for guinea-pigs may be starved by feeding on cabbage alone, but they show no signs of scurvy. There is an absence of something in the bread diet which they require. What it is, we do not know. This hypothetical body we call a vitamine. In all probability another deficiency disease is *Rickets*, a disease characterised by a failure of ossification in the bones. The disease is cured by change of diet. The ordinary process of *growth* is also dependent on the presence of these, at present mysterious, bodies, "growth vitamines" as we may call them. Rats, e.g., will not thrive on an artificially purified diet, but if a quantity of milk equal to only 1 per cent. of the artificial food be added, growth proceeds normally. This last fact raises a very important question. Growth we have seen is dependent on a vitamine. Now cancer is a pathological growth, and in all probability this growth is also a dependent on a vitamine. Now if the vitamines for ordinary and cancerous growths were *different*, it might be possible to construct a diet which would exclude the cancer vitamine, but this is visionary, for we are at present only at the beginning of our knowledge of vitamines. I would claim, then, that tropical medicine in this matter has not only extended its own sphere of knowledge, but has been of distinct service to medicine of temperate climes.

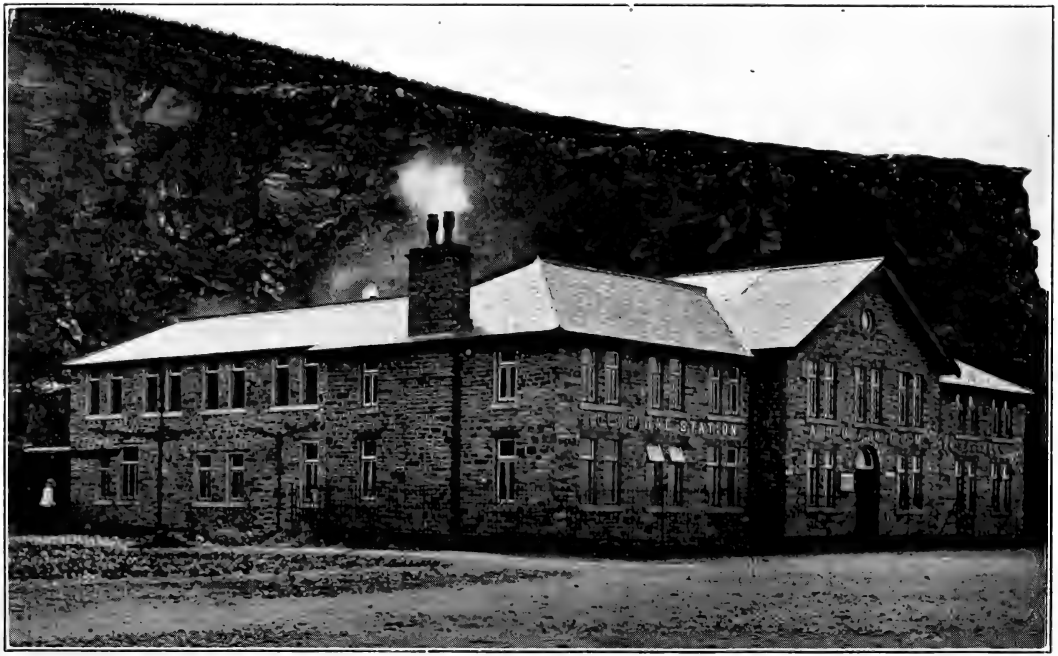


FIG. 1. The Port Erin Biological Station from the North East.
[From a photograph by Prof. F. J. Cole.

THE MARINE BIOLOGICAL STATION AT PORT ERIN BEING THE TWENTY-EIGHTH ANNUAL REPORT OF THE LIVERPOOL MARINE BIOLOGY COMMITTEE.

In this very exceptional year, when many concerns not directly connected with the necessities of existence or the conduct of a great war must suffer more or less, it is not surprising to find that we have a less successful year than usual to record at our Biological Station. Since early in August the thoughts and energies of most of us have been diverted to other channels; and although it is right that in the interests of others we should endeavour to keep all our affairs, so far as may be possible, running normally, still until more important and pressing matters are settled it is well that no unnecessary time, labour and expense should be devoted to what is not essential at the moment. Consequently the Committee and our other supporters and readers will, I am sure, understand and approve if the Report this year takes a shorter form than usual, and deals with little beyond the record of routine work carried out, especially during the earlier portion of the year, at the Port Erin Biological Station and elsewhere in the L.M.B.C. District.

The "Station Record" and the "Curator's Report" which follow show that during the Easter vacation and the Spring months, when both students and senior workers frequent our marine laboratory more than at any other time of the year, the numbers were larger than ever before and the work was abundant and of high quality. Last year we recorded about seventy researchers and students; this year up to the end of July we had a total of ninety occupying work-places in the

laboratory. It is interesting to note that of the three researchers at work during June and July one was a Frenchman from the University of Nancy and another a Belgian from Louvain. Since then we have had no one at work except our own staff. The effect of the war upon the number of visitors to the Aquarium was most striking. During the earlier part of the Summer the numbers steadily increased beyond those of previous years, and on Saturday, August 1st, the total was 1,054 in advance of that of the same date last year, but from that time the numbers declined rapidly. During August, which is usually our busiest month in the Aquarium, Port Erin was, comparatively speaking, deserted, and our total number of visitors for the year is now 12,031—a decrease of 4,576 compared with 1913. The number of "Guides" to the Aquarium sold to visitors is 657—a decrease of 383 compared with last year.

It may be useful to students and others proposing to work at Port Erin that the ground plan of the buildings showing the laboratory and other accommodation should be inserted here (see fig. 2, p. 23).

In regard to the educational work in the laboratories, the usual Easter vacation courses in Marine Biology were carried on during April under the guidance of the teachers in the Zoology and Botany departments of the University of Liverpool. Other groups of senior students came in parties or singly from at least a dozen Universities and Colleges. For example, a group of nine senior students and teachers led by Professor Cole came from University College, Reading, and another nine in charge of Dr. Stuart Thomson from the University of Manchester. A group of seven students from University College, Nottingham, attended a course under the direction of Mr. H. S. Holden, Lecturer in Botany. We had two workers from the University of Birmingham, and other Universities represented were Cambridge (including Newnham),

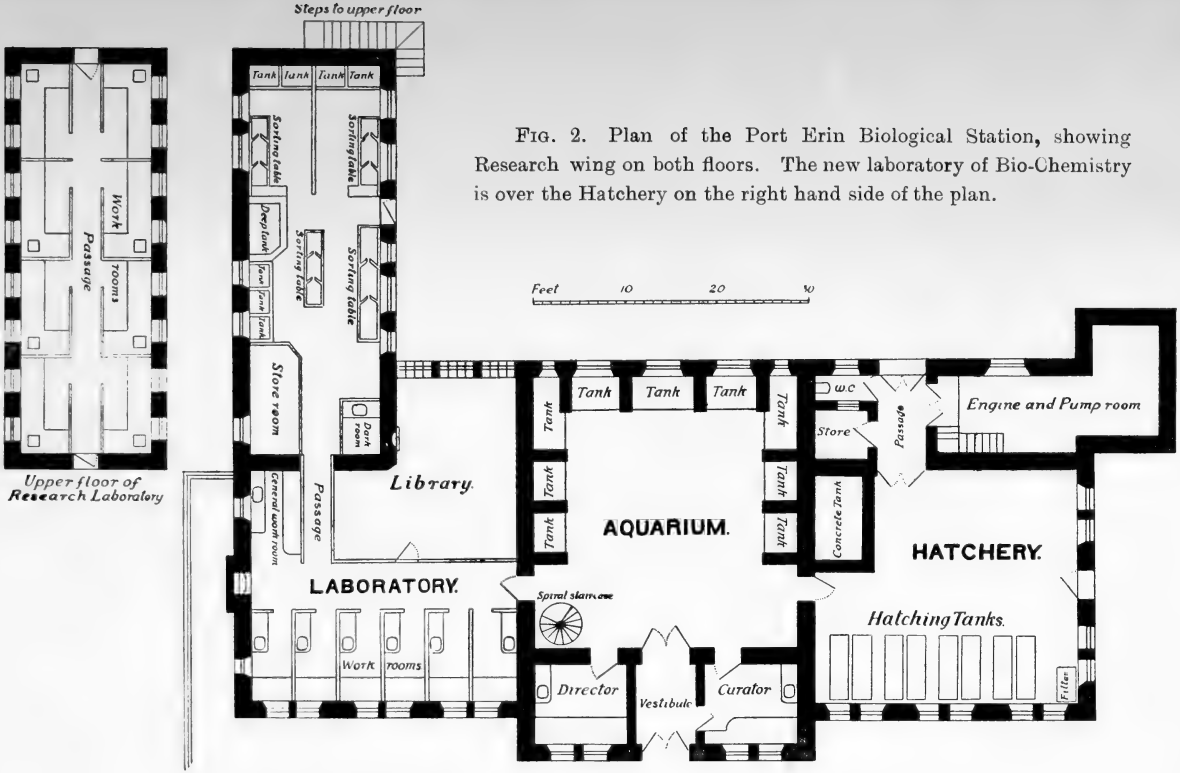


FIG. 2. Plan of the Port Erin Biological Station, showing Research wing on both floors. The new laboratory of Bio-Chemistry is over the Hatchery on the right hand side of the plan.

Harvard, Melbourne, Bangor, Cardiff, Nancy, Louvain, and the Imperial College of Science at South Kensington.

Collecting expeditions as usual, both at sea and along the shore at low tide, were arranged during the Easter vacation. During the remainder of the year the Curator and his staff made periodic collections from time to time as occasion offered, and plankton samples were taken across Port Erin Bay with regularity twice in each week throughout the year.

As on previous occasions, I shall first give the statistics as to the occupation of the "Tables" during the year, then will follow the "Curator's Report," and after that the reports that have been sent to me by various investigators on the work they have done.

The Station Record.

The past year has established another record as regards the number of researchers and students, no fewer than ninety having occupied our laboratories. Of this number eighty-five paid visits of varying duration from March 23rd to April 20th, while the remaining five came to conduct research at the beginning of the year and in the earlier part of the summer vacation.

List of Workers.

Dec. 19*th*, 1913 *to Jan.* 8*th*.	Mr. M. C. Vyvyan.—Marine Algæ.
Dec. 27*th*, 1913 *to Jan.* 5*th*.	Professor Herdman.—Official.
,,	Mr. G. A. Herdman.—Bio-Chemistry.
Feb. 27*th to March* 2*nd*.	Professor Herdman.—Official.
,,	Professor Moore.—Bio-Chemistry.
March 23*rd to April* 11*th*.	Mr. G. F. Sleggs.—General.
,,	Miss G. A. Platt.—General.
,,	Miss D. Thornton.—General.
,,	Miss M. M. Brew.—General.
,,	Miss A. S. Meeson.—General.
,,	Miss J. Price.—General.
March 23*rd to April* 4*th*.	Miss N. Lodge.—General.
,,	Miss E. Catlow.—General.
,,	Miss C. Whittaker.—General.
,,	Miss J. Curwen.—General.
,,	Miss J. Lord.—General.
,,	Miss D. Dobson.—General.
,,	Miss M. Tesh.—General.

March 23*rd to April* 11*th.*	Mr. S. T. Burfield.—Educational.
March 25*th to April* 11*th.*	Mr. R. D. Laurie.—Educational.
March 26*th to April* 9*th.*	Miss H. V. Davies.—General.
March 26*th to April* 11*th.*	Miss H. Clarke.—General.
March 30*th to April* 21*st.*	Professor R. J. Harvey Gibson.—Educational.
March 30*th to April* 24*th.*	Miss M. Knight.—Educational.
March 30*th to April* 13*th.*	Miss R. Holden.—Marine Algæ.
,,	Miss M. Jepps.—Marine Algæ.
,,	Professor Herdman.—Plankton.
March 31*st to April* 4*th.*	Miss M. Latarche.—General.
March 31*st to April* 11*th.*	Miss L. Baker.—Lichens.
,,	Miss P. McKie.—Lichens.
,,	Miss E. M. Blackwell.—Marine Algæ.
March 31*st to April* 13*th.*	Mr. H. Storey.—General.
April 1*st to* 21*st.*	Mr. H. G. Jackson.—Plankton.
April 1*st to* 14*th.*	Mr. A. J. Nicholson.—General.
April 1*st to* 21*st.*	Miss H. M. Duvall.—General.
April 2*nd to* 11*th.*	Mr. B. Sahni.—Marine Algæ.
April 3*rd to* 17*th.*	Miss E. Richmond.—General.
April 4*th to* 18*th.*	Dr. Stuart Thomson.—Educational.
,,	Mr. G. H. Crabtree.—General.
,,	Mr. A. W. Summersgill.—General.
,,	Mr. G. Talbot.—General.
,,	Miss A. Dixon.—General.
,,	Miss F. Lea.—General.
,,	Miss C. M. Lightbown.—General.
,,	Miss Williamson.—General.
,,	Miss E. N. Cowell.—General.
April 6*th to* 11*th.*	Mr. H. T. Cubbon.—General.
,,	Miss D. Jones.—Marine Algæ.
,,	Miss I. L. Millican.—Marine Algæ.
April 6*th to* 19*th.*	Mr. H. S. Holden.—Educational.
,,	Mr. E. Holden.—General.
,,	Mr. M. Straw.—Marine Algæ.
,,	Miss C. N. M. Brett.—Marine Algæ.
,,	Miss E. S. Hillman.—Marine Algæ.
,,	Miss J. M. McClatchie.—Marine Algæ.
,,	Miss D. Bexon.—Marine Algæ.
,,	Miss H. C. Bowser.—Marine Algæ.
,,	Miss H. H. Maguire.—Marine Algæ.
April 6*th to* 20*th.*	Mr. J. R. Bruce.—Marine Algæ.
,,	Miss G. H. Wood.—Marine Algæ.
,,	Miss E. M. Tate.—Marine Algæ.
,,	Miss E. Alexander.—Marine Algæ.
,,	Miss E. M. Mather.—Marine Algæ.
,,	Miss M. Bradley.—Marine Algæ.
,,	Miss G. Hanna.—Marine Algæ.
,,	Miss G. Wilkinson.—Marine Algæ.
,,	Miss D. Lamble.—Marine Algæ.
April 6*th to* 21*st.*	Miss F. Tozer.—General.
April 6*th to* 20*th.*	Miss O. V. Ellams.—General.
April 6*th to* 28*th.*	Miss G. V. Buchanan.—Plankton.
April 7*th to* 19*th.*	Miss R. Robbins.—Zostera.
,,	Miss R. C. Bamber.—Echinoderms.
April 7*th to* 14*th.*	Professor J. B. Farmer.—Mosses and Liverworts.
,,	Mr. R. J. Tabor.—Lichens.
,,	Miss H. Coburn.—Marine Algæ.
April 10*th to* 14*th.*	Miss H. C. New.—Marine Algæ.

April 8th to 15th.	Professor B. Moore.—Bio-Chemistry.
,,	Mr. E. Whitley.—Bio-Chemistry.
,,	Mr. J. Erik Hamilton.—General.
April 10th to 24th.	Professor F. J. Cole.—Educational.
,,	Mr. H. L. Hawkins.—Educational.
,,	Miss N. Eales.—Educational.
,,	Miss D. Crofts.—General.
,,	Mr. W. Baker.—General.
,,	Mr. F. D. Withers.—General.
,,	Mr. McKenzie.—General.
,,	Mr. M. Smith.—General.
,,	Mr. W. Kings.—General.
,,	Mr. W. H. Evans.—General.
April 13th to 19th.	Miss E. L. Gleave.—Archidoris
,,	Miss D. Hey.—Marine Algæ.
,,	Miss M. Shaw.—Marine Algæ.
June 5th to 18th.	Rev. S. Frappé.—Gills of Lamellibranchs.
June 5th to July 3rd.	Dr. P. Debaisieux.—Parasitology.
June 29th to July 11th.	Mr. H. S. Holden.—Himanthalia.

The "Tables"* in the Laboratory were occupied as follows :—

Liverpool University Table :—

Professor Herdman.	Mr. J. Erik Hamilton.	Miss M. Latarche.
Professor Harvey Gibson.	Mr. W. H. Evans.	Miss R. Robbins.
Professor Moore.	Miss M. Knight.	Miss R. C. Bamber.
Mr. R. D. Laurie.	Miss E. M. Blackwell.	Miss H. M. Duvall.
Mr. S. T. Burfield.	Miss E. L. Gleave.	Miss F. Tozer.

Liverpool Marine Biology Committee Tables :—

Mr. G. A. Herdman.	Dr. Paul Debaisieux.	Miss D. Hey.
Mr. H. T. Cubbon.	Mr. B. Sahni.	Miss L. Baker.
Mr. H. Storey.	Miss E. Richmond.	Miss P. McKie.
Rev. S. Frappé.	Miss R. Holden.	Miss G. V. Buchanan.
Mr. A. J. Nicholson.	Miss M. Jepps.	Miss M. Shaw.
Mr. R. J. Tabor.	Mr. M. C. Vyvyan.	Mr. E. Whitley.

Manchester University Table :—

Dr. Stuart Thomson.	Mr. G. Talbot.	Miss C. M. Lightbown.
Mr. G. H. Crabtree.	Miss A. Dixon.	Miss Williamson.
Mr. A. W. Summersgill.	Miss F. Lea.	Miss E. N. Cowell.

Birmingham University Table :—

Mr. H. Gordon Jackson. Mr. A. J. Nicholson.

University College, Reading, Table :—

Professor Cole.	Mr. F. D. Withers.	Mr. W. Kings.
M . H. L. Hawkins.	Mr. McKenzie.	Miss N. Eales.
Mr. W. Baker.	Mr. M. Smith.	Miss D. Crofts.

* Since the new research wing has been added several distinct apartments are generally available for the accommodation of the investigators assigned to any one of the University "Tables."

The following students of Liverpool University occupied the laboratory for periods varying from a fortnight to three weeks during the Easter vacation, and worked together under the supervision of Professor Harvey Gibson, Miss E. M. Blackwell, Miss M. Knight, Mr. R. Douglas Laurie and Mr. S. T. Burfield :—

Mr. R. J. Bruce.	Miss J. Curwen.	Miss E. M. Tate.
Mr. G. F. Sleggs.	Miss A. S. Meeson.	Miss E. Alexander.
Miss H. C. New.	Miss J. Price.	Miss E. M. Mather.
Miss G. A. Platt.	Miss J. Lord.	Miss M. Bradley.
Miss D. Thornton.	Miss D. Dobson.	Miss G. Hanna.
Miss N. Lodge.	Miss M. Tesh.	Miss D. Jones.
Miss M. M. Brew.	Miss H. V. Davies.	Miss I. L. Millican.
Miss E. Catlow.	Miss H. Clarke.	Miss G. Wilkinson.
Miss C. Whittaker.	Miss G. H. Wood.	Miss D. Lamble.

The following students of University College, Nottingham, attended a course in Marine Botany, conducted by Mr. H. S. Holden :—

Miss M. L. Straw.	Mr. E. S. Hillman.	Miss H. C. Bowser.
Miss C. N. M. Brett.	Miss D. Bexon.	Miss H. H. Maguire.
Miss J. M. McClatchie.		

Curator's Report.

Mr. Chadwick reports to me as follows on the various departments of the work :—

The Fish Hatchery.

"Hatching operations were begun this year on February 5th, an unusually early date, when a batch of 124,000 plaice eggs were put into the hatching boxes. The first batch of larvae, numbering 346,000, was set free on February 23rd. Quantities of eggs, exceeding half a million in number, were skimmed from the ponds on March 23rd and 30th respectively; and amongst the large batches of larvae set free by Professor Herdman from his steam yacht 'Runa' were three, numbering 749,700, 1,211,800 and 722,350 respectively. The spawning season closed on April 25th, the total number of eggs collected being 8,895,650.

"The Hatchery Record, giving the number of eggs collected, and of larval fish set free on the various days, is as follows:—

Eggs collected.	Date.	Larvae set free.	Date.
370,600	Feb. 5 to 12	346,050	Feb. 23
183,750	,, 16 to 20	162,750	March 7
33,600	,, 23	27,300	,, 11
469,350	,, 24 to March 3	397,950	,, 19
352,800	March 4 and 5	333,700	,, 23
426,300	,, 7 and 9	384,300	,, 27
741,600	,, 11 to 13	615,600	,, 31
530,250	,, 14 and 17	446,250	April 2
279,300	,, 19	248,850	,, 4
882,000	,, 20 and 23	749,700	,, 8
1,180,200	,, 24 to 28	1,211,800	,, 11
850,500	,, 30	722,350	,, 14
420,000	April 1	364,350	,, 15
409,500	,, 3	346,500	,, 17
525,000	,, 4 and 7	437,850	,, 18
252,000	,, 9	210,000	,, 20
508,000	,, 11 and 13	380,950	,, 22
348,600	,, 15 to 18	239,150	,, 28
132,300	,, 20 to 25	81,950	May 11
8,895,650	Total eggs.	7,707,350	Total larvæ.

"The early hatching season was probably due to the mild winter. The first plaice eggs were found on the surface of the pond on January 28th, and on February 3rd embryos at least a week old were obtained. In recent years the first fertilised eggs have generally been obtained on some date between the middle of February and the first week of March. So the present season is at least a fortnight earlier than usual.

Lobster Culture.

"The lobster hatching and rearing was this year undertaken by the Assistant Curator, Mr. T. N. Cregeen, and the increased success attained was due to his untiring and painstaking efforts. By personally interviewing local fishermen

he succeeded in obtaining 39 berried lobsters with nearly ripe eggs, a substantially larger supply than had hitherto been attainable. As the lobsters were brought in they were at once put into the pond, in which a number of hiding places, built of bricks and rough stone, had been prepared. The first newly-hatched larvae were taken from the pond on May 28th, and from that date to September 5th constant and sometimes large supplies of larvae were obtained, the total number being 40,500, an average of about 1,039 per adult lobster. Of these, 16,000 were set free in the first and second stages, and 24,500 were placed in the hatching boxes. By daily feeding with finely-minced fish, 'liver' of the edible crab and plankton, and the maintenance of scrupulous cleanliness in the hatching boxes, Mr. Cregeen succeeded in rearing 1,823 larvae to the fourth or 'lobsterling' stage (see fig. 3).

"As in all our previous experience of lobster culture, there was heavy mortality at the periods of ecdysis; but the loss due to cannibalism was this year very trifling. This was probably due to the plentiful supply of fresh food supplied. Quantities of plankton were taken with a coarse tow-net almost daily, and formed a considerable proportion of the food of the larvae. One thousand seven hundred and seventy-seven of the lobsterlings were set free at suitable parts of the coast line North and South of Port Erin Bay, and 46 were retained for further experiments in rearing. Of 13 lobsterlings hatched during the season of 1913 3 still survive, but their rate of growth has been slower than that of a young lobster of a similar age found in the pond several years ago.

The Aquarium.

"Until the oubreak of war the Aquarium attracted increasing numbers of visitors, and on August 1st the total number was 1,054 in advance of that of the corresponding

date last year. Thenceforward the numbers decreased; but, under the circumstances, the total for the year—12,031—is

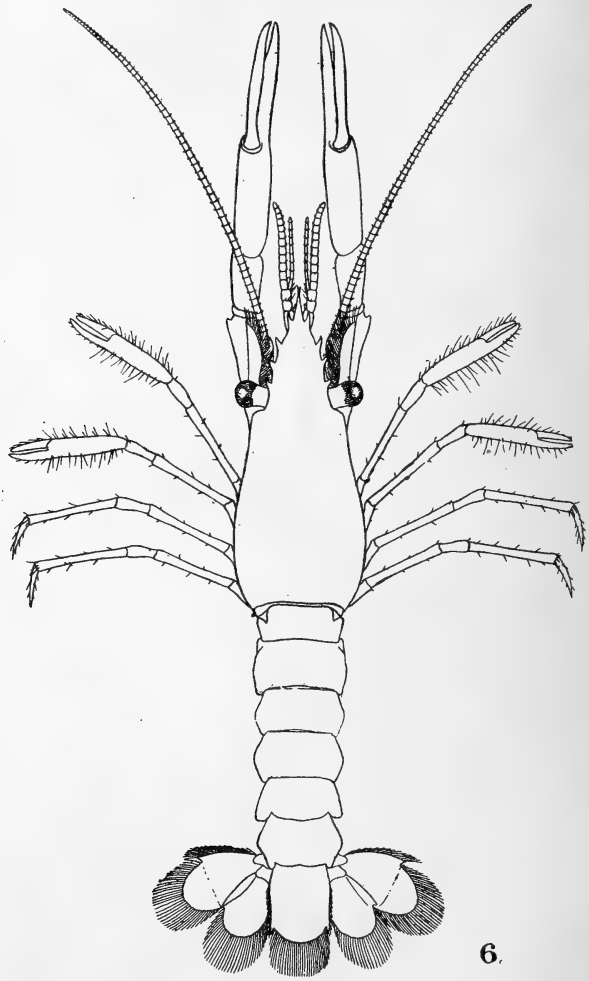


FIG. 3. Young Lobster or "Lobsterling" stage, after the fourth casting (magnified 5 times).

gratifying. It shows that under normal conditions the institution would have fully maintained its popularity. The

lobster culture in the Hatchery again attracted a large amount of attention on the part of the visitors, and there can be no doubt that it favourably influenced their numbers. During the latter half of the season a small series of dissections were made from common types such as the dog-fish, cod, octopus, etc., and exhibited in jars in the Aquarium. A few additions were made to the drawings which hang on the walls.

"The Curator resumed in October his lantern lectures and demonstrations to pupils of the Insular schools. Twenty-seven boys from the local Higher Education School have attended a systematic course of instruction in Nature Study on alternate Wednesday afternoons.

General.

"The only item of marine zoological interest which calls for notice is the extraordinary abundance of the medusa *Aurelia aurita* during the past summer. Large shoals, numbering hundreds of individuals, were seen on many occasions from the middle of June onwards, and it was not until the end of September that they finally disappeared."

H. C. Chadwick.

Other Reports on Work.

Professor Cole and his party of colleagues and students from Reading were chiefly occupied, as usual, in observing and collecting, and in making injected preparations of various invertebrata for their College Museum. Professor Cole writes to me:—"Our Easter party this year consisted of 9 persons, and the work was as usual largely educational."

Mr. Chadwick has sent me drawings of two remarkable cases of "twinning" in lobster larvae, which are reproduced in fig. 4. They occurred amongst the ordinary hatched larvae and were noticed while living. The figures are drawn

from the specimens preserved in formaline after their death. Both figures are magnified about 10 diameters.

Somewhat similar monstrosities to these have been described by Herrick* in the case of the American lobster (*Homarus americanus*), and that author's opinion is that "We have to do here with the fusion of two embryos which

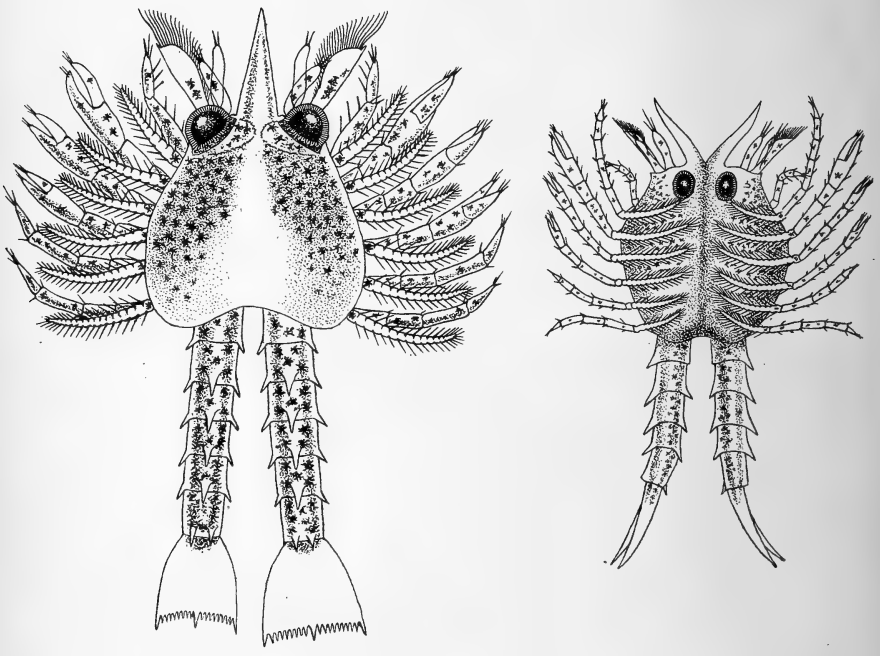


FIG. 4. Twinned Lobster larvae, hatched at Port Erin. × 10.

are practically distinct from the first." It is possible on the other hand that the doubling may be due to an injury or some abnormal condition affecting the ovum or embryo at an early stage. A pair of perfect twins are known to have been produced from one ovum in the case of the European lobster (Anderton).

* Bulletin of U.S. Fish Commission for 1895, p. 216.

Dr. Annie Porter has just published* a full account of the new parasitic Flagellate, *Herpetomonas patellae*, which she discovered in the limpet while working at Port Erin last winter. Herpetomonads are known as parasites in flies and other insects, but this is the first one to be found in a mollusc. The parasite was found in the alimentary canal and liver of eight per cent. of the limpets examined at Port Erin. Dr. Porter, Dr. Fantham, and others, have shown that other species of Herpetomonads found in some fleas are of pathogenic importance and may give rise to fatal diseases in rats, mice, or dogs, that may happen to swallow the fleas and so become infected with the parasite. When reduced to eating limpets it will be well to bear in mind that eight per cent. may contain this pathogenic organism, which, by the way, is closely related to the *Trypanosoma* which is the cause of " sleeping sickness." It is to be hoped that Dr. Porter will extend her investigations to other edible Mollusca so that eventually we may have some idea of the amount of risk, if any, that exists in eating uncooked such excellent food matters as the Oyster, the Mussel and the Scallop.

Miss H. M. Duvall, B.Sc., during the Easter vacation made some observations on the methods of feeding by means of ciliary currents in the Ascidian, *Clavelina lepadiformis*, which is found occasionally in rock pools on the Bradda side of Port Erin Bay. It was not easy to get the animals to expand fully and feed freely in captivity, but Miss Duvall succeeded in satisfying herself that a band of accumulated food particles and mucus forms in the interior of the branchial sac, neither along the endostyle, nor yet alongside the dorsal lamina (as described by Orton), but about the middle of the lateral walls midway between dorsal and ventral edges, and from that position is drawn posteriorly straight down into the oesophageal opening which terminates the branchial sac.

* Parasitology, November 9th, 1914.

Particles of carmine which were added to the water and were drawn in by the animal enabled this band to be seen very clearly extending from the oesophageal opening up to the middle of the branchial sac. Naturally this interesting observation ought to be repeated, both on the same and other species, and Miss Duvall hopes to return to the investigation at the earliest opportunity.

Dr. DEBAISIEUX, of Louvain, devoted his attention chiefly to a re-examination of *Merocystis kathae*, the Protozoan parasite found by Dr. Dakin in the renal organ of the whelk some years ago ; and he made a large collection of original drawings of the various stages of its life-history. He also examined various other molluscs and some fishes for Protozoan parasites.

Mr. S. T. BURFIELD, B.A., has continued his work on the remarkable pelagic Arrow-worm, *Sagitta bipunctata*, which he is investigating from every point of view with the object of producing an L.M.B.C. Memoir on the subject. He was mainly occupied at Port Erin in collecting material from the plankton samples and in fixing the best specimens for sectioning with a view to histological work. Attempts were also made to keep the adult animals alive in the laboratory with a view to obtaining eggs and early stages, but without much success so far. The work will be continued in all probability during next Easter vacation.

Miss R. C. BAMBER, M.Sc., and Mr. BURFIELD have started a series of observations on living Echinoderms with the view of ascertaining more precisely the direction of the water current through the madreporite. It is intended to repeat and extend these experiments in order to ascertain whether there are indications of an excretory function in the water-vascular system.

Mr. H. G. JACKSON, M.Sc., acted as my Assistant in the plankton investigation from the yacht during the Easter vacation. The results obtained will be given in full in the next

Lancashire Sea Fisheries Report and so need not be further referred to here. The rest of Mr. Jackson's time at Port Erin was occupied in collecting further material for his work on the larvae of Higher Crustacea—which are of importance not only for their own sake but also in relation to the feeding of fishes (see fig. 5). He examined the various plankton gatherings

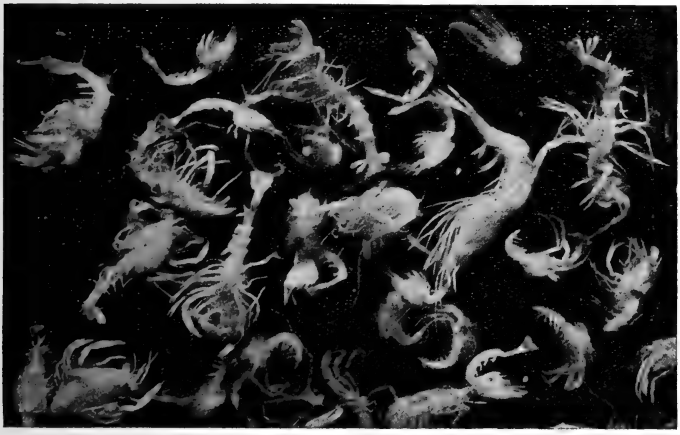


Fig. 5.—Larval Decapod Crustacea from the stomachs of Mackerel.
[From a Photo. by Mr. A. Scott.

with the object of tracing the young stages of crabs and lobsters, shrimps and prawns, and other allied animals throughout their life-histories, and also in their distribution over the district and throughout the year. Mr. Jackson published a first report on this subject in the Lancashire Sea Fisheries Report for 1912 and a second note in last year's Report (for 1913) and has in preparation a more detailed account.

Botanical Notes.

In addition to the courses of instruction on marine Algæ referred to above, several senior students spent some time in investigating the life-histories of selected seaweeds. Apart

from this more general work some experiments were made with a view to collecting data for an enquiry into the causes underlying certain marked features of algal distribution.

Miss L. Baker and Miss E. M. Blackwell spent some time investigating the lichens of the littoral and other regions, and produced a preliminary list. The mosses and liverworts of the district were collected and classified by Professor J. B. Farmer, F.R.S., and Mr. R. J. Tabor, of London.

It is hoped that several papers at present in course of preparation will be published as an outcome of the work done by the Botanical School at Port Erin during the Easter vacation, 1914.

Bio-Chemical Researches.

The researches carried out on the Bio-Chemical side during the year 1914 have been concerned with two main problems. In the first place the nutrition and metabolism of marine animals have been studied over prolonged intervals by means of the respiratory exchanges; and secondly the variations in alkalinity of sea-water at various periods in the year have been investigated. The second problem is related to the first, because the variations are mainly produced by changes in the balance between photo-synthetic activity of plants, and the metabolic oxidising activities of animals.

The results of the metabolic experiments have been published in two papers which appeared in the Transactions of the Liverpool Biological Society, Vol. XXVIII., 1914, viz. :—

1. The nutrition and metabolism of marine animals: The rate of oxidation and output of carbon-dioxide in marine animals in relation to the available supply of food in sea-water, by Professor B. Moore, Edward S. Edie and Edward Whitley.

2. The nutrition and metabolism of marine animals: The

effects in the lobster of prolonged abstention from food in captivity, by Professor B. Moore and George A. Herdman.

The main results may be summarised as follows :—

1. It is, in our view, definitely settled by experiment, that sea-water does not contain any appreciable amount of organic matter capable of acting as a nutrient medium for aquatic animals.

2. We have also obtained, over longer periods, figures indicating the rates of oxidation in larger marine animals, and have definitely shown that the preponderating amount of food consumed by such animals is utilised for increases of the animal by growth and for sexual reproduction, and that but a small fraction is oxidised for the metabolic needs of the animal in other activities than growth and reproduction.

3. Lobsters provided daily with a sufficient supply of fresh sea-water can be kept alive without food during a period of over seven months.

4. The live body-weight of such lobsters does not diminish during such a prolonged period of inanition. But while the actual weight of inorganic matter remains constant, the total dry weight and total organic weight are markedly diminished, and as a result the *percentage* of inorganic matter in the dry weight becomes increased.

5. As a result of the inanition the total oxidisable organic matter may fall to considerably less than one-half of the initial amount.

6. At the commencement of the period, protein, fat and carbohydrate are oxidised almost equally; later the carbohydrate becomes exhausted and, although fat is still present, nearly all the oxidation falls upon the proteins.

7. There is a satisfactory correspondence between the amount of oxygen consumed by the animals throughout the period and the amount of organic matter disappearing. The oxygen consumed corresponds very closely to that required

for oxidation of the organic matter disappearing, so that there is no reason to suppose that the animal utilises any dissolved organic matter which might hypothetically be present in the sea-water.

8. The rate of oxidation is throughout a slow one representable by 120 to 130 milligrams per lobster of 220-300 grams at the commencement, and dropping to about half this quantity towards the end of the experiment. This amount corresponds to a little over one-tenth of a gram of protein or carbohydrate daily.

The investigation of the alkalinity of sea-water has now been completed and the results will shortly appear. They show interesting chemical relationships corresponding to the known seasonal variations in the marine flora and fauna.

S.Y. "Runa," 1913.—Foraminifera.

It will be remembered that in last year's Report, in connection with the work done from the yacht "Runa," it was mentioned that a couple of dozen canvas bags of dredged sand, mud and other deposits, brought up from the bottom of the sea at various localities on the West Coast, had been sent for investigation to Mr. E. Heron-Allen and Mr. A. Earland, who are at present engaged on a monograph on the British Foraminifera. Mr. Heron-Allen has been good enough to send me the following note on the results obtained during the past year and supplementing what he enabled me to state in our last Report :—

"Since the date of the last Report the examination of the samples submitted to us has been proceeded with uninterruptedly, and is now approaching completion.

"The later samples have adequately fulfilled the promise of the first four, the majority of the Stations having furnished very extensive lists and affording valuable contributions to

the list of species recorded in British waters. Over 350 species and varieties have been identified.

"Station 20, 'Between Ru Ruag and Carr Point (off Gairloch), 20 fathoms,' has yielded remarkable quantities of fine specimens of *Cornuspira foliacea* (Philippi) and *Jaculella acuta* and *obtusa*, Brady, and of the rare *Ammodiscus charoides* (Jones and Parker).

"Several species have occurred which will require very careful diagnosis, and several species new to Science may be expected. Perhaps the most interesting discovery has been that of *Spiroloculina acutimargo*, Brady, var. *concava*, Wiesner MS., of which the only hitherto recorded specimens have been sent us by the author from Eiland Pomo in the Adriatic (195 fathoms) and which we have found at Station 4 (Loch Sunart, 12 fathoms).

"The last Station dredged by Professor Herdman was 'off Bradda, Isle of Man, 20 fathoms,' of which, at his request, we append a preliminary list of the species identified up to date.

"Station 23.—Material.—1 lb. 10 oz. of muddy shell and algal débris with large shells of bivalve Mollusca. Residue after removal of shells and stones, 10 oz. Residue after washing, 100 c.c. full of fragments of small crustacea. 112 species from the floatings."

List of species of Foraminifera dredged. Off Bradda Head, 20 fathoms.

1.—	Biloculina depressa, d'O.	Common.
2.—	,, bulloides, d'O.	Very rare.
3.—	,, elongata, d'O.	Frequent.
4.—	Spiroloculina planulata (Lam.)	Rare.
5.—	,, excavata, d'O.	Frequent.
6.—	Miliolina oblonga, d'O.	Very rare.
7.—	,, circularis (Born.)	Frequent.
8.—	,, subrotunda (Montagu)	Very rare.
9.—	,, seminuda (Reuss)	Very rare.
10.—	,, tricarinata (d'O.)	Very rare.
11.—	,, trigonula (Lam.)	Common.
12.—	,, seminulum (Linné)	Frequent.
13.—	,, candeiana (d'O.)	Very rare.
14.—	,, sclerotica (Karrer)	Rare.

15.—Miliolina fusca, Brady	Very rare.
16.— ,, laevigata (d'O.)	Rare.
17.— ,, bicornis (W. & J.)	Very rare.
18.— ,, brongniartii (d'O.)	Very rare.
19.— ,, rotundata (Montagu)	Very rare.
20.— ,, bosciana (d'O.)	Very rare.
21.— ,, pygmaea (Reuss)	Very rare.
22.—Ophthalmidium carinatum, B. & W.	Very rare.
23.—Cornuspira selseyensis, H.-A. & E.	Very rare.
24.— ,, involvens (Reuss)	Very rare.
25.—Bathysiphon argenteus, H.-A. & E.	Very rare.
26.—Haplophragmium pseudospirale (Will.)	Very rare.
27.— ,, canariense (d'O.)	Rare.
28.—Ammodiscus gordialis (J. & P.)	Very rare.
29.—Trochammina squamata J. & P.	Very rare.
30.—Textularia gramen, d'O.	Very rare.
31.— ,, conica, d'O.	Very rare.
32.—Verneuilina polystropha (Reuss)	Very rare.
33.—Spiroplecta biformis (P. & J.)	Very rare.
34.— ,, wrightii, Silvestri	Very rare.
35.—Bulimina pupoides, d'O.	Common.
36.— ,, elegans, d'O.	Common.
37.— ,, marginata, d'O.	Common.
38.— ,, elegantissima, d'O.	Very rare.
39.— ,, squamigera, d'O.	Very rare.
40.— ,, fusiformis, Will.	Very rare.
41.—Bolivina punctata, d'O.	Very rare.
42.— ,, textilarioides, Reuss	Very rare.
43.— ,, dilatata, Reuss	Rare.
44.— ,, plicata, d'O.	Rare.
45.— ,, variabilis (Will.)	Rare.
46.—Cassidulina laevigata, d'O.	Very rare.
47.— ,, crassa, d'O.	Very rare.
48.— ,, subglobosa, Brady	Very rare.
49.—Lagena globosa (Mont.)	Very rare.
50.— ,, lineata (Will.)	Very rare.
51.— ,, costata (Will.)	Frequent.
52.— ,, hexagona, Will.	Frequent.
53.— ,, squamosa (Mont.)	Frequent.
54.— ,, semistriata, Will.	Very rare.
55.— ,, striata (d'O.)	Very rare.
56.— ,, sulcata (W. & J.)	Very rare.
57.— ,, williamsoni (Alcock)	Common.
58.— ,, clavata (d'O.)	Frequent.
59.— ,, laevigata (Reuss)	Rare.
60.— ,, lucida (Will.)	Frequent.
61.— ,, marginata (Walker & Boys)	Very rare.
62.— ,, ornata, Will.	Very rare.
63.— ,, orbignyana (Seguenza)	Very rare.
64.— ,, apiculata, Reuss	Common.
65.— ,, bicarinata (Terquem)	Rare.
66.—Nodosaria pyrula, d'O.	Very rare.
67.— ,, communis, d'O.	Very rare.
68.— ,, scalaris (Batsch.)	Very rare.
69.— ,, obliqua (Batsch.)	Very rare.
70.— ,, consobrina (d'O.)	Very rare.
71.—Cristellaria crepidula (Fichtel & M.)	Very rare.
72.— ,, gibba, d'O.	Very rare.

73.—Cristellaria rotulata (Lam.)	Very rare.
74.—Polymorphina lactea (W. & J.)	Frequent.
75.— „ compressa, d'O.	Rare.
76.— „ rotundata (Born)	Very rare.
77.— „ gibba, d'O.	Very rare.
78.— „ sororia, Reuss	Frequent.
79.— „ amygdaloides, Reuss	Very rare.
80.—Uvigerina angulosa, Will.	Very rare.
81.—Globigerina bulloides, d'O.	Very rare.
82.— „ inflata, d'O.	Very rare.
83.—Spirillina vivipara, Ehrenberg	Very rare.
84.— „ limbata, Brady	Very rare.
85.—Patellina corrugata, Will.	Frequent.
86.—Discorbina nitida, Will.	Rare.
87.— „ mamilla, Will.	Common.
88.— „ rosacea, d'O.	Common.
89.— „ praegeri, H.-A. & E.	Common.
90.— „ turbo (d'O.)	Frequent.
91.— „ globularis, d'O.	Frequent.
92.— „ obtusa, d'O.	Common.
93.—Planorbulina mediterranensis, d'O.	Common.
94.—Truncatulina lobatula (W. & J.)	Rare.
95.— „ variabilis, d'O.	Very rare.
96.— „ ungeriana, d'O.	Very rare.
97.—Pulvinulina repanda (F. & M.)	Very rare.
98.— „ oblonga (Will.)	One.
99.— „ haliotidea, H.-A. & E.	Very rare.
100.—Rotalia beccarii (Linné)	Very rare.
101.— „ orbicularis, d'O.	Common.
102.— „ perlucida, H.-A. & E.	Very rare.
103.—Gypsina inhaerens (Schultze)	Very rare.
104.—Nonionina depressula (W. & J.)	Common.
105.— „ umbilicatula (Mont.)	Very rare.
106.— „ asterizans (F. & M.)	Very rare.
107.— „ stelligera (?) d'O.	Very rare.
108.— „ pauperata, B. & W.	Frequent.
109.—Polystomella striatopunctata (F. & M.)	Frequent.
110.— „ crispa, Linné	Very rare.
111.— „ macella (F. & M.)	Frequent.
112.—Operculina ammonoides (Gron.)	Very rare.

As this locality—off Bradda Head—is in our home-waters at Port Erin, liable to be dredged by students from the Biological Station at any time, I thought it advisable to ask Mr. Heron-Allen to allow me to print the above provisional list at the earliest opportunity so that it might be available for consultation by our workers.

Plankton Investigation at Port Erin.

In connection with the "intensive" study of the minute life of the sea which has been carried on for some years by Professor Herdman, Mr. Andrew Scott and Miss H. Mabel

Lewis, we find that 168 collections of the surface plankton in Port Erin Bay were made with the usual coarse and fine tow-nets during the ten months—January to the end of October, 1914. If we combine the volume of the catch of the coarse and fine nets and regard the result as a fairly accurate representation of the amount of pelagic life present at the time the hauls were made, we are able to give the following summary of the distribution throughout the period mentioned. The amount of plankton present in the Bay in January and February was very small. The average volume for eight hauls in January was 2·5 c.c. Seven hauls in February gave an average of 3·2 c.c. March showed a distinct increase with an average of 6·5 c.c. for eight hauls. In April there was a further marked increase, rising to 24 c.c. at the middle of the month and ending with 70 c.c. on April 30th: the average for nine hauls was 34·7 c.c. The first collection with the coarse and fine nets in May was taken on the 4th, when the combined volume of the two hauls was 88·5 c.c., the maximum quantity collected during 1914. Three days later it fell to 61·5 c.c. The plankton was found to have still further diminished on May 12th, and the volume of the catch made by the two nets then amounted to 26·2 c.c.: the average for eight hauls during May was 34·8 c.c. In June, although there was one large haul of 41 c.c. on the 11th, the average for nine collections was only 24·5 c.c. The amount taken by the two nets at the beginning of July was 21·2 c.c., at the end of the month it had decreased to 4·5 c.c., and the average for nine hauls was 12·7 c.c. There was a slight increase at the beginning of August compared with the conditions at the end of July, but this was not maintained. The quantity present during the last two weeks of the month fell from 11·3 c.c. to 2·6 c.c., and the average for nine hauls was 10·8 c.c. September also commenced with an increase compared with the end of the previous month. The volume diminished towards the middle of the month to 4·4 c.c. It

rapidly increased again, rising to 24·5 c.c. on the 25th. : the average for eight hauls amounted to 12·7 c.c. The plankton remained fairly abundant during the first three weeks of October, but a decrease from 14·6 c.c. on the 18th to 5·2 c.c. and 5·7 c.c. on the 23rd and 28th brought the average down to 11·3 c.c. for nine hauls.

The spring maximum was comparatively short and well defined. Although the greatest amount was obtained on May 4th, we may safely regard the period to have extended over the fortnight—April 24th to May 7th. The collection taken on April 20th contained 22·7 c.c. of plankton. On May 12th 26·2 c.c. were taken. Five collections taken between April 24th and May 7th contained an average of 69·6 c.c. each. The summer maximum was much more limited in duration than the spring one. The volume taken by the coarse and fine nets on June 11th amounted to 41 c.c. A few days before and after that date the volume collected amounted to 20 c.c. and 23 c.c. The autumn maximum occurred during the later part of September. The combined sample taken on September 25th contained 24·5 c.c. of plankton. This was the greatest volume collected by the two nets between the date of the summer maximum and the end of October.

Biddulphia mobiliensis (fig. 6) was never completely absent in any of the ten months. It was most abundant in March with a total of 287,700 for eight hauls. After March the numbers diminished rapidly, and only 50 were observed in the whole of the August collections. The *sinensis* form also attained its maximum in March, but was apparently absent in June, July and August. *Coscinodiscus concinnus* arrived at its maximum in April and the total for nine hauls was nearly $2\frac{1}{2}$ millions. The visitation was prolonged into June, but none were observed in July and August. *Lauderia*, *Rhizosolenia shrubsolii*, and *Chaetoceras debile*, *decipiens*, *teres* and *sociale* were the most abundant diatoms in the spring maximum. The four species of *Chaetoceras* mentioned were represented

by a total of over 166 millions for the eight hauls in May. *Chaetoceras decipiens* and *Biddulphia mobiliensis* were the only two diatoms that were never completely absent from the Bay plankton of the ten months. *Guinardia flaccida, Rhizosolenia shrubsolii* and *R. semispina* were the most conspicuous

FIG. 6. Plankton showing *Biddulphia mobiliensis* and *B. sinensis.*

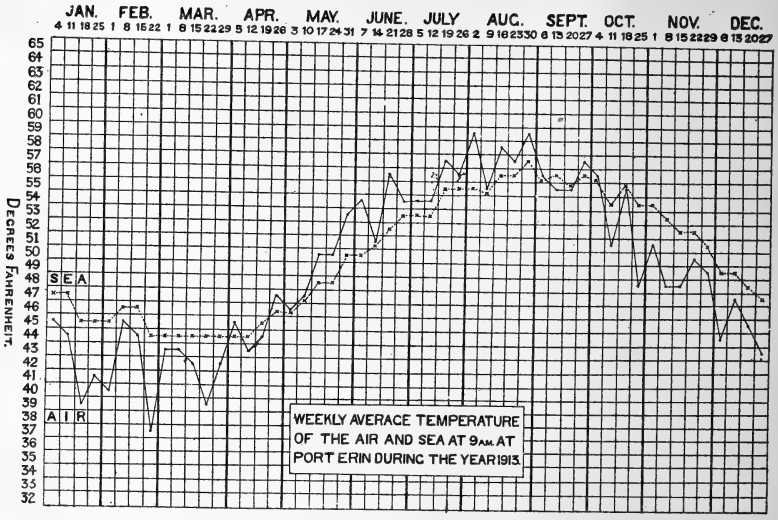
diatoms in the summer maximum. It was estimated that 3 millions of the first and over 17 millions of the second were present in the nine hauls of the June plankton. The diatom minimum occurred in August. Only three species were observed in the nine collections and the total number of individuals was 3,420.

Ceratium tripos was present throughout the ten months. The maximum of the Dinoflagellata was reached in May with nearly 300,000 in eight hauls. *Noctiluca* appeared to have been unusually abundant this year. It was present every month except August. It was estimated that the plankton

collected during May contained 28,680 *Noctiluca*. This represents the maximum, and it is considerably earlier than has been our experience of the organism in the Welsh and Lancashire coastal waters. *Noctiluca* is usually to be found in the plankton of North Wales from July onwards, but it is only in September that we find it to be abundant in Barrow Channel and off Walney Island. *Sagitta* was present throughout the ten months, but there was no well-defined maximum. The eight May collections gave 868 specimens, nine in June 828 and nine in August 847. Fish eggs were also represented in every one of the ten months ; and their maximum occurred in April.

The pelagic Copepoda arrived at their maximum in September. It was estimated that the eight collections with the coarse and fine nets contained 557,620 specimens, chiefly *Paracalanus* and *Oithona*, but *Pseudocalanus*, *Acartia*, *Calanus*, *Isias*, *Temora* and *Centropages* were also represented. *Calanus* reached its maximum in August, *Isias* in September when it was quite unusually abundant, *Pseudocalanus* in June, *Paracalanus* in September, *Temora* in April, *Centropages* in August, *Acartia* in July, and *Oithona* in July. *Anomalocera* made a sudden invasion in April, when the total for nine hauls was 5,500. It rapidly decreased during the next two months, and was not observed after the end of June. The other pelagic species *Microcalanus*, *Candacia*, *Metridia* and *Parapontella* appeared to make short spasmodic visits into the Bay from time to time. Eight *Candacia* were found in the January plankton and twenty-two in October. Our experience of *Metridia* in the Bay collections indicates that it lives below the surface. It is frequently observed in vertical hauls although it is absent from the surface collections taken at the same time.

A fuller account of the Bay plankton, along with the results of the Easter work at sea carried out on the yacht "Runa," will be published later on in the Lancashire Sea-Fisheries Annual Report.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DEGREES FAHRENHEIT.
SEA
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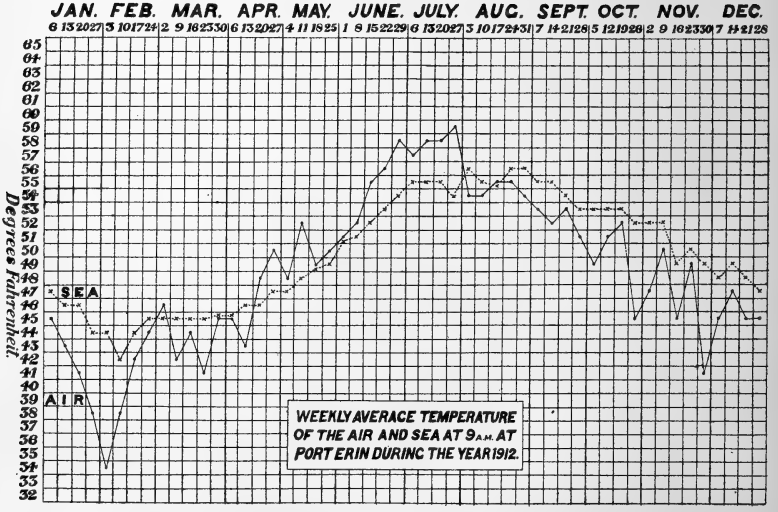
WEEKLY AVERACE TEMPERATURE OF THE AIR AND SEA AT 9AM AT PORT ERIN DURINC THE YEAR 1913.


JAN. FEB. MAR. APR. MAY. JUNE. JULY. AUG. SEPT. OCT. NOV. DEC.
Degrees Fahrenheit.
SEA
AIR
WEEKLY AVERACE TEMPERATURE OF THE AIR AND SEA AT 9AM AT PORT ERIN DURINC THE YEAR 1912.

The diagram of sea and air temperatures for 1914, compiled by Mr. Chadwick from his daily records, is not yet completed ; but those for the two preceding years, 1912 and 1913, are inserted here to show the general similarity of the two curves along with a few points of divergence, and to demonstrate again the manner in which the temperature of the sea lags behind that of the air in both winter and summer. The annexed charts show clearly the points of agreement and of difference between the two years.

L.M.B.C. Memoirs.

Since our last report was published, Memoir XXII., on the Echinoderm Larvæ of Port Erin, by Mr. H. C. Chadwick, has been issued to the public. Miss E. L. Gleave, M.Sc., has nearly completed her Memoir on Doris, the Sea-lemon ; Mr. Burfield is far advanced with Sagitta ; and still others are in preparation.

The following shows a list of the Memoirs already published or arranged for :

I. Ascidia, W. A. Herdman, 60 pp., 5 Pls.
II. Cardium, J. Johnstone, 92 pp., 7 Pls.
III. Echinus, H. C. Chadwick, 36 pp., 5 Pls.
IV. Codium, R. J. H. Gibson and H. Auld, 3 Pls.
V. Alcyonium, S. J. Hickson, 30 pp., 3 Pls.
VI. Lepeophtheirus and Lernæa, A. Scott, 5 Pls.
VII. Lineus, R. C. Punnett, 40 pp., 4 Pls.
VIII. Plaice, F. J. Cole and J. Johnstone, 11 Pls.
IX. Chondrus, O. V. Darbishire, 50 pp., 7 Pls.
X. Patella, J. R. A. Davis and H. J. Fleure, 4 Pls.
XI. Arenicola, J. H. Ashworth, 126 pp., 8 Pls.
XII. Gammarus, M. Cussans, 55 pp., 4 Pls.
XIII. Anurida, A. D. Imms, 107 pp., 8 Pls.
XIV. Ligia, C. G. Hewitt, 45 pp., 4 Pls.
XV. Antedon, H. C. Chadwick, 55 pp., 7 Pls.

XVI. Cancer, J. Pearson, 217 pp., 13 Pls.
XVII. Pecten, W. J. Dakin, 144 pp., 9 Pls.
XVIII. Eledone, A. Isgrove, 113 pp., 10 Pls.
XIX. Polychaet Larvæ, F. H. Gravely, 87 pp., 4 Pls.
XX. Buccinum, W. J. Dakin, 123 pp., 8 Pls.
XXI. Eupagurus, H. G. Jackson, 88 pp., 6 Pls.
XXII. Echinoderm Larvæ, H. C. Chadwick, 40 pp., 9 Pls.
XXIII. Doris, E. L. Gleave.
Sagitta, S. T. Burfield.
Actinia, J. A. Clubb.
Zostera, R. Robbins.
Halichondria and Sycon, A. Dendy.
Oyster, W. A. Herdman and J. T. Jenkins.
Sabellaria, A. T. Watson.
Ostracod (Cythere), A. Scott.
Asterias, H. C. Chadwick.
Botrylloides, W. A. Herdman.

In addition to these, it is hoped that other Memoirs will be arranged for, on suitable types, such as *Pontobdella*, a Cestode, a Nematode, a Cirripede, and a Pycnogonid.

As the result of a slight fire in the Zoology Department of the University, a portion of the stock of L.M.B.C. Memoirs has been partially destroyed. There are a certain number of damaged copies of some of the Memoirs which are stained or singed externally, but are still quite usable, and are suitable for laboratory work. The Committee has decided to offer these at prices ranging according to the condition from one-half to one-fourth of the published prices, as follows:—Memoir I., Ascidia, 6d. to 9d.; VI., Lepeophtheirus and Lernæa, 6d. to 1s.; VII., Lineus, 6d. to 1s.; XIII., Anurida, 1s. to 2s.; XIV., Ligia, 6d. to 1s.; XV., Antedon, 6d. to 1s. 3d.

Orders for these damaged copies should be sent to Professor Herdman, the University, Liverpool. New copies of any of the Memoirs should be ordered from Williams & Norgate.

We append to this Report :—

(A) The usual Statement as to the constitution of the L.M.B.C., and the Laboratory Regulations—with Memoranda for the use of students ;

(B) The Hon. Treasurer's Report, List of Subscribers, and Balance Sheet for the year.

Pennatula phosphorea, alive in a jar of sea-water : natural size.
[Photo. by Prof. R. Newstead.

Appendix A.

THE LIVERPOOL MARINE BIOLOGY COMMITTEE (1914).

His Excellency the Right Hon. Lord Raglan, Lieut.-Governor of the Isle of Man.

Rt. Hon. Sir John Brunner, Bart.

Prof. R. J. Harvey Gibson, M.A., F.L.S., Liverpool.

Mr. W. J. Halls, Liverpool.

Prof. W. A. Herdman, D.Sc., F.R.S., F.L.S., Liverpool. Chairman of the L.M.B.C., and Hon. Director of the Biological Station.

Mr. P. M. C. Kermode, Ramsey, Isle of Man.

Prof. Benjamin Moore, F.R.S., Liverpool.

Sir Charles Petrie, Liverpool.

Mr. E. Thompson, Liverpool, Hon. Treasurer.

Mr. A. O. Walker, F.L.S., J.P., formerly of Chester.

Mr. Arnold T. Watson, F.L.S., Sheffield.

Curator of the Station—Mr. H. C. Chadwick, A.L.S.

Assistant—Mr. T. N. Cregeen.

Junior Assistant—Mr. T. M. Cregeen.

CONSTITUTION OF THE L.M.B.C.

(Established March, 1885.)

I.—The OBJECT of the L.M.B.C. is to investigate the Marine Fauna and Flora (and any related subjects such as submarine geology and the physical condition of the water) of Liverpool Bay and the neighbouring parts of the Irish Sea and, if practicable, to establish and maintain a Biological Station on some convenient part of the coast.

II.—The COMMITTEE shall consist of not more than 12 and not less than 10 members, of whom 3 shall form a quorum ; and a meeting shall be called at least once a year for the purpose of arranging the Annual Report, passing the Treasurer's accounts, and transacting any other necessary business.

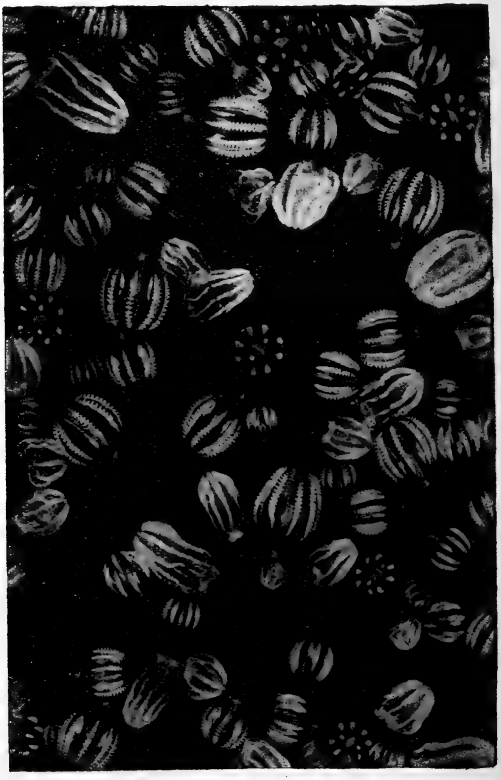
III.—During the year the AFFAIRS of the Committee shall be conducted by an HON. DIRECTOR, who shall be Chairman of the Committee, and an HON. TREASURER, both of whom shall be appointed at the Annual Meeting, and shall be eligible for re-election.

IV.—Any VACANCIES on the Committee, caused by death or resignation, shall be filled by the election at the Annual Meeting of those who, by their work on the Marine Biology of the district, or by their sympathy with science, seem best fitted to help in advancing the work of the Committee.

V.—The EXPENSES of the investigations, of the publication of results, and of the maintenance of the Biological Station shall be defrayed by the Committee, who, for this purpose, shall ask for subscriptions or donations from the public, and for grants from scientific funds.

VI.—The BIOLOGICAL STATION shall be used primarily for the Exploring work of the Committee, and the SPECIMENS collected shall, so far as is necessary, be placed in the first

instance at the disposal of the members of the Committee and other specialists who are reporting upon groups of organisms; work places in the Biological Station may, however, be rented by the week, month, or year to students and others, and duplicate specimens which, in the opinion of the Committee, can be spared may be sold to museums and laboratories.



A remarkable Plankton haul of the Ctenophore, *Pleurobrachia pileus*: natural size.

[Photo. by Mr. A. Scott.

LIVERPOOL MARINE BIOLOGICAL STATION

AT

PORT ERIN.

General Regulations.

I.—This Biological Station is under the control of the Liverpool Marine Biology Committee, the executive of which consists of the Hon. Director (Prof. Herdman, F.R.S.) and the Hon. Treasurer (Mr. E. Thompson).

II.—In the absence of the Director, and of all other members of the Committee, the Station is under the temporary control of the Resident Curator (Mr. H. C. Chadwick), who will keep the keys, and will decide, in the event of any difficulty, which places are to be occupied by workers, and how the tanks, boats, collecting apparatus, &c., are to be employed.

III.—The Resident Curator will be ready at all reasonable hours and within reasonable limits to give assistance to workers at the Station, and to do his best to supply them with material for their investigations.

IV.—Visitors will be admitted, on payment of a small specified charge, at fixed hours, to see the Aquarium and Museum adjoining the Station. Occasional public lectures are given in the Institution by members of the Committee.

V.—Those who are entitled to work in the Station, when there is room, and after formal application to the Director, are :—(1) Annual Subscribers of one guinea or upwards to the funds (each guinea subscribed entitling to the use of a work place for three weeks), and (2) others who are not annual subscribers, but who pay the Treasurer 10s. per week for the

accommodation and privileges. Institutions, such as Universities and Museums, may become subscribers in order that a work place may be at the disposal of their students or staff for a certain period annually; a subscription of two guineas will secure a work place for six weeks in the year, a subscription of five guineas for four months, and a subscription of £10 for the whole year.

VI.—Each worker is entitled to a work place opposite a window in the Laboratory, and may make use of the microscopes and other apparatus, and of the boats, dredges, tow-nets, &c., so far as is compatible with the claims of other workers, and with the routine work of the Station.

VII.—Each worker will be allowed to use one pint of methylated spirit per week free. Any further amount required must be paid for. All dishes, jars, bottles, tubes, and other glass may be used freely, but must not be taken away from the Laboratory. Workers desirous of making, preserving, or taking away collections of marine animals and plants, can make special arrangements with the Director or Treasurer in regard to bottles and preservatives. Although workers in the Station are free to make their own collections at Port Erin, it must be clearly understood that (as in other Biological Stations) no specimens must be taken for such purposes from the Laboratory stock, nor from the Aquarium tanks, nor from the steam-boat dredging expeditions, as these specimens are the property of the Committee. The specimens in the Laboratory stock are preserved for sale, the animals in the tanks are for the instruction of visitors to the Aquarium, and as all the expenses of steam-boat dredging expeditions are defrayed by the Committee, the specimens obtained on these occasions must be retained by the Committee (*a*) for the use of the specialists working at the Fauna of Liverpool Bay, (*b*) to replenish the tanks, and (*c*) to add to the stock of duplicate animals for sale from the Laboratory.

VIII.—Each worker at the Station is expected to prepare a short report upon his work—not necessarily for publication—to be forwarded to Prof. Herdman before the end of the year for notice, if desirable, in the Annual Report.

IX.—All subscriptions, payments, and other communications relating to finance, should be sent to the Hon. Treasurer. Applications for permission to work at the Station, or for specimens, or any communications in regard to the scientific work should be made to Professor Herdman, F.R.S., University, Liverpool.

Over 50 Dog-fishes caught in an hour with hook and line from the "Runa," in Loch Sunart.

Memoranda for Students and Others working at the Port Erin Biological Station.

Post-graduate students and others carrying on research will be accommodated in the small work-rooms of the ground floor laboratory and in those on the upper floor of the new research wing. Some of these little rooms have space for two persons who are working together, but researchers who require more space for apparatus or experiments will, so far as the accommodation allows, be given rooms to themselves.

Undergraduate students working as members of a class will occupy the large laboratory on the upper floor or the front museum gallery, and it is very desirable that these students should keep to regular hours of work. As a rule, it is not expected that they should devote the whole of each day to work in the laboratory, but should rather, when tides are suitable, spend a portion at least of either forenoon or afternoon on the sea-shore collecting and observing.

Occasional collecting expeditions are arranged under guidance either on the sea-shore or out at sea, and all undergraduate workers should make a point of taking part in these.

It is desirable that students should also occasionally take plankton gatherings in the bay for examination in the living state, and boats are provided for this purpose at the expense of the Biological Station to a reasonable extent. Students desiring to obtain a boat for such a purpose must apply to the Curator at the Laboratory for a boat voucher. Boats for pleasure trips are not supplied by the Biological Station, but must be provided by those who desire them at their own expense.

Students requiring any apparatus, glass-ware or chemicals from the store-room must apply to the Curator. Although the Committee keep a few microscopes at the Biological Station, these are mainly required for the use of the staff or for general

demonstration purposes. Students are therefore strongly advised, especially during University vacations, not to rely upon being able to obtain a suitable microscope, but ought if possible to bring their own instruments.

Students are advised to provide themselves upon arrival with the " Guide to the Aquarium " (price 3d.), and should each also buy a copy of the set of Local Maps (price 2d.) upon which to insert their faunistic records and other notes.

Occasional evening meetings in the Biological Station for lecture and demonstration purposes will be arranged from time to time. Apart from these, it is generally not advisable that students should come back to work in the laboratory in the evening ; and in all cases all lights will be put out and doors locked at 10 p.m. When the institution is closed, the key can be obtained, by those who have a valid reason for entering the building, only on personal application to Mr. Chadwick, the Curator, at 3, Rowany Terrace.

The Arrow-worm, *Sagitta bipunctata*.
[Photo. by Mr. A. Scott.

Appendix B.

HON. TREASURER'S STATEMENT.

The Balance Sheet for 1914 and list of subscriptions are shown in the following pages. Fortunately there is a small balance in hand which will be most useful, as there will be some heavy expenses to meet in the coming year.

One or more Memoirs will be published, and owing to the increasing number of students and workers at Port Erin, additional apparatus, books and sundry supplies will have to be bought.

The library at the Port Erin Biological Station is an extremely useful one and is for the use of students and other workers. Any donations for this, either of books or money, would be most welcome.

We have again received a grant from the Board of Agriculture for research work, which will be most useful next season.

Edwin Thompson,
Hon. Treasurer.

25, Sefton Drive,
Liverpool. *December* 16*th*, 1914.

SUBSCRIBERS.

	£	s.	d.
Browne, Edward T., B.A., Anglefield, Berkhamsted, Herts.	1	1	0
Brunner, Mond & Co., Northwich...	1	1	0
Brunner, Rt. Hon. Sir John, Bart., Silverlands, Chertsey	5	0	0
Brunner, J. F. L., M.P., 23, Weatherley Gardens, London, S.W.	2	2	0
Brunner, Roscoe, Belmont Hall, Northwich ...	1	1	0
Caton, Dr., 78, Rodney-street, Liverpool... ...	1	1	0
Chaudhuri, Dr. B. L., 120 Lower Circular-road, Calcutta	1	1	0
Clubb, Dr. J. A., Public Museums, Liverpool ...	0	10	6
Cole, Prof., University College, Reading	1	1	0
Crellin, John C., The late, J.P., Andreas, I. of Man	0	10	0
Dale, Sir Alfred, University, Liverpool	1	1	0
Dixon-Nuttall, F. R., J.P., F.R.M.S., Prescot ...	2	2	0
Gibson, Prof. R. J. Harvey, The University, Liverpool	1	1	0
Graveley, F. H., Indian Museum, Calcutta ...	0	10	6
Halls, W. J., 35, Lord-street, Liverpool	1	1	0
Herdman, Prof., F.R.S., University, Liverpool ...	2	2	0
Hewitt, David B., J.P., Northwich	1	1	0
Hickson, Prof., F.R.S., University, Manchester ...	1	1	0
Hill, Prof. J. P., University College, London ...	1	1	0
Holland, Walter, Carnatic Hall, Mossley Hill ...	1	1	0
Holt, Dr. Alfred, Dowsefield, Allerton	1	0	0
Holt, Mrs., Sudley, Mossley Hill, Liverpool ...	2	2	0
Holt, P. H., The late, Croxteth-gate, Sefton-park, Liverpool	1	1	0
Isle of Man Natural History Society	2	2	0
Jarmay, Gustav, Hartford, Cheshire	1	1	0
Livingston, Charles, 16, Brunswick-st., Liverpool	1	1	0
Manchester Microscopical Society...	1	1	0
Forward	£35	18	0

	£	s.	d.
Forward... ...	35	18	0
Meade-King, R. R., Tower Buildings, Liverpool...	0	10	0
Mond, R., Sevenoaks, Kent...	5	0	0
Monks, F. W., Warrington...	2	2	0
Mottram, V. H., The University, Liverpool ...	1	1	0
Muspratt, Dr. E. K., Seaforth Hall, Liverpool ...	5	0	0
O'Connell, Dr. J. H., Dunloe, Heathfield-road, Liverpool	1	1	0
Petrie, Sir Charles, Devonshire-road, Liverpool ...	1	1	0
Rathbone, Miss May, Backwood, Neston	1	1	0
Rathbone, Mrs., Green Bank, Allerton, Liverpool	2	0	0
Roberts, Mrs. Isaac, Thomery, S. et M., France ...	1	1	1
Robinson, Miss M. E., Holmfield, Aigburth, L'pool	1	0	0
Smith, A. T., 43, Castle-street, Liverpool... ...	1	1	0
Tate, Sir W. H., Woolton, Liverpool	2	2	0
Thompson, Edwin, 25, Sefton Drive, Liverpool ...	1	1	0
Thornely, Miss, Nunclose, Grassendale	0	10	0
Thornely, Miss L. R., Nunclose, Grassendale ...	2	2	0
Toll, J. M., 49, Newsham-drive, Liverpool ...	1	1	0
Walker, Alfred O., Ulcombe Place, Maidstone ...	3	3	0
Ward, Dr. Francis, 20, Park Road, Ipswich ...	2	2	0
Watson, A. T., Tapton-crescent Road, Sheffield ...	1	1	0
Whitley, Edward, Oxford	2	2	0
Yates, Harry, 75, Shudehill, Manchester	1	1	0
	£74	1	1
Deduct Subscriptions still unpaid *less* old Subscriptions received	6	1	0
	£68	0	1
Add Subscriptions for 1915	11	1	0
	£79	1	1

SUBSCRIPTIONS FOR THE HIRE OF "WORK-TABLES."

Victoria University, Manchester	£10	0	0
University, Liverpool	10	0	0
University, Birmingham	10	0	0
University College, London	2	2	0
Bedford College for Women, London	2	2	0
University College, Reading	2	2	0
	£36	6	0
Add old Subscriptions paid *less* Subscriptions still unpaid	5	16	0
	£42	2	0

THE LIVERPOOL MARINE BIOLOGY COMMITTEE.

Dr. IN ACCOUNT WITH EDWIN THOMPSON, HON. TREASURER. **Cr.**

1914.	£	s.	d.
To Printing and Stationery	24	6	3
,, Boat Hire	7	19	6
,, Books, Apparatus and Supplies at Port Erin Biological Station	53	13	10
, Postage, Carriage, &c.	6	18	2
,, Salary—Share of Curator's	85	0	0
,, ,, ,, Assistant's	27	6	0
,, Sundries	6	1	3
,, Balance in Hand, December, 1914	29	18	0
	£241	3	0

Endowed Invested Fund:—
British Workman's Public House Co. 90 Shares £1 each fully paid.

EDWIN THOMPSON,
HON. TREASURER.

Audited and found correct,
COOK & LEATHER,
Chartered Accountants.

LIVERPOOL, *December 16th, 1914.*

1914.	£	s.	d.
By Balance in hand, December, 1913	12	8	11
,, Subscriptions and Donations received	79	1	1
,, Amount received from Universities for hire of "Work Tables"	42	2	0
,, Laboratory Fees	13	7	0
,, Admissions to Aquarium, Share of	34	4	9
,, Interest on British Association (1896) Fund	37	10	10
,, Interest on Investment	3	9	11
,, Sale of Guides	8	5	0
,, ,, Post Cards and Annual Reports	0	15	8
,, ,, Specimens, Bottles, &c,	2	5	4
,, Bank Interest	7	12	6
	£241	3	0
Memoir Fund—Balance, December, 1913	130	1	6
By Sale of Memoirs	7	14	9
	£137	16	3
Cost of Echinoderm Larvae Memoir	13	10	3
	£124	6	0
Extension Fund:—Balance, as at December, 1913	£37	4	9
Grants from Board of Agriculture and Fisheries—			
Balance December, 1913	£175	0	0
Grant for 1914	100	0	0
	£275	0	0

REPORT ON THE INVESTIGATIONS CARRIED ON DURING 1914 IN CONNECTION WITH THE LANCASHIRE SEA-FISHERIES LABORATORY AT THE UNIVERSITY OF LIVERPOOL, AND THE SEA-FISH HATCHERY AT PIEL, NEAR BARROW.

EDITED BY

PROFESSOR W. A. HERDMAN, F.R.S.,
Honorary Director of the Scientific Work.

(With Plates, Charts and Text-Figures.)

CONTENTS.

INTRODUCTION.

The year 1914 falls into two very distinct and diverse parts. The first half, from January to July, was normal, and our investigations and results for that period are fairly comparable with those of other years; but the second half, from the beginning of August onwards, was in Sea-Fisheries, as in most other human affairs throughout Europe, quite

exceptional—and our work at sea ceased of necessity in September on the Fisheries Steamer, "James Fletcher," being taken over by the Admiralty for government service. The small tug-boat, chartered by the Committee to undertake some portion of the work of the "James Fletcher," and the Bailiffs' boats up and down the coast have been able to carry on some in-shore work; but hydrographic and planktonic observations at the off-shore stations, the fish-marking experiments, and several other series of records have all had to be abandoned since September.

It was hoped for some time that the use of some smaller steamer might be obtained for the purpose of carrying on at least the more important of the quarterly and monthly observations at sea, and the services of the steam-yacht "Runa"—a very suitable vessel for such work—were freely placed at the disposal of the Committee. Before, however, this offer could be definitely accepted, it was felt by all concerned that the difficulties and risks, resulting from the then state of war at sea, were such that the Committee would scarcely be justified in taking the responsibility. The proposed arrangement in regard to the "Runa" is, however, not abandoned but only postponed, and it is hoped that any month now conditions may have so far improved that it will be right to resume work at sea along some of our usual lines—so as to preserve, if possible, a thin thread of continuity in the series of observations, or what perhaps might better be described as a few scattered stepping-stones set down in the hope that they may help us to bridge the unfortunate gap between our records of past years and the investigations we hope to return to under happier circumstances in the future.

This gap, it is to be feared, will exist throughout all the observations in the European seas. The work of the International Council for the Exploration of the Sea

still continues in some laboratories on land, but co-ordinated observations at sea have practically ceased. I am informed by Professor C. Ostenfeld, of Copenhagen, that some of the neutral countries (Norway, Sweden and Denmark) are still carrying out a small portion of their programme—chiefly herring work, and in the case of Sweden and Denmark a certain amount of hydrography. It is eminently desirable in the interests of scientific fisheries investigation that every country that can should, whenever possible, get back to its normal programme of observations at sea, or any portion of it, in the hope that the gap may not be larger than is inevitable.

But even though the usual programme of work at sea is in abeyance, no investigators need remain idle. There is still much to do in the in-shore waters and on our neglected sea-beaches. Now seems an opportunity to concentrate attention upon the cultivation of the shallower seas, and there is no doubt that much still remains to be done in both the investigation and the exploitation of the industries of the coastal waters.

Now that considerable areas of our usual British fishing grounds are closed to trawlers, any increase of employment on the seashore and in shallow waters may be of direct and immediate advantage both to the men and to the country. Such industries as shellfish cultivation, shrimping and prawning, whitebait and sprat fishing, if extended and exploited judiciously, will add to employment, will increase the food supplies of the country, and may lead to the establishment of permanent industries of a profitable nature. On this coast we have been alive to such possibilities for some time past, and much of our work has been directed towards showing the improvements that might be introduced in connection with the shellfish industries. It has been shown in our reports how mussels and cockles can be fattened and greatly increased in value by transplanting to better feeding grounds, and

how, if reared in sewage-polluted waters, they can then be cleansed and purified before being sent to market.

The Lancashire and Western Joint Committee, realising the present opportunity of benefiting such deserving industries, have worked out several concrete cases where a moderate expenditure either in transplanting or in purifying, or both, would be likely to give immediate results, and have applied to the Development Commission, through the Board of Agriculture and Fisheries, for a grant to be expended during the present spring upon this useful work. It will be unfortunate if, at the present juncture, such directly productive expenditure, which may reasonably be expected to lead to the establishment of permanent shellfish industries, be prevented or delayed for want of the comparatively small grant which is necessary to start the work.

The present Report on the work which the Scientific Staff has been able to carry out during 1914 is, in the main, on the same lines as the reports for the last few years, with, of course, some necessary omissions caused by the interruption of work at sea during the absence of the steamer. The members of the Scientific Staff have, however, as will be seen from the articles in the report, been kept fully occupied with other work. Dr. Johnstone has been working at important questions in connection with schemes for the improvement of mussel cultivation on the Welsh Coast. Mr. Scott has been investigating samples of whitebait from the Menai Straits. Mr. Riddell has been fully occupied with the herring investigation which we undertook at the request of the Board of Agriculture and Fisheries.

I shall now give a brief account of these and the other investigations that are reported upon, but I should like to express the hope that readers will not be content with my imperfect editorial summary, but will read the details for themselves in the several authors' own words.

Work at the Piel Laboratory.

Mr. Scott makes the usual report as to the fishermen's classes, the visitors, the fish-hatching operations, and other matters relative to work at our Piel Laboratory and Hatchery in the Barrow Channel. The defensive operations in that neighbourhood necessitated by the outbreak of war restricted the work greatly in many directions. The usual fish-hatching operations were carried on in the spring of 1914, but, for various reasons, these will be impossible in the season of 1915. The practical classes for fishermen are, however, being carried on with some modifications. Increased attention is being given to the preparation of trawl fishermen for the Examinations of the Board of Trade. Mr. Scott's analysis of the whitebait samples, and his work on the fish-eggs and on the plankton of the Irish Sea were also carried out at the Piel Laboratory.

Work at the Liverpool Laboratory.

Diseases of Fishes.

Dr. Johnstone reports at length on several fishes which had been condemned as unfit for human food by Port Sanitary Officers and were sent on to the Laboratory for confirmatory evidence of the desirability of condemnation. The specimens received during 1914 illustrate very well some extreme cases of cancerous growths in valuable edible fishes and are worthy of a detailed report. Some other cases of obscure disease in fishes closely analogous to well-known human diseases are also described and illustrated.

Bacteriological Investigations.

Dr. Johnstone makes a lengthy report on investigations carried on partly at the Liverpool Laboratory, and partly at Piel and in the open, with respect to the best methods of

treating polluted mussels so that they may cleanse themselves from sewage bacteria. The shellfish were subjected to the cleansing action of a current of pure sea-water, and it was found that they were practically freed from sewage bacteria in 24 hours. Shellfish were also kept in standing pure sea-water, when it was found that the cleansing process was naturally slower, but that it might be accelerated by frequent changes of water: two to three days was then found to be the time that was necessary under these conditions. Shellfish were also subjected to treatment by sea-water containing chlorine. It has been known for some years that traces of chlorine sufficed practically to sterilise drinking water, and a commercial purification process of this nature has been in use in connection with small drinking water installations. The method is now going to be employed by the Conway Corporation, under the supervision of the Board of Agriculture and Fisheries, for the purpose of purifying the polluted mussels taken in the Conway Estuary. Dr. Johnstone found that mussels live well in sea-water containing five parts per million of chlorine, and could be cleansed from sewage bacteria by three changes of sea-water so dosed with chlorine. Sea-water containing sewage bacteria was sterilised by the addition of one part of chlorine per million. The actual time required for each change of water must, however, be worked out in the actual commercial plant employed for the purpose.

Mussels placed in rough enclosures on the foreshore at about half-tide level also cleanse themselves from sewage bacteria. Where enclosures cannot be made on account of the nature of the foreshore, a removable floating tank was used to store the mussels. Such experiments were made at Overton, Sunderland Point and Glasson, all in Morecambe Bay, and at Aberdovey and Barmouth in Cardigan Bay. These experiments were all quite successful, since it was found that a period of two to three days was enough for the process of

removal of over 95 per cent. of the sewage bacteria originally contained in the shellfish.

A practical working experiment was made at Overton in the Estuary of the Lune. Arrangements were also made for the certification of the treated mussels. Similar experiments on an industrial scale are being made at Aberdovey and Barmouth, in anticipation of the confirmation of the Order now being promoted by the Committee. It is important to notice that sewage bacteria rapidly die out when they enter sea-water. The various species, however, die out at different rates. Some observations with regard to this matter also have been made, and are reported on.

Mussels which are contaminated by sewage contain a number of different species of bacteria. Some of the latter are not of dangerous significance, i.e., they may not have come from the human alimentary canal; others have this origin. Most of these bacteria have hitherto been regarded somewhat loosely as being "Bacillus coli," though Dr. Alfred McConkey has shown that they ought not to be so confused. Dr. Johnstone now gives details of the reactions of a large number of bacteria isolated from sewage and, by applying McConkey's methods of analysis, shows that there is a considerable difference between the proportions of the various species of micro-organisms present in mussels and those present in human faecal matter. Probably most faecal micro-organisms die out on entering sea-water. This matter is obviously of great importance in regard to routine methods of bacteriological analysis and deserves much more investigation; but Dr. Johnstone's present paper will be found to be a valuable contribution to our knowledge.

Investigations on the Herring.

Mr. Riddell reports on the measurements of a number of variable characters of herrings made by him during 1914. This investigation is part of a larger scheme promoted and organised by the Board of Agriculture and Fisheries, and our

part of the work applies to samples of herrings obtained from the summer fishery round the south end of the Isle of Man, from the winter fishery in Carnarvon and Cardigan Bays, and from the steam trawler fishery off "The Smalls" in St. George's Channel. Over 1,000 herrings have been investigated during the year.

Mr. Scott reports on samples of sprats and whitebait sent to him from Morecambe Bay and Menai Straits. A very important and lucrative fishery for sprats and whitebait has been in progress at Morecambe for the last two or three winters. The sprats are mostly salted and are sent away in barrels, apparently for the purpose of being made into sardines. The smaller fish (which are both sprats and herrings) are riddled out from the general catch and are packed in boxes and sold as whitebait. The fish are caught both by seine-nets and in the "baulks." There is no sprat fishery of any consequence in the Menai Straits and the whitebait there are taken in the weirs. Sprats also come into the Mersey Estuary in the winter, and a large proportion of them are sexually mature fish.

Dr. Johnstone has examined all the samples of herring sent to the laboratory, with the object of estimating the variation in the amount of fat contained in the flesh. This rises to over 33 per cent. in the case of the Manx summer-caught fish and sinks to about 5 per cent. in the case of the Welsh winter-caught herrings. The fish that contain most fat are those that have their ovaries or testes ripened just at the time of maximum sea-temperature. The proportion of fat falls off very suddenly just about the time that the fish are spawning. It is well known that fish which are just spent are in the worst condition as regards human food.

Spawning Period of the Shrimp.

Mr. T. Monaghan has examined two large samples of shrimps during each month of the year 1914. These shrimps were caught in the Estuary of the Mersey by the New Brighton

police boat. They were sorted out into sexes, and the degree of maturity was observed in each shrimp. The spawning period is principally in the spring months, but some shrimps carrying eggs can be found at any time throughout the year.

Work at Sea, at Port Erin, and elsewhere.

Hydrographic Observations.

Dr. Bassett reports as usual on the observations and analyses of samples of water made during the first portion of the year, including September; but does not attempt to draw conclusions from his data, on account of the record being incomplete this year.

Plankton Investigations.

With the expert help of Mr. Scott and Miss H. M. Lewis, I have continued the "Intensive Study" of the samples of plankton taken off Port Erin (as a central point in the Irish Sea) throughout the year. We report upon our results as usual, but these are of a detailed nature and do not call for any general comment.

Photosynthesis and the Alkalinity of Sea-water.

Professor B. Moore and his two fellow-workers have given us an interesting series of studies of photosynthetic phenomena in sea-water, as the result of observations and experiments made at Port Erin at various times between August, 1912 and January, 1915. They discuss (1) the extent and nature of the seasonal variations in the alkalinity of the sea, and (2) the limits of photosynthesis by algæ as the alkalinity due to their action increases, and show that the increase in the alkalinity of the sea in spring is not due to increasing temperature disturbing the equilibrium between the carbon-dioxide of the sea and the atmosphere, but is caused by the photosynthesis of the innumerable minute plants (Diatoms) present at that time in the water.

W. A. HERDMAN.

Fisheries Laboratory,
University of Liverpool,
March 24th, 1915.

FISH HATCHING AT PIEL.

By Andrew Scott, A.L.S.

The hatching operations carried on in the Spring of 1914 resulted in the liberation of just over thirteen millions of fry, chiefly flounders as in former years. Adult plaice for the spawning tanks were obtained from Luce Bay in the late Autumn of 1913, by the kind permission of the Fishery Board for Scotland, and were kept in the tanks during the Winter. The flounders were collected in Barrow Channel a few weeks previous to the usual spawning time.

The spawning of both plaice and flounders was unusually late in beginning. The first fertilised eggs were not obtained till March 23rd, two days later than in any previous year, and nearly a month later than in 1913. So far as our records show, there was apparently no undue delay in fish spawning in the open sea under natural conditions. Plaice eggs were found to be present in the plankton collected in the central area of the Irish Sea on February 17th, and in Port Erin Bay on the 26th of the same month. The lateness of the spawning of the fish kept in our tanks compared with what takes place in the open-air spawning pond at the Port Erin Hatchery is probably largely due to the variation of the temperature of the sea-water at the two places. At low water in the neighbourhood of Piel large tracts of the shore are laid bare, and in places that do not dry the water becomes quite shallow. The exposed area soon becomes cooled down, especially during continued frosts or easterly winds, and when the flood tide sets in, any increased temperature that it may possess is quickly lost on coming into contact with the cooled area. In mild weather the early flood water, on the other hand, may be warmer than in the open sea. It is not always possible

for us to wait until high water, when the temperature will have become fairly constant, in order to commence pumping, owing to the somewhat limited capacity of our storage tanks.

The spawning of the fish continued for just over five weeks. During that time one million two hundred and twenty-five thousand plaice eggs were obtained, and thirteen million six hundred thousand flounder eggs. The eggs were incubated in the usual manner, and the resulting fry were liberated from time to time. The first batch of plaice embryos began hatching thirteen days after the eggs were fertilised. The time required to incubate the other batches varied from thirteen to fifteen days. The incubation of the flounder eggs took from nine to eleven days. The adult fish were set free in the Barrow Channel at the end of the hatching season.

The following tables give the number of eggs collected, and of the fry hatched and set free on the dates specified:—

PLAICE (*Pleuronectes platessa*, Linn.).

Eggs Collected.		Fry Set Free.	
March 23	35,000	30,000	April 12
„ 25	55,000	46,500	„ 16
„ 27	75,000	65,500	„ „
„ 29	80,000	69,500	„ 20
„ 31	85,000	74,500	„ „
April 2	85,000	74,500	„ 24
„ 4	95,000	84,000	„ „
„ 6	90,000	79,000	„ 28
„ 8	85,000	74,500	„ „
„ 10	80,000	69,000	May 2
„ 12	75,000	66,000	„ „
„ 14	75,000	66,000	„ 6
„ 16	70,000	59,500	„ „
„ 18	65,000	57,000	„ 10
„ 20	60,000	50,000	„ „
„ 22	55,000	46,500	„ 12
„ 26	35,000	30,000	„ 16
„ 29	25,000	20,000	„ „
Total Eggs	1,225,000	1,062,000	Total Fry.

Flounder (*Pleuronectes flesus*, Linn.).

	Eggs Collected.	Fry Set Free.		
March 23	... 350,000	300,000	... April	12
,, 25	... 550,000	475,000	... ,,	,,
,, 27	... 800,000	710,000	... ,,	16
,, 29	... 850,000	757,000	... ,,	,,
,, 31	... 950,000	845,000	... ,,	20
April 2	... 1,000,000	887,000	... ,,	,,
,, 4	... 1,100,000	975,000	... ,,	24
,, 6	... 1,100,000	975,000	... ,,	,,
,, 8	... 950,000	845,000	... ,,	28
,, 10	... 900,000	800,000	... ,,	,,
,, 12	... 850,000	757,000	... May	2
,, 14	... 850,000	757,000	... ,,	,,
,, 16	... 800,000	710,000	... ,,	6
,, 18	... 700,000	600,000	... ,,	,,
,, 20	... 650,000	575,000	... ,,	10
,, 22	... 550,000	475,000	... ,,	,,
,, 26	... 400,000	354,000	... ,,	16
,, 29	... 250,000	220,000	... ,,	,,
Total Eggs	13,600,000	12,017,000	Total Fry.	

Total Number of Eggs 14,825,000

Total Number of Fry 13,079,000

CLASSES, VISITORS, &c., AT PIEL.

By Andrew Scott, A.L.S.

The Education Committee of the Lancashire County Council voted the usual amount of money which allows forty-five studentships to be given to fishermen residing in the administrative area. These studentships enable the selected fishermen to attend at Piel and receive during two weeks a course of instruction in Marine Biology, or, if they are qualified, a combined course in Marine Biology and Navigation. The Education Committee also permitted Captain E. Barker Thornber, the County Navigation Instructor, to be at Piel for a period of six weeks. Captain Thornber gave instruction in Navigation and Seamanship to thirty-nine of the studentship holders who had put in the necessary sea time to qualify them to sit for the Board of Trade Certificates. The Education Committee of the County Borough of Southport sent four men, and the Education Committee of the County Borough of Blackpool again sent three men. The County Boroughs make special arrangements with the Sea Fisheries Committee for sending their men, and can either pay the studentships directly or through the Fisheries Committee. Altogether, fifty-two fishermen attended the classes held in 1914. The studentship holders were divided into four classes—one of eleven men, two of thirteen, and one of fourteen, as shown by the following lists:—

First Class, held March 2nd to 13th—L. Bettass, W. Chard, J. W. Crompton, J. Dawson, J. F. Flaxman, Jos. Jackson, W. A. Jackson, W. Kay, W. McLellan, J. Neave, W. Palmer, J. Rigby, E. Wales, and R. Wright, all of Fleetwood.

Second Class, held March 16th to 27th—S. Ainsworth, H. Ashton, J. R. Atkinson, L. Bethell, J. Croft, G. Douglas, C. Ellarby, J. Gornall, J. Iddon, J. Leadbetter, J. R. Wright, and T. Wright, all of Fleetwood.

Third Class, held March 30th to April 9th—C. A. Carpenter, L. Christian, C. Cullen, P. Ellarby, L. Larsen, C. McLean, M. McManus, J. Mead, E. Neave, N. Salthouse, J. Wright, and W. Wright, all of Fleetwood.

Fourth Class, held April 27th to May 8th—Frank Benson, Flookburgh; Thomas Hartley, Flookburgh; William Taylor, Bolton-le-Sands; J. W. Woodhouse, Morecambe; Samuel Bond, Morecambe; Thomas Rimmer, Blackpool; Albert Fenton, Blackpool; William Parr, Blackpool; Robert Johnson, Banks; James Evans, Southport; Henry Wright, Southport; William Wright, Southport; and Richard Rigby, Southport.

The first three classes were attended by fishermen from Fleetwood. They were students of the County Navigation School, and had served the required time at sea which enabled them to present themselves for examination for Board of Trade Certificates as second hand, skipper, or extra skippers of fishing vessels. The course of study was similar to that given on former occasions, and which has proved most suitable so far as time permitted. The morning lesson, lasting two and a half hours, was occupied by instruction in Marine Zoology having some relation to the fish and invertebrata captured in the trawl net. The afternoon lesson of three hours' duration was taken by Captain Thornber, and a very efficient course in Navigation and Seamanship was given. The students who were expecting to present themselves for examination immediately on their return to Fleetwood, usually returned to the laboratory for a further two hours practical work every evening and on

Saturdays. The extra instruction consisted of a revision of the afternoon lesson, chart work, *viva voce* examinations on rule of the road at sea, and use of the sextant. The additional work was purely voluntary on the part of the teacher and the students, but it showed that the men were most anxious to take every facility that was given to further their progress towards efficiency. The last class was attended by in-shore fishermen, such as musselers, cocklers, shrimpers, and men from second class fishing boats. Their course of instruction dealt with general Marine Biology relating to fish, economic shellfish, and other invertebrata of the shallow water.

The annual inspection of the classes was made by a party of members of the Sea-Fisheries Committee and the Education Sub-Committees of the Lancashire County Boroughs, under the leadership of the late Mr. J. Fletcher, Chairman of the Sea-Fisheries Joint Committee, on April 29th. Mr. Fletcher gave a useful address to the fishermen and the visitors on the objects of the Fishermen's Classes, and on the general work of the Sea-Fisheries Committee. The visitors then inspected the work that was being done, and afterwards returned to the steamer.

The Barrow Education Committee, with the permission of Mr. J. R. Ragdale, Chairman of the Sea-Fisheries Scientific Sub-Committee, organised an evening class in Nature Study for school teachers. The class met twice a week during part of the months of March, April and May. Sixteen teachers from the schools at Barrow attended, and went through a course of instruction dealing with the common objects of the sea-shore.

Mr. A. Harris, H.M. Inspector of Evening Schools for the district, paid official visits to the fishermen's classes and to the teachers' evening class, and inspected

the work that was going on. Dr. Jenkins, Sea-Fisheries Superintendent, and Mr. A. Hawcridge, Director of Education, Barrow-in-Furness, also visited the classes from time to time. The usual visits by members of local rambling clubs, and others interested in scientific work, were considerably reduced owing to the military restrictions enforced in the district.

Our thanks are due to the various departments and institutions mentioned in previous reports for additions to the library.

REPORT ON THE PERIODIC CRUISES.

By W. Riddell, M.A.

From various causes the work at sea has been very much interrupted during the past year. Owing to the steamer being laid up for survey and repairs, there were no cruises in January, March and August. Work at sea was finally suspended for the present at the end of September, when the steamer was taken over by the Admiralty.

Thus there have only been two quarterly scientific cruises, in February and May, and four monthly cruises, in April, June, July, and September. This means that for Station 14 there are only two plankton samples, and only six at each of the other three stations. Or, from another point of view, in the first part of the year, to the end of July, the quarterly cruises are complete, and of the monthly cruises two, January and March, were missed; while in the second part, since the outbreak of war, both quarterly and monthly cruises (with the exception of September) have had to be abandoned.

Surface samples have been taken from the steamer when employed on her ordinary duties, but these also represent little more than six months' actual collecting, and there are many gaps in the series.

Under these circumstances, and as I have been unable to find anything in the samples which calls for special mention, I do not propose to make any report upon the collections at present. When scientific work at sea is resumed under normal conditions, these samples will probably be found to be of importance as preserving some continuity in the series of observations, and will be utilised, as far as possible, in a future report.

DISEASED AND ABNORMAL CONDITIONS OF MARINE FISHES.

By James Johnstone, D.Sc.

(With seven Plates and seven Text-figures.)

Contents.

1. Piscine Sarcomata, with special reference to Cases of Multiple Tumours in Halibut and Cod.

On June 16th, 1914, Mr. F. Stokes, Port Sanitary Inspector at Grimsby, sent me a large part of a Halibut which had been caught by a steam trawler and landed at Grimsby. The whole fish had been put aside as unsuitable for human food, but the Inspector subsequently exercised a praiseworthy discretion in allowing the anterior part to pass into commerce in the usual way, and in sending me the posterior part for examination. The entire fish weighed about 16 stones, and considering the price of halibut, it must be admitted that condemnation of even half of this valuable product was an act of public duty involving some responsibility. The responsibility was honestly taken in this case, and, in my opinion, wisely so, for the part of the fish sent to me was certainly

unfit for human food. The flesh was "knobby" and "full of tumours." There was general discoloration along the ventral surfaces near the fin; and a large, black, soft area there showed that there was extensive destruction of muscular tissue. On cutting into this softer part of the fish it was seen that there was considerable necrosis, for a dense black, pultaceous fluid oozed out, leaving a large cavity. On cutting through the fish in various planes it was seen that there were very numerous tumours in the muscles of the abdominal region on either side of the ovaries, and above and below the latter in the muscles surrounding the vertical fin skeleton. The tail of the fish was also very greatly eroded and swollen, and there were very many small tumours in the tissues beneath the integument in this region.

Malignant Tumours in Marine Fishes.

A fairly large number of cases of the occurrence of malignant growths in fishes have now been described, and one may say something as to the general character of these abnormalities. Benign tumours are not very uncommon in marine fishes. Usually these growths are such as would be called "wens" in man. They occur just beneath the integument, on nearly all parts of the body, and they are sometimes found in the body cavity, as growths of the peritoneal tissues. They are fibrous in nature, "hard" or "soft," and they are always very distinctly capsulated so that they can easily be "shelled-out." Examination of a large number of sea fishes exposed in public markets may reveal one or two cases of growths of this nature.

Malignant tumours are more rare, but even these may be found by examining fairly large numbers of fish. Those that I have seen myself have occurred in Skates,

Rays, Cod and Halibut. They are, probably, no less uncommon in other edible sea fishes, but the above species are large and fairly valuable fish, and more attention is naturally paid to them. In speaking of "malignancy," one means rather less than he would in referring to the malignancy of human affections. In a fish suffering from a tumour there is nothing in the nature of a "clinical history," and it would be very difficult to make any really good experiments with the object of investigating the effects on metabolism, or general health, or bodily functioning, of the presence of a progressive tumour. One can only investigate the morbid anatomy and compare this with human pathological conditions. It is difficult also to be sure whether or not a tumour has any effect on the health of the fish in which it occurs. There are indications of normality or abnormality of functioning in what one, rather vaguely, calls the "condition" of the animal. There may be great emaciation, or the lack of maturation of ovaries or testes; the flesh may be thin and watery, and tasteless if cooked; there may be a poorness in fat if the fish is one which, like the Halibut, Salmonidae or Clupeidae, is rich in fat at certain seasons of the year; or there may be a large deviation from the average weight which one regards as characteristic of fishes of various sizes and at different seasons of the year. Usually one can say whether or not a fish is "in good condition," meaning by that whether it has been well fed or not; but that is all that is to be said as to the general health of the animal. Post-mortem examination accompanied by detailed microscopic investigation may show whether or not internal organs are diseased, but we do not know enough about the naked-eye appearance of these organs to say at once whether they are normal or abnormal. Fishermen, who are very observant, take

much notice of the appearance of the liver in fishes which they gut, and judge of the condition of the animals by the size and pigmentation of this organ. In fishes like Cod and Elasmobranchs, at all events, the liver undergoes seasonal changes, and a bio-chemical study of these variations would be one of much interest.

By "malignancy," then, we mean the same set of anatomical conditions that are to be observed in cases of human progressive growths or neoplasms, where a clinical history affords data for attributing general ill-health or death to the agency of the growths. Malignant tumours in fishes are therefore such growths as are progressive; which are not capsulated and diffuse; or which, if they are capsulated show indications of breakdown of this structure; and which are multiple. "Metastases" occur in such cases, that is, one seldom sees a single malignant tumour: usually there are many such, but whether or not the metastatic growths have proceeded from a single "focus" one cannot easily say. Some indications point to an infective origin for these growths in fishes; and to certain conditions in the environment which favour or promote their occurrence, but this is a question which is not easily investigated.

All the malignant tumours which I have seen in fishes are sarcomata: I have seen nothing in the least resembling a carcinoma. In one or two cases details of histology resembling acinose structures have been observed, but these have never been so marked as to justify us in speaking of the growths as other than sarcomatous ones. Abnormal tissue growth, other than that of the connective tissues, does not seem to occur, that is, I have never seen such structures as myomata, osteomata, gliomata, and the like. When a malignant tumour arises in a fish it is always an overgrowth of

connective tissue, and it is almost always the sub-integumentary connective tissues of the animal that are affected. I have seen what was apparently a malignant growth of the choroidal gland in a Flounder, with destruction of the remaining parts of the eye; and I have also seen fibrous nodules in the liver of a Conger, probably local proliferative growth of Glisson's capsule. But, in general, malignant tumours in fishes are abnormal developments of the sub-integumentary tissues. Not enough is known as to the nature of the skin and underlying tissues in fishes to speak more definitely than this. The layers beneath the epidermis differ greatly in the various orders of fishes. But below the layer from which the scales originate is always a layer of more or less strongly developed coarse connective tissue, and this is the *locus* of the malignant tumours of which I speak. In Adami's terminology these fish growths are *hylic tumours of mesenchymal origin.*

The tumour may be restricted to this situation, growing as a swelling visible from without, beneath a stretched or ruptured epidermis. But the tumour may also be invisible from without since it may extend along the connective tissue septa between the myotomes, or between the smaller muscle bundles, and even between the individual muscle fibres. The muscular tissue itself seems never to be affected. When this growth to the interior occurs, the tumours may only be recognised when the flesh of the fish is cut into.

The malignant tumour in fishes is usually a melanotic sarcoma, or fibro-sarcoma, consisting of round or spindle cells, or both, with a more or less abundant fibrous stroma. Young tumours may be colourless, or pink (apparently a stage in the formation of melanin), but they are usually tinged grey or are dense black. The

more advanced is the tumour, the more deeply is it charged with melanin. The latter may at first be recognised as definite granules laid down within cells of recognisable outline, but as the tumour grows the deposit of melanin becomes denser, and finally the cells are no longer apparent. The melanin formation is to be connected, in some way, with the tendency for pigment formation in the integuments of most fishes. If it is a "metabolic habit" of this nature it may be more typical, in man, of morbid growths in coloured races, or in brunettes, than in white races, or in blondes. I do not know whether or not this is the case. Melanism of malignant growths has been observed in Skates, Rays, Halibut and Cod, but it is more characteristic of the two former species than of the two latter.

Necrosis always occurs when the tumours attain some size. Even in small growths, say in those of two or three centimetres in diameter in Skates and Rays, there is always very considerable softening and disintegration of tissue. Melanin formation then attains a maximum, and the tumour appears to be coal-black in section. When it becomes much larger than this its central parts liquefy. Usually there are no blood vessels in these malignant tumours, and autolysis probably occurs. When the larger tumours, say those of 5 to 10 cms. in diameter, are cut open, the contents run out as an inky-black fluid with the consistency of pus. A soft or liquid tumour of this stage does not set hard when preserved in alcohol: its central parts always remain soft and pasty.

Distribution of the Tumours.

Such are the general characters of malignant growths in fishes. The specimen which I have now to describe illustrates very well all these characters, and particularly

the means of spreading by breakdown of previously capsulated growths. In this Halibut there were very many tumours varying in diameter, from about one-half to ten centimetres. The larger tumours were probably the result, not only of individual growth, but also of the confluence of originally distinct growths. The larger tumours were always loaded with melanin; the smaller ones were colourless or faint pink. The general appearance of these smaller tumours is represented in Text-figs. 1 and 2. Text-fig. 1 (part of a longitudinal

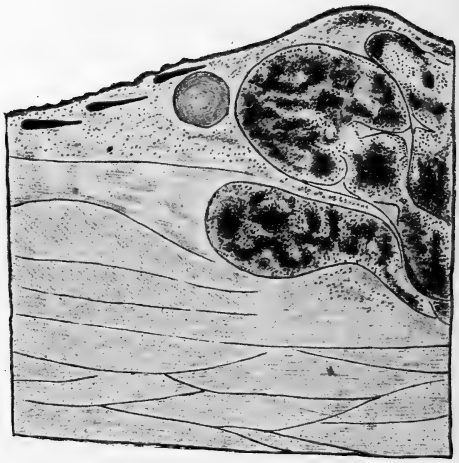


Fig. 1.

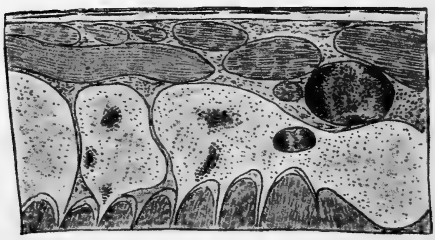


Fig. 2.

Figs. 1 and 2. Hand sections through the flesh of a Halibut containing Melanotic Sarcomata. The upper fig. (1) is part of a longitudinal section, and the lower fig. (2) is part of a transverse section.

section) shows several growths which are apparently independent of each other, that is, each is surrounded by a distinct thin capsule. In hand sections of the fresh material these tumours show as either creamy white or grey in colour, with black mottling due to melanin impregnation. Text-fig. 2, which is part of a transverse section through the fish, shows two small rounded tumours containing some melanin, and also several larger ones which are nearly colourless. These figures represent fairly well the appearances disclosed by making hand sections, in various planes, through the flesh of the fish behind the anus. Almost everywhere there were rounded tumours which were usually to be recognised by their pink or grey or black colour, but which were sometimes colourless and recognisable only by their texture. Usually they could be seen to possess definite boundaries, that is, distinct thin capsules. They were generally situated immediately beneath the skin, but sometimes they were apparently embedded in the muscles. Some were surrounded by adipose tissue. In the Halibut, unlike most Pleuronectid fishes, there is, in large, well-fed fishes, a rather thick layer of fat near the bases of dorsal and ventral fins, on either side of the axonosts. Sometimes there were small tumours in the middle of this fatty tissue.

Encapsulation and Boundaries of the Tumours.

Hand sections made through the flesh of the fish by means of a sharp razor showed that the tumours were usually definitely bounded. This definite outline suggested the presence of a capsular structure, but when microtome sections were made, stained and mounted, it was seen that such a capsule was not always present, and when it was present there were often indications of its

breakdown, and the proliferation of the tissue of the tumour into the surrounding structures. The details of the capsulation are, therefore, of considerable interest.

Fig. 1 of Plate II shows the place of apposition of three small rounded tumours. No fine histological details of the tissues of the tumour are represented, the latter being indicated only by the fine stippling of the figure. The boundaries of the tumours are formed by a fine, thin layer of connective tissue, just a little denser than the adjacent tissue. Between the tumours is a loose kind of connective tissue containing some coarser fibres with some bundles of muscle fibres, and round the tumours this tissue seems only to become a little more compact and to be roughly arranged in a concentric manner.

Fig. 2 of Plate II represents quite a different kind of boundary. Here the tumour is embedded in a mass of adipose tissue which is situated near the base of the ventral fin. Fig. 6 of Plate III represents the normal appearance of the tissues in this region as seen under high magnification. There are loose and irregularly-running striped muscle fibres and small bundles of such, with bundles of fibrous connective tissue, blood capillaries, areolar connective tissue, and fat cells. The latter may be relatively much more numerous than is represented in this figure. In fig. 2 of Plate II the adipose tissue is much denser, and here there appears to have taken place a double progressive change: first, a development of fibrous connective tissue among the fat cells, and then the development of a typical, small-celled sarcoma along this region of fibrosis. The locus of the sarcoma is represented by the darkly stippled area, while the skeleton of uniformly tinted material round the sarcomatous nodule represents the area in which the process of fibrosis has occurred. Under an apochromatic lens this fibrous bar-

work is seen to be a dense felt-work of very fine interlacing fibres without any obvious nuclei. In Mallory preparations it is seen to be greatly altered in places, becoming almost uniform in structure. There is no distinct boundary between it and the adjacent fat cells, and some of the latter seem to be actually embedded in the felt-work. Apparently it is the result of the progressive development of the connective tissue stroma still existing between the fully formed fat cells. Then along this region of fibrosis the cells of the sarcoma appear to be proliferating, so that we have a series of changes in the original normal tissues in this part of the fish. These are (1) the replacement of the areolar connective tissue by adipose tissue, producing the structure represented at the marginal parts of the figure; (2) the fibrosis of this adipose tissue, that is, a progressive development of the stroma that remained after fat-formation had attained its maximum; and (3) the multiplication of the connective tissue cells remaining in the stroma, and the proliferation of those cells along the regions of fibrosis as a small-round-celled sarcoma. The alteration in the fibrosed tissues we may regard as due to the influence of the fully-developed sarcoma. In the fresh condition the latter was pink in colour, and as there were already small groups of melanin granules scattered throughout it, we may, perhaps, regard the pink colour as indicating the first stages in the process of melanisation of the tumour.

Figs. 1 and 2 of Plate II represent, therefore, the contrasted conditions of capsulated and non-capsulated tumours. Figs. 3 to 7 of the same plate represent conditions where tumours already distinctly capsulated are actively proliferating by the breakdown, in places, of the capsules. Fig. 3 is a low-power drawing. The stippled area represents the locus of the tumour, which

is here a mixed-cell sarcoma. Round most of this nodule is a thin fibrous capsule similar to those represented in fig. 1, but at the place shown in fig. 3 this has quite broken down. There is no indication here of connective tissue fibres, except the arrangement of the elongated nuclei. Round the nodule are ordinary striped muscle fibres, and these have quite broken down at one place, and the sarcoma is actively growing here and is invading the interspaces between the muscle fibres, spreading along the connective tissue stroma among the latter. This seems to be quite typical of the manner in which these fish sarcomata proliferate. Fig. 1 of Plate III represents a small part of the same field drawn in fig. 3 of Plate II, but seen under an apochromatic lens, and greatly magnified. On the lower margin of the figure are parts of two unchanged striped muscle fibres, while the rest of the figure represents the changes undergone by other similar fibres as the result of the proximity of the sarcoma. What we have here is apparently a condition resembling the Zenkerian degeneration of striped muscle fibres induced as the result of various causes: it is a special kind of necrotic change. The sarcolemma appears to remain intact, but the striation of the fibre quite disappears, and its substance seems to become replaced by some hyaline, non-staining, waxy material. But here also we have a multiplication of the nuclei of the fibre, or perhaps the invasion of the latter by extraneous cells. The space within the sarcolemma is filled by large numbers of small oval cells. These may have resulted from the multiplication of the original muscle nuclei, but they may also have migrated in the individual fibres from without. It is doubtful whether much of the tissue of the sarcomatous nodule has originated in this way: it is more likely that this replacement of the muscle fibres by

aggregations of small oat-shaped cells occurs only at the margin of the tumour and as the result of a stimulus exerted by some substance secreted by the latter.

Figs. 4 to 7 of Plate II represent the proliferation of sarcomatous tissue along a region of fibrosis. Fig. 4 is a low-power drawing and shows the growing point of a nodule, which elsewhere is surrounded by a feeble capsule. Round the nodule is loose areolar connective tissue with loosely arranged muscle fibres running in various directions. A large area of the section, of which the figure represents only a small part, is occupied by a rather dense fibrous tissue, similar to that found in benign, capsulated fibromata. Fig. 7 represents the general nature of this tissue, and it is seen to consist of bundles of fine fibres, pursuing a straight or slightly convoluted course. Among them are some coarser fibres staining red with Mallory's stain. These are shown to the left in fig. 4. Towards the right in this figure they are seen to be undergoing disintegration, and there also are to be seen the remains of the original capsule of the tumour. The stippling shows the locus of the truly sarcomatous cells, and it is clear that there is no distinct boundary between these and the adjacent fibrous tissue. This is the growing margin of the tumour—the region of active proliferation, and it is shown, highly magnified, in fig. 6. Here we see the fibrous tissue with some of its own nuclei, and in it there are large numbers of very small round cells. Fig. 7 represents part of the section still further removed from the growing margin of the tumour, and here the tissue represented is, for the most part, that of the fibroma along which the sarcoma is proliferating. The nuclei of this fibrous tissue are shown, but there are also a few small round or ellipsoidal cells, which are evidently those of the sarcoma. Fig. 5

represents a part of the section shown in fig. 4, and it should be referred to the extreme right of that figure. It shows the fully developed sarcomatous cells.

Here, then, as in the case of the tumour appearing among the fat cells, we have a secondary process. A previous process of fibrosis of connective and muscular tissue has occurred, and then this fibrosed region has been invaded, and finally is being replaced by a truly sarcomatous, or malignant (an actively growing) tissue.

Histology of the Tumours.

The nodules are strictly non-vascular; absolutely no traces of blood or lymph vessels can be made out in their substance. They belong to no single type; thus figs. 2 to 5 of Plate III all represent parts of a single section of one tumour.

Fig. 5 represents the tissue near the margin of the nodule. It is not part of the capsule; that consists of a thin layer of fine fibres containing relatively few nuclei. Near to the margin these fibres pass into a network of loosely interlaced fibres, each of which is a connective tissue cell, spindle-shaped, but with the extremities of the cell greatly prolonged. Some of these cells possess two nuclei. The latter are ellipsoidal in shape. Accompanying this network of connective tissue cells is a fine stroma, but this is greatly reduced.

This tissue passes into that represented in fig. 2, which consists of a rather dense mass of typical spindle-cells. These run in various directions. Among them are, apparently, a few smaller, spherical cells, but such are probably spindle-cells like the rest, but seen end-on. The stroma is almost entirely absent. The growth here is obviously a typical spindle-celled sarcoma.

Parts of the same tumour are formed from cells such as those represented in fig. 3. These are all small and round. There are typical nuclei with very little cell-

substance. The stroma is entirely absent. The tumour here is what has been described as a small-round-celled sarcoma.

Finally the central part of this, and of most other nodules, consists of a tissue represented in fig. 4. This is, perhaps, the most characteristic tissue of the growths here investigated. In it are found large numbers of small spherical cells, but there are many other kinds. Some are spindle-shaped, larger than those represented in fig. 4, and with blunt, rounded ends. The nuclei of these, and the small spherical cells, are apparently quite normal. There are numbers of larger cells, oval, round, or irregular in shape: many of these contain two nuclei. Then there are structures which one may call "giant cells," and these are not all the same. Some have a large central nucleus with several smaller peripheral nuclei. Others can simply be called multinuclear cell masses. Others again appear to be syncytia containing a number of small nuclei distributed quite irregularly. Apparently they have arisen from irregular nuclear division which has not been followed by cell division, rather than from the fusion of adjacent single-nucleus cells. Along with all these elements are cell fragments, cells containing melanin granules, and apparently loose melanin. Such a section represents the conditions in a tumour which is beginning to undergo necrotic degeneration. In the central parts of the larger tumours some of the same elements can be recognised, but here the process of autolysis has gone on to a remarkable extent, and the substance of the tumour consists of a thick liquid containing disintegrated cells, with melanin granules. The latter are the more abundant the further the process of necrosis has proceeeded.

Summing up, then, the specimen here described is a good example of a fish suffering from multiple, melanotic

sarcomata. These may have arisen by metastatic growth, fragments of an original tumour having been distributed through the lymph channels. But the general appearance of the specimen suggests rather that the separate sarcomatous nodules have arisen *in situ* independently of each other, and one may perhaps suggest that this affection, and perhaps most of those which I have previously described as sarcomata in marine fishes, are to be compared with the disease described by Gaylord and Marsh* in Salmonoid fishes in American waters. Here we have a condition of carcinoma of the thyroid becoming endemic in fishes living free in populated waters. Some fishes are immune to the infection. The latter is apparently produced by some agent, or substance, contained in the water inhabited by the fishes; and notably in the tanks of hatcheries in which the fishes may be bred or confined.

It is, of course, difficult, or perhaps impossible, to investigate such a possible causation in the case of such fishes as Halibut or Rays, but it appears to be possible that the origin of sarcomatous growths in marine fishes living in the wild may be due to infection of some kind.

Degeneration of Sarcomatous Tumours.

There are no observations that I know with respect to the rate of growth of malignant tumours in marine fishes; and we do not know positively that such growths as those which I have described ultimately cause the death of the animals. Possibly sarcomata in marine fishes may be of very slow growth, and the ordinarily-caught marine edible fish is a short-lived animal—at least, its duration of life prior to capture is usually a

* "Carcinoma of the Thyroid in the Salmonoid Fishes." H. R. Gaylord and M. C. Marsh; *Bulletin of the Bureau of Fisheries*, Vol. XXXII, 1912; pp. 365-524; Pls. LVI-CX. Washington, 1914.

matter of only a few years. Probably, then, those specimens which one sees are very seldom fully developed. Now and then, however, I have seen marine fish which show, by their extreme emaciation, the morbid influence of sarcomatous growths exhibited by them. In such cases there is always more or less breakdown of the tissues of the tumours.

Such a case is very well illustrated by a cod sent to me by Mr. F. Stokes, Port Sanitary Inspector at Grimsby, and it may be useful to describe this fish in some detail. It was a full-grown fish rather over 3 feet in length. It is represented in Text-fig. 3, which, however, minimises the emaciation of the fish. The degree of emaciation was extreme, and was best seen by looking at the fish from the dorsal aspect. The tumour was about 8 inches in diameter, well raised, and occupying the right side of the fish between the second dorsal and the first ventral fin. It was a (relatively) huge growth. It looked far larger than the photograph indicates, but its volume can be estimated by looking at the degree of curvature of the lateral line. The fish had been gutted, but fragments of the viscera remaining behind showed that it was a sexually mature and ripe male. The tumour had no connection with the body cavity, though it bulged into the latter.

It was very soft to the touch, and its substance was evidently almost liquid. On feeling it carefully, fairly hard objects could be made out quite unattached, and floating in the liquid part. On cutting into it the substance flowed out. It had exactly the colour and consistency of newly-cooked oatmeal porridge, but there were irregularly solid bodies in this viscous mass about an inch or so in diameter. These had the consistency of ripe Gorgonzola cheese. They were easily broken down,

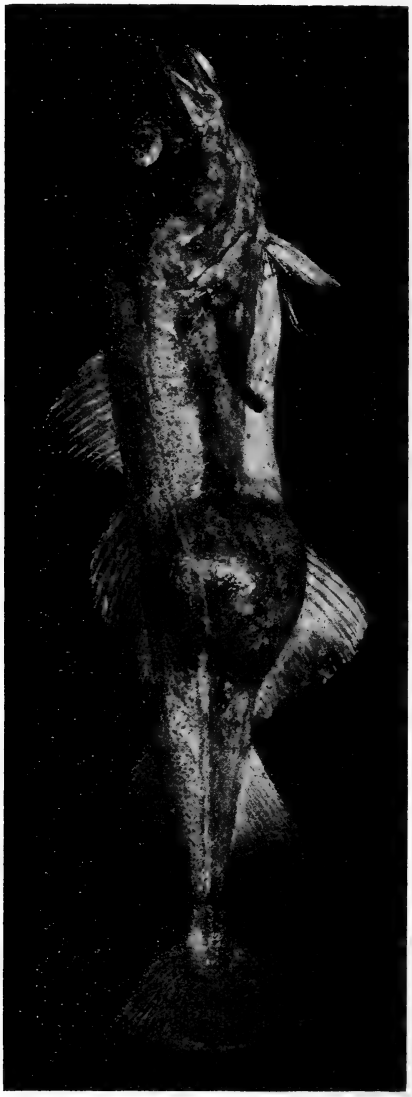


FIG. 3. Photograph of a Cod exhibiting a non-melanotic Sarcoma in process of necrosis.

but were harder in their central parts. Microscopic examination of this fresh tumour substance showed little definite detail. There was much fat or oil present in the form of small globules. Small spherical bodies resembling sporozoan cysts, containing rounded cell-like structures, and surrounded by definite capsules, could be seen. These suggested that the tumour was one caused by the enormous multiplication of some Protozoan, but no spores could be seen. For the rest the substance, when fresh, consisted of unrecognisable débris.

Smears were, however, made, fixed in Zenker's fluid, hardened and stained by iron haematoxylin and Orange G. Some of the nodular masses were also preserved in Zenker's and Bouin's fixatives, and pieces of the adjacent muscle substance were cut out and fixed in the same way. The tumour was then examined more attentively. It was scraped out, and it was then seen that it occupied a cavity in the flesh of the animal. This cavity was not bounded by any capsular structure: what appeared at first to be a capsule was the connective tissue muscle dissepiments. Some of these run roughly concentric to the skin, and those near to the latter remained, although most had been involved in the substance of the tumour. They had almost no muscle tissue between them, and being forced together by the pressure of the growing tumour, they looked like a capsule. This was, however, not the case; the tumour was not delimited from the surrounding tissues—it was truly malignant.

Later examination showed clearly that it was a sarcoma undergoing profound autolytic degeneration. The evidence for this may, however, be stated, since the conditions were very interesting. First of all we may look at the stained smear. This is represented in fig. 2 of Plate VII; the drawing shows, not a definite field as

seen under the microscope, but rather a selection of all the different kinds of cells which could be seen at the same time by an oil-immersion lens. There was a large amount of fine cell-débris, but neglecting this, we see that we have the elements of a mixed-cell sarcoma. The commonest cells present were the small spherical ones with large nuclei represented at A in the figure. There were a few spindle cells (B), but not many. Here and there were cells containing in addition to one or two typical nuclei a spherical body surrounded by a concentric space. These inclusions did not take nuclear stains. I think they belong to the category of formations represented by "Russell's bodies," and have no significance except that they represent morbid changes in the cancer cells. There were other cells (D) containing numbers of chromidial bodies: these represent nuclear fragmentation or nucleolar formations. Then there were the rather extraordinary cells represented at E. These are polymorphic, or "amoeboid." They may contain more than one nucleus, though usually one only. Their cytoplasm was usually highly vacuolated. They would be of interest if one could be sure that they existed, as they are drawn, in the fresh, unfixed tissue, but a dried smear is a type of preparation from which one may expect anything, so I think the forms of these cells are artificial and due to the process of smearing. Finally there were multinuclear cells such as are represented at F. Thus we have a substance containing cell-débris, unaltered cells of various forms, and cells altered as the result of the degeneration of the sarcoma and the process of fixation.

One of the nodular bodies contained in the tumour was fixed, hardened and cut. Figs. 4 and 5 of Plate VI show the structure of these bodies. They were "cheesy" in consistence, but cut in the microtome after embedding

in paraffin without difficulty. Fig. 4 is quite typical, and shows a substance consisting of a granular matrix in which are numerous isolated cells. Some of these cells are "spindles," often lying with their long axes in the same direction, but not arranged in any way. There are larger spherical or irregularly shaped cells, and many small spherical ones. That is the general structure of the nodule, except that here and there the matrix becomes hyaline, and the cells disappear or are represented only by nuclear fragments.

For the most part, both cytoplasm and nuclei of the cells stain rather lightly with methyl-blue eosin (more lightly by far than one sees in sarcomatous tissues when employing this stain). Here and there were small rounded bodies consisting apparently of a fused mass of cells and surrounded with a rather definite fibrous capsule. These are shown in fig. 5 of Plate VI. Evidently they are the cyst-like structures seen in the examination of the fresh material.

What we have, therefore, in the semi-liquid and nodular parts of the tumour is an autolysing mixed-cell sarcoma. Degeneration has proceeded very far, with the result that little of the original structure of the tumour has persisted.

Parts of the muscle substance cut out from the margins of the tumour were now embedded and cut, but most of these showed no sarcomatous tissue, only muscle fibres, attenuated, so to speak, and separated from each other by spaces larger than normal. But on examining these tissues some were found in which the sarcoma could be detected by its difference in consistency. A section through such a part is shown in Text-fig. 4.

Here we have, plainly, a sarcomatous tissue which has probably ceased to infiltrate the normal tissue. On

the lower right of the figure is a muscle septum or dissepiment with "attenuated" muscle fibres lying beneath it. Above this is the broken-down sarcoma. There are spaces which were doubtless filled with a semi-liquid cell mass, which became lost on cutting out the tissue. On the upper left of the figure is part of a nodule. There is a very distinct limiting layer or capsule to this nodule. The figure is magnified about

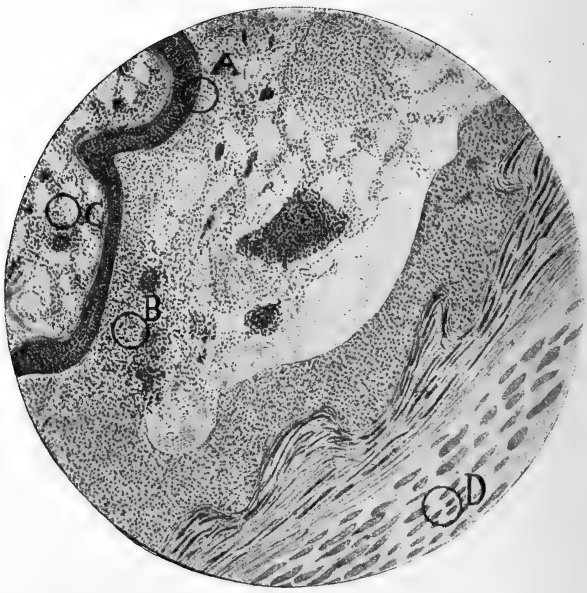


FIG. 4. Part of a section through the muscle substance adjacent to the large tumour shown in Text-fig. 3.

twenty diameters, and the small circles A to D drawn on it represent the fields shown under an oil-immersion lens and drawn as figs. 4, 6, 7 and 8 of Plate VI.

Fig. 7 is the field represented by D in Text-fig. 4; it shows the "attenuated" muscle fibres. These are difficult to draw. They are evidently greatly altered.

Some are homogeneous and with a distinct sarcolemma, but with no trace of striation, while others show the muscle-fibre substance in a state of fragmentation and with either no trace of sarcolemma, or with this investment in tatters, so to speak. Here and there are traces of the utter disintegration of the fibres. What we have here is, therefore, the degeneration of the surrounding un-infiltrated muscle substance as the result of the products of the necrosis of the sarcoma.

The field B, that is fig. 4, is a small part of the otherwise broken-down sarcoma, where some remains of structure can be seen. It is here a "spindle-celled" or an "oat-cell" sarcoma, and little autolytic change or structural disintegration is evident. There are two "giant-cell" masses, that is, groups of "clumped" or "agglutinated" cells. These are not surrounded by a capsule, but one can see a roughly concentric arrangement of the spindle cells round them. They are probably the same kind of bodies as are represented in fig. 5, except that in the latter case a process of fibrosis has taken place round the cell masses. Fig. 4 probably represents the original structural form of the sarcoma.

Fig. 6 represents the details of the capsule shown at A in Text-fig. 4. The capsule is a structure consisting of small spindle cells, but the proportions of the "spindles" vary greatly. The lower part of the figure represents the left part of the capsule as seen in Text-fig. 4. Here the spindle cells break down, or fuse together, so that the cell outlines become obscure or quite absent; and the nuclei disintegrate. A Mallory-stained preparation showed that the inner part of the capsule was greatly altered. The upper part of the figure represents the semi-liquid cell débris already spoken of.

Thus the sarcoma was originally a multiple one.

There were many centres of growth. The part provided by this capsule was originally one of these multiple tumours. The nodules found in the degenerate tumour were most probably the remains of the originally separate tumours. In the central part of the whole mass these separate tumours quite broke down as the result of necrosis, and their capsules disappeared and are represented only by scattered fibrous remains.

Finally fig. 8 of Plate VI, representing the field C of Text-fig. 4, that is, the interior of the mass bounded by the capsule A shows evidently the same structure as that represented by fig. 4, that is, simply a broken-down, partially necrosed sarcomatous tissue.

The specimen is worth description in detail since it illustrates fairly well the penultimate fate of a large piscine sarcomatous growth. Summarising, we find that a multiple sarcoma of the mixed-cell type has grown to impossible dimensions. Deficient vascular supply (for the paucity of blood vessels in such tumours is notable) has prevented the removal of products of katabolism, and has starved the cells. "Stewing in their own juice," the sarcomatous cells are being killed and are disintegrating into "mush." Accompanying this process of poisoning of the cells is probably the process of true autolysis, or self-digestion of cells and capsules by their own enzymes. Products of cell excretion diffusing into the adjacent muscle substance have affected the latter injuriously, so that degeneration of the fibres is going on.

These excretory products entering the blood stream of the fish have produced the general marked emaciation of the animal. The ultimate phase of the development of the growth would probably have been the rupture of the tumour with the formation of a huge abscess. This would certainly itself have been fatal to the fish. But it

is unfortunate that this definitive stage could not have been observed.

2. Haemangioma in the Eye of a Stickleback.

A three-spined Stickleback (*Gasterosteus purigitius*) caught among some shrimps was very noticeable because of a tumour on the left eye. The pupil was surrounded by a raised annular swelling, brown in colour, and the cornea was mottled with opaque specks, while small arborescent growths stood out from its central parts. The thing looked so queer that sections were made. The fish had been preserved in formalin, but the tissues were in an excellent state for histological examination.

Fig. 1 of Plate VII represents a section passing transversely through about the middle of both eyes. The lenses are entire, but the retina and choroid have become retracted away from the sclerotic. The stippled area between choroid and sclera is the choroidal gland. Bones and blood vessels are shaded dense black. The two bones on the dorsal side of the head are obviously the frontals, and that just above the pharynx is the parasphenoid. The others I have not identified.

The tumour is represented in the figure by the darkly-stippled area outside the eye on the left (the section is seen from behind). The cornea of this eye is also seen to be thickened, and there are growths on the conjunctiva. The tumour extends dorsally along beneath the skin to the region of the right frontal, and ventrally, also beneath the skin, to the level of the pharynx. There are two large spaces beneath the eyes, lymph-spaces, but I have not traced them, and these are partially filled with a loose tissue consisting of blood capillaries and numerous lymphocytes. All parts of the tumour stain, as does a knot of blood capillaries.

Fig. 1 of Plate VI represents a field of the denser part of the tumour as seen under an oil-immersion lens. What we have here is simply a dense plexus of blood capillaries. There is no other tissue present, and the vessels are gorged with blood corpuscles, so that hardly anything but the nuclei of the latter embedded in an almost homogeneous matrix is to be seen. Fig. 2 of the same plate shows a looser region of the tumour where the blood corpuscles have been partially removed in the process of washing the section. The walls of the capillaries can now be seen, with many contained red blood corpuscles. The latter are quite normal, and so are the vessels themselves. There is no interstitial tissue, and the walls of the capillaries are only very slightly thickened. Their calibre is, however, greater than normal.

We may, in a loose way, call this tumour a haemangioma, but it is hardly the same thing as a human tumour of this kind regarded as a true blastoma, or malignant new growth. Undoubtedly there is growth of the vessels in the sense that they are very abundant in a region where they are normally scanty, and we may call the structure a tumour since it has produced a very noticeable external swelling. But the vessels themselves are almost normal in structure—they are simply very numerous. Perhaps the nearest approach to this condition among human pathological ones is that of a varicose vein or a haemorrhoid. Like the latter structure, this tumour has probably been induced by some obstruction in the circulation. The condition, however, is quite interesting.

It will be noticed that secondary changes have been induced in the cornea. This region of the eye in Teleost fishes differs in some particulars from that in the higher

mammal. Text-fig. 5 represents the histology of the corneal region in the normal and abnormal eyes of this stickleback.

In such a fish as this the structures in the neighbourhood of the corneal region of the eye are as follows (from without inwards):—(1) The conjunctiva composed of an outer limiting membrane, or cuticle, a stratum of epithelium from about two to six cells deep, and an inner limiting membrane. The cells are rounded

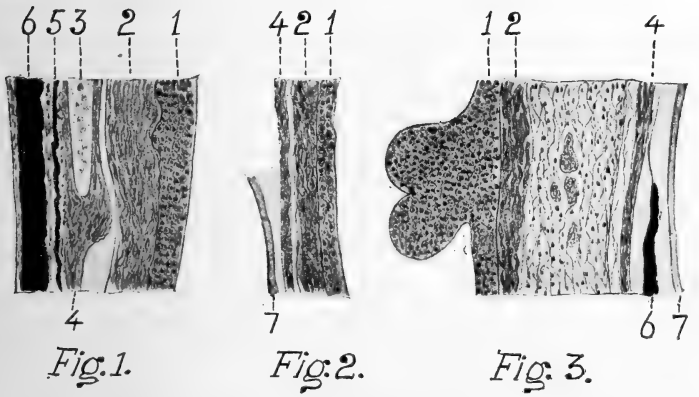


FIG. 5. (1) The normal corneo-sclerotic junction; (2) The normal cornea over the central part of the lens; (3) The abnormal cornea at the marginal part of the pupil. The numbers 1-6 in the three figures refer to layers which are continuous over the bulbus oculi within the limits indicated.

or polygonal, horny, and show no evident tendency to become squamous. (2) The cornea proper, which consists of a rather close layer of thick, transparent, elastic fibres, running in a slightly convoluted direction, and bounded internally by a very thin limiting membrane. (3) The sclerotic. This is partly cartilaginous in a Teleostean fish. The cartilaginous part ceases at the corneo-sclerotic junction (fig. 1), and the remaining layer (4) consists of elastic fibres which lie mainly external to the

cartilaginous layer in the posterior part of the bulbus. They are continued over the lens. (5 and 6) The "supra-choroid" and choroid, which are the pigment layers betwen the retina and the choroidal gland. They are produced anteriorly to the internal margin of the iris. (7) The capsule of the crystalline lens.

If we compare 3 with 2 in fig. 4 we shall see that a new layer has been intercalated between the conjunctival and sclerotic elements of the cornea. This is fairly thick, and consists of loose areolar tissue containing lymphocytes in its interstices. Here and there, there are small groups of blood capillaries; some of these are shown in the figure, and evidently they are capillaries that are growing centrally from the main tumour in the marginal parts round the eye.

This abundant blood supply round the eye and in the cornea itself is doubtless responsible for the growth of the epithelial layer of the cornea. The small knob-like, or even arborescent, growths on the conjunctiva have the same structure as the latter layer, that is, they consist of horny (in the sense that they have evident walls) cells, spherical or polygonal in shape. They do not become squamous towards their free surface, but are everywhere of the same form. We cannot call these little growths epitheliomata or papillomata since they have no core of connective tissue. They are solid outgrowths from the conjunctiva—conjunctival warts we may perhaps call them.

3. Papillary Cystadenoma in a Ling (*Molva molva*).

In March, 1914, Mr. T. Bailey sent me a roe taken from a Ling landed at Fleetwood by a steam-trawler. The structure did not look in the least like a roe, but

Mr. Bailey's identification was based on the conclusion that it could not be anything else, since all the other viscera could be identified. Familiarity with the appearance of the organs in the body cavity of a fish, acquired by seeing very many animals gutted, renders such a "process of elimination" a very certain one in its results. As a matter of fact, the specimen is an interesting example of an ovarian cystadenoma.

The ovary of a Gadoid fish is a closed sac communicating with the exterior by a short, wide oviduct (a different thing, morphologically, from the oviduct of such a fish as an Elasmobranch). Numerous lamellae project from the inner surface of this sac radially towards the centre. These lamellae, and the internal surface of the ovarian sac, are clothed with germinal epithelium, and as the ovary ripens the ova form in this epithelium, enlarge, and finally dehisce into the cavity, where they pass through the final stages of maturation. After spawning, the ovary becomes a flaccid, thin-walled, almost transparent sac, consisting of thin germinal epithelium, with, of course, added layers of connective tissues and peritoneum. The extent of alteration in this particular organ can be gathered by comparing this description with Plate IV and Text-fig. 6.

Plate IV represents, one-half natural size, the whole ovary. When fresh it was a repulsive-looking structure, evidently a mass of cysts of various sizes, and with walls of varying thickness. On cutting into these cysts a clear glairy, albuminous fluid ran out, and the cyst walls collapsed. In many of these cysts were inclusions, two of which are represented as fig. 2 of Plate V.

Text-fig. 6 represents a hand section through the middle of the ovary. We see now that it is a hollow sac with enormously thickened walls, in the interior of

which the cysts are developed: the latter may, of course, also have been formed by fusion, or coalescence of parts of the ovarian lamellae. The solid parts are the locus of a process of fibrosis, and the black, irregular objects in some of the cysts are sections of inclusions. As a rule the cyst walls are thin, and the massive tissue occupies the angular spaces between cysts.

Fig. 2 of Plate V represents two of the inclusions, very nearly natural size. They are very irregular in size and shape; yellowish in colour; very hard, so that

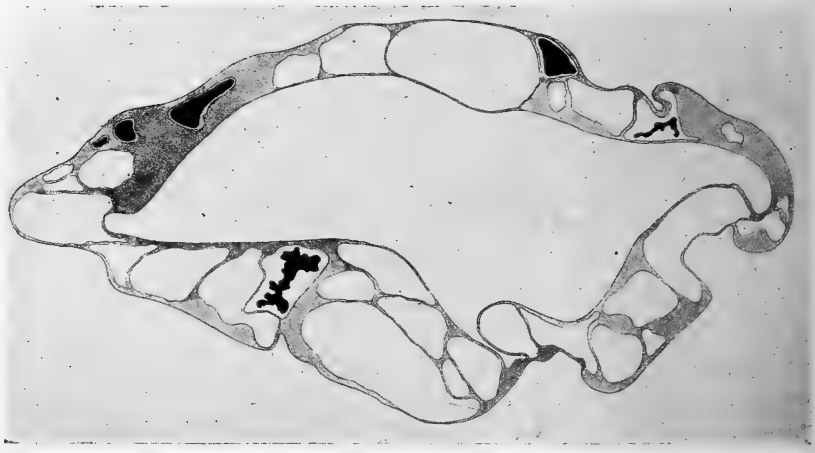


FIG. 6. Transverse Hand Section of the abnormal ovary of a Ling; half natural size.

they spoil the edge of a razor when it is attempted to cut a hand section. They are evidently calcified to some extent, for they become softer after immersion for a day in decalcifying fluid. Then hand sections may be made, but the study of such, after staining lightly with borax-carmine, gives very little information. They are structureless, in the sense that there are no organised cell-groupings or layers, nor any fibrous tissue. They are

"cheesy," and present an obvious lamination in their marginal parts. Generally they are very similar to the substance of the ovarian concretion which I described in last year's Report*; and like that, are probably composed, typically, of a sclero-proteid substance.

Plate I illustrates the minute anatomy of this ovary. First of all, the massive tissues are composed of fibrous tissue such as that represented in fig. 4. The fibres are short and thick, and usually slightly convoluted. In some places they tend to run concentrically round nuclei of formation. Among them are a few cells of various sizes. The greater part of the structure is therefore a fibroma.

Fig. 3 represents the structure of the epithelium lining the mucous cysts. The specimen had been fixed in formalin only, so that the finer details of structure are not favourably exhibited. Probably some disintegration of the epithelium has taken place. It appears, indeed, as if a goblet-celled layer may have been present, and had latterly been broken up with the process of secretion of the liquid filling the cysts. In the sections made there is only a structureless layer containing, here and there, some migrant leucocytes. Below this is a layer of areolar tissue of the usual kind. This is, in places, infiltrated with leucocytes. Below this is the fibrous tissue described above.

Fig. 2 represents a part of a section of the massive tissue containing a cavity, evidently of the same nature as those of the mucus-containing cells. Within it is the section of a body of different nature. Fig. 1 represents the marginal part of a section of this body. Centrally it is very similar in structure to the

* *Ann. Rept. Lancashire Sea Fish. Laboratory for* 1913; pp. 41-48, pl. III. Liverpool, 1914. (In *Trans. Liverpool Biological Soc.*, Vol. 28, 1914.)

tissue of the massive parts of the ovary, but the fibres are thicker and more convoluted. On the whole they suggest a process of alteration, and a progressive destruction of morphological detail. The epithelium is represented in fig. 1: it consists of ellipsoidal cells which flatten out towards the free margin very much as those of an epidermal layer; indeed, this epithelium is very like an epidermis, except that the "prickles" between the cells are quite absent.

It is difficult to resist the notion that we have here the earlier stage of one of the inclusions contained in the cysts. The latter are horny, and have evidently been altered in the direction of the complete obliteration of their finer structure, but the section just described may represent what they were like during their stages of growth.

The specimen seems, therefore, to be a multilocular cystoma, and it presents many points of resemblance with the papillary cystadenomas described in works on human pathology. Fibrosis of the tissues of the ovary has probably preceded the process of cyst formation. The inclusions in the cysts are probably invaginations of the walls of the latter with original modification of structure. Later on these invaginations become pinched off from the cyst-walls, and their vascular connections become lost. Alteration—some kind of hyaline degeneration—then occurs so as to produce the hard, partially calcified inclusions.

4. A Plaice with One Eye.

A small plaice about 10 cms. in length, sent to us in a sample of other fish, attracted instant attention on account of the complete absence of the left eye. The head of the fish is represented in fig. 1 of Plate V, and it

will be seen that the right eye is quite normal, whereas the left orbit is covered over by skin and has a little raised part at its centre. It was expected that something interesting might be disclosed on a more minute investigation, so a series of sections through the orbital region were made. One of these sections passing nearly through the centre of the right eye is represented in Text-fig. 7.

Both eyes ought to appear, nearly meridionally, in the same section. The left eye is, however, wanting, and the connective tissue which lies immediately underneath the integument occupies very much the position which would have been that of the sclera of the left eye had it been present. The eye-muscles are present, and are nearly normal in their development. The ophthalmic artery (that is, the efferent pseudobranchial vessel, or the afferent vessel of the choroidal gland) is also present, with its vein. The optic nerve is present, although it is greatly reduced distally. In a transverse section of a normal plaice having the plane of that of Text-fig. 7, both optic nerves should have nearly the same area of cross section. Further back than the plane of section of Text-fig. 7 the eye-muscles and nerves of both sides approach each other and enter the common eye-muscle canal together, precisely as in the case of a normal fish. Tracing the eye-muscles and other structures forward in the series of sections, they are seen to die out. They end in the mass of fibrous connective tissue shown in the figure just dorsal to the muscles. The ophthalmic vessels become occluded and disappear from the sections in the same tissue.

The integument lining the orbit of the missing eye is modified. It becomes very much thicker, and there is a great development of sub-epidermal fibrous tissue (not a real corium). The epidermis contains very numerous

"goblet" mucus-secreting cells, far more than are contained on adjacent regions of the head. In the central parts of the orbit this epidermis is thrown into complex folds forming a rosette-shaped excrescence, part of which is represented in the figure.

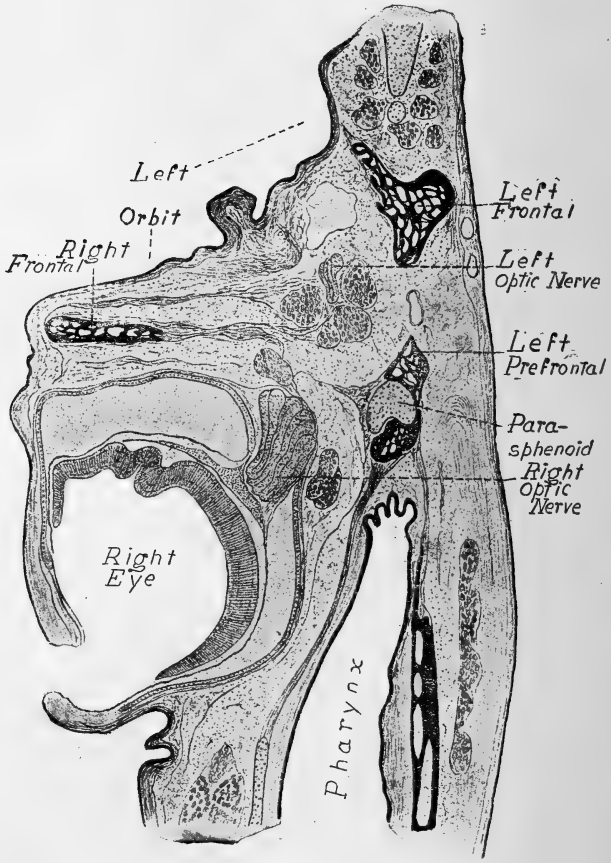


FIG. 7. Transverse section through the orbital region of the head of the Plaice represented in Fig. 1, Pl. V. The dorsal and ventral parts of the head have been cut away for convenience in making the series of sections, and the lens of the right eye has been removed. The drawing is slightly diagrammatic in that the minute structure of the tissues is not represented. The proportions and positions of the various parts are accurately represented.

It is hardly possible that the absence of the eye is congenital since the optic nerve and the eye-muscles could not have attained the development indicated. It is possible, however, that a failure to form the lens in early development may have led to this abnormality. In the absence of the ectodermal invagination forming the lens the bulbus oculi may have degenerated: there is no reason why the optic nerve should have degenerated since the trophic centre is in the brain. That this happened seems, on the whole, more likely than that a traumatic lesion led to the disappearance of the whole eye, since in the latter case there ought to have been a much greater development of scar tissue than the sections show. As it is, there is nothing in them to suggest granulation tissue, and the development of mucus-secreting cells in the integument lining the empty orbit is very striking.

EXPLANATION OF THE PLATES.

Plate I. Papillary Cystadenoma in a Ling.

Fig. 1. Section through the marginal part of the inclusion represented in fig. 2. Superficially there is an epithelium of ellipsoidal cells becoming flattened towards the surface. Centrally there is coarse fibrous tissue undergoing hyaline degeneration. Oil-immersion lens.

Fig. 2. Section through part of the massive tissue of the cystadenoma, and through a cavity containing an inclusion. The latter is probably a stage in the formation of the hard, cheesy inclusions represented in fig. 2, Pl. V.

Fig. 3. Section through the epithelium lining a cyst. The upper margin of the figure represents the internal surface of the cyst. There is a basement membrane, and beneath this a layer of areolar connective tissue with leucocytes. A goblet-cell epithelium was probably present, but has disappeared. Oil-immersion lens.

Fig. 4. Part of the tissue of the massive part of the cystadenoma. Coarse fibrous tissue. Oil-immersion lens.

Plate II. Multiple Sarcomata in Halibut.

Fig. 1. The edges of three distinctly capsulated tumours. The stippled areas represent the sarcomatous growths, which are here mainly a fibrous tissue with very numerous round and spindle cells. The latter pass into a fibrous tissue. The capsules are fibrous, but are not strongly developed. Mag. about 25 dia.

Fig. 2. An unencapsulated sarcomatous nodule among adipose tissue near the ventral fins. Fibrous tissue becoming replaced by fat cells. The sarcoma is invading the coarser fibrous tissue between the fat cells. There is no trace of a capsule. Mag. about 25 dia.

Fig. 3. Edge of a sarcomatous nodule showing the breakdown of the capsule. The nodule is surrounded by muscle fibres, and has at most of its periphery a fibrous capsule. At the place shown the capsule disappears, and the sarcomatous tissue is infiltrating the surrounding muscle-tissue. The stippled area represents closely-crowded ellipsoidal or spherical cells. See also Pl. III, fig. 1. The diameter of the whole field is about 2 mm.

Fig. 4. The growing edge of a sarcomatous nodule infiltrating dense fibrous tissue. To the right is the typical sarcoma; to the left is dense fibrous tissue with coarse bundles. Degenerating muscle fibres are represented above. Diameter of the whole field about 2 mm.

Fig. 5. A small part of the tumour shown in fig. 4, where the sarcoma is typically developed. Small, round cells; larger irregularly rounded cells; some spindle cells. Cell débris, and melanin granules. Apochromatic lens.

Fig. 6. A small part of the growing edge of the tumour shown in fig. 4. Small, round cells infiltrating connective tissue. Apochromatic lens.

Fig. 7. A small part of the dense, fibrous tissue which lies to the left in fig. 4. It is well away from the growing margin of the tumour, but some small ellipsoidal and round cells are contained

in the interstices of the fibrous bundles. Connective tissue nuclei are shown. Apochromatic lens.

Plate III. Multiple Sarcomata in Halibut.

Fig. 1. A small part of the growing margin of the tumour shown in fig. 3, Pl. II. Small oat-shaped cells, about 0·0065 mm. in longest diameter. Muscle fibres undergoing degeneration. Some unchanged muscle fibres. Apochromatic lens.

Fig. 2. Part of the same tumour near the margin. Short spindle cells with a few round cells. Apochromatic lens.

Fig. 3. Part of the same tumour near the centre. The sole tissue, in places, consists of small round cells. Apochromatic lens.

Fig. 4. Part of the same tumour in the central part where there is melanin formation, and softening due to necrosis. The tumour here is a "mixed-cell" one. Few spindle cells. Many small cells, larger irregularly-rounded cells, multinuclear cells, "giant cells," some cell débris and melanin deposits. Apochromatic lens.

Fig. 5. Part of the same tumour near the periphery, but not a part of the capsule. The spindle cells shown in fig. 2, Pl. III, pass here and there into a fibrous tissue in which the cells seem to be produced to form coarse, loosely interlaced fibres. This is represented in the present figure. Apochromatic lens.

Fig. 6. Normal tissue such as forms the *locus* of these tumours. Essentially muscular tissue with much loose fibrous tissue between the muscle bundles. There is usually extensive fat formation. A vessel, blood-capillary or gorged lymph-capillary, is shown. Apochromatic lens.

PLATE IV. OVARIAN CYSTADENOMA IN A LING.

The whole ovary, about $\frac{1}{2}$ natural size.
Photo. by A. Scott.

PLATE V.

Fig. 1. Plaice with one eye. Natural size.

Fig. 2. Two inclusions from the cysts shown in the photograph above (Pl. IV). Slightly enlarged.

PLATE VI.

Fig. 1. Haemangioma in a Stickleback. Section through the fully-developed tumour. Engorged and dilated blood capillaries without interstitial tissue. Oil-immersion lens.

Fig. 2. The same. Part of the same section where the capillary plexus was looser. Some of the contents of the vessels are removed in the process of preparing the section. The vessels contain typical red blood corpuscles. Oil-immersion lens.

Fig. 3. Sarcoma in a Cod. Section of the tumour in the muscles adjacent to the degenerated tumour. Spindle and oat-shaped cells, with nuclei due to coalescence of the cells. Oil-immersion lens.

Fig. 4. The same. Part of the section in an area of necrosis. Matrix of cell débris. Spindle-cells, oat-shaped cells, and spherical cells lying loosely. Oil-immersion lens. Field B in Text-fig. 4.

Fig. 5. The same. Part of a nodule lying loose in the necrosed tissue of the tumour. "Giant-cell" masses are surrounded by strong fibrous investments. Oil-immersion lens.

Fig. 6. The same. Part of a section of the sarcoma in the muscle tissue adjacent to the necrosed mass. The figure shows the details of a capsule surrounding a broken-down mass of sarcomatous cells. The upper part of the section represents almost completely broken-down tumour cells. Oil-immersion lens. Field A in Text-fig. 4.

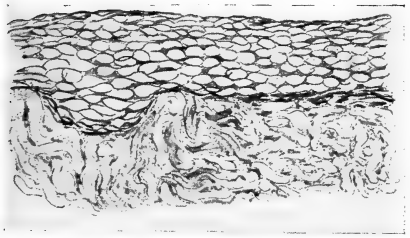
Fig. 7. The same degenerating muscle fibres in the vicinity of the main tumour. Oil-immersion lens. Field D in Text-fig. 4.

Fig. 8. The same. Necrosed sarcomatous tissue in the muscle tissue adjacent to the main tumour. Oil-immersion lens. Field C in Text-fig. 4.

Plate VII.

Fig. 1. Haemangioma in a Stickleback. Transverse section through the head of the fish in the region of the eyes. The tumour is represented by the darkly-stippled area round the eye on the left of the figure.

Fig. 2. Sarcoma in a Cod. Cell-elements found in a smear made from the semi-liquid part of the necrosed tumour. Oil-immersion lens.



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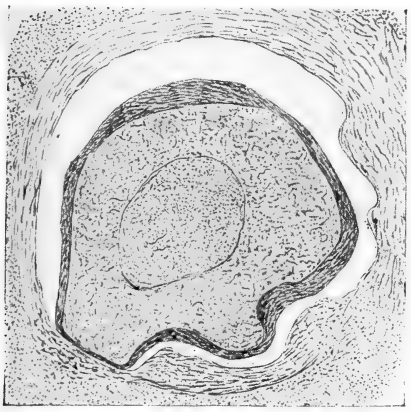
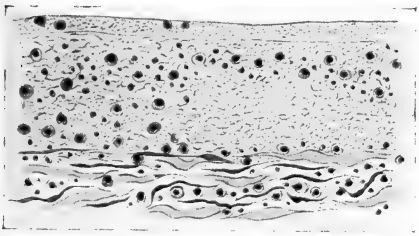


FIG. 2.



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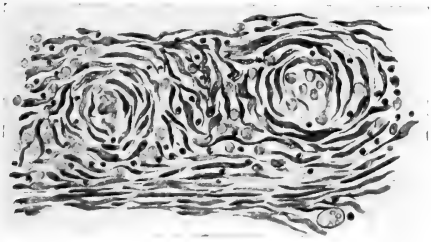
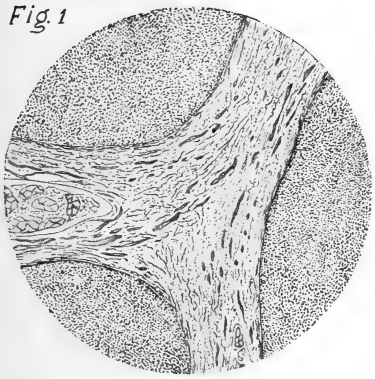
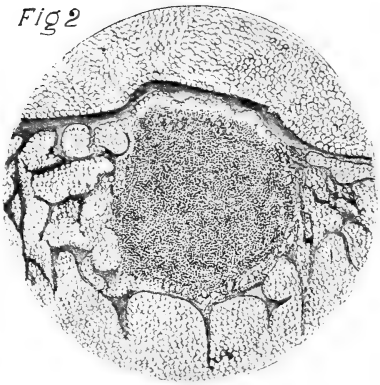
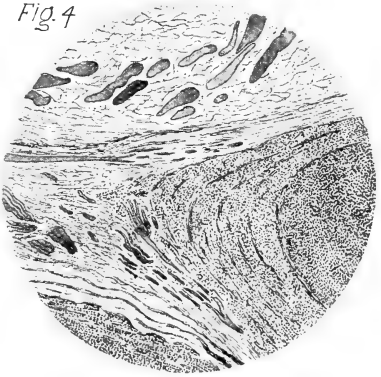


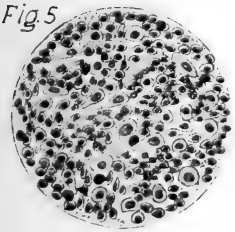
FIG. 4.

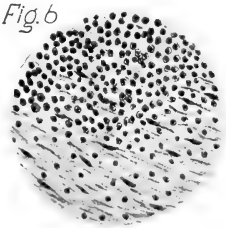
PAPILLARY CYSTADENOMA IN A LING.



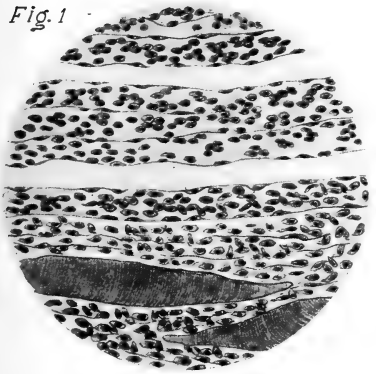


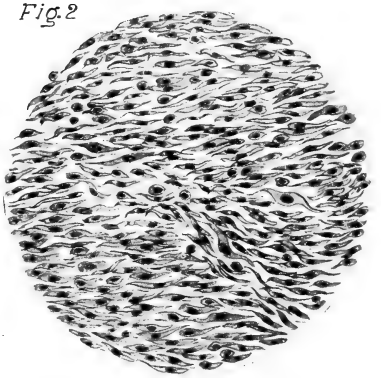


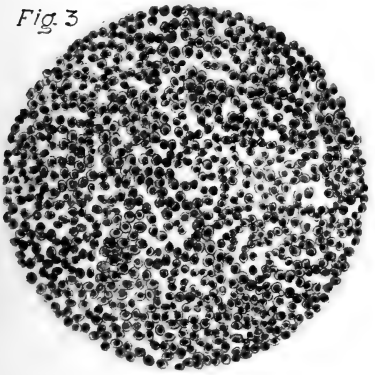


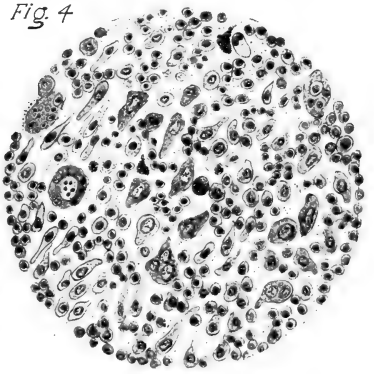


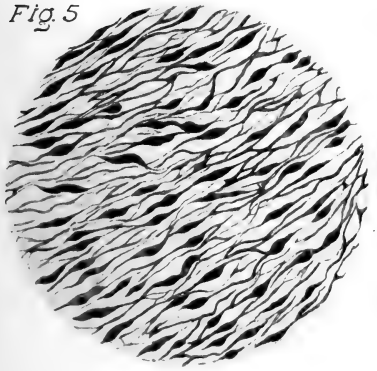
MULTIPLE SARCOMATA IN A HALIBUT.

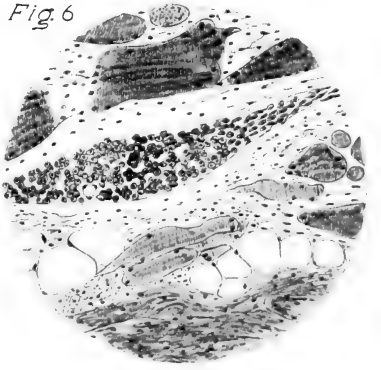




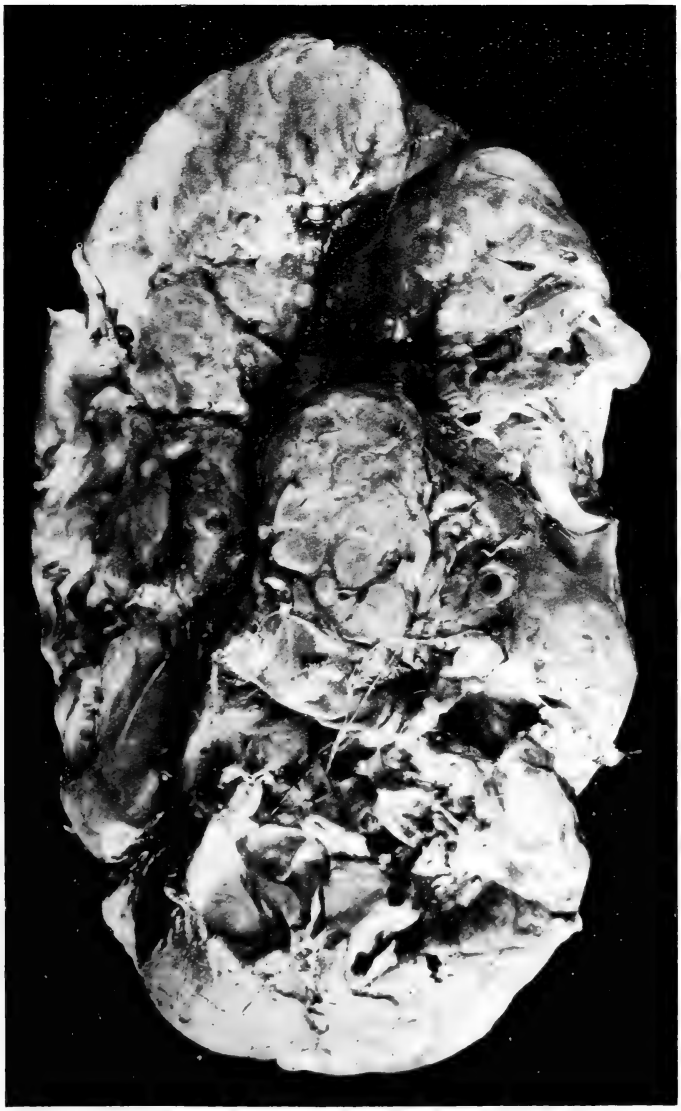








MULTIPLE SARCOMATA IN A HALIBUT.



PAPILLARY CYSTADENOMA IN A LING.

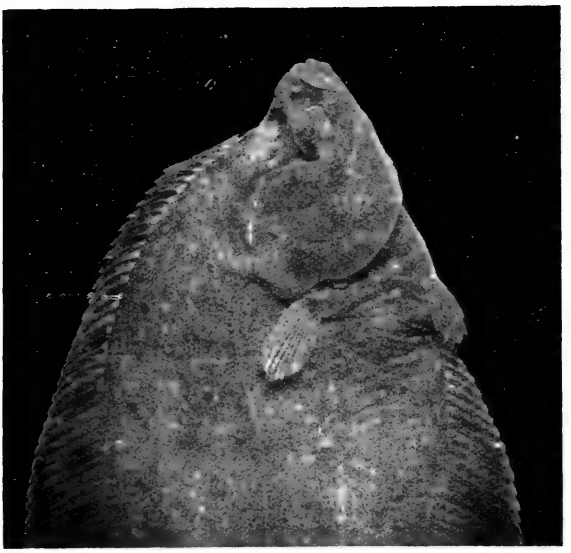


FIG. 1.

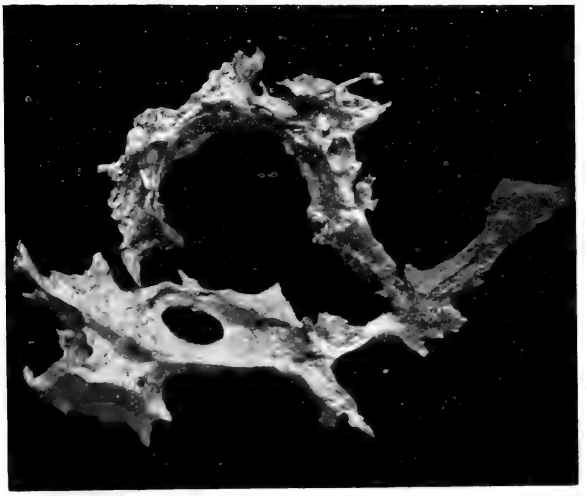


FIG. 2.

FIG. 1. Plaice with only one eye.
FIG. 2. Inclusions from Papillary Cystadenoma in a Ling.

PLATE VI.

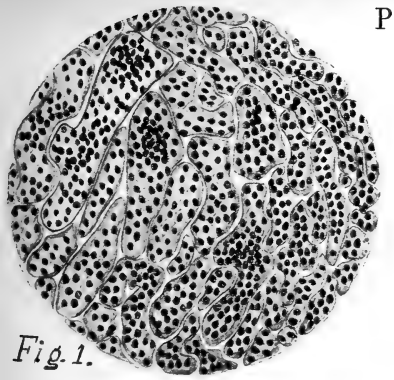


Fig. 1.

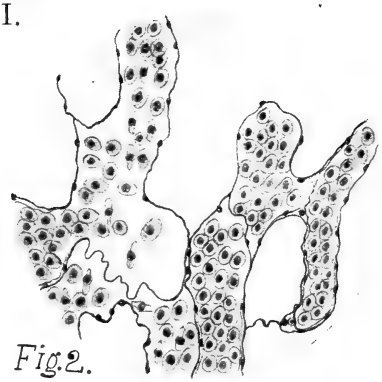


Fig. 2.

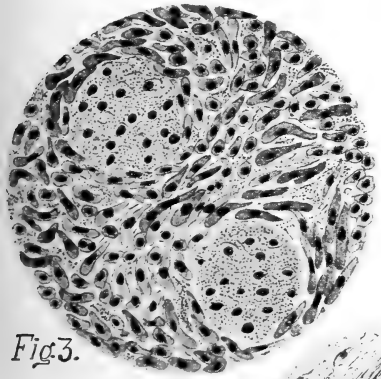


Fig. 3.

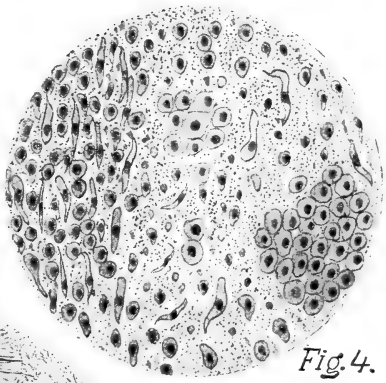


Fig. 4.

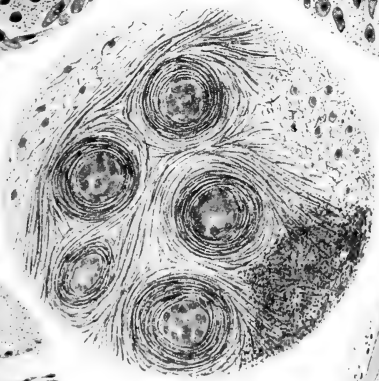


Fig. 5.

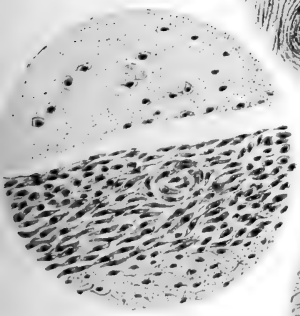


Fig. 6.

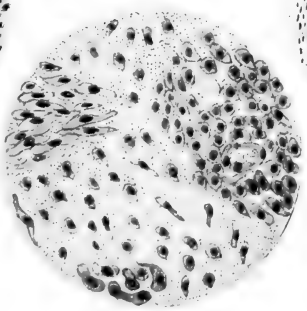


Fig. 8.

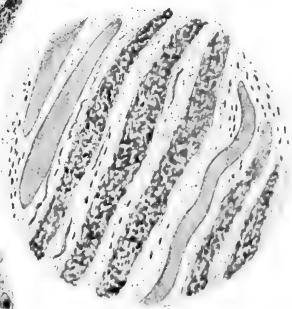


Fig. 7.

HAEMANGIOMA AND SARCOMA IN FISHES

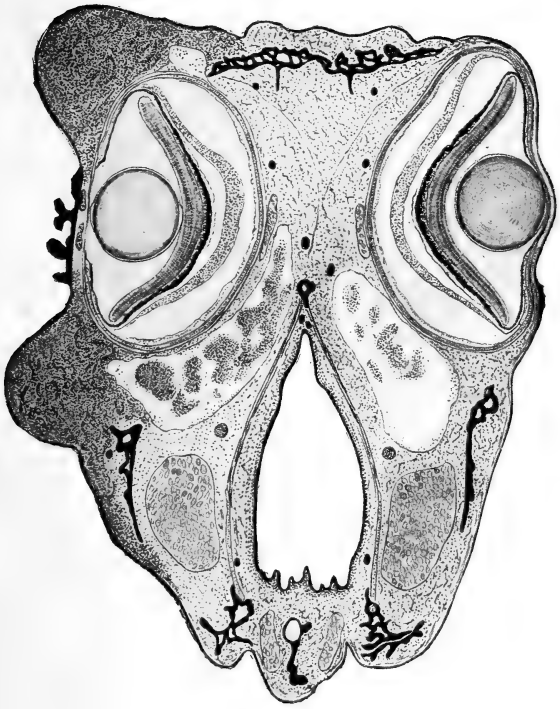


FIG. 1.

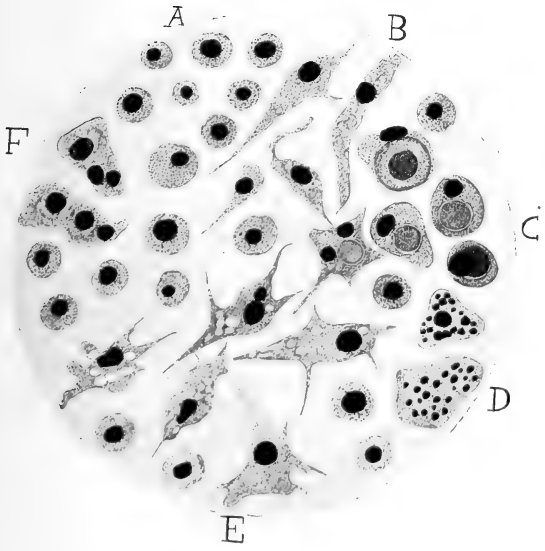


FIG. 2.

FIG. 1. Haemangioma in Head of Stickleback.
FIG. 2. Sarcomatous Cells from Cod.

THE METHODS OF CLEANSING LIVING MUSSELS FROM INGESTED SEWAGE BACTERIA.

By James Johnstone, D.Sc.

(With a Chart and two Plates.)

Contents.

Introduction.

Some time ago the Chairman of the Scientific Sub-Committee requested me to make some investigations with regard to practicable means of cleansing mussels from sewage bacteria. Some experiments of this kind were made in the past with respect to one definite locality—The Estuary of the Conway—and favourable results were obtained. In these experiments some boxes containing mussels were simply laid down on the beach, being fixed so that the tide could not wash them away. The idea underlying the experiments was to expose polluted mussels to sea-water of the last two hours or so of flood, and the first few hours of the ebb-stream. Flood-stream water is practically unpolluted, and the first of the ebb-stream water may also be regarded as unpolluted. In these circumstances the mussels rapidly cleansed themselves of about 90 % of the sewage bacteria originally contained in them. The results were thus very satisfactory, but they

have not general application. Other conditions, varying in every channel and estuary, must be considered. The nature of the foreshore, and the facilities for transport to and from the cleansing grounds are important factors, and they determine whether or not this method of cleansing is practicable.

About the same time the Conway Corporation made proposals for cleansing the mussels taken from the grounds under their control by treatment with sterilised sea-water. There were many practical objections to the proposal for relaying the mussels on the part of the foreshore where I had made my experiments, and at no other place could unpolluted sea-water be obtained. The Corporation suggested, therefore, to pump sea-water into large tanks, and then to sterilise this water by adding chlorine solution. After removal of the chlorine the sterile water was to be made to flow over the mussels in the cleansing tanks. The Chairman also requested me to test this process on a small scale so as to obtain some indications of the minimal proportion of chlorine necessary to destroy the sewage bacteria, and the maximal proportion which the mussels could stand without injury to their normal functioning. The length of time required for cleansing, and some other points, were also to be investigated. Accordingly Mr. A. Scott and I made some experiments at Piel during last April and May, and I repeated these experiments, with some others, at Liverpool later in the year.

Only a few direct experiments were, however, made. It was soon seen that it was theoretically possible to cleanse mussels by means of sea-water rendered practically sterile by dosage with chlorine. This knowledge, however, goes only a little way, and it does not seem possible to anticipate, merely by laboratory experiments, the difficulties that are sure to be encountered in working the process on the commercial scale. Some time afterwards, the Conway Corporation obtained a grant from the Development Commissioners to make this

experiment on a large scale, and the conduct of the operations was undertaken by the Board of Agriculture and Fisheries. The works at Conway, now in course of erection, will therefore provide the proper means of investigating the practical details of the process to be adopted.

While the few experiments to which I refer above were being made some other questions suggested themselves. In spite of the close attention that is now apparently directed in this country to public health problems, precise information with regard to the pollution of shellfish is difficult to obtain. One finds no agreement among public health bacteriologists as to what ought to be meant by "*Bacillus coli*." There are no generally recognised methods of analysis, and there is no generally recognised "standard of permissible impurity." On the one hand bacteriologists like Houston, Klein, Savage and others speak of "typical" and "atypical" *Bacillus coli*, while on the other hand workers like MacConkey and Clemesha have shown that faeces, sewage and polluted materials generally contain a number of micro-organisms, all of which are what a biologist would regard as "specifically distinct" from each other. These micro-organisms are separable from each other by the application of a number of fermentation tests, and a careful analysis shows that the old "*Bacillus coli*" of Houston, and others, is really a category of half a dozen to a dozen distinct micro-organisms. If all these bacteria had the same significance from the public health point of view, that is, if they all indicate faecal pollution, there is no need of distinguishing between them. Further, all these bacteria may live normally and multiply in faeces, but it is not known whether they live and multiply in estuarine sea-water, and in the bodies of shellfish. Even if we did not know that many of the micro-organisms which inhabit the human alimentary canal die out when they pass into sea-water this would be probable. One would expect that the profound change of

habitat so experienced would influence the "bacterial flora" characteristic of faeces. Some organisms might be very resistant, and would maintain themselves, and multiply in sea-water and estuarine shellfish, just as they did in the alimentary canal of man; while others, being more delicate, that is, more habituated to a truly parasitic mode of life, would speedily die out when they enter a new medium.*

The organisms which inhabit shellfish must therefore be precisely diagnosed by the application of the fermentation reactions recommended by MacConkey, and their original habitat must be traced. Conversely the organisms inhabiting the human alimentary canal must also be differentiated from each other and diagnosed, and then we must study what is their behaviour in media like fresh water, estuarine sea-water, and the bodies of shellfish. A systematic description of the species of bacteria present in faeces, and an estimate of the relative abundance of the various forms ought to be made, and compared with a similar description and estimate of the species found in shellfish. There will be a difference, and this difference will be due to the change of habitat. The various faecal organisms ought then to be isolated in pure cultures, and their individual behaviour in sea-water and shellfish studied.

It must be confessed that it is easy to make such a programme of research, but difficult to carry it out. It has, in fact, been little more than suggested in this Report. The work is simply impossible for anyone who has anything else to do. The results here described merely suggest that much more valuable ones might be obtained if a really adequate investigation could be made.

* See MacConkey, "Further observations on the differentiation of lactose-fermenting bacilli, with special reference to those of intestinal origin," *Journal of Hygiene*, Vol. IX, No. 1, April, 1909. Clemesha, "The bacteriology of surface waters in the tropics," Calcutta, 1912.

(1) The Identity of the Micro-Organisms isolated.

There has been no opportunity for making a systematic and prolonged investigation of the species of lactose-fermenting bacteria inhabiting shellfish, and the data given in the following tables are only those obtained in the routine examination of samples which were examined with special objects in view. The regular method adopted in these analyses was to make an emulsion of the soft parts of five mussels, and then make up this emulsion with sterile water to a volume of 250 c.c. One c.c., containing 1/50th part of a single mussel, was then plated in neutral-red, bile salt, lactose agar and incubated at 37° C. for 24 hours. Five such plates were usually made, so as to get a reliable mean estimate of the numbers of lactose-fermenting organisms per mussel. A number of colonies from each plate were then isolated and subcultures were made on nutrient agar. After incubation, for a period of 4 to 6 hours, the motility, or lack of motility, of the organisms was observed, and after further incubation for 2 days the fermentation tests were applied. The various media were inoculated and incubated for 4 days before the results were recorded. The symbol + in the tables means that acid and gas formation took place in the sugars; that a distinct pink colour was observed in a peptone water culture on the addition of paradimethylamidobenzaldehyde (the method being that of Marshall, *Journal of Hygiene*, Vol. VII, p. 581); and that a peptone-glucose culture became pink within 24 hours after the addition of strong caustic potash solution. If a sugar medium was merely discoloured, or rendered acid, without gas formation, the reaction is described as negative. These results are clear and definite, but this is hardly the case with the observations of the presence or absence of motility in the organisms studied. Sometimes there was no doubt at all whether or not an organism was motile or immotile, but in very many cases it was not easy to be certain. In the attempt to identify the various organisms

motility is not one of the characters considered—in the meantime at least.

The Tables, Appendix I, give the results of these subcultures. It has been thought necessary to give the individual results in detail. The last Table is a summary of the results of examination of 72 organisms made previously, in former analyses. In these analyses indole formation was not observed, so that the results are not strictly comparable with those of 1914. But, on comparing them, it is fairly easy to see that the same species of bacteria have been isolated; and I have little hesitation in incorporating these former results with the latter ones, so as to present a first provisional description of the relative abundance of the different species of lactose-fermenting micro-organisms inhabiting sewage-polluted shellfish. In attempting to identify the species, I have made use of MacConkey's table given in the paper already cited. I reproduce this table here for easier reference. Not all MacConkey's tests have been cited, since I have only applied some of those used by him. But a rough comparison of his results and mine are clearly possible.

Lactose-fermenting Bacilli studied by MacConkey.

Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Name.
	+	−	−	+	−	+	−	−	*B. acidi lactici.*
	+	−	−	−	+	−	+	+	*B. levans.*
	+	−	−	−	−	+	−	+	*B. grünthal.* *B. sulcatus gasoformans.* *B. castellus.*
	+	−	−	−	−	+	−	−	*B. vesiculosus.*
	+	−	−	−	−	−		−	*B. coli mutabilis.*
	+	−	+	−	−	+	−	+	*B. coli communis.* *B. cavicida.*
	+	−	+	−	−	+	−	−	*B. schafferi.*
	+	+	+	+	+	+	+	−	*B. oxytocus perniciosus.*
	+	+	+	+	−	−	−	−	*B. rhinoscleroma.* *B. friedländer.*
	+	+	+	−	−	+	−	−	*B. neapolitanus.*
	+	+	−	+	−	−	+	−	*B. lactis aerogenes.* *B. dysenteriae vitulorum.* *B. capsulatus* (Pfeiffer).
	+	+	−	+	−	−	±	−	*B. gasoformans non-liquefaciens.*
	+	+	—	—	—	+	—	—	*B. coscoroba.*
	+	+	−	−	−	−	+	+	*B. cloacae.*
	+	+	−	+	−	+	−		Nos. 100, 101.

Table I.

Reactions of 197 organisms isolated from Mussels.

Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer reaction.	No. of organisms.	Percentage.		Name.
+	+	−	−	−	−	+	−	47	24	1	*B. grünthal* group. *B. vesiculosus.*
+	+	+	−	+	−	+	−	20	10	2	MacConkey's Nos. 100, 101.
+	+	−	−	+	−	+	−	18	9	3	*B. acidi lactici.*
+	+	−	+	−	−	+	−	14	7	4	*B. coli communis.* *B. cavicida.*
+	+	+	−	+	−	−	+	12	6	5	*B. lactis aerogenes.* *B. dysenteriae.* *B. capsulatus.*
+	+	+	+	−	−	+	−	9	4	6	*B. neapolitanus.*
+	+	+	−	−	−	−	+	8	4	7	*B. cloacae.*
+	+	+	+	+	−	+	+	8	4	8	
+	+	+	−	+	−	−	−	7	3	9	
+	+	+	+	−	−	−	−	7	3	10	
+	+	+	−	−	−	−	−	6	3	11	
+	+	+	+	+	−	−	+	5	3	12	
+	+	+	+	+	−	+	−	5	3	13	
+	+	+	−	−	−	+	−	4	2	14	*B. coscoroba.*
+	+	+	+	+	−	−	−	3	1	15	*B. rhinoscleroma.*
+	+	+	−	+	−	+	+	2	1	16	
+	+	−	+	+	−		−	2	1	17	
+	+	−	−	+	−	−	−	1		18	
+	+	+	+	−	−	−	+	1		19	
+	+	−	+	−	−	−	+	1		20	
+	+	+	+	−	−	+	+	1		21	
+	+	−	−	−	−	−	+	1		22	
+	+	−	−	−	−	+	+	1		23	
+	+	+	+	−	+		−	1		24	

Include also 13 organisms which do not ferment both glucose and lactose.

Table I shows that 37 distinct organisms have been separated out from the 200 isolated, that is, "distinct" if we are to understand that the presence or absence of the reaction with any one of the test substances used implies specific distinctness. It is impossible to say yet whether or not this is the case. If it is regarded as essential to being *Bacillus coli* that an organism should ferment glucose and lactose, it can also be urged that the ability to ferment any one of the other

sugars used, or to produce indole, or to produce the Vosges and Proskauer reaction, are also characters that differentiate species from species. It may be that under certain conditions the same bacillus may or may not ferment a certain substance, but in the absence of extensive research into the natural history of the organism we are not yet justified in assuming this. If we include motility of a bacillus (that is the presence of locomotory appendages, or flagella—a definite morphological character) as also a diagnostic specific feature, the number of species would be still greater. Now this character does seem to be one which has not the same value as the fermentation tests. There is some doubt as to the exact conditions in which motility ought to be looked for. MacConkey recommends that the nutrient agar culture should be examined after 4—6 hours' incubation, while Clemesha suggests the examination of an 18 hours' broth culture. It appears that motility may, therefore, be exhibited at one stage of a culture and not at other stages, and we cannot assume that cultures of different bacilli, in the same medium, and at the same time of incubation are strictly equivalent with regard to the morphology of the organisms. Further, motility is not always easy to observe, that is, it is not easy, at times, to be sure that the motion observed is not simply the Brownian movement of immotile particles. For these and other reasons I have not included this character among those regarded as diagnostic, though its presence or absence is recorded in the Appendix relating to the individual organisms.

But it is clear that the presence or absence of the fermentation reactions, with sugars other than glucose and lactose, are not simply matters of chance; that is, the Table does not merely show the permutations of characters theoretically possible. This is what MacConkey shows, and my own results are very much the same as his. Table I shows that four combinations of reactions, Nos. 1 to 4, are exhibited by half

of all the organisms studied. These, we may hold, are therefore really distinct species, or at least small groups of such. Comparing this Table with that published by MacConkey we also see that very much the same kinds of organisms have been found in both cases.

Group I, in my table, includes the species called, by MacConkey, *B. grünthal*, *B. sulcatus gasoformans*, *B. castellus*, and *B. vesiculosus*. The first three organisms differ from the latter one only in that they are motile while it is not so. No. 2 in my table appears to correspond with the organisms numbered 100 and 101 by MacConkey. No. 3, called *B. acidi lactici*, is very rare in MacConkey's list. *B. coli communis* and *B. cavicida* are much more abundant in MacConkey's list than in mine. Many of the other unnamed organisms may be found in both lists. The two groups of organisms may best be compared as follows :—

MacConkey. Organisms abundant in human faeces.		**Organisms present in Sewage polluted Mussels.**
B. neapolitanus	30 %	4 %
B. No. 71		
B. vesiculosus	22 %	24 %
B. grünthal		
B. sulcatus gasoformans		
B. castellus		
B. coli communis	20 %	7 %
B. cavicida		
B. schafferi	6 %	0 %
B. lactis aerogenes	4 %	6 %
B. dysenteriae		
B. capsulatus		
Organisms rare in human faeces.		
B. acidi lactici	0·5 %	9 %
B. coscoroba	0·5 %	2 %
B. cloacae	0·0 %	0 %
B. Nos. 100 *and* 101	0·5 %	10 %
B. rhinoscleroma	0·5 %	1 %

Thus we see at once that the organisms which appear normally to inhabit human faeces, and those which may be

found in sewage polluted shellfish, are not necessarily the same. Only one group—that formed by *B. grünthal* and its congeners are about equally abundant in both lists. MacConkey's organisms, Nos. 100 and 101, rare organisms in faeces, are fairly abundant in my samples of mussels, and the same is to be said of the lactic acid bacillus. Then it is very notable that *B. coli communis*, the third most abundant group in faeces in MacConkey's list, is much rarer in mussels. Generally, one sees that a notable change has occurred in the relative proportions of the bacteria present between the time when they leave the human intestine and the time when they may be found in marine shellfish. Some of the organisms are fairly resistant and appear to withstand the change of habitat, while others cease to multiply and die out more or less rapidly. It is also to be noted that organisms, having a general resemblance to "*B. coli*," and which incomplete analyses might easily identify as "atypical" forms of that bacillus, are to be found abundantly in shellfish, but are either absent or very rare in human faeces.

(2) The Longevity of Intestinal Bacteria in Sea-Water.

What we must do, therefore, is to isolate, one by one, the most characteristic faecal organisms, and then study their natural history, that is, their rate of reproduction in fresh water, in sea-water, in sewage, in shellfish, in soil, in diffuse or bright light, and so on. Hardly any of this kind of work has been done, although it appears to be quite essential if bacteriological methods are to be employed in public health work with respect to the recognition of intestinal bacteria in open natural conditions. Not to do this investigation would be much the same as carrying on fishery regulations with only a knowledge of the morphology of fishes as it is taught in the schools, and without knowing anything about the distribution, migrations and habits of fishes in the open.

Some bacteriological work of the kind I mention has indeed been carried out, notably by Clemesha in India, and by several workers in this country, with respect to the longevity of "*B. coli*" and "*B. typhosus*" in sea-water. In these latter cases the characters of the organisms studied are, unfortunately, not given in detail, and the observations made are not very precise.

I therefore give here some data relating to the longevity of intestinal bacteria in sea-water, premising that no real opportunity for a satisfactory investigation of the questions suggested above has been afforded, and that the results here given are to be regarded only as indicative of the possible methods which might be adopted.

An organism was isolated from human faeces having the following characters :—

Glucose +, lactose +, cane sugar +, dulcite +, adonite −, inulin −, indole +, Vosges and Proskauer's reaction −, motility −. It was kept on nutrient agar for a month or two and was then re-cultivated in all the above media: the same series of reactions were again exhibited by it. A few drops of the dulcite-broth culture (which had been incubated for about four days) were then added to about half a litre of sea-water of normal salinity (sp. gr. = 1·024 at 18° C.), and the numbers of bacteria in this liquid were estimated. The flask containing the culture was then kept in a cupboard at ordinary laboratory temperature (about 16° C.) and in ordinary diffuse light, and the number of bacteria in it was estimated from day to day. The results were as follows :—

Experiment I.

Days.	1	2	3	4	5	6	7	8	9	10	11	12
No. of bacteria per 1/1000 c.c.	(1321)	309	106	45	29	20	6	...	...	...	...	...
No. of bacteria per 1/100 c.c.	...	...	...	...	...	...	...	70	70	65	59	44

The number (1,321) at the beginning of the first day was not actually observed, since the plate was so crowded as to be impossible to count. This number has been extrapolated, as will be discussed later on.

At the beginning of the ninth day the plate (containing 70 organisms) was put aside and incubated for a day longer in the cold. Ten colonies were then subcultured on nutrient agar and re-cultivated in the same series of media. The same reactions were given as in the case of the original colony. A few drops of the dulcite-broth culture, after incubation for four days, were added to sterilised sea-water as before, and the number of bacteria contained in this liquid were again estimated from day to day. The results obtained were :—

Experiment II.

Days.	1	2	3	4	5
No. of bacteria per 1/10000 c.c....	21	...	...	...	...
,, ,, 1/1000 c.c. ...	231	84	4	...	...
,, ,, 1/100 c.c.....	(2310)	886	43	36	2

The count (2,310) for 1/100 c.c. for the first day (that is, at the beginning of the experiment) was not observed. It is the count for 1/1000 c.c., 231, multiplied by 10.

The plate counted on the third day (with 43 colonies) was incubated for a day longer in the cold, and eight colonies from it were subcultured on nutrient agar. These were again subcultured in the various media, with again the same results as were given by the original colony. One of the dulcite-broth tubes was again taken, after incubation for four days, and a few drops from it were added to half a litre of sterile sea-water as before. The number of bacteria present was estimated from day to day with the following results :—

Experiment III.

Days.	1	2	3	4	5	6	7
No. of bacteria per 1/1000 c.c.	30	...	...	...	...	...	...
„ „ 1/100 c.c....	...	37	5	...	...	...	...
„ „ 1/50 c.c. ...	...	...	...	0	...	...	...
„ „ 1 c.c.	...	...	...	...	3	1	...
„ „ 5 c.c.	...	...	...	...	...	...	0

The experiment was now discontinued.

A further experiment was made with the object of checking the original one. A similar culture to that employed above was used, but in a different manner. A fresh culture on nutrient agar was made from the stock culture (which had been kept for several months), and a few c.c. of sterile broth having been poured into the tube the culture was rubbed up into the liquid so as to make an emulsion of the bacteria. About 1 c.c. of this emulsion was then added, as before, to about half a litre of sterile sea-water contained in a flask, kept as before. The number of bacteria in the culture was estimated from day to day with the following results :—

Experiment IV.

Days.	1	2	3	4	5	6	7	8	9	10	11
No. of bacteria per 1/10,000 c.c.	279	319	129	58	24	32	10	...	...	...	...
„ „ 1/1,000 c.c.	...	...	...	...	...	...	...	45	54	11	...
„ „ 1/100 c.c. ...	...	...	...	...	...	...	...	...	...	...	56

Now, considering the first three experiments, we see from these rough results that the number of bacteria per unit volume of culture first of all undergoes a very rapid reduction, and then the rate of reduction becomes very much less. It is quite clear that a graph of these ungraduated figures would show that the curve would fall asymptotically close to the axis of y, and these would approach the axis of x in the same way.

We have to deal, in fact, with the decrease of the numbers of bacteria expressed by the gas-volume law, $pv = constant$. So much is clear merely from the inspection of the experimental data. The law of diminution may, however, be more minutely studied from smoothed or graduated figures, and these I give in the table below. The method of graduation is explained in Appendix II. Text-figure 1 is a graph of these numbers.

Days.	Expt.	1	2	3	4	5	6	7	8	9	10	11	12
Nos. of bacteria in unit volume of culture as percentages of the original number	I	100	18	7·4	3·5	2·7	2·2	0·9	0·6	0·5	0·4	0·3	0·2
	II	100	7·6	1·7	0·5	0·2	...	...	...	...	...	...	...
	III	100	2·1	0·2	0·04	0·001	0·004	...	...	...	...	...	...
	IV	...	...	...	...	...	...	...	...	...	...	...	...

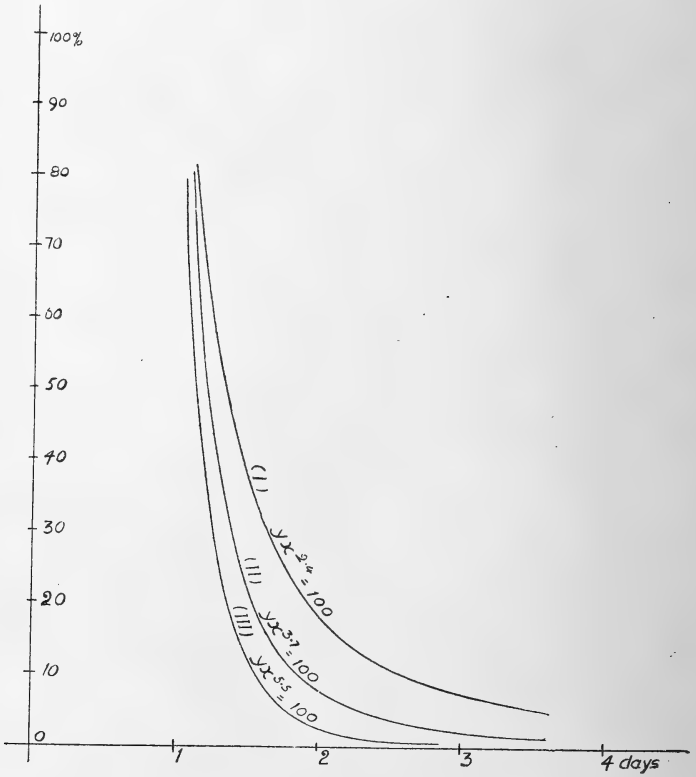


FIG. 1. Curves showing the rate of disappearance of intestinal bacteria from a sea-water culture. The points plotted represent the graduated figures of the table above.

The percentages for the fourth experiment cannot be given since the mode of reduction is not quite the same as in the first three experiments. The numbers of bacteria rise from the first to the second days and then fall, and after this initial rise the mode of diminution is the same as in the earlier experiments. The difference is due to the fact that a certain quantity of food-medium was added with the bacilli inoculated, so that an initial multiplication took place. Afterwards, however, the same manner of fall is to be seen from the figures.

The graphs of these three experiments are rectangular hyperbolas, and are :—

I. $yx^{2.41} = \text{constant.}$
II. $yx^{3.7} = \text{constant.}$
III. $yx^{5.54} = \text{constant.}$

The index of x, that is, the slope of the curve, the measure of the rate at which the bacteria die off during the experiment, is greatest in I and least in III. This means that the successive cultivation of the same strain through sea-water has affected the organisms, so that their resistant power to the change medium has become less in the course of each experiment. The constant, of course, only affects the scale to which the results are plotted, or otherwise expressed.

(3) The disappearance of intestinal bacteria from Mussels subjected to a cleansing process.

It has been known for some time that intestinal bacteria disappear from shellfish allowed to stand in still, or running sea-water. The first experiments of this kind were made by Klein at St. Bartholomew's Hospital as long ago as 1905. Oysters, mussels and cockles were dosed with enormous quantities of typhoid bacilli, and allowed to stand in wooden tubs containing sterile sea-water. After several days a very marked diminution in the numbers of the contained bacilli

was experienced, and Prof. Klein came to the conclusion that a "quarantine" period of about four days duration would usually be sufficient for the cleansing of polluted shellfish. In 1908 I made several such experiments in the open at Conway and obtained similar results. In four days there was a very marked reduction in the numbers of the contained bacteria, amounting usually to well over 90 per cent. About the same time Mr. A. Scott made similar experiments by keeping mussels in the water of the tanks at Piel Hatchery, and obtained much the same results. Since these first experiments others have been made, and at the beginning of 1914, when I was requested by the Chairman to report generally on the subject, some more precise observations were begun. I regret that it has simply been impossible to follow up the many obvious further questions suggested by these observations, and since the opportunity to do so is not likely soon to occur, I wish to record here the results obtained.

a. The cleansing of mussels by sterile sea-water.

By "sterile sea-water" is to be understood sea-water taken from the flood stream, and containing no micro-organisms in 1 c.c. capable of growing on neutral-red, bile-salt, lactose agar. The object of this experiment was to ascertain the time required to wash out sewage bacteria from the alimentary canal of the mussel.

Mussels, regarded as objectionably polluted, were taken from the foreshore in Barrow Channel, near the Piel Laboratory. An analysis made showed that the mean number of sewage bacteria contained per shellfish was 7,400. (The individual counts of 6 plates made each from 1 c.c. of an emulsion made from the soft parts of 5 mussels were : 120, 122, 140, 150, 151, 172. Each c.c. of this emulsion represented 1/50th part of a mussel). The mussels were put into glass aquarium tanks of about 10 litres capacity, and sea-water was run through these

tanks at the rate of about 1 litre per five minutes. Samples of the mussels were taken after treatment of this kind for one, two, and four days.

First Sampling. After 1 day.

1/50th mussel contained 0 intestinal bacteria.
" " 0 " "
" " 1 " "
Mean per mussel = 16·6.

Second Sampling. After 2 days.

1/50th mussel contained 4 intestinal bacteria.
" " 2 " "
" " 1 " "
Mean per mussel = 116.

Third Sampling. After 4 days.

1/50th mussel contained 6 intestinal bacteria.
" " 1 " "
" " 0 " "
Mean per mussel = 116.

Colourless colonies appeared in the plates made from the crude mussels, but none was seen in the plates made from the treated mussels.

The sewage bacteria present were therefore reduced by 98·5 per cent.

This experiment was not repeated. The same results had been obtained, as on a former occasion, by Mr. Scott, and little more information was to be attained by laboratory trials. It might have been possible to reduce still further, the time necessary for sufficient cleansing, and it might also be desirable to observe what degree of cleansing takes place when mussels are allowed to remain in various volumes of standing sea-water, and for variable periods. But on considering the difficulties which are likely to be encountered in applying the principles of these methods on a large scale,

it was felt that further laboratory experiments would afford little additional information, and that the details of a commercial process would have actually to be studied in the working plant itself.

b. Experiments in the open sea.

It was felt desirable to repeat, in new localities, the experiments in the open already made; and, in fact, a wish had been expressed by the fishermen at various musselling centres that cleansing works should be started. In September of 1914, Dr. Jenkins and I met Mr. R. W. B. Gardner at Sunderland Point, in the estuary of the Lune, and we then proceeded to visit a place which Mr. Gardner regarded as suitable for relaying mussels for cleansing purposes. This place, with its immediate surroundings, is marked on the chart reproduced at the end of this paper, and described in the explanation, so that I need not further refer to it. There were already some mussels there, and a preliminary analysis showed that these were relatively unpolluted. Mr. Gardner, therefore, laid down a quantity of mussels at this spot, and I made a series of analyses. The mussels were taken from the training wall in the Lune Estuary, a place known to be highly polluted with sewage. A sample examined prior to being laid down for cleansing gave the following results :—

1/50th mussel	Plate 1,	261	red colonies,	1	colourless colony.	
	,, 2,	200	,,	3	,,	,,
	,, 3,	267	,,	4	,,	,,
	,, 4,	200	,,	1	,,	,,
	,, 5,	237	,,	2	,,	,,

Mean number of sewage bacteria per mussel = 11,650.

First Sampling. Relaid for 2 tides.

5 Plates were made. There was no reduction in the numbers of bacteria contained in the sample.

Second Sampling. Relaid for 4 tides.

Plate 1, 16 red colonies, 0 white colonies.
„ 2, 28 „ 0 „ „
„ 3, 25 „ 2 „ „

Mean number of sewage bacteria per mussel = 1,150.

Third Sampling. Relaid for 6 tides.

Plate 1, 3 red colonies, no colourless colonies.
„ 2, 12 „ 0 „ „
„ 3, 12 „ 0 „ „

Mean number of sewage bacteria per mussel = 450.

This experiment is exceptional in that no reduction was experienced after 1 day's relaying. Most of the sewage bacteria were eliminated after 2 days' relaying, and after three days the reduction amounted to 99·6 per cent.

Photographs of typical cultures obtained in these experiments are reproduced in Plate I.

At the same time a sample of mussels taken from the same place (the training wall) was laid down on the foreshore at Sunderland Point (see the chart). These mussels had not been examined before relaying, but they must have been as greatly polluted as those dealt with in the above experiment. After being relaid for two days they were sampled with the following results :—

1/50th mussel { Plate 1, 7 red colonies, no colourless colonies.
„ 2, 8 „ „ „
„ 3, 9 „ „ „

The reduction experienced was, therefore, similar to that of the first experiment.

Yet a further experiment was made with River Lune mussels. Some of the fishermen at Glasson Dock wished me to try relaying mussels at a point near to the railway station there (see the chart). The place did not seem to me to be at all suitable, but it was tried. A sample of mussels taken from the same source, the Training Wall, was laid down.

Before relaying, five mussels from the sample were examined. Three plates were made :—

1/50th mussel { Plate 1, 1,800 red colonies, 20 colourless colonies. ,, 2, ,, 3, } The colonies were so numerous that the plates were uncountable.

These mussels therefore contained at least 90,000 sewage bacteria each.

First Sampling. Relaid for four tides.

Plate 1, 370 red colonies, 15 colourless colonies.
,, 2, 627 ,, 23 ,, ,,
,, 3, 432 ,, 47 ,, ,,

A considerable reduction of contained bacteria was therefore experienced by these mussels. But reinfection doubtless occurred, for the results are not satisfactory. The place proposed was rejected as the result of the experiment.

In these experiments the mussels were laid down on the foreshore itself, on a beach of sand and gravel. There was no enclosure of any kind. The places selected were out of the main tidal streams, and were not exposed to any sea, so that the mussels remained undisturbed—they had, in fact, previously been employed by local fishermen for storing mussels prior to sending them to market. The question then arose as to the practicability of relaying mussels in places like the Estuaries at Aberdovey and Barmouth, where the tidal streams are rapid, where there is a certain exposure to the sea, and where the shore slopes down very steeply from high-water mark. Construction of enclosures, designed to hold mussels while being cleansed, would be very difficult and expensive in such circumstances; and to meet this difficulty, Mr. G. Hazlehurst, a member of the Committee, made the very practical suggestion that a floating tank should be used for relaying. A tank boat was therefore made, big enough to hold several bags of mussels. The bow and stern were open, being guarded by strong wire gauze, and movable

bulkheads were placed at each end so that the further entrance of water could be prevented after the tank had been filled. The tank could be moored at any place selected. A sample of mussels from Aberdovey Channel, near the Pier, was taken and placed in the tank, and the latter was then moored above Trevri Point. (See Mr. Durlacher's Chart in the Report for 1913 for these localities.) After being relaid for three tides the mussels were sampled. The shellfish lived well in the tank, and had attached themselves to each other and to the wooden sides.

First sampling. The original, un-relaid mussels.

1/50th mussel	Plate 1,	13	red colonies,	no	colourless colonies.
	,, 2,	15	,,	1	,, ,,
	,, 3,	16	,,	1	,, ,,

Mean number of sewage bacteria per mussel = 735.

Second sampling. Relaid for three tides.

1/50th mussel	Plate 1,	13	red colonies,	no	colourless colonies.
	,, 2,	10	,,		,, ,,
	,, 3,	13	,,		,, ,,

Mean number of sewage bacteria per mussel = 600.

The experiment was repeated later in the year. A sample of mussels was dredged from near Penhelyg Point, and was relaid in the tank, the latter being moored on the beach near the life-boat house at Aberdovey (see Mr. Durlacher's chart). At both this place and the one in the other Aberdovey experiment, water circulated through the tank for about four to six hours during each tide. The tank came adry during ebb-tide, and it was expected that before the time when the number of sewage bacteria had increased notably in the water of the channel, the tank would be out of the water.

First sampling. The original, un-relaid mussels.

1/50th mussel	Plate 1,	13	red colonies,	no	colourless colonies.
	,, 2,	8	,,	0	,, ,,
	,, 3,	8	,,	0	,, ,,

Mean number of sewage bacteria per mussel = 485.

Second sampling. Relaid for three tides.

1/25th mussel	Plate 1,	13	red colonies,	no	colourless colonies.
	,, 2,	5	,,	0	,, ,,
	,, 3,	6	,,	0	,, ,,

Mean number of sewage bacteria per mussel = 200.

There was very little reduction in the number of contained bacteria in these experiments, but that some reduction did occur was quite certain. The mussels originally were not polluted to a significant extent; they were certainly quite suitable as human food, in my opinion. The object of the experiment was to test, in a preliminary way, the practicability of utilising a floating movable tank for the purpose of containing mussels undergoing cleansing, in such places where the nature of the foreshore makes it difficult or expensive to construct tanks or ponds, or other artificial enclosures. The experiments are being repeated at other places on the Welsh coast.

A further experiment of the same kind was made at Barmouth in February, 1915. The same difficulties presented themselves here as did at Aberdovey, that is, the area of foreshore suitable for the construction of a tidal cleansing pond or tank is very limited. The floating tank used in the last experiment was, therefore, again used in this one. It will be seen from Mr. Durlacher's chart, published at p. 352 of last year's Report, that the floats used for the drift experiments passed directly in front of the inlet, called Aberamffra Harbour. But at half flood-tide we might expect the sewage coming up from the Harbour to be very greatly diluted, while the ebb-tide would contain but little contaminating matter at any state; at all events the experiment was so arranged, and the tank so moored that it dried, that is, it came aground, for about four hours on each tide. Therefore, the first of the flood-tide, which was the water most likely to be contaminated, did not reach the mussels, while the last of the ebb-tide, water which

might also be polluted from the upper part of the Estuary, did not come near them.

Mussels were, therefore, collected from the bed of the River Mawddach, between Aberamffra and the Railway Bridge, the place usually fished, and these mussels were put into the floating tank. The latter was moored at about half tide level in Aberamffra Harbour. Samples were taken before treatment, and after four and six tides, that is, after two and three days.

First sampling. The original, un-relaid mussels.

1/50th mussel:
Plate 1, 10 red colonies.
,, 2, 20 ,,
,, 3, 8 ,,
,, 4, 6 ,,

Mean number of sewage bacteria per mussel = 440.

Second Sampling. Relaid for four tides.

1/50th mussel:
Plate 1, 2 red colonies.
,, 2, 1 ,,
,, 3, 0 ,,
,, 4, 0 ,,

Mean number of sewage bacteria per mussel = 37.

Third Sampling. Relaid for six tides.

1/50th mussel:
Plate 1, 1 red colony.
,, 2, 1 ,,
,, 3, 1 ,,
,, 4, 0 ,,

Mean number of sewage bacteria per mussel = 37.

That is, there was a reduction of sewage bacteria amounting to about 92 % of the number originally contained in the mussels.

c. Experiments with water sterilised by means of Chlorine.

These natural difficulties are so great, in some places, that the problem of naturally cleansing the mussels by exposure to clean flood-tide water is insoluble ; or the cost of so treating

the shellfish would be so great as to be prohibitive. In these circumstances the possibility of sterilising the water used for cleansing, by some chemical means, may be considered. Early in 1914 the Committee requested me to meet an Inspector of the Board of Agriculture and Fisheries and discuss with him, and the officials of the Conway Corporation, the methods which were in contemplation, in that district, for the cleansing of the local mussels. I did so, and made a report to the Committee, when I was requested to report further on the practicability of the method proposed to be adopted at Conway —that of the sterilisation of the water used for cleansing by means of dosage by chlorine solution. In April and May of 1914 Mr. Scott and I accordingly carried out these experiments, which I subsequently repeated. Some reference to the results obtained was made in my report to the November Meeting of the Scientific Sub-Committee, but I have been further requested to make a more complete statement.

Several distinct points had to be investigated (1) The concentration of chlorine in the sea-water necessary for sterilisation; (2) The concentration of chlorine which the mussels could withstand without interference with their normal functioning; and (3) The length of time necessary for the removal of the bacteria contained in the cavities of the bodies of the mussels. It was already known that a very small quantity of chlorine in water sufficed for almost complete sterilisation—this was, in fact, the principle of the Candy Process of water purification, and numerous experiments had been made, to say nothing of data recorded in the text-books on Public Hygiene. It was, however, desirable to assure oneself personally of the fact of this sterilisation, so five 100 c.c. flasks were filled with normal sea-water and sterilised, and then the same volume of a culture of bacilli fermenting glucose, lactose, cane sugar, dulcite, adonite, but not inulin, forming indole and giving a positive Vosges and Proskauer reaction,

and exhibiting motility, was added to each. The number of bacteria in one of the flasks was estimated, it was about 3,000. Chlorine water was then added to each of the other flasks in quantity enough to make solutions of 1, 3, 5, and 7 parts per million, and the flasks were allowed to stand at ordinary laboratory temperature for 24 hours. One c.c. of the culture was then taken from each and plated in neutral-red, bile-salt, lactose agar. All the plates were sterile.

One part per million of chlorine seems, therefore, to be enough to secure the destruction of most of the ordinary bacteria present in sea-water, though there is no object in using so very dilute a solution. The next point was to determine what concentration the mussels could stand without injury. Some bleaching powder solution was first of all made, and this was added to aquaria containing mussels, but these rough trials were unsatisfactory. Finally chlorine water was made from a mixture of potassium dichromate and hydrochloric acid, and after washing in tapwater, the gas was absorbed in distilled water. A standard solution of thiosulphate (and one of pot. dichromate for standardisation of the thiosulphate) were made, and the strength of the chlorine water was estimated before each experiment. Several glass aquaria were filled with sea-water—each of them held about ten litres—and then dilute chlorine water was added so as to produce solutions having, very approximately, the concentrations of 4, 6, 8, and 10 per million. The chlorine solution and the sea-water were mixed, and mussels were added, and the behaviour of the shellfish noted from hour to hour. There was some doubt as to whether those in the solution of 10 per million opened their shells—I think they may have done so—but all the others functioned normally, even spinning byssus threads and attaching themselves to the glass of the aquaria. Even in the 4 per million solution, the smell of chlorine could be detected at the beginning of the experiment. Evidently,

therefore, chlorine in sea-water to the extent of 5 parts per million does not interfere with the functioning of mussels—I did not expect that it would, since the chlorine throat washes used are really stronger than this. It was, however, desirable actually to make the trial.

A tank with chlorinated sea-water, of concentration, 5 per million, was now prepared, and mussels were placed in it. These mussels were part of the same sample used in making the experiment of cleansing by a water circulation—they contained on the average 7,400 sewage bacteria each. The tank was allowed to stand in full daylight, though not in direct full sunlight, and samples of the mussels were taken after one and two days.

First Sampling. Mussels in 5 per million chlorine sea-water for one day.

1/50th mussel { Plate 1, 17 red colonies.
,, 2, 28 ,,
,, 3, 14 ,,

Mean number of sewage bacteria per mussel = 985.

Second Sampling. After two days.

1/50th mussel { Plate 1, 11 red colonies.
,, 2, 14 ,,
,, 3, 11 ,,

Mean number of sewage bacteria per mussel = 600.

The mussels were not removed from the tank, but the water was now run off and replaced by sea-water freshly dosed with chlorine so as to make a strength of 5 per million. This water was allowed to stand another day, when the mussels were again examined.

Third Sampling. After 1 day, the water being changed.

1/50th mussel } 3 plates, all of which were sterile.

The mussels were therefore freed from sewage contamination.

The experiment was repeated at Liverpool in July. Short

glass specimen cylinders of about 6 litres capacity were used as tanks. It was thought, during the Piel experiments, that the mussels opened their shells more freely in the dark than in full daylight, so these glass jars were covered with black paper and were fitted with cardboard lids. A solution of chlorine in water was made as before, the strength of this was determined, and it was diluted to a convenient concentration. Titrations of the chlorine solution, and of the thiosulphate standard solution, were made immediately before each experiment.

A similar jar containing only sea-water was used as a control on the chlorinated water jar. The latter contained sea-water dosed with chlorine to the concentration of 5 per million. Mussels which had been sent from Barrow Channel were placed in each jar, and then a sample of the same lot of mussels was examined. The results of this analysis were :—

First Sampling. Original, untreated mussels.

1/50th mussel

Plate 1, 76 red colonies.
,, 2, 78 ,,
,, 3, 58 ,,
,, 4, 49 ,,
,, 5, 138 ,,

Mean number of sewage bacteria per mussel = 3,990.

Second Sampling. After one day in chlorinated sea-water.

1/50th mussel

Plate 1, 19 red colonies.
,, 2, 33 ,,
,, 3, 19 ,,
,, 4, 17 ,,
,, 5, 42 ,,

Mean number of sewage bacteria per mussel = 1,300.

Third Sampling. After one day in unchlorinated sea-water.

1/50th mussel

Plate 1, 50 red colonies.
,, 2, 53 ,,
,, 3, 50 ,,
,, 4, 56 ,,
,, 5, 50 ,,

Mean number of sewage bacteria per mussel = 2,590.

The chlorinated sea-water in the first jar was re-dosed so as to make it of concentration 5 per million—the water itself not being changed. This solution was allowed to stand another day.

Fourth Sampling. After two days in chlorinated sea-water.

1/50th mussel	Plate 1,	8 red colonies.
	,, 2,	8 ,,
	,, 3,	4 ,,
	,, 4,	10 ,,
	,, 5,	8 ,,

Mean number of sewage bacteria per mussel = 380.

The water still being unchanged, fresh chlorine solution was again added so as to bring up the strength to 5 per million. This water, with its contained mussels, was allowed to stand another day.

Fifth Sampling. After three days in chlorinated sea-water.

1/50th mussel	Plate 1,	2 red colonies.
	,, 2,	2 ,,
	,, 3,	3 ,,

Mean number of sewage bacteria per mussel = 116.

Fifteen mussels had been put into the jar originally. Two of them died.

It is clear, therefore, that mussels may live, open their shells, and circulate water through the mantle cavity when that water contains 5 parts per million of dissolved chlorine. In such a medium mussels cleanse themselves from ingested sewage bacteria.

(4) The effect of change of medium on the biological characters of intestinal bacteria.

It has been suggested that micro-organisms living in the human alimentary canal, and exhibiting certain definite fermentation reactions with certain sugars, might no longer do so when they inhabit a widely different medium, say,

sea-water, or the alimentary canal of a shellfish. In particular, do dulcite-fermenting organisms isolated from faeces continue to ferment dulcite after they have been living in sea-water for some time? This question was suggested to me by Dr. MacConkey as a possible subject for experiment. I regret that I have few data to give which might conceivably answer the question.

Some mussels, taken from Roosebeck Scar, were put into a clean tank in the Piel Hatchery. These mussels are very clean, so that no micro-organisms, capable of growing on neutral red, bile-salt, lactose agar, can usually be found in 1/50th part of a single animal. A culture of an organism isolated from faeces was made in dulcite broth. This organism had the following characters:—Glucose +, lactose +, cane sugar +, dulcite +, adonite −, inulin −, indole +, Vosges and Proskauer's reaction −. About a dozen mussels were taken, and then, the valves of the shell being very slightly forced apart, about 1 c.c. of the culture was injected, by means of a hypodermic syringe, into the mantle cavities. The mussels were then kept out of water for about six hours, so as to allow the culture to be taken into the alimentary canal, when they were placed in a small glass aquarium through which clean sea-water was circulated at the rate of about 1 litre per five minutes.

After a period of eighteen hours, five mussels were taken out and the soft parts were emulsified in 250 c.c. of sterile water. 1 c.c. of the emulsion was inoculated in each of five plates, and the latter were incubated. The colonies were so very dense that they formed a very fine haze in the medium. There must have been very many thousands of colonies on each plate. The remainder of the mussels were kept in the aquarium for another five days, when they were again sampled. Two plates were made from an emulsion of the soft parts of five mussels in 250 c.c. of water, and each plate contained

1/50th mussel. There were 257 and 418 red colonies on these plates, thus showing a very considerable amount of reduction of the contained bacteria. Ten colonies were selected from the first plate, and pure subcultures were made. Nos. 1 to 9 gave the reactions:—Glucose, lactose and dulcite all +; cane sugar, acid; adonite and inuline −; indole +; Vosges and Proskauer's reaction −. No. 10 gave the same series of reactions except that dulcite was not fermented. There was thus, apparently, a slight change in the biological characters of the organism, but without further confirmatory work of the same kind one cannot positively assert that this change is a real one.

There remains the series of experiments already quoted. A dulcite + organism isolated from faeces was inoculated in sea-water and cultivated there for seven days. Then it was recultivated on neutral red, bile-salt, lactose agar, and then on nutrient agar. Ten colonies gave the original reactions. The organism was again inoculated in sea-water and grown there for three days, and again passed through neutral red and nutrient agar. Eight colonies all gave the original series of reactions.

Thus no change in the characters of this organism was produced by a rather long sojourn in sea-water. So far as they go, these two experiments do not support the notion that faecal micro-organisms undergo any change of biological characters, when they enter sea-water or the alimentary canals of marine shellfish. They die out, of course, but so long as they live they exhibit their original powers of fermenting carbohydrates. Of course the number of observations made is far too few to serve as the basis for any general statement with regard to this point.

(5) Summary.

(1) The process of self-cleansing of sewage polluted shellfish by placing them for some days in sea-water, which

is free, or nearly so, from sewage bacteria, depends on two things. (*a*) The ingested bacteria are merely washed out from the mantle cavities, and internal cavities, of the animals by the stream of water which is continually being circulated through these passages. (*b*) The bacteria rapidly die out in a medium which they are unable to use as a source of energy.

(2) Truly intestinal bacteria are probably highly specialised organisms. They are not so much saprophytes as parasites. Their optimum temperature is about 37° C., that is, the temperature of the interior of the body of mammals, while the temperature of sea-water, and that of the interior of the body of marine shellfish, varies from about 3° C. to 15° C. It is doubtful whether there is any initial multiplication of the bacteria on entering the shellfish—to prove that there is we should have to show that the numbers of bacteria per unit volume of shellfish was generally greater than the numbers per unit volume of the surrounding water. The concentration of bacteria in the latter varies enormously with state of tide and other conditions, and the condition of greatest concentration would be that which ought to be compared with the concentration of bacteria in the shellfish.

(3) Faecal micro-organisms may disappear very rapidly when introduced into sea-water, the destruction of 90 % of the organisms originally present being effected during the first two days.

(4) Mussels may be cleansed from ingested sewage bacteria by keeping them in water sterilised by the addition of chlorine. A concentration of chlorine in sea-water of 5 parts per million is sufficient, practically, to sterilise the water, while it does not interfere with the ordinary functioning of the shellfish.

The action of chlorine is twofold. It may simply render a polluted sea-water practically free from sewage bacteria, and then this sterile water washes out the ingested bacteria from the alimentary canals and mantle cavities of the shellfish,

and does not re-infect the animals. It may also actually destroy the micro-organisms present in the bodies of the shellfish, but it does not appear likely that the action of this kind may be of importance. The trace of chlorine present in the water would immediately enter into combination with constituents of the mucus covering the body, and would not really act on ingested micro-organisms.

(5) Mussels may be very quickly cleansed, to the extent of getting rid of 90 % of their contained sewage organisms, by exposure to running sea-water, or to repeated changes of standing water dosed with chlorine. Laboratory experiments indicate that the cleansing process need not require more than one day. On a commercial scale the time required would depend on the perfection of the water circulation, and it cannot be estimated except by experiment with the actual plant suggested.

(6) Faeces, sewage, polluted estuarine sea-water, and shellfish all contain a mixture of species of glucose and lactose fermenting bacilli. All these have been called "*Bacillus coli*," or "coliform" organisms, or "typical," or "atypical" coli. Some of them are of equal significance with the true *B. coli communis*, in that they are of exclusive intestinal origin, but others may have sources of origin of vastly less significance. This mixture of species differs in faeces and in polluted shellfish.

These species probably mostly cease to multiply, and finally die out, when they enter estuarine sea-water, or the bodies of marine shellfish. But the rate at which reproduction falls off and the rate of mortality probably differ according to the species. It is, therefore, of practical importance that analyses should indicate the proportions of the various species, or categories of related species, present in a sample of polluted shellfish, since this may indicate whether the pollution was recent, and therefore of possible danger, or remote, and therefore of little significance.

(7) There is urgent need for investigation into the specific nature of the various kinds of micro-organisms present in faeces and in polluted waters and in shellfish. The natural history of these organisms also imperatively demands investigation.

APPENDICES.

I. Reactions of micro-organisms isolated from Mussels.

Mussels from Piel Shore (6.6.1913). *Mean number of sewage bacteria per mussel was* 1,510. *The characters of* 10 *organisms were as follows :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Gelatine.	Morphology.
1	+	+	+	*a*	+	—	+	—	+	0	Bacilli.
2	+	+	+	*a*	+	—	+	—	—	0	Cocci.
3	+	+	+	*a*	+	—	+	—	—	0	Long slender bacilli.
4	+	+	+	*a*	+	—	+	—	—	0	Bacilli.
5	*a*	*a*	—	*a*	*a*	—	+	—	+	0	,,
6	+	+	+	*a*	+	—	—	+	—	0	,,
7	*a*	*a*	—	*a*	*a*	—	—	—	—	0	,,
8	+	+	+	*a*	+	—	+	—	+	0	Short bacilli.
9	*a*	*a*	—	*a*	*a*	—	—	—	+	0	Long slender bacilli.
10	*a*	*a*	—	*a*	*a*	—	—	—	+	0	Bacilli.

(10.7.1914). *Mean number of sewage bacteria per mussel* = 3,900

1	+	+	+	—	+	—	—	+			
2	+	+	+	—	+	—	+	—			
3	+	+	+	—	—	—	—	+			
4	+	+	+	+	—	—	+	—			
5	+	+	+	—	+	—	+	+			
6	+	+	—	+	—	—	+	—			
7	+	+	+	+	+	—	+	+			
8	+	+	+	+	+	—	+	+			
9	+	+	+	+	+	—	+	+			
10	+	+	+	+	+	—	+	+			

Mussels from Aberdovey (24.6.1913). *Mean number of sewage bacteria per mussel* = 20,000. *The characters of* 10 *organisms were as follows :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Gelatine.	Morphology.
1	+	+	−	−	−	−	+	−	−	0	Short bacilli.
2	+	*a*	*a*	−	−	−	+	−	+	+	Bacilli.
3	+	+	+	−	+	−	−	+	−	0	Long bacilli.
4	+	+	−	−	−	−	+	−	−	0	Short bacilli.
5	+	+	+	−	+	−	+	−	−	*g*	Moderately long bacilli.
6	+	*a*	*a*	−	−	−	+	−	+	+	,, ,,
7	+	+	+	*a*	+	*a*	+	−	−	*g*	Short bacilli.
8	*a*	*a*	*a*	−	−	−	+	−	+	0	Moderately long bacilli.
9	+	+	+	−	+	−	+	−	−	0	Very long bacilli.
10	+	+	+	−	+	*a*	−	+	−	0	Moderately long bacilli.

Mussels from Barmouth (22.7.1913). *Mean number of sewage bacteria per mussel* = (*a*) 2,420, (*b*) 6,200. *The characters of* 14 *of these organisms are as follows :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Gelatine.	Morphology.
1	+	+	−	−	−	−	+	−	−	0	Bacilli.
2	+	+	+	−	−	−	+	−	−	0	,,
3	+	+	−	−	−	−	−	+	−	0	Long slender bacilli.
4	+	+	+	−	+	−	+	−	−	0	Bacilli.
5	+	+	+	*a*	−	−	−	−	−	0	Long slender bacilli.
6	+	+	+	−	+	−	−	−	−	0	Bacilli.
7	+	+	+	+	+	−	+	−	−	0	,,
	(*a*) Nos. 1 to 7 are from Trwyn y Gwaith.										
8	+	+	+	+	−	−	+	−	+	0	,,
9	−	+	+	−	+	−	+	−	+	0	,,
10	+	+	−	−	−	−	+	−	−	0	,,
11	+	+	+	−	+	−	−	−	−	0	,,
12	+	+	+	−	+	−	+	−	−	0	,,
13	+	+	+	+	−	−	+	−	+	0	,,
14	+	+	+	+	+	−	+	+	+	+	,,

(*b*) Nos. 8 to 14 are from Aberamffra Harbour.

Mussels from Ribble Channel (8.10.1913). *Mean number of sewage bacteria per mussel* = 21,000. *Reactions of* 10 *of these organisms as follows :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Gelatine.
1	−	+	−	−	−	−	+	−	+	0
2	+	+	−	−	+	−	+	−	−	0
3	+	+	+	+	+	−	−	−	+	0
4	+	+	+	+	+	−	−	+	+	0
5	+	+	−	−	+	−	+	−	−	0
6	+	+	−	−	+	−	−	−	−	0
7	+	+	−	−	+	−	+	−	−	0
8	+	+	−	−	+	−	+	−	−	0
9	+	+	−	−	+	−	+	−	+	0
The above are organisms from the mussels.										
10	+	+	−	−	+	−	+	−	−	0
11	+	+	+	+	−	−	+	+	−	0
12	+	+	+	−	+	−	−	−	−	0

The organisms 10 to 12 were isolated from the water of the Channel.

Roosebeck Scar mussels kept in large tank in running sea-water (5.1914). *Mean number of sewage bacteria per mussel* = 590.

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Morphology.
1	+	+	+	−	−	−	−	+	+	Long bacilli.
2	+	+	+	−	−	−	−	+	−	Very long bacilli.
3	+	+	+	−	−	−	−	−	−	Long bacilli.
4	+	+	+	+	−	−	−	−	−	Short bacilli.
5	+	+	−	−	−	−	+	−	−	Very short bacilli, or cocci.
6	+	+	−	−	−	−	+	−	+	Short bacilli.
7	+	+	+	−	+	−	−	+	−	Very long bacilli.
8	+	+	+	−	*a*	−	−	−	+	Long bacilli.
9	+	+	−	−	+	−	+	−	+	Very short bacilli.
10	+	+	−	+	−	−	+	−	+	Very short bacilli.

Mussels from Barrow Channel (10.7.1914). *Kept in chlorinated water for* 18 *hours. Characters of* 6 *organisms :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer Reaction.	Motility.
1	+	+	+	−	−	−	+		+
2	+	+	−	−	−	−	+	−	−
3	+	+	+	−	−	−	−	+	−
4	+	+	−	+	−	−	+	−	−
5	+	+	+	−	−	−	+	+	+
6	+	+	+	+	+	−	+	−	−

Mussels from Barrow Channel (10.7.1914). *Kept in sea-water for* 12 *hours. Characters of* 10 *organisms :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer Reaction.	Motility.
1	+	+	−	−	−	−	+	−	+
2	+	+	+	+	+	−	−	−	+
3	+	+	−	+	−	−	+	−	+
4	+	+	+	−	−	−	−	+	+
5	+	+	+	−	−	−	−	−	+
6	+	+	+	+	−	−	−	−	+
7	+	+	−	+	−	−	+	−	+
8	+	+	+	+	−	−	+	−	+
9	+	+	+	−	−	−	−	−	+
10	+	+	+	+	−	−	−	−	+

Mussels from Conway taken from Bar (18.9.1914). *Mean number of intestinal bacteria per mussel* = 2,760. *Reactions of* 12 *of these organisms as follows :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Morphology.
1	+	+	−	−	+	−	+	−	+	Small bacilli.
2	+	+	+	−	+	−	−	−	−	Long bacilli.
3	+	+	+	−	+	−	−	+	−	,,
4	+	+	+	−	+	−	+	+	−	Small bacilli.
5	+	+	+	−	+	−	−	+	−	,,
6	+	+	+	−	+	−	−	−	−	Long bacilli.
7	+	+	+	−	+	−	+	−	−	Very long bacilli.
8	+	+	+	−	+	−	−	+	−	Bacilli.
9	+	+	+	−	+	−	−	+	−	Small bacilli.
10	+	+	+	−	+	−	−	+	−	Bacilli.
11	*a*	*a*	*a*	−	−	−	+	−	+	Very small bacilli.
12	+	*a*	+	−	−	−	+	−	+	,, ,,

Mussels from the Estuary of the Lune (26.10.1914). *Re-laid at Overton for* 6 *tides. Mean number of sewage bacteria per mussel* = 450. *Reactions of* 10 *of these organisms as follows :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Morphology.
1	−	−	−	−	−	−	−	−	+	Bacilli.
2	+	+	+	−	+	−	−	−	−	,,
3	+	+	+	−	*d*	*d*	+	−	−	,,
4	+	+	+	−	−	−	−	+	+	,,
5	+	+	−	−	+	−	+	−	−	Small bacilli.
6	+	+	−	−	−	−	+	−	+	Bacilli.
7	+	+	−	−	−	−	+	−	+	Small bacilli.
8	+	+	+	+	+	−	−	+	−	Bacilli.
9	+	+	−	+	−	−	+	−	−	Very small bacilli.
10	+	+	*a*	−	−	*d*	+	+	−	Very long bacilli.

Mussels from the Estuary of the Dovey (11.12.1914). *Mean number of sewage bacteria per mussel* = 200. *Reactions of* 16 *of these organisms as follows :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Morphology.
1	+	+	+	+	+	−	−	−	−	Bacilli.
2	+	+	+	+	+	−	+	+	−	Chains.
3	+	+	+	−	−	−	−	+	−	Bacilli.
4	+	+	+	+	+	−	+	−	−	,,
5	+	+	+	+	+	−	−	+	−	Chains.
6	+	−	−	−	−	−	−	−	−	,,
7	+	+	+	+	−	−	−	+	−	Very short bacilli.
8	+	+	+	−	−	−	−	+	−	Chains
9	+	+	+	+	+	−	+	+	−	Very short bacilli.
10	+	+	+	+	+	−	−	+	−	,, ,,
11	+	+	+	+	+	−	−	+	−	Bacilli.
12	+	+	−	+	−	−	−	+	−	Very short bacilli.
13	+	−	+	−	−	−	−	+	+	Bacilli.
14	+	+	+	+	−	−	+	+	+	,,
15	+	+	+	+	+	−	+	+	−	Chains.
16	+	+	+	+	−	−	−	−	−	Bacilli.

Mussels from the Estuary of the Conway (12.1.1915). *Mean number of sewage bacteria per mussel* = 3,670. *Reactions of* 18 *of these organisms as follows :—*

	Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer.	Motility.	Morphology.
1	+	+	−	−	−	−	+	−	−	Small bacilli.
2	+	+	*a*	−	+	−	+	−	−	Bacilli.
3	+	+	+	−	+	−	+	−	−	Long bacilli.
4	+	+	+	−	+	−	+	−	−	,,
5	+	+	+	−	+	−	−	−	−	,,
6	+	+	+	−	+	−	+	−	−	,,
7	+	+	−	+	−	−	+	−	−	Bacilli.
8	+	+	+	−	+	−	+	−	−	Chains.
9	+	+	+	−	+	−	+	−	−	Bacilli.
10	+	+	−	+	−	−	+	−	−	,,
11	+	+	−	+	−	−	+	−	+	Small bacilli.
12	+	+	−	−	−	−	+	−	−	Bacilli.
13	+	+	+	−	+	−	−	−	?+	Small bacilli.
14	+	+	+	+	−	−	+	−	?+	,,
15	+	+	−	−	−	−	+	−	+	Bacilli.
16	+	+	−	−	+	−	+	−	+	,,
17	+	+	−	−	−	−	+	−	−	,,
18	+	+	+	+	+	−	+	−	+	,,

Reactions of 72 organisms isolated from mussels. (These reactions are not contained in the previous tables.)

Glucose.	Lactose.	Cane Sugar.	Dulcite.	Adonite.	Inulin.	Indole.	Vosges and Proskauer Reaction.	Mannite.	No. of Organisms.
+	+	−	−	−	−	...	−	+	32
+	+	−	−	+	−	...	−	+	7
+	+	−	+	−	−	...	−	+	7
+	+	+	+	−	−	...	−	+	7
+	+	+	−	+	−	...	−	+	3
+	+	+	−	−	−	...	−	+	2
+	+	−	+	+	−	...	−	+	2
+	+	+	+	+	−	...	−	+	1
+	+	+	+	−	+	...	−	+	1
+	+	−	−	−	−	...	−	−	1
									63

9 other organisms did not ferment both glucose and lactose.

II. The graduation of the rough data of the Experiments I, II and III (see pp. 128-133)

It may be well worth while to discuss the method of graduation of the rough figures obtained in the above experiments, as they illustrate a frequent difficulty in biological work involving series of numerical values. It is quite evident, merely from a glance at the figures, that they are to be fitted to a curve, the form of which is that of the rectangular hyperbola.

But if we try to draw this curve, after plotting the points represented by the figures, we shall find that to do this by inspection is not easy; and if we try it several times, without looking at the graphs previously made, we may find that the results are not the same. This is generally true of the method of making curves representing biological statistics. The data are rough, and allow us considerable latitude in making their graphs. *We can, quite unconsciously it may be, make these*

graphs go as best suits our argument. We ought to have a method of making them which removes personal bias.

An easy application of Pearson's method of moments enables us to do this in the present case.

Assume, then, that the general equation representing the drop in numbers of the bacteria in the cultures is :—

$$yx^n = a$$

that is, the equation to a hyperbola.

Now taking logarithms and transposing we get from this equation :—

$$\log y = -n \log x + \log a,$$

that is, the equation to a straight line, $-n$ being the tangent which the line makes with the axis of x, and *log a* the intercept on the axis of y; we have now to find these constants n and a.

If we plot these points given by the equation we find that they are irregular, and we have the same difficulty in drawing a straight line through them as we had in the original data. If the points lay very near a straight line we could find both n and a by inspection of the graph. But since this cannot be done with certainty, we have to find the constants analytically. Let us regard the logs of y and x, not as logs, but simply as y and x co-ordinates. They form a distribution, and we have to find the mean of this, and the first moment of all the frequencies in it about the mean. The graph of the distributions shows us a series of trapezoids on unequal bases. To find the moments we must therefore calculate the areas of these trapezoids, and then the mid-ordinates. We must suppose the areas to be concentrated round the mid-ordinates. If the y- co-ordinates are y , y_2, y_3, &c., and the x- co-ordinates, x_1, x_2, x_3, &c., we find the areas from :—

$$(x_2 - x_1)\frac{(y_2 + y_1)}{2} \quad \text{&c.}$$

while the corresponding mid-ordinates are given by $\frac{x_2 - x_1}{2}$

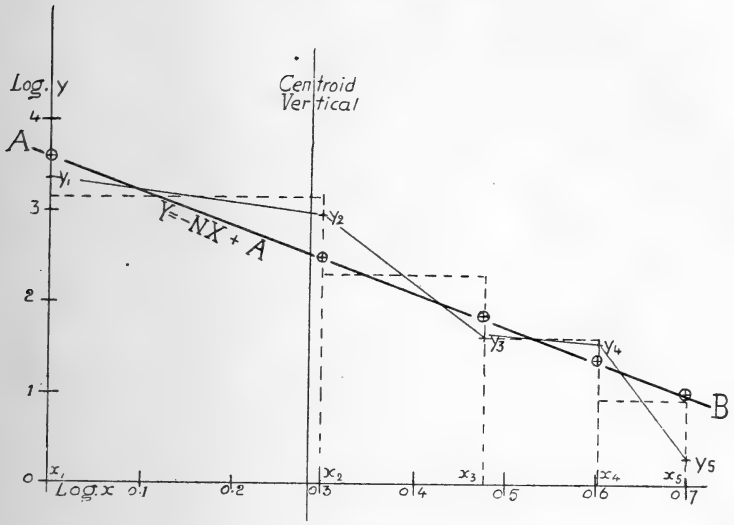


FIG. 2. Graduated and ungraduated data of the example on p. 160.

We now take some one ordinate (not a mid-ordinate) near the mean, and we find the moments of inertia of the mid-ordinates about this arbitrary origin. This gives us ν_1, the first moment about the arbitrary origin. Subtracting this value from, or adding it to, the value of the arbitrary origin, according to its sign, we get the mean ; and dividing it by the area we get μ_1 the first moment of the whole distribution about the mean. We find that the latter is always zero, but we must calculate it in order that we may find the limits (l_1 and l_2) of the distribution about the mean.

The area of the whole distribution is, of course, simply the sum of those of the trapezoids.

Now if the equation to the straight line is $y = -nx + a$

$\int_{l_1}^{l_2} (nx + a)\, dx$ = area of the distribution. In finding the first moment what we do is to multiply each frequency by x so that $\int_{l}^{l_2} [x\,(nx + a)]\, dx$ = first moment about the mean = 0.

We have now two equations and by solving these simultaneously we find the constants n and a.

It may be worth while to give an actual example of the calculation.

1 Days. x	2 Nos. of bacteria in unit volume. y	3 Logs of x	4 Logs of y	5 Values of mid-ordinates	6 Bases of trapezoids.	7 Areas. (5 × 6) a	8 Distance from arbitrary origin. d	9 1st moments about arbitrary origin. $a \times d$	y
1	2,310	0·0000	3·3636						3·58
				3·1555	0·3010	0·9498	− 0·1505	− 0·1429	
2	886	0·3010	2·9474						2·47
				2·2904	0·1761	0·4033	+ 0·088	+ 0·0355	
3	43	0·4771	1·6335						1·82
				1·5949	0·1250	0·1994	+ 0·2386	+ 0·0476	
4	36	0·6021	1·5563						1·35
				0·9286	0·0969	0·0899	+ 0·3495	+ 0·0314	
5	2	0·6990	0·3010						1·00
						1·6424		+ 0·1145	
								− 0·1429	
								− 0·0284	

Area of whole distribution = 1·6424.

$$\nu_1 = \frac{-\ 0{\cdot}0284}{1{\cdot}6424} = -\ 0{\cdot}0172$$

Mean = arbitrary origin − 0·0172 = 0·3010 − 0·0172 = 0·284

Limits are therefore − 0·284 and 0·6990 − 0·284 = + 0·415

$$\text{Area} = 1{\cdot}6424 = \int_{x=-0{\cdot}284}^{x=0{\cdot}415} (nx + a)\, dx = 0{\cdot}0458\, n + 0{\cdot}699\, a$$

$$\mu_1 = 0 = \int_{x=-0{\cdot}284}^{x=0{\cdot}415} [x(nx + a)]\, dx = 0{\cdot}0314\, n + 0{\cdot}0458\, a$$

These equations give $y = -3{\cdot}7x + 2{\cdot}535$

We have now found the equation to the straight line which fits best the irregularly placed points which we have quoted. The area beneath this straight line, and between the first and last ordinates, is equal to that formed by the series of trapezoids. We calculate the new ordinates, $-n$ being

the tangent which the line makes with the axis of x, and a the intercept on the centroid vertical of the graph. Remembering that these are really logarithms we now find the smoothed values of the original data.

We also find the equation to the hyperbola. Converting the logs back again into natural numbers we find the value of n, now the index of x and positive; a is, of course, merely a scale constant. It represents the (graduated) number of bacteria per unit volume which was present in the culture at the beginning of the experiment. We find the constants in the equation $yx^n = a$.

III. The Counts of Colonies on Plates.

Some matters of interest with regard to methods may be noticed here. As a rule one finds different kinds of colonies on the same plate. In cultures of badly polluted or recently polluted mussels in neutral red, bile-salt, lactose agar we find both crimson and colourless colonies. The latter are always on the surface of the plates; at least, if they are present in the depth of the medium one cannot see them. They differ greatly in their ability to ferment sugars from the crimson colonies; as a rule they do not ferment glucose and lactose. They are hardly ever seen in cultures from mussels which are not badly polluted, and which have been taken from places at some considerable distance from the source of pollution. They are always absent in cultures made from mussels which have been re-laid. I attach some importance to the presence of such colourless colonies as indicating recent pollution. I have subcultured very few of them—none of these subcultures are described in this Report. As a matter of both theoretical and practical interest they ought to be closely investigated.

The crimson colonies are present both on the surface

and in the depth of the plate. Those on the surface are usually flat with raised centres, and of varying shades of crimson. The central part is usually deeper in colour than the margin. Sometimes they have very little colour, but one can always distinguish them from the transparent colourless colonies. The latter are generally slightly irridescent, whereas the others (the crimson colonies) are slightly opaque even if they have very little colour. The colonies in the depths are always much smaller, and their colour is much deeper. The larger ones are egg-shaped when seen from the surface, and there is generally a slight haze round them, easily seen if the medium is clear. In cultures from mussels, made as I have described, we inoculate a complex physical mixture, a suspension, largely of broken-down solid tissue, and this often prevents the haze round the deep colonies from being visible. These deep colonies are really lenticular in shape, with their long diameters perpendicular to the surface of the plate. They assume this position, I suppose, because they grow more easily towards the surface than parallel to it.

Now these differences in the naked-eye appearance of the red colonies are of little or no significance: we cannot tell, except by subculturing a colony, what micro-organism it represents. The differences are produced by the varying positions of the colonies in the medium, and by the degree of aggregation of the colonies. Consider what is the physical structure of an agar plate. It is an emulsoid consisting of two liquid phases. In a dilute solution of agar, like that employed in the preparation of "solid" nutritive media, cooling throws out of true solution a phase consisting of droplets of a relatively concentrated solution of agar in water (or water in agar). These droplets coalesce together to form a meshwork in the interstices of which is the second phase, the relatively dilute solution of agar in water. The liquids of the two phases do not separate into layers, but remain

permanently in the form of a jelly, or a viscous liquid, according to the concentration. The interstices of the meshwork are, nevertheless, so fine as to prevent the transport of even such small particles as the bacilli. How these spread through the jelly is a subject for microscopical investigation by sections of the hardened substance—I do not know if this has been attempted. The agar jelly we may imagine to be like a sponge containing liquid, and the liquid consists of the dilute agar solution, and of the solutions of the nutritive and other substances entering into the composition of the medium. In a very concentrated agar jelly, the phase containing much water would consist of droplets enclosed by the phase containing little water, so that diffusion of electrolytes through the jelly would be very slow. But in the dilute agar jelly there would be little more resistance to diffusion than if the whole were a true molecular solution.

The surface of the jelly is always moist, since it is contracting and liquid is being forced out. A colony on the surface will, therefore, grow more rapidly than one in the depth of the plate, since it will take up the dissolved nutritive substance round itself, and the concentration of the latter will be renewed by diffusion from surrounding areas of the surface as rapidly as it is lowered. Therefore, these superficial colonies will be larger, and slightly different in appearance.

Diffusion will be rather slower in the depth of the plate, and adjacent colonies will act as centres of absorption for the dissolved nutritive substances. If the colonies are numerous they ought to be smaller, since they are growing at the expense of the liquid food substances added to the jelly rather than the latter itself. And this is what we actually find. The colonies in the crowded plates are always smaller than those in plates which have been inoculated by only a few bacteria.

Further, and this is a matter of considerable practical importance in methods, a crowded plate ought to indicate

a higher degree of pollution than the actual count appears to indicate. For the colony in the depth is only visible when it concentrates the stain added to the medium, either physically, or by a process of *intra-vitam* staining—this is a point of theoretical interest that might be investigated—and numerous colonies will deplete the jelly of the added nutritive substances. The more rapidly growing colonies will, therefore, "starve out" those which grow more slowly: we may safely assume that there are such individual differences among bacteria of even the same species and culture.

As a matter of fact this is easily proved by making a series of decimal dilutions of the same culture. In the higher dilutions we shall find that the numbers of colonies in the plates are multiples, or sub-multiples, by 10 of some number. But the counts of the lower dilutions, if the original culture were sufficiently concentrated, are not so much as 10, 100, times those of the middle dilutions. It is important to note, therefore, that the estimates of the number of bacteria in the substance being investigated should be based on the higher rather than on the lower dilutions; and that dilutions should be made so as to have few colonies on the plate, apart altogether from the question of ease in making the counts.

IV. Methods of Sampling.

Precautions in taking and forwarding samples of mussels usually take the form of keeping the shellfish cool by packing the vessels containing them in ice. It is assumed that the micro-organisms contained in the shellfish may multiply if the temperature is raised, and that this may, indeed, be the case can often be shown by keeping the sample for a day or more at laboratory temperature. But if it is the case that intestinal bacteria do not continue to live and reproduce in sea-water, as the experiments described on pp. 128-133.

show, then it may also be the case that a reduction in the numbers of the micro-organisms contained in shellfish may be experienced as the result of keeping the sample for a day or two after collection. I have obtained such a result in at least one case. It is probable that micro-organisms contained in the alimentary canal of the shellfish tend to be extruded into the mantle cavity, and if the temperature rises, or the mussels are kept too long, the water in the mantle cavity may be lost by the opening of the shells. In such a case the estimate of the bacteria per mussel may be less than it ought to be. Obviously the samples ought to be stored in small sterilised vessels which hold no more than the precise number of shellfish which are to be examined, and the water in the vessel ought to be added to the mixture, which is prepared from the soft parts of the shellfish. It is as well that this possibility of a reduction in the number of bacteria should be considered in cases where apparently anomalous results are obtained.

The Barmouth purification experiment of 1915, referred to on p. 140, is a case in point. Here we have to deal with an estuarine area subject to pollution, as the chart published in last year's report shows. In July of 1913 I visited the estuary and made an inspection of the mussel beds and took samples. These gave unequivocal evidence of most undesirable pollution. A typical plate, made from 1/50th part of a mussel was photographed, and is reproduced as fig. 2 of Plate II in the present report. In February of this year (1915) the Fishery Officer at Aberdovey made the experiment to which I refer above. The samples were sent to me by parcel post. Fig. 1 of Plate II represents a typical plate made from 1/50th mussel, and the difference in the apparent pollution is very marked. Such a degree of pollution as that represented by fig. 1 I am inclined to regard as of little or no significance. The question of the extent to which the mussels underwent

self-cleansing is, of course, quite a different one, and is unaffected by any question of irregular sampling, for all the samples were taken and forwarded in the same way, and whatever applies to the original sampling also applies to the sampling after the relaying. But we have to consider the difference in the apparent bacterial contents of two samples of mussels sent from the same area. In summer the population of Barmouth is a holiday one (say about 5,000), whereas in winter it may be reduced to about half that number. In summer the volume of fresh water in the river is small, whereas in the months of January and February it may be considerable on account of rains. The smaller population and the increased amount of flushing of the river may perhaps account for the difference in the apparent pollution, but perhaps the difference may be due, to some extent, to the conditions of sampling. I mention this particular case as illustrating the contention that the actual conditions, as regards pollution, of a particular natural shellfish area, at a particular time, may not be accurately estimated merely from samples collected and transmitted by persons other than those making the analysis. In cases where the results of analysis are important, the collection and further history of the sample prior to analysis is equally important.

Explanation of the Chart.

Entrance to the Lune Estuary.

The sketch chart has been reduced from the 6-inch ordnance map (Lancashire Sheet XXXIV). Sandbanks uncovered by ordinary tides are represented by fine stippling. Hard, stony foreshore is represented by coarser stippling and small circles. The extensive salt marshes of this district are represented by short horizontal lines. The positions of the mussel beds and the places where mussels were re-laid are indicated by coarse cross-hatching.

The narrow strip of clear water is the main channel leading to Lancaster and Glasson Dock. At low water of ordinary tides it is only a few feet in depth, and it is still shallower at low water of spring tides. It is about 300 feet in width on the average. Small, shifting channels run from it towards the "becks," or brooks, ashore, and through the salt marshes. Many of these smaller channels are not shown on the sketch chart.

The main course of the stream is along the deepest part of the estuary during the first part of flood tide. When the banks are covered the stream passes over them, in the main following the trend of the estuary, but with numerous irregularities in direction and velocity due to the conformation of the sea bottom. When the banks are covered by the flood tide, the water on them may be regarded as bacteriologically pure, since it comes in past Sunderland Point from the Main Lune Channel, a passage widening out into Lune Deep, practically the open sea. The flood stream runs for four to five hours, the ebb stream for seven to eight hours. The higher parts of the banks are soon uncovered, so that the water ebbing from off them has not had time to become polluted from the upper parts of the river. There is hardly

any pollution of the smaller channels on the north side of the estuary since there are only a few houses here and there along this shore.

The main sources of pollution of the estuary are Lancaster and Stodday, that is, a population of over 40,000 persons. All this sewage is untreated. It enters the channel at various points higher up than the area represented on the Sketch Chart. Two sewers are shown, those at Glasson and Conder Green. These serve a population of about 1,600 persons. The sewage is untreated and discharges directly into the sea, the Glasson sewer into the main channel, and the Conder Green sewer into a brook which flows through a salt marsh into the main channel.

The mussels taken from the Lune Estuary come from various places. Most are taken from the training wall at Bazil Point and the adjacent part of the channel. This wall is a rubble structure which comes adry at low water, so that it is washed by the water coming down the channel at the lowest states of the tide. The mussels here are thoroughly polluted. The numbers M 90,000, 11,650, and 3,700 represent the results of recent analyses—they are the mean numbers of sewage bacteria contained in a mussel. Some mussels are taken from Crook Skear, which is the area of rough foreshore opposite Sunderland Point. Others are taken from the Skears at Abbey Lighthouse, and from the bottom of the Channel near there.

The water in the estuary at low water is greatly polluted. The numbers W 83 and W 82 on the sketch chart represent the numbers of sewage bacteria per cubic centimetre as found in some recent analyses. These numbers would probably vary greatly from time to time according to the state of tide and the amount of fresh water in the river.

In the cleansing experiments the mussels were taken from the training wall, and the adjacent bottom of the channel not

far from Bazil Point. They were re-laid, first of all, at the place marked a little way north-east from Bazil Point. The sand banks are very high here, so that by the time the flood-tide has reached this part of the foreshore the sewage in the lower part of the channel will have become enormously diluted. Whatever water ebbs from off this foreshore, and down the little channel passing it, will therefore be clean. As the tide ebbs out the water in the channel will become more and more foul, but it can no longer come near the re-laid mussels. The experiments were made on 7th, 8th, 9th, and 10th October, 1914. The highest spring tides were on the 4th (an 18 foot 6 inch tide at Liverpool); when they were begun the mussels covered at 11 a.m. and uncovered at 3 p.m. At low water they would be covered for about two to three hours on each tide. The experiment made at Sunderland Point was made under much the same conditions. In these experiments the number of bacteria contained in a mussel was reduced from about 12,000 to about 400.

The place selected at Glasson was, as the chart shows, much less favourable. It is very close to the outflow of sewage from the River Conder. It is well up a steep beach, so that it was covered for about three and a half hours on each tide (two to five days after lowest neaps, a 12 foot 9 inch tide at Liverpool). There was much rain, and the river was in flood. The contamination was reduced from about 90,000 to 24,000 sewage bacteria per mussel. The results might have been more favourable given better conditions, but the place is, evidently, far from suitable.

Explanation of Plate I.

Cultures of 1/50th part of a mussel in neutral-red, bile-salt, lactose agar. (See pp. 136-7.)

Fig. 1. The original sample of mussels taken from the training wall in the River Lune.

Fig. 2. The same mussels re-laid for six tides.

Fig. 3. The same mussels re-laid for four tides.

Explanation of Plate II.

Fig. 1. Culture of 1/50th part of a mussel from Barmouth, winter of 1915. (See p. 140.) The liquid inoculated was very turbid, so that the medium is greatly discoloured. There were eight red colonies in the plate.

Fig. 2. A similar culture made from Barmouth mussels from a neighbouring bed, taken in the summer of 1913. The red colonies are very numerous and there is a large patch of small, partially fused colourless colonies.

Plate III.

Lower part of the Estuary of the Lune.

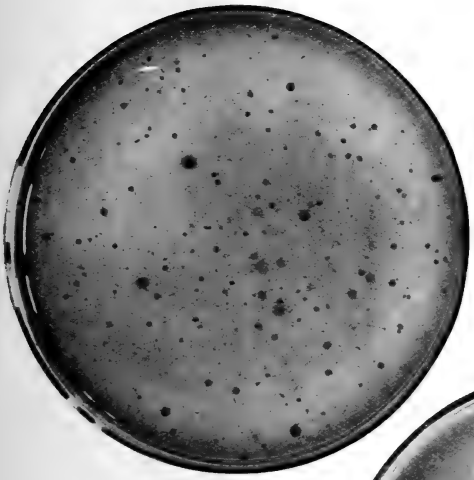


FIG. 1.

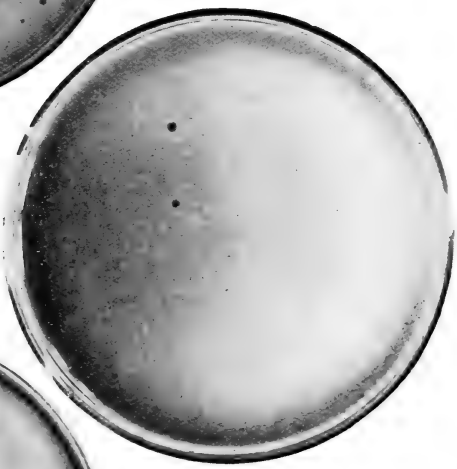


FIG 3.

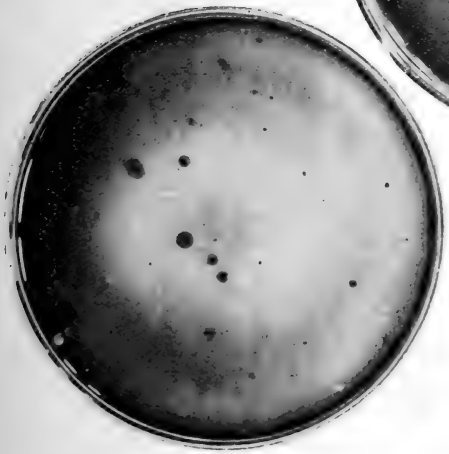


FIG. 2.

Cultures of Faecal Bacteria from Mussel Relaying Experiments.

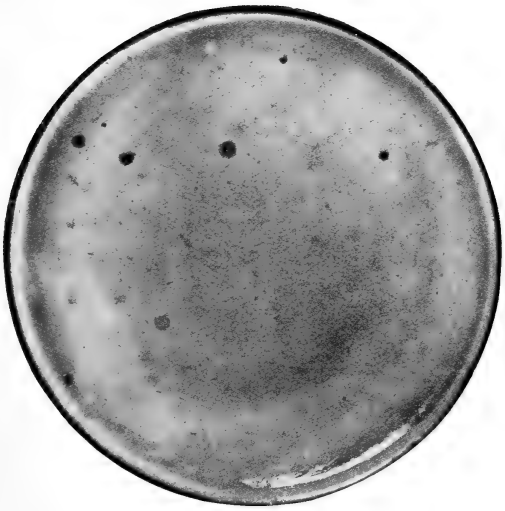


FIG. 1.

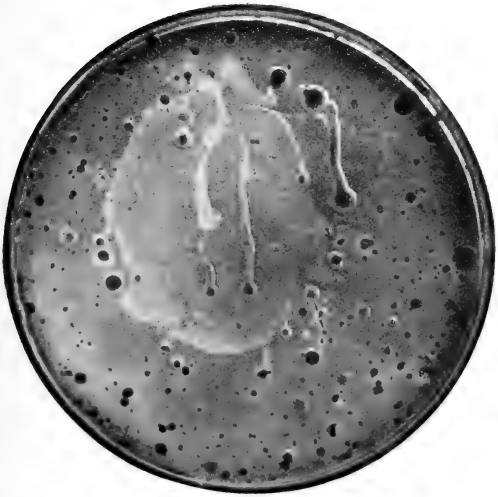
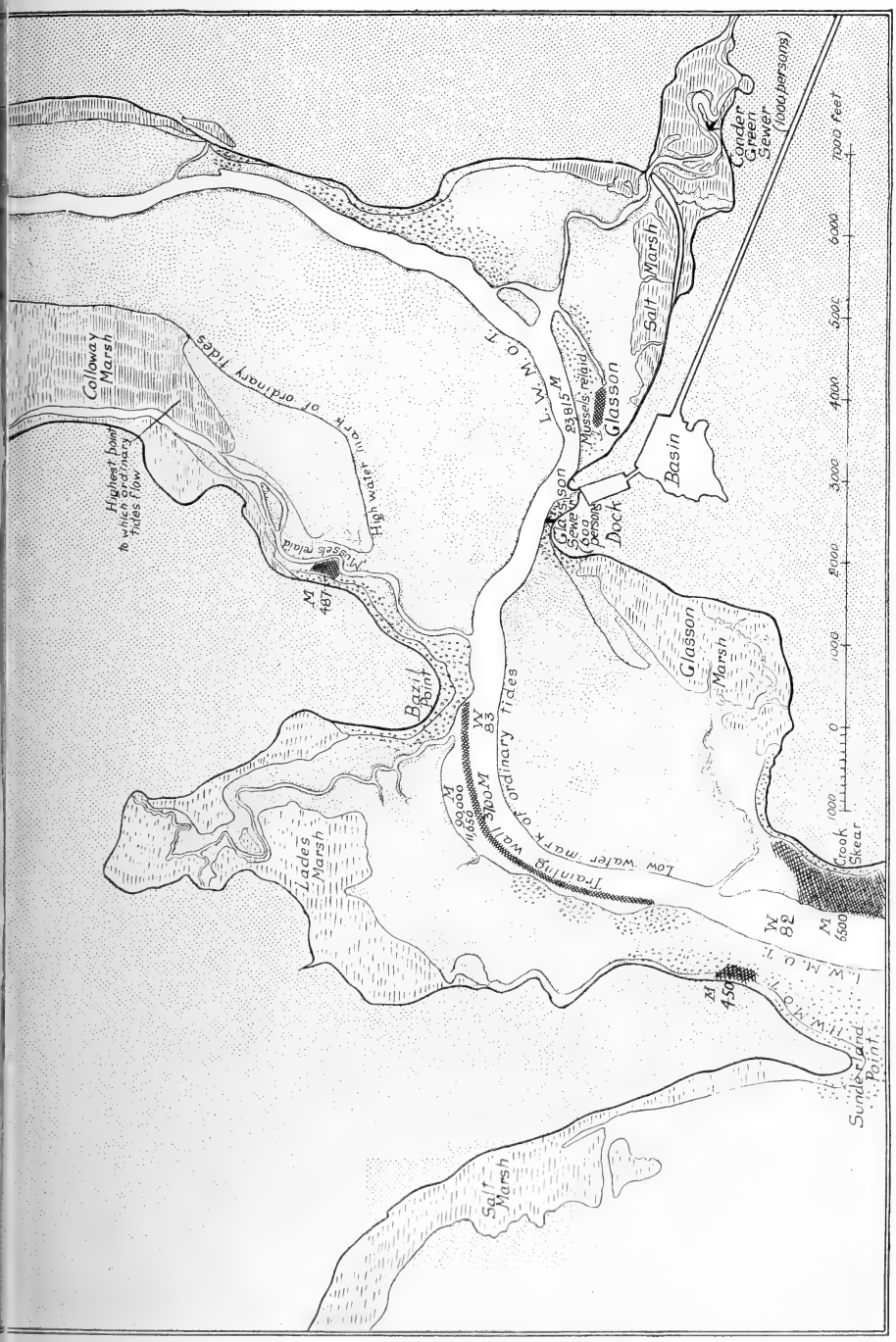


FIG. 2.

Cultures of Faecal Bacteria from Aberdovey Mussels.



Lower part of the Estuary of the Lune.

REPORT ON HERRING MEASUREMENTS.

By W. Riddell, M.A.

During the past year 19 samples of herring have been examined, amounting to 1,068 fish. Particulars of these will be found in Tables III-V.

As some errors have been found in the tables published in last year's Report, these have been checked, and the revised figures are here re-published as Tables I and II.

In Tables I and II, and sample (1) of Table III, the length *T* is measured to the line joining the tips of the lobes of the caudal fin when this is in its natural position; *A* is measured to the anus. After this, at the request of the Board of Agriculture and Fisheries, these measurements were slightly altered. *T* is now measured to the tip of the dorsal lobe of the caudal fin when extended in line with the body, and *A* is taken to the beginning of the anal fin. The other characters are :—

T.cd. Total length to base of tail.
D. Length from tip of snout to beginning of dorsal fin.
V. Length from tip of snout to beginning of pelvic fin.
l.cp.l. Lateral length of head.
K_2. Number of keeled scales between anus and pelvic fins.
s. Sex.
g. Condition of gonads on Hjort's scale.

I do not consider that the measurements recorded up to now present any definite evidence either for or against the question of local races.

There are differences between samples from different areas, but samples from the one area differ just as much among

themselves. Thus one would expect the two trawled samples to represent one race, but these two samples differ markedly. The same thing may be seen in the samples from Port Erin.

At first sight this might be regarded as against any division into races, but there are two defects in the figures which make any such conclusion untrustworthy.

In the first place, the sampling has, in many cases, been quite insufficient. This is especially the case in regard to the fish from the Welsh Coast. The samples for the 1913-1914 season are much too small, and while those for 1914-1915 are better, they are still, I consider, insufficient. This seems to have been a bad season on the Welsh coast; it was impossible to get large samples, and no fish were received after the end of 1914. In the second place, many of the samples were kept in cold storage until time allowed of their being examined. This, I am now sure, renders any measurements made upon such samples quite untrustworthy. I do not believe that any correction can be applied to compensate for the distortion caused by this method of preservation. This is the only conclusion I can draw from my own results, and Williamson's* experiments upon mackerel point to the same thing. Future samples must be examined in a fresh condition. It is possible that other modes of preservation may not be open to the same objection. Heincke applies a correction to some of his samples which were preserved in spirit. But I think that, until direct experiment has shown that such a correction gives satisfactory results, all measurements made on preserved fish must be regarded with suspicion, and not used for comparison with measurements made on fresh material.

This does not apply, of course, to the examination of skeletal characters, such as vertebrae. So far I have not examined the vertebrae of a sufficient number of fish to be able to draw any conclusion from them. The keeled-scales,

* *Fishery Board, Scotland, 18th Annual Report.*

also, are unaffected; but here, as regards the present figures, we are faced with the other difficulty of insufficient sampling.

Further samples from all districts must be examined before the question of local races can be discussed with any confidence.

One point on which something may, perhaps, be said is the relative numbers of the sexes. Heincke states that he has the same impression as Ewart, that the females come to the spawning grounds earlier than the males, and also leave earlier. He also says that he believes that among spawning herring the females are in the majority.

My figures lead me to believe that males are more abundant than females. Even omitting Stages I, II and VII, so as to remove any possibility of a mistake in determining the sex, there are more males than females in my samples.

This would agree with Fulton's statement* that among fish with demersal eggs females are, as a rule, fewer than the males, although his figures for the herring† show practical equality in numbers between the sexes, females being in a slight majority. Ewart's‡ account of the spawning of the herring would also seem to indicate a preponderance of males.

* *Fishery Board, Scotland, 9th Annual Report.*
† *Fishery Board, Scotland, 8th Annual Report.*
‡ *Fishery Board, Scotland, 2nd Annual Report.*

Table I.

(All measurements in the tables are in millimetres.)

½ mile E. of New Quay Head, November 7th, 1913. 4″ mesh.

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
1	262	230	117	50·87	131	56·95	172	74·78	46	20·00	14	♀	V
2	260	222	116	52·25	122	54·99	170	76·57	46	20·72	14	♀	V
3	256	221	119	53·84	125	56·56	174	78·73	46	20·81	13	♀	V
4	255	223	116	52·01	123	55·15	166	74·43	46	20·62	14	♀	V
5	252	221	112	50·67	123	55·65	170	76·92	47	21·26	15	♀	V
6	250	220	111	50·45	122	55·45	169	76·81	47	21·36	14	♀	V
7	250	219	115	52·51	122	55·70	165	75·34	45	20·54	12	♀	V
8	250	218	113	51·83	119	54·58	164	75·22	45	20·64	14	♀	V
9	250	217	112	51·61	120	55·29	165	76·03	45	20·73	13	♀	V
10	250	213	115	53·99	119	55·86	164	76·99	45	21·12	14	♀	V
11	249	212	109	51·41	114	53·77	163	76·88	43	20·28	15	♀	V
12	248	214	114	53·27	117	54·67	163	76·16	46	21·49	14	♀	V
New Quay, November 11th, 1913. 4″ mesh.													
13	276	239	123	51·46	128	53·55	182	76·15	53	22·17	13	♀	V
14	274	242	125	51·65	137	56·61	189	78·09	50	20·66	14	♀	V
15	268	237	124	52·32	133	56·11	183	77·21	47	19·83	15	♀	V
16	264	229	120	52·40	128	55·89	178	77·72	49	21·39	13	♀	V
17	259	224	114	50·89	124	55·34	176	78·57	51	22·76	14	♀	V
18	258	227	116	51·10	120	52·86	174	76·65	47	20·70	14	♀	V
19	257	225	111	49·33	119	52·88	172	76·44	46	20·44	14	♀	V
20	257	224	115	51·33	119	53·12	167	74·55	45	20·09	13	♀	V
21	256	223	115	51·56	121	54·26	171	76·68	46	20·62	13	♀	V
22	255	221	120	54·29	120	54·29	172	77·82	45	20·36	15	♀	V
23	253	221	113	51·13	122	55·20	174	78·73	45	20·36	14	♀	V
24	250	225	113	50·22	124	55·11	173	76·88	48	21·33	13	♀	V
25	250	218	112	51·37	119	54·58	167	76·60	47	21·56	13	♀	V
26	246	216	108	50·00	117	54·16	166	76·85	45	20·83	15	♀	V
27	239	210	104	49·52	113	53·80	158	75·23	43	20·47	13	♀	V
Penrhyn Weir, Bangor, November 17th, 1913.													
28	276	244	127	52·04	131	53·68	184	75·40	50	20·49	15	♀	V
29	262	231	122	52·81	129	55·84	175	75·75	50	21·64	15	♀	V
30	255	225	118	52·44	126	56·00	172	76·44	47	20·88	15	♀	V
31	253	225	116	51·55	128	56·88	172	76·44	48	21·33	15	♀	V
32	253	223	117	52·46	119	53·36	165	73·99	48	21·52	15	♀	V
33	249	217	114	52·53	125	57·60	165	76·03	47	21·65	13	♀	V
34	248	217	114	52·53	119	54·83	167	76·95	50	23·04	15	♀	V
35	248	214	112	52·33	121	56·54	164	76·63	47	21·96	15	♀	V
36	246	218	118	54·12	125	57·33	168	77·06	49	22·47	14	♀	V
37	242	213	108	50·70	116	54·46	158	74·17	45	21·12	14	♀	V
Moelfre, November 20th, 1913.													
38	272	239	121	50·62	139	58·15	184	76·98	52	21·75	15	♀	VII
39	268	235	124	52·76	133	56·59	183	77·85	54	22·97	14	♀	VII
40	264	234	120	51·28	131	55·98	172	73·50	52	22·22	14	♀	V-VI
41	263	230	120	52·17	130	56·52	172	74·78	52	22·60	15	♀	V-VI
42	262	235	121	51·48	130	55·31	175	74·46	50	21·27	14	♀	VII
43	262	234	120	51·28	132	56·41	179	76·49	51	21·79	14	♀	VII
44	259	226	120	53·09	127	56·19	175	77·43	49	21·68	14	♀	VII
45	255	226	114	50·44	128	56·63	170	75·22	45	19·91	14	♀	V-VI
46	250	223	111	49·77	122	54·70	168	75·33	48	21·52	15	♀	VII
47	249	213	114	53·52	118	55·39	164	76·99	47	22·06	15	♀	VII

Table I—*Continued.*

Moelfre, November 20th, 1913—*Continued.*

s.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
48	246	216	110	50·92	120	55·55	162	75·00	46	21·29	14	♂♀	VII
49	245	214	114	53·27	120	56·07	163	76·16	48	22·43	14	♀	V-VI
50	245	214	111	51·87	119	55·60	164	76·63	48	22·43	15	♂	VII
51	239	209	106	50·71	114	54·54	159	76·07	44	21·05	15	♂	V-VI
52	238	209	110	52·63	118	56·45	162	77·51	45	21·53	13	♂	V-VI
53	237	207	106	51·20	115	55·55	157	75·84	46	22·22	15	♀	VII
54	234	207	107	51·69	115	55·55	155	74·87	47	22·70	14	♂♀	VII
55	232	204	110	53·92	114	55·88	152	74·51	47	23·03	15	♂	VII
				1 mile N.E. of New Quay, November 24th, 1913. 4″ mesh.									
56·	272	240	127	52·91	136	56·66	185	77·08	52	21·66	14	♀	V
57	270	242	124	51·23	130	53·71	179	73·96	48	19·83	14	♀	V
58	266	235	121	51·48	129	54·89	179	76·17	48	20·42	14	♂	V
59	265	233	118	50·64	130	55·79	179	76·82	50	21·45	15	♀	V
60	264	236	122	51·69	130	55·08	178	75·42	50	21·14	14	♀	V
61	260	229	117	51·09	127	55·45	171	74·67	48	20·96	14	♀	V
62	256	226	119	52·65	127	56·19	173	76·54	48	21·23	15	♂	V
63	256	225	114	50·66	127	56·44	171	76·00	48	21·33	13	♂	V
64	254	227	118	51·98	123	54·18	171	75·33	45	19·82	14	♀	V
65	254	221	116	52·48	127	57·46	172	77·82	50	22·62	15	♂	V
66	253	220	114	51·81	123	55·91	168	76·36	48	21·81	15	♀	V
67	251	222	114	51·35	119	53·60	168	75·67	48	21·62	15	♂	V
				Penrhyn Weir, Bangor, November 24th, 1913.									
68	271	241	125	51·86	132	54·77	185	76·76	50	20·74	14	♀	V
69	271	239	123	51·46	130	54·39	181	75·73	48	20·08	16	♀	V
70	260	229	124	54·14	127	55·45	175	76·41	51	22·27	14	♂	V
71	259	229	115	50·21	122	53·27	172	75·10	48	20·96	14	♀	V
72	256	226	117	51·77	124	54·86	174	76·99	49	21·68	15	♀	V
73	250	221	118	53·39	124	56·10	168	76·01	49	22·17	14	♀	V
74	248	217	114	52·53	122	56·22	167	76·95	48	22·12	13	♂	V
75	246	217	114	52·53	121	55·76	166	76·49	48	22·12	14	♂	V
76	241	214	112	52·33	118	55·14	157	73·36	46	21·49	14	♂	V
77	219	197	98	49·74	105	53·29	144	73·09	43	21·82	14	♀	V
78	218	185	94	50·81	102	55·13	138	74·59	39	21·08	14	♀	I-II
79	203	177	99	55·93	101	57·06	134	75·70	41	23·16	13	♀	I-II
80	202	179	94	52·51	99	55·30	137	76·53	42	23·46	14	♂	I-II
				½ mile from New Quay Head, December 8th, 1913. 4″ mesh									
81	282	252	130	51·58	145	57·53	190	75·39	53	21·03	16	♀	V
82	275	244	126	51·63	134	54·91	182	74·59	51	20·90	14	♀	V
83	267	240	124	51·66	133	55·41	181	75·41	49	20·41	14	♀	V
84	266	236	119	50·42	133	56·35	181	76·69	50	21·14	15	♀	V
85	266	234	120	51·28	130	55·55	175	74·78	49	20·94	14	♂	V
86	264	235	121	51·48	130	55·31	179	76·17	50	21·27	16	♂	V
87	258	229	119	51·96	129	56·33	173	75·54	50	21·83	14	♂	V
88	254	224	117	52·23	127	56·69	171	76·33	47	20·98	14	♂	V
89	252	221	119	53·84	125	56·56	174	78·73	48	21·71	14	♂	V
90	250	223	117	52·46	123	55·15	172	77·13	47	21·07	15	♂	V
91	249	221	116	52·48	124	56·10	171	77·37	47	21·26	15	♂	V
92	243	213	109	51·17	118	55·39	162	76·05	48	22·53	15	♀	V

Table I—*Continued.*

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
Moelfre, December 8th, 1913.													
93	254	224	115	51·33	123	54·91	170	75·89	50	22·32	13	♂	VII
94	253	224	116	51·78	123	54·91	170	75·89	48	21·42	13	♂♂	VII
95	253	223	114	51·12	122	54·70	168	75·33	50	22·42	13	♂♂	VI
96	249	221	115	52·03	122	55·20	170	76·92	47	21·26	14	♂♂	VII
97	242	213	112	52·58	119	55·86	168	78·87	45	21·12	14	♂♂	VI
98	241	212	111	52·35	116	54·71	162	76·41	45	21·22	13	♀♀	VI
99	240	211	108	51·18	116	54·97	159	75·35	45	21·32	14	♀♀	VII
100	239	212	111	52·35	119	56·13	162	76·41	47	22·17	14	♀♀	VII
101	239	211	107	50·71	116	54·97	159	75·35	44	20·85	13	♂♀	VII
102	237	208	105	50·48	117	56·25	159	76·44	46	22·11	14	♂♀	VII
Penrhyn Weir, December 9th, 1913.													
103	254	226	118	52·21	124	54·86	170	75·22	48	21·23	14	♀	V
104	254	225	116	51·55	124	55·11	170	75·55	47	20·88	14	♀♀	V
105	254	222	116	52·25	122	54·99	170	76·57	50	22·51	14	♂♀	V
106	252	223	115	51·56	121	54·26	165	73·99	49	21·97	14	♂♀	III
107	238	211	107	50·71	114	54·03	155	73·45	45	21·32	13	♂♀	V
108	238	210	108	51·42	116	55·24	161	76·66	45	21·43	14	♀	V
109	214	186	97	52·15	103	55·37	140	75·26	44	23·65	12	♀♀	I-II
110	206	181	96	53·03	101	55·80	136	75·13	42	23·20	14	♂♀	I-II
111	205	181	93	51·38	102	56·35	133	73·48	41	22·65	13	♀♀	I-II
112	204	183	92	50·27	98	53·55	134	73·22	41	22·40	12	♀♀	I-II
113	202	174	94	54·02	96	55·17	129	74·13	41	23·56	14	♀♀	I-II
114	201	174	90	51·72	97	55·74	131	75·28	40	22·98	12	♀♀	I-II
115	199	172	88	51·16	94	54·65	128	74·41	40	23·25	12	♀♀	I-II
116	198	174	88	50·57	96	55·17	130	74·71	40	22·98	14	♂♀	I-II
117	198	173	91	52·60	96	55·49	128	73·98	41	23·69	14	♂♂	I-II
118	195	174	93	53·44	97	55·74	131	75·28	41	23·56	13	♂♂	I-II
119	192	171	89	52·04	94	54·97	128	74·85	40	23·39	14	♂♀	I-II
120	184	161	84	52·17	88	54·65	120	74·53	38	23·60	13	♀	I-II
Tremadoc Bay, December 18th, 1913.													
121	261	230	118	51·30	126	54·78	172	74·78	50	21·73	15	♀♀	VII
122	255	227	118	51·98	123	54·18	170	74·89	47	20·70	14	♀♀	VI
123	255	226	115	50·88	120	53·09	166	73·44	45	19·91	15	♂♀	VI
124	250	222	114	51·35	117	52·70	166	74·77	45	20·27	15	♂♀	VII
125	249	220	115	52·27	121	55·00	167	75·90	46	20·90	14	♂♀	VII
126	246	218	112	51·37	121	55·50	164	75·22	47	21·56	14	♂♀	VII
127	246	217	112	51·61	116	53·45	162	74·65	47	21·65	15	♀♀	VII
128	245	217	111	51·15	118	54·37	164	75·57	44	20·27	15	♀♀	VI
129	242	218	111	50·91	117	53·67	162	74·31	45	20·64	15	♀♀	VI
130	241	213	110	51·64	116	54·46	157	73·70	46	21·69	13	♂♂	VII
131	236	210	109	51·90	111	52·85	155	73·81	45	21·43	15	♂	VII
Moelfre, January 6th, 1914													
132	268	236	121	51·27	129	54·66	177	75·00	47	19·91	14	♂♀	VI
133	257	231	119	51·51	127	54·97	174	75·32	49	21·21	14	♂♀	VI
134	246	220	113	51·36	122	55·45	170	77·27	46	20·90	15	♂♂	VII
135	245	215	112	52·09	118	54·88	163	75·81	48	22·32	13	♂♂	VII
136	244	214	107	50·00	116	54·20	160	74·76	46	21·49	13	♂♀	VII
137	243	214	112	52·33	119	55·60	162	75·70	47	21·96	14	♂♀	VII
138	238	212	108	50·94	116	54·71	159	75·00	45	21·22	13	♂♀	VII
139	233	204	103	50·49	113	55·39	155	75·98	44	21·56	13	♂♀	VII
140	223	197	103	52·28	110	55·83	149	75·63	45	22·84	13	♂♀	VII-I
141	222	197	101	51·26	109	55·33	146	74·11	44	22·33	13	♂♀	VII-I
142	222	193	101	52·33	105	54·40	145	75·13	43	22·28	14	♂	VII-I
Mean				51·78		55·22		75·79		21·54	14·03		

Table II.

150 Herring : trawled off The Smalls, October 25th, 1913.

(Kept in cold storage until January 12th, 1914.)

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
1	311	280	147	52·50	153	54·64	209	74·64	59	21·07	14	♂	V
2	309	272	144	52·94	152	55·88	207	76·10	58	21·32	14	♂	V
3	301	262	139	53·05	149	56·87	200	76·33	61	23·28	13	♀	V
4	300	262	134	51·14	147	56·10	199	75·95	58	22·13	13	♀	V
5	298	260	141	54·22	149	57·30	200	76·92	61	23·46	13	♂	V
6	292	257	139	54·08	144	56·03	198	77·04	57	22·17	15	♂	V
7	292	257	139	54·08	148	57·58	198	77·04	57	22·17	13	♂	V
8	292	256	140	54·68	143	55·85	198	77·34	59	23·04	14	♀	V
9	292	255	136	53·33	141	55·29	195	76·47	57	22·35	13	♀	V
10	292	255	140	54·94	148	58·03	196	76·86	58	22·74	14	♀	V
11	291	252	132	52·38	140	55·55	192	76·19	59	23·41	13	♀	V
12	290	257	136	52·91	142	55·25	192	74·70	55	21·40	13	♂	V
13	290	257	136	52·91	144	56·03	196	76·26	56	21·79	15	♀	V
14	288	256	136	53·12	143	55·85	196	76·56	57	22·26	14	♂	V
15	288	252	136	53·96	141	55·95	189	75·00	54	21·42	14	♀	V
16	288	250	130	52·00	139	55·60	191	76·40	58	23·20	14	♂	V
17	287	252	130	51·58	138	54·76	189	75·00	54	21·42	14	♂	V
18	287	251	131	52·19	142	56·57	190	75·69	55	21·91	13	♀	V
19	285	254	133	52·36	144	56·69	191	75·19	56	22·04	13	♂	V
20	285	248	125	50·40	136	54·83	188	75·80	53	21·37	14	♂	V
21	284	250	136	54·40	139	55·60	188	75·20	58	23·20	13	♂	V
22	284	249	130	52·20	141	56·62	191	76·70	55	22·08	14	♂	V
23	284	249	132	53·01	137	55·02	189	75·90	55	22·08	14	♂	V
24	283	251	135	53·78	137	54·58	185	73·70	53	21·11	14	♀	V
25	282	253	133	52·56	141	55·73	194	76·68	57	22·52	15	♀	V
26	282	252	135	53·57	141	55·95	195	77·38	60	23·80	15	♂	V
27	282	244	132	54·09	138	56·55	189	77·45	54	22·13	14	♀	V
28	281	250	132	52·80	139	55·60	188	75·20	57	22·80	14	♀	V
29	280	249	138	55·42	141	56·62	193	77·51	58	23·29	14	♀	V
30	280	248	132	53·22	137	55·24	191	77·01	57	22·98	15	♂	V
31	280	245	131	53·46	138	56·32	192	78·36	55	22·44	14	♂	V
32	280	243	127	52·26	138	56·79	188	77·36	53	21·81	14	♂	V
33	279	242	130	53·71	136	56·19	183	75·61	53	21·90	14	♀	V
34	278	244	124	50·81	139	56·96	187	76·63	53	21·72	14	♂	V
35	277	246	129	52·43	134	54·47	181	73·57	55	22·35	15	♀	V
36	277	246	132	53·65	138	56·09	186	75·61	53	21·54	14	♀	V
37	277	245	131	53·46	134	54·69	183	74·69	57	23·26	14	♀	V
38	277	245	130	53·06	136	55·51	189	77·14	54	22·04	14	♂	V
39	277	245	130	53·06	139	56·73	191	77·94	55	22·44	15	♀	V
40	277	245	130	53·06	137	55·91	189	77·14	52	21·24	14	♂	V
41	277	245	131	53·46	135	55·10	188	76·73	56	22·85	14	♂	V
42	277	245	130	53·06	139	56·73	187	76·32	56	22·85	14	♂	V
43	277	241	128	53·11	135	56·01	182	75·51	53	21·99	14	♂	V
44	276	244	127	52·04	132	54·09	184	75·40	48	19·67	14	♀	V
45	276	240	127	52·91	134	55·83	184	76·66	56	23·33	12	♂	V
46	276	240	127	52·91	134	55·83	183	76·25	55	22·91	15	♀	V
47	275	244	125	51·22	130	53·27	186	76·22	54	22·13	14	♀	V
48	275	243	127	52·26	133	54·73	183	75·28	52	21·39	14	♀	V
49	275	243	125	51·44	132	54·32	183	75·28	54	22·22	13	♂	V
50	275	242	130	53·71	133	54·95	187	77·27	53	21·90	15	♀	V

Table II—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
51	275	242	130	53·71	133	54·95	184	76·03	56	23·14	14	♂♀	V
52	275	241	127	52·69	133	55·18	180	74·68	54	22·40	14	♀	V
53	275	240	128	53·33	139	57·91	187	77·91	54	22·50	14	♂♂	V
54	274	245	134	54·69	141	57·55	190	77·55	57	23·26	13	♂♂	V
55	273	243	127	52·26	136	55·96	185	76·13	55	22·63	14	♂♂	V
56	273	243	124	51·02	134	55·14	185	76·13	52	21·39	14	♂♂	V
57	273	241	126	52·24	137	56·84	183	75·93	57	23·65	14	♀	V
58	273	239	127	53·13	136	56·90	183	76·56	52	21·75	14	♂♀	V
59	272	244	127	52·04	136	55·73	184	75·40	52	21·31	14	♀	V
60	272	242	125	51·65	133	54·95	182	75·20	52	21·48	14	♂	V
61	272	242	129	53·30	138	57·02	187	77·27	54	22·31	14	♂♂	V
62	272	241	124	51·45	135	56·01	186	77·17	55	22·82	14	♂♂	V
63	272	241	128	53·11	132	54·77	181	75·06	56	23·23	15	♂♂	V
64	272	239	128	53·55	135	56·48	184	76·98	52	21·75	15	♂♀	V
65	272	239	126	52·71	134	56·06	182	76·10	55	23·01	14	♂♀	V
66	272	236	125	52·96	134	56·77	179	75·84	52	22·03	14	♂♀	V
67	270	240	129	53·75	133	55·41	183	76·25	54	22·50	14	♀	V
68	270	237	127	53·59	132	55·69	181	76·37	52	21·93	13	♀	V
69	270	236	124	52·54	131	55·50	181	76·69	49	20·76	15	♀	V
70	269	236	126	53·39	131	55·50	182	77·11	52	22·03	14	♀	V
71	268	237	128	54·00	131	55·27	182	76·79	56	23·62	15	♀	V
72	268	236	125	52·96	133	56·35	182	77·11	49	20·76	14	♂♀	V
73	268	236	125	52·96	131	55·50	176	74·57	53	22·45	14	♂♀	V
74	267	236	126	53·59	133	56·35	180	76·27	53	22·45	14	♂♀	V
75	267	235	127	54·04	135	57·44	180	76·59	55	23·40	13	♂	V
76	267	233	122	52·36	131	56·22	177	75·96	49	21·03	13	♂♂	V
77	266	236	124	52·54	131	55·50	179	75·84	54	22·88	13	♂♂	V
78	266	235	123	52·34	131	55·74	180	76·59	52	22·12	14	♂♂	V
79	265	238	125	52·52	132	55·46	184	77·31	53	22·26	14	♂	V
80	265	237	125	52·75	134	56·53	179	75·52	55	23·20	14	♂♀	V
81	265	235	120	51·06	128	54·46	170	72·34	52	22·12	15	♂	V
82	265	235	125	53·19	131	55·74	180	76·59	53	22·55	15	♂♀	V
83	265	232	121	52·15	133	57·32	179	77·15	51	21·98	13	♂♀	V
84	265	230	121	52·60	126	54·78	175	76·09	51	22·17	15	♀	V
85	264	235	123	52·34	131	55·74	176	74·89	51	21·70	14	♀	V
86	264	233	122	52·36	131	56·22	177	75·96	53	22·74	15	♀	V
87	264	231	125	54·11	130	56·27	179	77·48	51	22·07	14	♀	V
88	264	229	123	53·71	130	56·76	170	74·23	54	23·58	13	♂♂	V
89	264	229	123	53·71	126	55·02	171	74·67	49	21·39	14	♂♂	V
90	263	236	128	54·23	134	56·77	181	76·69	54	22·88	12	♂♂	V
91	263	235	128	54·46	131	55·74	179	76·17	53	22·55	14	♂♂	V
92	263	233	125	53·64	127	54·50	177	75·96	53	22·74	14	♀	V
93	263	232	123	53·01	129	55·60	178	76·72	51	21·98	14	♀	V
94	263	231	123	53·24	130	56·27	172	74·45	53	22·94	13	♂♀	V
95	263	231	121	52·38	133	57·57	177	76·62	53	22·94	13	♂♀	V
96	262	233	123	52·78	132	56·65	178	76·39	52	22·31	14	♂♀	V
97	262	232	125	53·87	129	55·60	177	76·29	50	21·55	14	♂♂	V
98	262	231	121	52·38	132	57·14	180	77·92	51	22·07	15	♂♂	V
99	262	229	124	54·14	128	55·89	169	73·80	52	22·70	13	♂♀	V
100	261	230	126	54·78	130	56·52	179	77·82	53	23·04	14	♀	V
101	261	229	120	52·40	126	55·02	173	75·54	51	22·27	14	♂♀	V
102	261	227	117	51·54	124	54·62	173	76·21	53	23·34	13	♀	V
103	260	231	124	53·68	126	54·54	175	75·75	52	22·51	15	♀	V
104	260	231	122	52·81	129	55·84	173	74·89	52	22·51	13	♀	V

Table II—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
105	260	226	120	53·09	124	54·86	172	76·10	53	23·45	14	♀	V
106	259	234	125	53·41	131	55·98	181	77·35	50	21·36	14	♂	V
107	259	230	120	52·13	125	54·34	173	75·21	55	23·91	14	♂	V
108	259	229	119	51·96	130	56·76	175	76·41	50	21·83	14	♂	V
109	259	229	119	51·96	125	54·58	170	74·23	50	21·83	14	♂	V
110	256	228	120	52·63	125	54·82	172	75·43	51	22·36	13	♂	V
111	255	230	123	53·43	128	55·65	177	76·95	52	22·61	14	♀	III-IV
112	255	227	121	53·30	125	55·06	173	76·21	50	22·02	14	♂	V
113	255	225	119	52·88	124	55·11	169	75·11	50	22·22	15	♂	V
114	255	224	118	52·67	128	57·14	170	75·89	52	23·21	14	♂	V
115	255	222	116	52·25	122	54·99	165	74·32	50	22·51	15	♂	V
116	255	220	122	55·45	126	57·27	171	77·72	50	22·72	14	♂	V
117	254	234	124	52·99	130	55·55	178	76·06	50	21·36	14	♀	V
118	254	228	119	52·19	125	54·82	169	74·12	48	21·05	14	♀	V
119	254	222	120	54·05	124	55·85	171	77·02	51	22·97	15	♂	V
120	253	225	119	52·88	125	55·55	172	76·44	48	21·33	14	♂	V
121	253	224	115	51·33	121	54·01	172	76·78	53	23·66	14	♂	V
122	251	223	120	53·81	128	57·39	175	78·47	48	21·52	14	♂	IV
123	251	222	117	52·70	124	55·85	170	76·57	49	22·07	13	♂	IV
124	249	222	118	53·15	122	54·99	169	76·12	49	22·07	14	♀	V
125	249	218	113	51·83	122	55·96	165	75·68	50	22·93	13	♂	V
126	248	224	121	54·01	125	55·80	171	76·33	49	21·87	15	♀	V
127	248	221	114	51·58	119	53·84	166	75·11	46	20·81	13	♀	III
128	248	218	118	54·12	122	55·96	167	76·60	50	22·93	13	♀	III
129	248	217	115	52·99	119	54·83	165	76·03	44	20·27	14	♂	IV
130	247	222	114	51·35	122	54·99	169	76·12	49	22·07	14	♀	V
131	245	220	113	51·36	119	54·09	166	75·45	48	21·81	14	♀	V
132	245	215	114	53·02	118	54·88	160	74·41	48	22·32	14	♀	V
133	245	215	112	52·09	117	54·41	163	75·81	45	20·93	14	♂	V
134	244	219	118	53·88	121	55·25	165	75·34	48	21·91	13	♀	IV
135	244	217	110	50·69	115	52·99	162	74·65	48	22·12	13	♀	IV
136	244	217	112	51·61	120	55·29	165	76·03	45	20·73	13	♀	V
137	244	215	112	52·09	117	54·41	161	74·88	49	22·79	14	♂	V
138	244	211	112	53·08	117	55·45	164	77·72	45	21·32	15	♀	IV
139	243	216	114	52·77	119	55·09	162	75·00	46	21·29	14	♀	V
140	242	213	110	51·64	117	54·92	161	75·58	47	22·06	14	♂	V
141	241	213	108	50·70	117	54·92	164	76·99	48	22·53	13	♂	V
142	239	213	109	51·17	116	54·46	163	76·52	45	21·12	15	♀	IV
143	238	211	111	52·60	116	54·97	161	76·30	45	21·32	14	♂	V
144	238	211	109	51·65	117	55·45	161	76·30	45	21·32	15	♂	V
145	238	211	113	53·55	117	55·45	156	73·93	47	22·27	12	♂	V
146	237	210	107	50·95	114	54·28	159	75·71	45	21·43	14	♀	V
147	236	210	109	51·90	116	55·24	155	73·81	47	22·38	14	♂	IV
148	236	208	112	53·84	116	55·76	161	77·40	47	22·59	14	♂	IV
149	235	210	109	51·90	122	58·09	162	77·14	48	22·85	13	♂	IV
150	221	196	104	53·06	107	54·59	147	75·00	43	21·93	14	♂	II
Mean				52·85		55·68		76·07		22·23	13·91		

Table III.

Herring from Port Erin, Isle of Man.

(1) Received June 3rd, 1914 (arrived in bad condition).

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
1	282	241	131	54·35	134	55·60	184	76·34	55	22·82	15	♂	I-II
2	281	241	126	52·24	129	53·52	179	74·27	55	22·82	15	♀	I-II
3	278	240	125	52·08	134	55·83	178	74·16	52	21·66	15	♂	I-II
4	278	239	127	53·13	132	55·23	179	74·89	52	21·75	14	♀	I-II
5	272	236	123	52·11	132	55·93	176	74·57	51	21·61	15	♂	I-II
6	272	236	122	51·69	132	55·93	176	74·57	52	22·03	15	♂	I-II
7	271	236	122	51·69	130	55·08	175	74·15	52	22·03	14	♂	I-II
8	270	237	119	50·21	129	54·43	175	73·83	50	21·09	15	♂	I-II
9	270	236	122	51·69	128	54·23	175	74·15	50	21·14	17	♂	I-II
10	270	235	120	51·06	128	54·46	179	76·17	50	21·27	15	♀	I-II
11	270	230	116	50·43	125	54·34	173	75·21	50	21·73	16	♂	I-II
12	269	232	121	52·15	126	54·31	172	74·13	51	21·98	13	♂	I-II
13	268	233	122	52·36	132	56·65	176	75·53	52	22·31	15	♀	I-II
14	268	232	121	52·15	129	55·60	172	74·13	55	23·70	15	♂	I-II
15	268	232	122	52·58	128	55·17	172	74·13	52	22·41	14	♀	I-II
16	267	233	120	51·50	128	54·93	174	74·68	52	22·31	14	♂	I-II
17	267	233	119	51·07	129	55·36	175	75·10	48	20·60	14	♂	I-II
18	267	233	118	50·64	127	54·50	170	72·96	51	21·88	14	?	I-II
19	267	230	121	52·60	123	53·43	171	74·34	49	21·30	15	♂	I-II
20	267	229	122	53·27	129	56·33	174	75·98	49	21·39	15	♂	I-II
21	266	232	121	52·15	127	54·74	177	76·29	50	21·55	16	♂	I-II
22	266	230	122	53·04	127	55·21	173	75·21	49	21·30	14	♂	I-II
23	265	227	122	53·74	124	54·62	168	74·00	47	20·70	16	♂	I-II
24	264	228	121	53·07	122	53·50	174	76·31	50	21·92	16	♂	I-II
25	264	227	118	51·98	124	54·62	167	73·56	45	19·82	15	♂	I-II
26	264	227	118	51·98	126	55·50	160	70·48	50	22·02	16	♀	I-II
27	263	230	120	52·13	127	55·21	173	75·21	48	20·87	15	♀	I-II
28	263	230	118	51·30	128	55·65	172	74·78	49	21·30	14	♂	I-II
29	262	226	118	52·21	125	55·30	167	73·84	51	22·56	14	♂	I-II
30	262	225	118	52·44	121	53·77	171	76·00	49	21·77	17	♀	I-II
31	261	228	123	53·94	129	56·57	170	74·56	52	22·80	15	♂	I-II
32	261	227	120	52·86	126	55·50	168	74·00	49	21·58	15	♀	I-II
33	261	227	116	51·10	122	53·74	167	73·56	50	22·02	15	♀	I-II
34	261	226	121	53·53	124	54·86	168	74·33	50	22·12	15	♀	I-II
35	261	224	114	50·89	122	54·46	163	72·76	48	21·42	14	♀	I-II
36	260	228	118	51·75	129	56·57	170	74·56	50	21·92	16	♂	I-II
37	259	226	117	51·77	117	51·77	162	71·68	48	21·23	15	♂	I-II
38	259	226	117	51·77	123	54·42	169	74·77	48	21·23	15	♀	I-II
39	259	225	117	52·00	124	55·11	168	74·66	47	20·88	16	♂	I-II
40	259	223	115	51·56	123	55·15	169	75·78	50	22·42	14	♀	I-II
41	258	223	115	51·56	121	54·26	162	72·64	46	20·62	14	♂	I-II
42	257	222	114	51·35	119	53·60	162	72·97	49	22·07	14	♂	I-II
43	255	224	113	50·44	122	54·46	168	75·00	47	20·98	15	♀	I-II
44	255	224	114	50·89	122	54·46	163	72·76	48	21·42	16	♀	I-II
45	255	220	114	51·81	122	55·45	163	74·09	47	21·36	14	♂	I-II
46	254	223	112	50·22	120	53·81	161	72·19	49	21·97	15	♀	I-II
47	254	222	114	51·35	117	52·70	163	73·42	44	19·82	12	♀	I-II
48	254	221	112	50·67	118	53·39	161	72·85	45	20·36	15	♂	I-II
49	254	221	115	52·03	123	55·65	167	75·56	47	21·26	14	♀	I-II
50	254	219	114	52·05	120	54·79	167	76·25	50	22·83	14	♂	I-II
51	254	217	110	50·69	119	54·83	162	74·65	44	20·27	14	♀	I-II
52	253	221	116	52·48	120	54·29	159	71·94	51	23·07	14	♀	I-II
53	253	220	113	51·36	120	54·54	160	72·72	48	21·81	14	♀	I-II

Table III—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
54	253	220	111	50·45	120	54·54	163	74·09	54	24·54	14	♀	I-II
55	253	217	113	52·07	123	56·68	161	74·19	48	22·12	14	♂♀	I-II
56	253	217	110	50·69	115	52·99	162	74·65	49	22·58	15	♂♀	I-II
57	252	222	115	51·80	122	54·99	167	75·22	49	22·07	15	?	I-II
58	252	221	114	51·58	122	55·20	162	73·30	48	21·71	14	♂♂♀	I-II
59	252	221	111	50·22	120	54·29	161	72·85	44	19·91	15	♂♂♀	I-II
60	252	220	112	50·91	118	53·63	164	74·54	47	21·36	16	♂♂♀	I-II
61	252	218	111	50·91	122	55·96	163	74·77	48	22·01	14	♂♂♀♀	I-II
62	252	217	112	51·61	125	57·60	163	75·06	48	22·12	15	♂♂♀♀	I-II
63	252	216	111	51·38	121	56·01	163	75·46	47	21·75	15	♂♂♀♀	I-II
64	251	222	113	50·90	120	54·05	163	73·42	47	21·17	14	♂♂♀♀	I-II
65	251	218	110	50·45	118	54·12	163	74·77	49	22·47	15	♂♂♀♀	I-II
66	251	217	115	52·99	118	54·37	164	75·57	47	21·65	16	♂♂♀♀	I-II
67	250	224	117	52·23	120	53·57	166	74·10	48	21·42	15	♂♂♀♀	I-II
68	250	222	115	51·80	125	56·30	165	74·32	47	21·17	15	♂♂♀♀	I-II
69	250	220	115	52·27	120	54·54	165	75·00	49	22·27	16	♂♂♀♀	I-II
70	250	220	108	49·09	115	52·27	163	74·09	45	20·45	15	♂♂♂♀	I-II
71	250	219	114	52·05	120	54·79	162	73·97	48	21·91	15	♂♂♂♀	I-II
72	250	219	112	51·14	118	53·88	159	72·60	45	20·54	14	♂♂♂♀	I-II
73	250	216	112	51·85	123	56·94	165	76·38	45	20·83	16	♂♂♂♀	I-II
74	250	212	108	50·94	117	55·18	157	74·05	46	21·69	13	♂♂♂♀	I-II
75	249	219	111	50·68	118	53·88	160	73·08	48	21·91	14	♂♂♂♀	I-II
76	249	215	111	51·62	116	53·95	159	73·95	49	22·79	17	♂♂♂♀	I-II
77	249	214	113	52·80	120	56·07	160	74·76	48	22·43	15	♂♂♂♀	I-II
78	248	217	113	52·07	117	53·91	162	74·65	47	21·65	14	♂♂♀♀	I-II
79	248	215	111	51·62	119	55·34	161	74·88	45	20·93	16	♂♂♀♀	I-II
80	248	215	110	51·16	113	52·55	159	73·95	45	20·93	15	♂♂♀♀	I-II
81	248	214	111	51·87	119	55·60	159	74·29	50	23·36	14	♂♂♀♀	I-II
82	248	214	110	51·40	117	54·67	160	74·76	45	21·02	15	♂♂♀♀	I-II
83	248	212	109	51·41	122	57·54	160	75·47	47	22·17	15	♂♂♀♀	I-II
84	247	216	116	53·70	119	55·09	162	75·00	48	22·22	15	♂♂♀♀	I-II
85	247	216	111	51·38	115	53·24	157	72·68	47	21·75	15	♂♂♀♀	I-II
86	247	216	108	50·00	116	53·70	158	73·14	46	21·29	14	♂♂♀♀	I-II
87	247	215	112	52·09	117	54·41	160	74·41	46	21·39	16	♂♂♀♀	I-II
88	247	215	106	49·30	113	52·55	158	73·48	46	21·39	15	♂♂♀♀	I-II
89	247	213	111	52·11	116	54·46	160	75·11	47	22·06	14	♂♂♀♀	I-II
90	246	219	112	51·14	118	53·88	164	74·88	49	22·37	15	♂♂♀♀	I-II
91	246	213	112	52·58	118	55·39	156	73·24	46	21·59	15	♂♂♀♀	I-II
92	246	213	106	49·48	117	54·92	158	74·17	45	21·12	14	♂♂♀♀	I-II
93	246	213	110	51·64	119	55·86	159	74·64	47	22·06	15	♂♂♀♀	I-II
94	246	212	111	52·35	118	55·66	156	73·58	46	21·69	14	♂♂♀♀	I-II
95	245	219	113	51·59	118	53·88	160	73·08	45	20·54	15	♂♂♀♀	I-II
96	245	216	109	50·46	119	55·09	155	71·76	48	22·22	—	♂♂♀♀	I-II
97	245	213	112	52·58	115	53·99	154	72·30	46	21·59	16	♂♂♀♀	I-II
98	245	212	109	51·41	115	54·24	158	74·52	46	21·69	15	♂♂♀♀	I-II
99	245	211	110	52·13	121	57·34	158	74·88	48	22·74	14	♂♂♀♀	I-II
100	245	211	109	51·65	116	54·97	158	74·88	43	20·38	14	♂♂♀♀	I-II
101	245	210	111	52·85	116	55·24	159	75·71	49	23·33	15	♂♂♀♀	I-II
102	244	215	109	50·69	115	53·48	153	71·16	45	20·93	14	♂♂♀♀	I-II
103	244	211	108	51·18	114	54·03	160	75·83	45	21·32	15	♂♂♀♀	I-II
104	243	213	110	51·64	114	53·52	156	73·24	46	21·59	14	♂♂♀♀	I-II
105	243	211	107	50·71	115	54·50	154	72·98	46	21·80	14	♂♂♀♀	I-II
106	243	210	107	50·95	116	55·24	155	73·81	44	20·95	13	♂♂♀♀	I-II
107	242	213	110	51·64	114	53·52	157	73·70	47	22·06	14	♂♂♀♀	I-II
108	242	211	110	52·13	114	54·03	150	71·09	46	21·80	14	♂♂♀♀	I-II
109	242	211	109	51·65	119	56·39	159	75·35	44	20·85	14	♂♀?	I-II

Table III—*Continued.*

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
110	242	211	107	50·71	114	54·03	154	72·98	45	21·32	14	♀♂♂♂♂♂♀♂♂♀♂♂♀♂♂♀♂♂♂♂	I-II
111	242	210	105	50·00	116	55·24	154	73·33	45	21·43	14		I-II
112	242	209	110	52·63	116	55·50	154	73·68	46	22·01	14		I-II
113	242	209	109	52·15	114	54·54	155	74·16	44	21·05	14		I-II
114	242	209	107	51·19	118	56·45	158	75·59	44	21·05	15		I-II
115	242	208	108	51·92	117	56·25	156	75·00	49	23·55	14		I-II
116	241	209	107	51·19	116	55·50	153	73·20	46	22·01	14		I-II
117	241	208	109	52·40	114	54·80	150	72·11	48	23·07	15		I-II
118	240	210	107	50·95	112	53·33	154	73·33	44	20·95	15		I-II
119	240	209	106	50·71	113	54·06	156	74·64	45	21·53	15		I-II
120	240	208	110	52·88	117	56·25	150	72·11	44	21·15	14		I-II
121	240	207	107	51·69	114	55·07	150	72·46	44	21·25	14		I-II
122	239	207	104	50·24	114	55·07	154	74·39	45	21·73	13		I-II
123	239	205	104	50·73	111	54·14	155	75·61	43	20·97	15		I-II
124	237	206	107	51·94	114	55·34	153	74·27	46	22·33	15		I-II
125	237	206	103	50·00	111	53·88	152	73·78	42	20·38	16		I-II
126	235	201	106	52·73	113	56·21	150	74·62	45	22·38	14		I-II
127	233	204	106	51·96	107	52·45	149	73·04	44	21·56	16		I-II
128	231	198	101	51·01	106	53·53	146	73·73	43	21·71	14		I-II

(2) Received June 13th, 1914.

[From this onward T is measured to the end of the upper lobe, and A to the beginning of the anal fin.]

1	274	237	125	52·75	131	55·27	183	77·21	53	22·36	16	♀♂♂♂♂♂♂♂♂♂♂♂♂♂♂♂♂	II
2	271	232	128	55·17	129	55·60	180	77·58	55	23·70	14		I-II
3	267	231	124	53·68	128	55·45	180	77·92	51	22·07	15		II
4	264	230	121	52·60	124	53·95	175	76·09	55	23·91	14		III
5	263	228	121	53·07	128	56·14	177	77·63	49	21·49	—		II
6	259	223	117	52·46	125	56·05	174	78·02	50	22·42	14		I-II
7	258	222	117	52·70	125	56·30	173	77·92	47	21·17	14		I-II
8	258	221	116	52·48	119	53·84	166	75·11	49	22·17	15		II
9	254	221	114	51·58	121	54·75	167	75·56	48	21·71	14		? II
10	254	221	114	51·58	121	54·75	172	77·82	49	22·17	14		II
11	254	221	116	52·48	122	55·20	172	77·82	50	22·62	14		II
12	253	219	117	53·42	122	55·70	169	77·16	49	22·37	14		II
13	252	221	115	52·03	119	53·84	170	76·92	48	21·71	15		II
14	252	220	117	53·17	120	54·54	166	75·45	49	22·27	16		II
15	252	220	114	51·81	122	55·45	170	77·27	48	21·81	16		II
16	252	217	111	51·15	124	57·14	170	78·34	47	21·65	15		II
17	251	216	113	52·31	118	54·63	165	76·38	49	22·68	15		II
18	251	216	113	52·31	116	53·70	166	76·85	48	22·22	14		II
19	250	217	112	51·61	122	56·22	168	77·41	47	21·65	14		II
20	250	216	114	52·77	119	55·09	167	77·31	46	21·29	15		II
21	250	216	113	52·31	119	55·09	164	75·92	50	23·14	14		II
22	249	223	114	51·12	123	55·15	173	77·57	48	21·52	16		II
23	249	218	112	51·37	117	53·67	167	76·60	47	21·56	—		II
24	249	218	115	52·75	118	54·12	165	75·68	50	22·93	15		? II
25	249	217	116	53·45	119	54·83	166	76·49	47	21·65	14		II
26	249	217	115	52·99	121	55·76	166	76·49	48	22·12	13		II
27	249	217	110	50·69	119	54·83	164	75·57	46	21·19	14		II
28	249	216	114	52·77	119	55·09	166	76·85	45	20·83	14		II
29	248	216	112	51·85	115	53·24	163	75·46	46	21·29	16		II

Table III—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
30	248	216	111	51·38	114	52·77	164	75·92	45	20·83	14	♀	II
31	248	213	116	54·46	120	56·33	164	76·99	50	23·47	15	♀	II
32	247	215	110	51·16	115	53·48	163	75·81	47	21·86	—	♀	II
33	247	214	113	52·33	118	55·14	164	76·63	46	21·49	14	♂♀	II
34	246	214	109	50·93	114	53·27	168	78·50	46	21·49	14	♂♀	IV
35	246	213	112	52·58	118	55·39	165	77·46	48	22·53	14	♀	II
36	246	212	110	51·88	116	54·71	162	76·41	49	23·11	14	♀	II
37	245	215	111	51·62	117	54·41	164	76·27	46	21·39	14	♂	II
38	245	212	109	51·41	117	55·18	164	77·35	45	21·22	15	♂	II
39	245	212	109	51·41	114	53·77	166	78·30	46	21·69	14	♂	II
40	244	213	110	51·64	116	54·46	164	76·99	49	23·00	14	♂	II
41	244	210	110	52·38	115	54·76	161	76·66	46	21·90	15	♂	II
42	243	216	109	50·46	114	52·77	162	75·00	47	21·75	14	♂	II
43	243	214	115	53·73	118	55·14	165	77·10	49	22·89	14	♂	II
44	243	213	111	52·11	118	55·39	165	77·46	47	22·06	15	♂	II
45	243	212	112	52·83	114	53·77	164	77·35	48	22·64	14	♂	II
46	243	212	109	51·41	115	54·24	159	75·00	47	22·17	14	♂♀	II
47	243	212	109	51·41	115	54·24	160	75·47	45	21·22	16	♂	II
48	243	211	106	50·23	114	54·02	161	76·30	45	21·32	14	♂♀	II
49	243	210	110	52·38	114	54·28	161	76·66	48	22·85	13	♂	II
50	243	210	111	52·85	117	55·71	161	76·66	49	23·33	14	♂	II
51	242	211	110	52·13	116	54·97	162	76·77	45	21·32	14	♂	II
52	242	209	111	53·11	116	55·02	165	78·94	47	22·48	14	♂	II
53	242	207	106	51·20	113	54·59	160	77·29	47	22·70	15	♂	II
54	242	207	107	51·69	114	55·07	157	75·84	45	21·73	15	♂	I
55	242	207	110	53·14	115	55·55	162	78·26	46	22·22	14	♂	I
56	242	207	108	52·16	110	53·14	157	75·84	48	23·18	15	♂	II
57	241	211	111	52·60	115	54·50	164	77·72	47	22·27	16	♂	II
58	241	210	107	50·95	112	53·33	158	75·23	46	21·90	15	♂	II
59	241	209	109	52·15	116	55·02	160	76·55	48	22·96	14	♂	II
60	241	208	108	51·92	115	55·28	163	78·36	45	21·63	13	♂	I
61	240	209	110	52·63	114	54·54	160	76·55	47	22·48	14	♂	II
62	240	209	110	52·63	114	54·54	159	76·07	48	22·96	13	?	? I
63	240	208	109	52·40	112	53·84	158	75·96	45	21·63	14	♂	II
64	240	206	105	50·97	109	52·91	157	76·21	44	21·36	14	♂	II
65	239	209	108	51·67	113	54·06	157	75·12	45	21·53	14	♂	II
66	239	208	106	50·96	112	53·84	161	77·40	49	23·55	14	♂	II
67	239	208	106	50·96	112	53·84	160	76·92	45	21·63	16	♂	II
68	239	207	107	51·69	116	56·03	157	75·84	47	22·70	15	♂	I
69	239	207	106	51·20	114	55·07	156	75·36	46	22·22	14	♂	I
70	238	208	106	50·96	115	55·28	161	77·40	44	21·15	14	♂	II
71	237	206	109	52·91	111	53·88	159	77·18	48	23·30	16	♂	II
72	237	206	105	50·97	114	55·34	159	77·18	45	21·84	—	♂	II
73	237	204	107	52·45	113	53·39	155	75·98	45	22·05	15	♂	II
74	236	203	108	53·20	112	55·17	158	77·83	47	23·15	14	♂	I
75	235	204	107	52·45	110	53·92	159	77·94	47	23·03	14	♂	II
76	235	202	105	50·98	110	54·45	157	77·72	47	23·26	14	♂	II
77	234	205	107	52·19	111	54·14	153	74·63	44	21·46	14	♂	I
78	234	205	105	51·22	109	53·17	153	74·63	48	23·41	14	♂	II
79	234	203	106	52·21	109	53·69	155	76·35	44	21·67	14	♂	II
80	234	202	108	53·46	112	55·44	156	77·22	44	21·78	14	♂	II
81	234	199	104	52·26	111	55·77	155	77·88	45	22·61	14	♂	II
82	233	204	107	52·45	114	55·88	157	76·96	45	22·05	15	♂	II
83	233	204	106	51·96	113	55·39	157	76·96	45	22·05	15	♂	I
84	233	200	106	53·00	110	55·00	156	78·00	46	23·00	15	♂	II
85	232	209	106	50·71	111	53·11	159	76·07	45	21·53	13	♂	II

Table III—*Continued.*

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
86	232	200	105	52·50	113	56·50	156	78·00	46	23·00	14	♂♀	II
87	232	200	106	53·00	108	54·00	155	77·50	45	22·50	14	♂	II
88	230	202	107	52·97	114	56·43	157	77·72	45	22·27	15	♂	II
89	230	201	103	51·24	112	55·72	156	77·61	44	21·88	14	♂	II
90	230	201	105	52·23	109	54·22	149	74·12	44	21·88	15	♂	II
91	230	200	105	52·50	111	55·50	152	76·00	44	22·00	14	♂	II
92	228	198	100	50·50	107	54·04	150	75·75	44	22·22	14	♂	II
93	228	198	103	52·02	111	56·06	152	76·76	46	23·23	14	♂	II
94	227	197	104	52·79	107	54·31	147	74·61	45	22·84	14	♂	II
95	226	194	102	52·57	106	54·63	147	75·77	44	22·68	14	♂	II
96	225	196	109	55·61	111	56·63	151	77·04	45	22·95	16	♂♀	II
97	225	196	101	51·53	108	55·10	151	77·04	44	22·44	14	♂♀	II
98	225	194	103	53·09	108	55·67	148	76·28	43	22·16	14	♂	II
99	220	190	98	51·57	104	54·73	146	76·84	43	22·63	16	♂	II
100	214	188	99	52·66	102	54·25	144	76·59	42	22·34	14	♂♀	II

(3) Received June 25th, 1914.

1	272	230	123	53·47	127	55·21	177	76·95	52	22·61	14	♂	III
2	270	232	123	53·01	128	55·17	171	73·70	51	21·98	14	♂♀	III
3	270	230	121	52·60	128	55·65	182	79·13	52	22·61	17	♀	IV
4	269	230	121	52·60	131	56·95	179	77·82	51	22·17	14	♀	III
5	269	228	117	51·31	125	54·82	179	78·50	50	21·93	14	♀	III
6	267	230	122	53·04	130	56·52	179	77·82	50	21·73	14	♂♀	IV
7	266	230	119	51·73	129	56·08	181	78·69	51	22·17	15	♂♀	IV
8	263	227	118	51·98	124	54·62	175	77·09	50	22·02	15	♂♀	III
9	262	226	117	51·77	127	56·19	175	77·43	52	22·90	15	♂♀	IV
10	261	225	119	52·88	125	55·55	172	76·44	48	21·33	14	♂♀	III
11	261	223	114	51·12	123	55·15	171	76·68	49	21·97	15	♂♀	III
12	258	223	115	51·56	125	56·05	172	77·13	47	21·07	13	♂♀	III
13	257	223	117	52·46	122	54·70	174	78·02	49	21·97	15	♀	II
14	256	221	115	52·03	120	54·29	164	74·20	48	21·71	14	♂♀	III
15	255	218	117	53·67	121	55·50	170	77·98	50	22·93	14	♂♀	III
16	254	221	116	52·48	118	53·39	170	76·92	52	23·57	16	♂♀	III
17	254	220	115	52·27	120	54·54	166	75·45	49	22·27	15	♂♀	III
18	252	217	116	53·45	120	55·29	168	77·41	49	22·58	15	♂♀	III
19	252	216	112	51·85	121	56·01	166	76·85	48	22·22	14	♂♀	III
20	252	215	115	53·48	124	57·67	170	79·07	49	22·79	15	♂♀	IV
21	252	214	112	52·33	115	53·73	166	77·57	46	21·49	15	♂♀	III
22	251	222	115	51·80	121	54·50	172	77·47	48	21·62	16	♂♀	IV
23	249	216	112	51·85	121	56·01	161	74·53	47	21·75	14	♂♀	III
24	249	215	112	52·09	120	55·81	168	78·14	46	21·39	15	♂♀	III
25	248	217	112	51·61	120	55·29	169	77·88	46	21·19	14	♂♀	III
26	248	213	107	50·23	119	55·86	162	76·05	48	22·53	14	♂♀	III
27	247	216	112	51·85	116	53·70	166	76·85	45	20·83	16	♂♀	III
28	247	214	107	50·00	117	54·67	162	75·70	44	20·56	14	♂♀	III
29	247	213	111	52·11	118	55·39	166	77·93	45	21·12	16	♂♀	III
30	247	213	110	51·64	115	53·99	162	76·05	45	21·12	16	♂♀	III
31	247	211	112	53·08	116	54·97	163	77·25	45	21·32	14	♂♀	III
32	246	213	113	53·05	118	55·39	163	76·52	47	22·06	14	♂♀	III
33	246	213	111	52·11	116	54·46	168	78·87	44	20·65	16	♂♀	III
34	246	212	110	51·88	117	55·18	162	76·41	46	21·69	15	♂♀	IV
35	246	212	109	51·41	115	54·24	164	77·35	47	22·17	15	♂	III
36	246	210	111	52·85	117	55·71	161	76·66	46	21·90	15	♂	III

Table III—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
37	245	215	110	51·16	115	53·48	160	74·41	46	21·39	15	♂	III
38	245	213	112	52·58	116	54·46	164	76·99	47	22·06	15	♂♀	III
39	245	212	110	51·88	116	54·71	162	76·41	47	22·17	14	♀	III
40	245	212	110	51·88	117	55·18	161	75·94	44	20·75	13	♀	III
41	245	210	109	51·90	113	53·81	165	78·57	45	21·43	15	♀	III
42	244	212	110	51·88	116	54·71	161	75·94	48	22·64	15	♂♀	III
43	244	212	111	52·35	115	54·24	163	76·88	47	22·17	15	♂♀	III
44	244	212	110	51·88	119	56·13	164	77·35	48	22·64	15	♂♀	III
45	244	211	109	51·65	116	54·97	164	77·72	46	21·80	15	♀	III
46	244	210	109	51·90	115	54·76	161	76·66	45	21·43	14	♀	III
47	244	209	107	51·19	113	54·06	158	75·59	44	21·05	15	♀	III
48	243	211	109	51·65	119	56·39	164	77·72	46	21·80	14	♂♀	III
49	243	210	108	51·42	113	53·81	159	75·71	45	21·43	15	♂♀	IV
50	242	212	110	51·88	114	53·77	160	75·47	44	20·75	15	♀	III
51	242	210	110	52·38	116	55·24	163	77·61	47	22·38	14	♀	III
52	242	210	106	50·47	115	54·76	163	77·61	45	21·43	15	♀	III
53	242	208	107	51·44	110	52·88	159	76·44	46	22·11	14	♀	I
54	242	207	109	52·65	115	55·55	159	76·81	44	21·25	15	♂♀	III
55	242	206	106	51·45	116	56·31	162	78·64	44	21·36	15	♂♀	III
56	241	208	106	50·96	112	53·84	158	75·96	45	21·63	14	♀	III
57	241	207	108	52·16	114	55·07	160	77·29	42	20·29	15	♀	III
58	241	206	109	52·91	114	55·74	161	78·15	45	21·84	15	♀	III
59	240	207	110	53·14	114	55·07	159	76·81	42	20·29	14	♂♀	III
60	240	207	107	51·69	116	56·03	156	75·36	45	21·73	14	♂♀	II
61	240	206	109	52·91	113	54·85	161	78·15	45	21·84	15	♀	III
62	240	205	106	51·70	111	54·14	159	77·56	44	21·46	15	♀	IV
63	240	204	107	52·45	111	54·41	160	78·43	46	22·54	14	♀	III
64	239	206	108	52·42	115	55·82	158	76·69	46	22·33	14	♀	III
65	239	205	109	53·17	112	54·63	161	78·53	45	21·95	15	♀	III
66	239	204	106	51·96	114	55·88	158	77·45	44	21·56	16	♂♀	III
67	238	205	104	50·73	109	53·17	156	76·09	45	21·95	15	♂♀	IV
68	238	204	109	53·43	114	55·88	158	77·45	45	22·05	13	♂♀	III
69	237	205	108	52·68	115	56·09	157	76·58	46	22·43	13	♂♀	III
70	237	205	104	50·73	110	53·65	159	77·56	45	21·95	16	♂♀	III
71	235	204	106	51·96	110	53·92	157	76·96	46	22·54	15	♂♀	IV
72	235	203	104	51·23	114	56·15	158	77·83	44	21·67	14	♀	III
73	235	202	104	51·48	109	53·96	152	75·24	43	21·27	14	♀	III
74	235	201	103	51·24	108	53·73	152	75·62	46	22·88	13	♀	III
75	234	202	105	50·98	115	56·93	158	78·21	42	20·79	15	♂♀	III
76	234	201	105	52·23	111	55·22	156	77·61	45	22·38	15	♂♀	III
77	233	203	103	50·73	109	53·69	154	75·86	43	21·18	14	♂♀	III
78	233	202	105	51·98	107	52·97	153	75·74	47	23·26	16	♂	III
79	232	199	106	53·26	112	56·28	152	76·38	44	22·11	15	♂♀	II
80	231	199	105	52·76	110	55·27	153	76·88	45	22·61	15	♀	III
81	231	199	105	52·76	114	57·28	154	77·38	45	22·61	14	♀	III
82	231	199	103	51·75	111	55·77	152	76·38	43	21·60	14	♀	IV
83	230	199	104	52·26	108	54·27	152	76·38	44	22·11	15	♂♀	III
84	230	197	100	50·76	108	54·82	152	77·15	44	22·33	14	♂♀	II
85	230	196	102	52·04	109	55·61	147	75·00	41	20·91	14	♂♀	III
86	229	198	102	51·51	110	55·55	151	76·26	44	22·22	14	♀	III
87	229	198	104	52·52	110	55·55	155	78·28	44	22·22	15	♂♀	III
88	228	198	107	54·04	109	55·05	150	75·75	45	22·72	14	♂	III
89	227	197	102	51·77	110	55·83	153	77·66	44	22·33	15	♂♀	III
90	226	197	102	51·77	107	54·31	151	76·65	43	21·82	15	♂	III
91	226	193	103	53·36	107	55·43	150	77·72	44	22·79	17	♂♀	III
92	225	192	100	52·08	107	55·72	150	78·12	43	22·39	14	♀	II

Table III—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
93	224	193	101	52·33	105	54·40	143	74·09	43	22·28	13	♂	III
94	223	193	99	51·29	104	53·88	148	76·68	42	21·76	14	♂♂	III
95	220	191	97	50·78	106	55·49	148	77·48	42	21·99	14	♂♀	III
96	219	188	98	52·12	106	56·27	140	74·46	42	22·34	13	♀	II
97	217	187	96	51·33	100	53·47	141	75·40	41	21·92	15	♂	III
98	215	187	96	51·33	102	54·54	144	77·00	40	21·38	14	♂♂	II
99	214	183	95	51·91	102	55·73	140	76·50	40	21·85	13	♂♂	III
100	207	177	92	51·97	99	55·93	134	75·70	40	22·59	15	♂	III
						(4) Received July 9th, 1914.							
1	265	230	117	50·87	129	56·08	182	79·13	49	21·30	16	♂♀	III
2	265	228	122	53·50	125	54·82	177	77·63	53	23·24	14	♀♀	IV
3	265	227	121	53·30	125	55·06	171	75·33	49	21·58	15	♀♀	IV
4	264	225	120	53·33	125	55·55	175	77·77	50	22·22	14	♀♀	IV
5	263	227	121	53·30	125	55·06	175	77·09	52	22·90	14	♀♀	IV
6	263	225	117	52·00	125	55·55	173	76·88	48	21·33	13	♀	V
7	260	226	119	52·65	125	55·31	175	77·43	50	22·12	15	♂	V
8	260	224	114	50·89	123	54·91	175	78·12	53	23·66	15	♂♀	V
9	260	221	118	53·39	121	54·75	173	78·28	53	23·98	15	♂♀	IV
10	258	225	110	48·88	125	55·55	170	75·55	50	22·22	14	♀	IV
11	258	222	120	54·05	120	54·05	173	77·92	50	22·51	14	?	? II
12	258	221	117	52·94	127	57·46	173	78·28	51	23·07	15	♂	V
13	257	222	115	51·80	123	55·40	171	77·02	49	22·07	13	♀	V
14	255	220	115	52·27	122	55·45	172	78·18	48	21·81	14	♂♀	IV
15	254	221	115	52·03	119	53·84	170	76·92	48	21·71	15	♀	IV
16	253	220	114	51·81	122	55·45	169	76·81	49	22·27	14	♂♀	IV
17	253	220	114	51·81	126	57·27	173	78·63	46	20·90	14	♂♀	IV
18	253	218	116	53·21	118	54·12	169	77·52	47	21·56	14	♂	III
19	252	222	111	50·00	121	54·50	170	76·57	46	20·72	15	♂♂	V
20	252	220	116	52·72	122	55·45	165	75·00	46	20·90	15	♂♀	IV
21	252	219	113	51·59	118	53·88	166	75·79	50	22·83	14	♂	III
22	252	215	112	52·09	119	55·34	165	76·74	47	21·86	14	?	? II
23	251	219	113	51·59	118	53·88	166	75·79	49	22·37	14	♀	III
24	251	218	113	51·83	121	55·50	168	77·06	49	22·47	14	♂♂	V
25	251	216	113	52·31	116	53·70	166	76·85	47	21·75	14	♂♀	V
26	250	218	111	50·91	115	52·75	167	76·60	45	20·64	15	♀	IV
27	250	217	113	52·07	117	53·91	167	76·95	48	22·12	13	♂♂	V
28	250	216	119	55·09	122	56·52	167	77·31	50	23·14	15	♂♂	IV
29	250	216	116	53·70	125	57·87	167	77·31	49	22·68	13	♂♂	V
30	249	216	115	53·24	119	55·09	168	77·41	50	23·14	14	♂♀	IV
31	249	214	111	51·87	119	55·60	168	78·50	47	21·96	14	♂	IV
32	249	214	109	50·93	112	52·33	160	74·76	43	20·09	14	♀	IV
33	247	216	111	51·38	119	55·09	164	75·92	49	22·68	14	♀	IV
34	247	215	110	51·16	121	56·27	166	77·20	48	22·32	15	♂	IV
35	246	215	112	52·09	116	53·95	165	76·74	47	21·86	15	♂♂	III
36	246	214	109	50·93	119	55·60	166	77·57	47	21·96	14	♂♂	IV
37	246	211	108	51·18	119	56·39	160	75·83	47	22·27	14	♂♂	III
38	245	215	148	50·23	115	53·48	161	74·88	47	21·86	14	♂♂	IV
39	245	212	109	51·41	115	54·24	165	77·83	49	23·11	15	♂♂	III
40	245	212	108	50·94	117	55·18	162	76·41	48	22·64	16	♂♂	V
41	245	212	107	50·47	115	54·24	159	75·00	48	22·64	15	♂♂	IV
42	245	210	111	52·85	120	57·14	166	79·04	47	22·38	13	♂♂	V
43	245	209	109	52·15	115	55·02	160	76·55	46	22·01	14	♂	V

Table III—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
44	244	211	110	52·13	116	54·97	163	77·25	47	22·27	14	♀	IV
45	243	214	111	51·87	121	56·54	164	76·63	48	22·43	14	♀	IV
46	243	212	111	52·35	115	54·24	162	76·41	48	22·64	14	♀	IV
47	243	212	109	51·41	114	53·77	162	76·41	48	22·64	16	♀	IV
48	243	209	110	52·63	117	55·98	167	79·90	47	22·48	15	♀	IV
49	243	208	108	51·92	116	55·76	164	78·84	47	22·59	14	♀	IV
50	243	207	106	51·20	115	55·55	162	78·26	45	21·73	13	♀	IV
51	242	212	110	51·88	116	54·71	164	77·35	48	22·64	14	♂♀	III
52	242	212	109	51·41	114	53·77	162	76·41	47	22·17	16	♂♀	V
53	242	210	107	50·95	114	54·28	159	75·71	44	20·95	13	♂♂	V
54	241	212	112	52·83	113	53·30	159	75·00	46	21·69	14	♂♂	V
55	241	211	108	51·18	113	53·55	162	76·77	46	21·80	13	♂♂	V
56	241	209	110	52·63	115	55·02	163	77·99	45	21·53	13	♂♀	III
57	240	211	109	51·65	110	52·13	163	77·25	43	20·38	15	?	? II
58	240	207	108	52·16	112	54·10	160	77·29	44	21·25	17	♂	V
59	240	206	108	52·42	116	56·31	160	77·67	48	23·30	16	♂♀	IV
60	240	206	106	51·45	118	57·28	165	80·09	45	21·84	16	♂♀	IV
61	240	205	108	52·68	115	56·09	161	78·53	45	21·95	16	♂♀	V
62	240	204	105	51·47	109	53·43	156	76·47	45	22·05	15	♂♀	IV
63	240	203	108	53·20	112	55·17	158	77·83	47	23·15	16	♂♂	IV
64	239	208	109	52·40	113	54·32	160	76·92	45	21·63	15	♂♂	V
65	239	207	107	51·69	117	56·52	160	77·29	48	23·18	16	♂♀	IV
66	238	209	107	51·19	114	54·54	160	76·55	49	23·44	15	♀	III
67	238	206	102	49·51	111	53·88	157	76·21	46	22·33	15	♀	III
68	237	208	111	53·36	113	54·32	157	75·48	46	22·11	15	♂♂	IV
69	237	205	105	51·22	114	55·61	157	76·58	43	20·97	15	♂♂	IV
70	237	202	107	52·97	110	54·45	157	77·72	45	22·27	14	♂♂	V
71	236	207	106	51·20	113	54·59	162	78·26	45	21·73	14	♂♂	IV
72	236	205	102	49·75	108	52·68	155	75·61	40	19·51	14	♂♂	V
73	236	204	106	51·96	114	55·88	154	75·49	46	22·54	14	♂♂	V
74	235	204	107	52·45	112	54·90	160	78·43	43	21·07	15	♂♂	III
75	235	203	105	51·72	108	53·20	160	78·81	42	20·89	15	♂♂	IV
76	235	201	107	53·23	109	54·22	157	78·10	45	22·38	15	♂♂	IV
77	235	201	104	51·74	110	54·72	153	76·11	44	21·88	14	♂♂	IV
78	235	201	103	51·24	114	56·71	154	76·61	46	22·88	14	♂♂	IV
79	235	200	100	50·00	111	55·50	156	78·00	46	23·00	14	♂♀	IV
80	234	203	103	50·73	110	54·68	153	75·36	46	22·66	15	♂♀	V
81	234	201	102	50·74	111	55·22	154	76·61	44	21·88	15	♂♀	III
82	232	202	105	51·98	111	54·95	156	77·22	44	21·78	15	♂♀	III
83	232	200	98	49·00	108	54·00	152	76·00	45	22·50	14	♂♀	IV
84	232	199	105	52·76	109	54·77	150	75·37	42	21·10	14	♂♀	III
85	231	200	102	51·00	104	52·00	151	75·50	40	20·00	14	♂♂	IV
86	231	199	104	52·26	108	54·27	149	74·87	43	21·60	14	♂♂	V
87	231	199	103	51·75	110	55·27	152	76·38	43	21·60	14	♂♂	V
88	231	199	101	50·75	109	54·77	153	76·88	43	21·60	15	♂♂	IV
89	230	202	103	50·99	108	53·46	151	74·75	46	22·77	16	♂♂	III
90	230	200	104	52·00	108	54·00	152	76·00	45	22·50	14	♂♀	III
91	230	200	103	51·50	106	53·00	149	74·50	42	21·00	14	♂♀	IV
92	230	200	106	53·00	106	53·00	153	76·50	46	23·00	14	♂♀	IV
93	230	196	101	51·53	112	57·14	152	77·55	47	23·97	13	♂♀	IV
94	228	198	101	51·01	112	56·56	154	77·77	46	23·23	16	♀	III
95	227	196	101	51·53	110	56·12	150	76·53	46	23·46	17	♀	IV
96	227	194	103	53·09	104	53·60	148	76·28	42	21·64	13	♂♂	III
97	223	193	101	52·33	106	54·92	145	75·13	44	22·79	14	♂♂	III
98	223	192	101	52·60	109	56·77	148	77·08	41	21·35	16	♂♀	III
99	219	187	100	53·47	109	58·28	150	80·21	43	22·99	14	♂♀	III
100	211	183	93	50·81	97	53·00	137	74·86	40	21·85	14	♂♀	II

Table III –*Continued.*

(5) Received July 31st, 1914.
(In cold storage until September 23rd.)

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
1	263	226	122	53·98	126	55·75	182	80·53	50	22·12	15	♀♀♀♀♀♀♀	III
2	260	226	119	52·65	126	55·75	176	77·87	49	21·68	13	♀♀♀♀♀♀♀	V
3	256	222	117	52·70	120	54·05	176	79·27	51	22·97	14	♀♀♀♀♀♀♀	IV
4	256	220	118	53·63	123	55·91	170	77·27	48	21·81	15	♀♀♀♀♀♀♀	IV
5	255	222	116	52·25	121	54·50	174	78·37	50	22·51	13	♀♀♀♀♀♀♀	III
6	254	220	116	52·72	121	55·00	174	79·09	46	20·90	14	♀♀♀♀♀♀♀	IV
7	253	222	115	51·80	124	55·85	172	77·47	50	22·51	15	♀♀♀♀♀♀♀	V
8	253	219	113	51·59	118	53·88	170	77·62	46	21·00	14	♀♀♀♀♀♂♂	IV
9	253	217	112	51·61	123	56·68	170	78·34	48	22·12	14	♀♀♀♀♀♂♂	IV
10	252	218	112	51·37	121	55·50	171	78·44	45	20·64	15	♀♀♀♀♀♂♂	V
11	250	217	112	51·61	118	54·37	168	77·41	47	21·65	14	♀♀♀♀♀♂♂	IV
12	250	216	114	52·77	117	54·16	170	78·70	50	23·14	15	♀♀♀♀♀♀♀	IV
13	250	216	112	51·85	119	55·09	167	77·31	45	20·83	14	♀♀♀♀♀♀♀	IV
14	250	216	112	51·85	119	55·09	160	74·07	46	21·29	15	♀♀♀♀♀♂♂	III
15	250	216	110	50·92	122	56·48	171	79·16	45	20·83	15	♀♀♀♀♀♂♂	V
16	250	215	113	52·55	122	56·74	168	78·14	46	21·39	14	♀♀♀♀♀♂♂	V
17	248	212	108	50·94	117	55·18	165	77·83	45	21·22	13	♀♀♀♀♀♂♂	III
18	246	214	112	52·33	124	57·94	170	79·43	44	20·56	14	♀♀♀♀♀♂♂	IV
19	246	210	108	51·42	116	55·24	162	77·14	45	21·43	15	♀♀♀♀♀♀♀	IV
20	245	212	111	52·35	115	54·24	161	75·94	46	21·69	14	♀♀♀♀♀♀♀	III
21	244	212	107	50·47	115	54·24	168	79·24	46	21·69	14	♀♀♀♀♀♀♀	IV
22	243	211	115	54·50	117	55·45	160	75·83	48	22·74	16	♀♀♀♀♀♀♀	IV
23	243	210	111	52·85	116	55·24	161	76·66	45	21·43	13	♀♀♀♀♀♀♀	III
24	242	211	108	51·18	116	54·97	158	74·88	47	22·27	14	♀♀♀♀♀♀♀	IV
25	242	209	108	51·67	118	56·45	162	77·51	42	20·09	14	♀♀♀♀♀♀♀	III
26	241	206	107	51·94	115	55·82	163	79·12	45	21·84	13	♀♀♀♀♀♀♀	III
27	240	208	110	52·88	114	54·80	161	77·40	45	21·63	15	♀♀♀♀♀♀♀	IV
28	240	207	106	51·20	112	54·10	162	78·26	45	21·73	14	♀♀♀♀♀♀♀	IV
29	240	206	107	51·94	113	54·85	160	77·67	45	21·84	13	♀♀♀♀♀♂♂	III
30	240	206	108	52·42	114	55·74	161	78·15	47	22·81	14	♀♀♀♀♀♂♂	IV
31	239	207	110	53·14	116	56·03	163	78·74	46	22·22	15	♀♀♀♀♀♂♂	III
32	239	207	108	52·16	113	54·59	160	77·29	43	20·77	14	♀♀♀♀♀♂♂	IV
33	238	208	108	51·92	116	55·76	166	79·80	42	20·19	14	♀♀♀♀♀♂♂	III
34	238	204	106	51·96	114	55·88	160	78·43	41	20·09	14	♀♀♀♀♀♂♂	III
35	237	205	110	53·64	114	55·61	162	79·02	46	22·43	13	♀♀♀♀♀♂♂	IV
36	237	204	106	51·96	116	56·86	162	79·40	45	22·05	14	♀♀♀♀♀♀♀	III
37	237	203	104	51·23	110	54·68	157	77·34	44	21·67	14	♀♀♀♀♀♀♀	IV
38	236	206	108	52·42	113	54·85	160	77·67	42	20·38	14	♀♀♀♀♀♂♂	III
39	236	205	103	50·24	107	52·19	156	76·09	42	20·48	14	♀♀♀♀♀♂♂	IV
40	236	199	105	52·76	110	55·27	158	79·39	42	21·10	14	♀♀♀♀♀♂♂	III
41	235	205	103	50·24	106	51·70	156	76·09	45	21·95	14	♀♀♀♀♀♀♀	II
42	235	202	106	52·47	110	54·45	153	75·74	41	20·29	16	♀♀♀♀♀♂♂	III
43	235	202	104	51·48	109	53·96	154	76·43	45	22·27	14	♀♀♀♀♀♀♀	IV
44	235	200	108	54·00	114	57·00	156	78·00	45	22·50	14	♀♀♀♀♀♂♂	III
45	234	201	111	55·22	117	58·20	163	81·09	45	22·38	14	♀♀♀♀♀♀♀	IV
46	234	197	104	52·79	108	54·82	154	78·17	43	21·82	13	♀♀♀♀♀♂♂	III
47	233	204	106	51·96	112	54·90	158	77·45	41	20·09	15	♀♀♀♀♀♀♀	IV
48	233	202	105	51·98	109	53·96	156	77·22	44	21·78	15	♀♀♀♀♀♂♂	III
49	233	200	105	52·50	111	55·50	152	76·00	45	22·50	15	♀♀♀♀♀♀♀	IV
50	232	202	102	50·49	110	54·45	153	75·74	45	22·27	15	♀♀♀♀♀♀♀	IV
51	232	200	103	51·50	110	55·00	158	79·00	43	21·50	14	♀♀♀♀♀♀♀	III
52	232	200	104	52·00	110	55·00	153	76·50	41	20·50	14	♀♀♀♀♀♀♀	IV
53	231	200	108	54·00	110	55·00	151	75·50	43	21·50	13	♀♀♀♀♀♂♂	IV

Table III—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
54	231	198	104	52·52	109	55·05	154	77·77	43	21·71	13	♂	IV
55	230	202	107	52·97	113	55·94	162	80·19	41	20·29	14	♂♀	IV
56	230	200	106	53·00	109	54·50	151	75·50	46	23·00	13	♂♀	IV
57	230	200	100	50·00	108	54·00	152	76·00	42	21·00	15	♂♀	III
58	230	199	107	53·76	109	54·77	157	78·89	44	22·11	14	♂♀	III
59	229	199	106	53·26	110	55·27	154	77·38	43	21·60	15	♂♀	III
60	229	198	102	51·51	107	54·04	151	76·26	40	20·20	—	♂♀	II
61	229	196	101	51·53	106	54·08	149	76·02	44	22·44	13	♂♀	III
62	228	200	100	50·00	107	53·50	155	77·50	43	21·50	13	♂♀	III
63	228	195	100	51·28	104	53·33	152	77·94	41	21·02	14	♂♀	III
64	227	198	102	51·51	106	53·53	156	78·78	40	20·20	15	♂♀	III
65	227	197	104	52·79	107	54·31	152	77·15	43	21·82	—	♀	II
66	227	196	103	52·54	105	53·57	152	77·55	41	20·91	16	♂♀	III
67	227	196	105	53·57	108	55·10	149	76·02	45	22·95	14	♂♀	III
68	227	195	100	51·28	108	55·38	156	80·00	41	21·02	14	♂♀	III
69	226	196	107	54·59	109	55·61	152	77·55	43	21·93	14	♂♀	IV
70	226	193	102	53·12	106	54·92	150	77·72	40	20·72	15	♂♀	III
71	225	196	101	51·53	106	54·08	152	77·55	43	21·93	14	♂♀	III
72	225	194	100	51·54	104	53·60	151	77·83	40	20·61	14	♂	III
73	224	196	102	52·04	107	54·59	151	77·04	42	21·42	14	♂	III
74	224	194	100	51·54	108	55·67	150	77·31	42	21·64	14	♂	II
75	223	193	99	51·29	105	54·40	151	78·23	43	22·28	15	♂	II
76	223	192	99	51·56	103	53·64	148	77·08	40	20·83	13	♂♀	III
77	222	192	100	52·08	104	54·16	150	78·12	43	22·39	—	♂	III
78	222	190	101	53·15	107	56·31	149	78·42	40	21·05	13	♂♀	III
79	222	189	99	52·38	101	53·43	147	77·77	41	21·69	—	♀	II
80	220	190	101	53·15	107	56·31	151	79·47	40	21·05	14	♂♀	IV
81	219	190	97	51·05	100	52·63	149	78·42	39	20·52	16	♂♀	III
82	218	192	100	52·08	106	55·20	148	77·08	43	22·39	14	♂♀	III
83	218	186	97	52·15	105	56·44	145	77·95	44	23·65	13	♂	III
84	216	187	99	52·94	101	54·01	142	75·93	39	20·85	15	♂	II
85	215	189	99	52·38	102	53·97	145	76·72	39	20·63	14	♂♀	III
86	214	181	96	53·03	99	54·69	139	76·79	43	23·75	14	♂	III
87	212	183	98	53·55	100	54·64	143	78·15	38	20·76	15	♂	III
88	212	183	97	53·00	101	55·19	144	78·69	39	21·31	13	♂	II
89	208	179	95	53·07	99	55·30	141	78·77	40	22·34	—	♂♀	II
90	200	172	91	52·89	96	55·81	133	77·32	39	22·67	13	♂	II
				(6) Received August 21st, 1914.									
				(Cold storage until September 28th.)									
1	270	232	125	53·87	131	56·46	181	78·01	49	21·12	13	♂	IV
2	267	230	121	52·60	130	56·52	186	80·87	51	22·17	13	♂	IV
3	266	229	119	51·96	126	55·02	178	77·72	49	21·39	15	♂♀	IV
4	265	229	123	53·71	127	55·45	177	77·28	50	21·83	14	♂	IV
5	263	230	119	51·73	125	54·34	181	78·69	47	20·43	14	♂♀	IV
6	263	229	119	51·96	124	54·14	177	77·28	48	20·96	14	♂♀	IV
7	263	228	117	51·31	123	53·94	177	77·63	48	21·05	15	♂♀	IV
8	261	227	115	50·66	122	53·74	176	77·53	49	21·58	16	♂♀	IV
9	261	225	117	52·00	123	54·66	176	78·22	46	20·44	14	♂	IV
10	260	226	116	51·32	121	53·53	174	76·99	48	21·23	15	♂♀	IV
11	260	225	119	52·88	124	55·11	178	79·11	47	20·88	14	♂	IV
12	259	224	112	50·00	118	52·67	172	76·78	46	20·53	15	♂	III
13	258	225	117	52·00	123	54·66	174	77·33	49	21·77	14	♂	IV

Table III—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
14	257	221	115	52·03	125	56·56	171	77·37	49	22·17	14	♀	IV
15	256	222	114	51·35	120	54·05	168	75·67	51	23·42	15	♂	III
16	256	220	117	53·17	124	56·36	174	79·09	44	20·00	14	♂	IV
17	255	223	112	50·22	118	52·91	170	76·23	45	20·17	15	♂	III
18	255	220	115	52·27	121	55·00	173	78·63	45	20·45	15	♂	IV
19	255	218	112	51·37	121	55·50	166	76·14	47	21·56	14	♀	III
20	254	222	120	54·05	123	55·40	174	78·37	47	21·17	16	♂	IV
21	254	219	114	52·05	119	54·15	170	77·62	48	21·91	15	♂	IV
22	254	218	115	52·75	122	55·96	173	79·35	44	20·18	14	♂	IV
23	254	218	113	51·83	120	55·04	166	76·14	46	21·10	13	♀	IV
24	253	222	120	54·05	125	56·30	175	78·82	47	21·17	14	♂	IV
25	253	221	116	52·48	123	55·65	171	77·37	47	21·26	14	♀	IV
26	253	221	116	52·48	119	53·84	173	78·28	45	20·36	16	♀	IV
27	253	221	113	51·13	122	55·20	173	78·28	48	21·71	13	♀	IV
28	253	220	114	51·81	119	54·09	169	76·81	45	20·45	14	♂	IV
29	253	219	111	50·68	117	53·42	169	77·16	45	20·54	14	♂	III
30	252	220	116	52·72	121	55·00	168	76·36	48	21·81	15	♂	V
31	252	216	115	53·24	120	55·55	170	78·70	45	20·83	15	♀	IV
32	251	215	110	51·16	116	53·95	163	75·81	46	21·39	14	♀	IV
33	248	215	111	51·62	114	53·02	169	78·60	46	21·39	15	♂	V
34	248	214	113	52·33	118	55·14	168	78·50	45	21·02	14	♀	IV
35	247	217	115	52·99	120	55·29	169	77·88	45	20·73	15	♂	IV
36	246	221	112	50·67	117	52·94	171	77·37	46	20·81	15	♀	IV
37	245	217	112	51·61	119	54·83	166	76·49	49	22·58	14	♀	IV
38	245	213	112	52·58	117	54·93	162	76·05	44	20·65	14	♀	IV
39	244	213	110	51·64	117	54·93	169	79·34	44	20·65	14	♀	IV
40	243	210	109	51·90	114	54·28	162	77·14	46	21·90	14	♀	IV
41	235	217	114	52·53	122	56·22	168	77·41	48	22·12	14	♂	IV
42	233	213	110	51·64	115	53·99	162	76·05	47	22·06	14	♀	III
43	233	203	102	50·24	113	55·66	155	76·35	42	20·89	14	♂	IV
44	230	213	110	51·64	120	56·33	165	77·46	47	22·06	14	♀	IV
45	230	198	101	51·01	107	54·04	150	75·75	41	20·70	14	♂	IV
46	225	191	98	51·30	102	53·40	145	75·91	44	23·03	14	♂	IV
47	222	207	106	51·20	113	54·59	155	74·87	44	21·25	15	♂	IV
48	221	190	99	52·10	102	53·68	146	76·84	39	20·52	14	♂	II
49	217	190	100	52·63	104	54·73	146	76·84	41	21·58	16	♂	III
50	215	203	104	51·23	111	54·68	155	76·35	41	20·19	14	♂	IV

(7) Received September 4th, 1914.)
(Cold storage until October 2nd.)

1	273	233	121	51·93	129	55·36	182	78·11	51	21·88	14	♀	IV
2	271	235	127	54·04	133	56·59	184	78·29	53	22·55	13	♀	V
3	270	232	120	51·72	128	55·17	183	78·87	51	21·98	15	♂	III
4	270	229	120	52·40	130	56·76	181	79·03	53	23·14	16	♂	V
5	268	231	120	51·94	125	54·11	177	76·62	49	21·21	14	♂	IV
6	266	229	121	52·83	122	53·27	178	77·72	49	21·39	14	♂	III
7	264	229	121	52·83	127	55·45	178	77·72	48	20·96	15	♂	V
8	264	228	118	51·75	125	54·82	177	77·63	51	22·36	14	♂	III
9	263	226	121	53·53	126	55·75	177	78·31	51	22·56	13	♂	IV
10	263	225	119	52·88	124	55·11	172	76·44	52	23·11	15	♀	V
11	262	224	119	53·12	124	55·35	178	79·46	47	20·98	15	♂	III
12	261	226	121	53·53	124	54·86	177	78·31	49	21·68	16	♂	V

Table III—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
13	259	219	114	52·05	118	53·88	171	78·08	50	22·83	14	♀	IV
14	258	220	118	53·63	121	55·00	175	79·54	50	22·72	16	♀	V
15	256	223	115	51·56	120	53·81	169	75·78	50	22·42	14	♀	V
16	256	221	116	52·48	124	56·10	172	77·82	46	20·81	14	♀	IV
17	256	219	118	53·88	123	56·16	170	77·62	48	21·91	15	♀	V
18	256	219	111	50·68	118	53·88	172	78·53	48	21·91	15	♀	V
19	256	218	116	53·21	120	55·04	171	78·44	48	22·01	15	♀	IV
20	255	221	113	51·13	121	54·75	171	77·37	46	20·81	14	♀	IV
21	255	220	115	52·27	120	54·54	171	77·72	49	22·27	14	♀	IV
22	255	220	115	52·27	125	56·81	173	78·63	46	20·90	13	♀	IV
23	255	219	115	52·51	119	54·15	168	76·71	50	22·83	15	♀	V
24	255	219	114	52·05	118	53·88	169	77·16	49	22·37	14	♀	IV
25	255	218	113	51·83	120	55·04	170	77·98	49	22·47	15	♀	IV
26	254	219	119	54·15	123	56·16	170	77·62	50	22·83	14	♀	IV
27	254	219	114	52·05	121	55·25	169	77·16	47	21·46	15	♀	V
28	253	220	118	53·63	124	56·36	173	78·63	49	22·27	—	♀	IV
29	252	219	111	50·68	120	54·79	170	77·62	45	20·54	14	♀	IV
30	252	217	116	53·45	119	54·83	168	77·41	45	20·73	16	♀	IV
31	252	217	114	52·53	119	54·83	170	78·34	47	21·65	14	♀	V
32	252	217	112	51·61	124	57·14	168	77·41	47	21·65	14	♀	V
33	252	216	113	52·31	119	55·09	167	77·31	48	22·22	14	♀	V
34	251	216	112	51·85	116	53·70	165	76·38	47	21·75	13	♀	IV
35	250	216	112	51·85	119	55·09	166	76·85	48	22·22	14	♀	IV
36	249	215	110	51·16	116	53·95	171	79·53	44	20·46	15	♀	IV
37	248	215	110	51·16	115	53·48	163	75·81	47	21·86	15	♀	IV
38	248	214	110	51·40	115	53·73	165	77·10	48	22·43	14	♀	V
39	245	212	112	52·83	115	54·24	165	77·83	48	22·64	15	♀	V
40	245	211	108	51·18	114	54·02	160	75·83	45	21·32	14	♀	IV
41	244	211	110	52·13	115	54·50	165	78·19	43	20·38	14	♀	IV
42	244	209	105	50·23	114	54·54	160	76·55	45	21·53	14	♀	IV
43	242	209	109	52·15	110	52·63	159	76·07	44	21·05	15	♀	IV
44	240	207	108	52·16	114	55·07	161	77·77	45	21·73	15	♀	IV
45	240	205	105	51·22	114	55·61	159	77·56	44	21·46	14	♀	IV
46	238	206	108	52·42	111	53·88	159	77·18	44	21·36	14	♀	IV
47	238	206	106	51·45	113	54·85	160	77·67	45	21·84	14	♀	IV
48	238	205	107	52·19	109	53·17	159	77·56	44	21·46	15	♀	IV
49	237	206	108	52·42	113	54·85	156	75·72	46	22·33	14	♀	III
						(8) Received October 2nd, 1914.							
1	280	238	125	52·52	127	53·36	179	75·21	54	22·68	15	♀	VI
2	278	239	127	53·13	135	56·48	188	78·66	55	23·01	14	♀	VII
3	278	239	124	51·88	133	55·64	185	77·40	55	23·01	14	♀	VI
4	276	239	125	52·30	134	56·06	187	78·24	52	21·75	16	♀	VII
5	276	237	120	50·63	127	53·59	181	76·37	54	22·78	14	♀	VI
6	275	236	124	52·54	132	55·93	182	77·11	52	22·03	13	♀	VII
7	270	240	127	52·91	136	56·66	186	77·50	52	21·66	14	♀	VI
8	270	236	122	51·69	131	55·50	185	78·39	52	22·03	14	♀	VII
9	267	232	122	52·58	133	57·32	182	78·44	52	22·41	15	♀	VII
10	267	231	118	51·08	130	56·27	179	77·48	52	22·51	15	♀	VII
11	266	228	121	53·07	127	55·70	181	79·38	51	22·36	15	♀	VII
12	265	230	123	53·43	128	55·65	180	78·26	51	22·17	14	♀	VII
13	265	230	120	52·13	122	53·04	175	76·09	49	21·30	14	♀	IV
14	265	228	115	50·43	127	55·70	174	76·31	49	21·49	15	♀	VII
15	265	227	116	51·10	124	54·62	178	78·41	50	22·02	15	♀	VII

Table III—*Continued.*

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
16	263	228	123	53·94	128	56·14	177	77·63	51	22·36	13	♂	VII
17	263	227	121	53·30	125	55·06	177	77·97	51	22·46	14	♂	V-VI
18	263	226	117	51·77	126	55·75	177	78·31	50	22·12	15	♂	VII
19	262	226	120	53·09	124	54·86	170	75·22	50	22·12	14	♂	III
20	262	225	115	51·11	126	56·00	174	77·33	45	20·00	14	♂	V-VI
21	262	224	118	52·67	120	53·57	176	78·57	47	20·98	15	♀	V-VI
22	261	226	115	50·88	125	55·30	173	76·54	50	22·12	14	♂	VII
23	260	224	112	50·00	122	54·46	171	76·33	50	22·32	15	♀	III
24	260	223	114	51·12	120	53·81	172	77·13	49	21·97	14	♂	II
25	259	224	114	50·89	124	55·34	173	77·23	50	22·32	14	♀	VII
26	259	223	116	52·01	121	54·26	170	76·23	50	22·42	15	♂	VII
27	259	222	117	52·70	120	54·05	168	75·67	48	21·62	15	♀	III
28	259	221	118	53·39	122	55·20	172	78·28	49	22·17	14	♂	VII
29	258	225	115	51·11	126	56·00	174	77·33	50	22·22	14	♀	VII
30	258	224	119	53·12	125	55·80	172	76·78	49	21·87	14	♂	VII
31	255	218	113	51·83	119	54·58	166	76·14	49	22·47	16	♂	V
32	254	219	114	52·05	117	53·42	165	75·34	50	22·83	14	♂	V
33	254	218	111	50·91	119	54·58	168	77·06	47	21·56	14	♂	VII
34	254	218	114	52·29	118	54·12	168	77·06	49	22·47	14	♀	IV
35	252	217	115	52·99	119	54·83	169	77·88	49	22·58	14	♂	V
36	252	217	114	52·53	120	55·29	168	77·41	48	22·12	15	♀	VII
37	251	219	110	50·22	117	53·42	170	77·62	46	21·00	14	♀	III
38	250	215	111	51·62	121	56·27	170	79·07	47	21·86	15	♂	VI
39	250	215	110	51·16	118	54·88	167	77·67	49	22·79	15	♀	VII
40	249	217	112	51·61	117	53·91	164	75·57	48	22·12	14	♂	V
41	249	216	110	50·92	119	55·09	169	78·24	44	20·37	14	♀	IV
42	248	215	111	51·62	116	53·95	166	77·20	49	22·79	14	♀	VII
43	247	213	113	53·05	122	57·27	167	78·40	47	22·06	15	♂	VII
44	247	213	111	52·11	118	55·39	167	78·40	48	22·53	14	♂	VII
45	247	212	109	51·41	117	55·18	161	75·94	48	22·64	15	♂	V
46	245	211	110	52·13	118	55·92	167	79·14	47	22·27	14	♂	III
47	244	220	113	51·36	117	53·17	166	75·45	50	22·72	14	♀	II
48	242	210	110	52·38	114	54·28	162	77·14	45	21·43	14	♂	VII
49	240	207	111	53·62	115	55·55	162	78·26	45	21·73	14	♂	IV
50	235	204	105	51·47	110	53·92	156	76·47	48	23·52	14	♂	V
Mean				51·98		54·87		77·15*		21·84	14·45		

* Excluding Sample 1.

Table IV.

Trawled off The Smalls. Received October 15th, 1914.

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
1	303	260	132	50·77	141	54·22	199	76·53	55	21·15	15	♂	V
2	300	256	136	53·12	140	54·68	196	76·56	55	21·48	14	♀	IV
3	297	256	140	54·68	145	56·64	201	78·51	58	22·65	14	♂	V
4	297	255	133	52·15	141	55·29	196	76·86	55	21·56	14	♂	V
5	297	255	131	51·37	142	55·68	200	78·43	55	21·56	15	♂	V
6	297	253	134	52·96	142	56·12	193	76·28	56	22·13	14	♀	IV

Table IV—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
7	297	253	132	52·17	142	56·12	196	77·47	55	21·73	14	♀♂♂	IV
8	297	252	130	51·58	140	55·55	194	76·98	55	21·82	15	♂♂	V
9	295	255	136	53·33	141	55·29	194	76·07	54	21·17	14	♂♂	V
10	295	253	133	52·56	142	56·12	197	77·86	54	21·34	14	♂♂	V
11	294	254	132	51·96	138	54·33	197	77·55	56	22·04	15	♂♂	V
12	293	251	131	52·19	141	56·17	193	76·89	55	21·91	16	♂♂	V
13	292	251	131	52·19	141	56·17	195	77·68	54	21·51	16	♂♂	V
14	291	251	132	52·58	140	55·77	194	77·29	56	22·31	15	♂♂	IV
15	291	247	130	52·63	142	57·49	193	78·13	55	22·26	14	♂♂	V
16	289	250	130	52·00	138	55·20	191	76·40	54	21·60	15	♂♂	V
17	289	248	127	51·21	135	54·43	192	77·41	52	20·96	14	♂♂	V
18	289	244	126	51·63	135	55·32	187	76·63	53	21·72	15	♂♂	IV
19	288	248	128	51·61	137	55·24	192	77·41	53	21·37	14	♀♂	IV
20	288	246	127	51·62	135	54·87	192	78·04	55	22·35	15	♀♂	V
21	288	245	133	54·28	138	56·32	194	79·18	54	22·04	14	♂♂	V
22	287	249	133	53·41	139	55·82	191	76·70	59	23·69	14	♂♂	V
23	286	246	129	52·43	138	56·09	190	77·23	55	22·35	13	♀♂	IV
24	286	246	129	52·43	135	54·87	191	77·64	55	22·35	14	♂♂	V
25	285	246	130	52·84	135	54·87	189	76·82	56	22·76	15	♂♂	IV
26	285	244	128	52·45	133	54·50	186	76·22	52	21·31	15	♂♂	V
27	285	243	129	53·08	139	57·20	189	77·77	51	20·98	15	♀♂	IV
28	284	244	128	52·45	137	56·14	191	78·27	53	21·72	15	♂♂	V
29	284	244	129	52·86	136	55·73	190	77·86	53	21·72	15	♂♂	IV
30	283	243	129	53·08	135	55·55	183	75·28	52	21·39	14	♂♂	V
31	283	242	127	52·47	135	55·78	191	78·92	54	22·31	16	♂♂	IV
32	283	238	123	51·68	129	54·20	186	78·15	52	21·84	15	♂♂	IV
33	282	239	123	51·46	129	53·97	183	76·56	53	22·17	15	♂♂	V
34	282	239	123	51·46	135	56·48	187	78·24	53	22·17	14	♂♂	IV
35	281	244	130	53·27	133	54·50	189	77·45	54	22·13	14	♀♂	IV
36	281	243	125	51·44	135	55·55	187	76·95	51	20·98	15	♂♂	IV
37	280	241	127	52·69	133	55·18	186	77·17	51	21·16	14	♂♂	V
38	280	240	128	53·33	132	55·00	186	77·50	50	20·83	15	♂♂	IV
39	280	239	125	52·30	133	55·64	184	76·98	54	22·59	14	♂♂	V
40	280	238	125	52·52	132	55·46	183	76·89	54	22·68	14	♀♂	V
41	280	237	125	52·75	134	56·53	182	76·79	52	21·93	14	♀♂	V
42	279	239	123	51·46	129	53·97	181	75·73	53	22·17	13	♀♂	III
43	278	239	122	51·04	128	53·55	183	76·56	52	21·75	15	♂♂	V
44	278	238	123	51·68	131	55·04	183	76·89	53	22·26	13	♂♂	IV
45	277	237	124	52·32	130	54·85	183	77·21	52	21·93	15	♂♂	V
46	277	236	123	52·11	130	55·08	181	76·69	53	22·45	14	♀♂	IV
47	277	235	123	52·34	132	56·17	182	77·44	51	21·70	14	♀♂	IV
48	276	237	127	53·59	133	56·11	182	76·79	52	21·93	13	♀♂	IV
49	276	235	122	51·91	128	54·46	181	77·02	52	22·12	14	♀♂	IV
50	275	237	124	52·32	131	55·27	180	75·94	53	22·36	14	♂♂	V
51	275	237	124	52·32	131	55·27	183	77·21	52	21·93	15	♂♂	IV
52	275	236	126	53·39	133	56·35	182	77·11	50	21·14	13	♂♂	IV
53	275	236	125	52·96	131	55·50	184	77·96	50	21·14	15	♂♂	V
54	275	236	122	51·69	128	54·23	179	75·84	52	22·03	15	♂♂	V
55	275	235	125	53·19	130	55·31	182	77·44	51	21·70	13	♀♂	IV
56	275	234	120	51·28	129	55·12	180	76·92	53	22·64	15	♂♂	IV
57	275	232	124	53·44	129	55·60	181	78·01	52	22·41	14	♀♂	IV
58	273	236	121	51·27	131	55·50	182	77·11	49	20·76	14	♂♂	V
59	273	233	122	52·36	127	54·50	177	75·96	50	21·45	14	♂♂	V
60	273	233	121	51·93	131	56·22	180	77·25	50	21·45	14	♂♂	IV
61	273	232	124	53·44	130	56·03	179	77·15	51	21·98	14	♂♂	IV
62	272	241	127	52·69	136	56·43	184	76·34	51	21·16	14	♂♂	IV
63	272	236	120	50·84	125	52·96	177	75·00	50	21·14	14	♂♂	V
64	272	233	125	53·64	128	54·93	181	77·68	52	22·31	15	♂♂	IV

Table IV—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
65	272	231	119	51·51	127	54·97	177	76·62	52	22·51	15	♂♀	IV
66	272	231	115	49·78	125	54·11	174	75·32	53	22·94	13	♀♂	IV
67	272	230	121	52·60	126	54·78	179	77·82	51	22·17	13	♂♂	V
68	271	233	120	51·50	127	54·50	178	76·39	49	21·03	14	♂♂	V
69	271	232	122	52·58	126	54·31	177	76·29	50	21·55	14	♂♂	IV
70	270	233	117	50·21	126	54·07	177	75·96	52	22·31	14	♂♂	V
71	270	232	123	53·01	127	54·74	180	77·58	50	21·55	15	♂♀	IV
72	270	231	121	52·38	125	54·11	176	76·19	50	21·64	15	♂♀	IV
73	270	230	123	53·43	128	55·65	179	77·82	50	21·73	14	♂♀	IV
74	270	229	120	52·40	126	55·02	176	76·85	50	21·83	14	♂♀	IV
75	269	232	118	50·86	124	53·44	174	75·00	51	21·98	15	♂♂	V
76	269	231	120	51·94	130	56·27	177	76·62	50	21·64	14	♂♂	V
77	269	230	120	52·13	124	53·95	178	77·39	49	21·30	14	♂♂	V
78	269	230	115	50·00	124	53·95	176	76·52	50	21·73	14	♂♀	IV
79	268	233	121	51·93	125	53·64	179	76·82	50	21·45	15	♂♂	V
80	268	231	119	51·51	126	54·54	178	77·05	48	20·77	13	♂♂	V
81	268	225	117	52·00	120	53·33	173	76·88	50	22·22	14	♂♀	IV
82	267	230	124	53·95	131	56·95	180	78·26	50	21·73	14	♂♂	V
83	267	230	120	52·13	125	54·34	174	75·65	50	21·73	14	♂♀	IV
84	267	229	117	51·09	126	55·02	176	76·85	47	20·52	15	♂♀	IV
85	267	225	118	52·44	124	55·11	173	76·88	49	21·77	14	♂♂	IV
86	266	227	120	52·86	125	55·06	175	77·09	51	22·46	13	♂♀	IV
87	265	227	121	53·30	128	56·34	176	77·53	51	22·46	15	♀♀	V
88	264	224	112	50·00	123	54·91	168	75·00	47	20·98	13	♂♀	IV
89	263	231	119	51·51	124	53·67	174	75·32	50	21·64	14	♂♂	V
90	263	227	121	53·30	125	55·06	177	77·97	49	21·58	14	♂♂	IV
91	263	227	118	51·98	124	54·62	176	77·53	50	22·02	14	♂♀	IV
92	263	227	114	50·22	122	53·74	174	76·65	47	20·70	14	♂♂	V
93	263	225	120	53·33	126	56·00	173	76·88	49	21·77	14	♂♀	IV
94	263	225	117	52·00	120	53·33	172	76·44	48	21·33	15	♀♀	IV
95	263	225	115	51·11	123	54·66	174	77·33	49	21·77	14	♂♂	IV
96	262	226	116	51·32	122	53·98	173	76·54	48	21·23	15	♂♂	V
97	262	225	117	52·00	126	56·00	174	77·33	51	22·66	15	♂♀	IV
98	262	225	117	52·00	127	56·44	176	78·22	48	21·33	15	♀♀	IV
99	262	224	119	53·12	123	54·91	171	76·33	49	21·87	14	♂♀	IV
100	262	223	117	52·46	122	54·70	171	76·68	49	21·97	15	♂♀	IV
101	262	221	114	51·58	122	55·20	169	76·47	49	22·17	14	♂♂	V
102	261	228	117	51·31	127	55·70	171	75·00	51	22·36	13	♂♂	II
103	261	222	118	53·15	121	54·50	173	77·92	49	22·07	15	♂♂	V
104	260	225	118	52·44	122	54·22	172	76·44	48	21·33	14	♂♀	IV
105	259	222	116	52·25	122	54·99	171	77·02	50	22·51	15	♂♂	V
106	258	222	118	53·15	123	55·40	173	77·92	48	21·62	14	♂♂	IV
107	258	222	115	51·80	122	54·99	170	76·57	49	22·07	13	♂♂	IV
108	258	222	115	51·80	122	54·99	171	77·02	49	22·07	14	♂♂	V
109	257	222	115	51·80	118	53·15	169	76·12	50	22·51	14	♂♀	IV
110	257	220	112	50·91	117	53·17	166	75·45	48	21·81	15	♂♀	IV
111	257	219	118	53·88	123	56·16	171	78·08	46	21·00	14	♀♀	V
112	257	219	113	51·59	119	54·15	166	75·79	48	21·91	14	♀♀	IV
113	256	220	119	54·09	123	55·91	170	77·27	48	21·81	13	♀♀	IV
114	255	221	113	51·13	117	52·94	173	78·28	49	22·17	14	♂♀	V
115	255	220	116	52·72	120	54·54	165	75·00	49	22·27	12	♀♀	IV
116	254	219	109	49·77	116	52·96	166	75·79	45	20·54	14	♀♀	IV
117	254	218	114	52·29	120	55·04	168	77·06	45	20·64	13	♂♀	V
118	253	220	109	49·54	115	52·27	165	75·00	46	20·90	14	♂♀	IV
119	253	215	113	52·55	115	53·48	164	76·26	49	22·79	13	♂♀	IV
120	239	206	101	49·02	110	53·39	157	76·21	43	20·87	14	♂♀	III
Mean				52·15		55·01		76·92		21·79	14·21		

Table V.

Herring from Welsh Coast.

(1) From Aberdovey. Received October 22nd, 1914.

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
1	277	240	126	52·50	130	54·16	182	75·83	51	21·25	15	♂	IV
2	272	240	122	50·83	129	53·75	183	76·25	49	20·41	15	♂♂	V
3	265	231	118	51·08	125	54·11	175	75·75	47	20·34	15	♂♀	VII
4	265	230	118	51·30	126	54·78	180	78·26	49	21·30	14	♀	V
5	258	223	113	50·67	121	54·26	171	76·68	49	21·97	14	♀♀	V
6	252	219	110	50·22	120	54·79	163	74·42	49	22·37	14	♂♀	VII
7	250	216	109	50·46	119	55·09	163	75·46	49	22·68	13	♂♀	VII
8	244	214	109	50·93	120	56·07	160	74·76	47	21·96	13	♀	VII
9	240	211	106	50·23	114	54·02	157	74·40	45	21·32	12	♀	IV
10	237	206	104	50·48	111	53·88	158	76·69	44	21·36	15	♀	IV
11	232	202	104	51·48	110	54·45	149	73·76	44	21·78	13	♂	IV
12	232	200	100	50·00	110	55·00	154	77·00	43	21·50	13	♂♂	III
13	229	199	101	50·75	107	53·76	149	74·87	45	22·61	15	♂♀	III
14	223	194	101	52·06	107	55·15	145	74·74	45	23·19	12	♀	III
15	221	193	98	50·77	102	53·12	142	73·57	42	21·76	—	♂♀	IV
16	221	191	97	50·78	103	53·92	142	74·34	43	22·51	14	♀	III
17	221	189	95	50·26	103	54·49	141	74·60	41	21·69	15	♂♂	IV
18	219	189	97	51·32	104	55·02	142	75·13	42	22·22	14	♂♂	IV
19	219	188	96	51·06	104	55·32	143	76·06	42	22·34	14	♂	IV

(2) New Quay Bay, 4 in. mesh. Received October 28th, 1914.

1	284	246	125	50·81	136	55·28	189	76·82	52	21·13	14	♀	IV
2	274	236	123	52·11	132	55·93	183	77·54	53	22·45	14	♀	IV
3	270	234	120	51·28	126	53·84	180	76·92	48	20·51	14	♀	V
4	264	228	120	52·63	126	55·26	179	78·50	49	21·49	15	♀	V
5	261	229	116	50·65	124	54·14	175	76·41	49	21·39	16	♂	V
6	260	228	117	51·31	123	53·94	173	75·87	50	21·92	14	♂	V
7	260	224	113	50·44	118	52·67	171	76·33	47	20·98	16	♀	IV
8	259	225	112	49·77	121	53·77	169	75·11	47	20·88	13	♀	IV
9	259	223	118	52·91	122	54·70	175	78·47	51	22·87	15	♂♀	V
10	251	217	113	52·07	117	53·91	169	77·88	44	20·27	14	♂♀	IV
11	250	216	111	51·38	115	53·24	165	76·38	44	20·37	15	♂	V
12	245	212	109	51·41	114	53·77	160	75·47	46	21·69	16	♀	IV

(3) Moelfre. Received November 19th, 1914.

1	273	234	122	52·13	125	53·41	179	76·49	51	21·79	15	♀	VII
2	265	230	118	51·30	122	53·04	177	76·95	48	20·87	15	♂	VI
3	264	228	116	50·87	124	54·38	176	77·19	48	21·05	14	♂♀	V
4	264	226	116	51·32	120	53·09	176	77·87	48	21·23	14	♂	V
5	258	223	113	50·67	119	53·36	170	76·23	48	21·52	14	♂	VII
6	258	219	115	52·51	116	52·96	170	77·62	48	21·91	14	♂	VII
7	256	224	119	53·12	122	54·46	172	76·78	49	21·87	13	♂♀	VII
8	253	228	117	51·31	123	53·94	171	75·00	48	21·05	13	♀	VII
9	250	213	112	52·58	118	55·39	165	77·46	46	21·69	13	♂♂	VII
10	246	212	107	50·47	115	54·24	162	76·41	45	21·22	13	♂♂	VII
11	245	212	110	51·88	116	54·71	161	75·94	45	21·22	14	♂♀	VII
12	244	210	106	50·47	114	54·28	160	76·19	46	21·90	15	♂♂	V
13	234	202	103	50·99	109	53·96	150	74·25	42	20·79	14	♂	III
14	228	194	100	51·54	104	53·60	150	77·31	40	20·61	14	♂	II

Table V—*Continued.*

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
(4) Aberdovey. Received November 19th, 1914.													
1	270	235	122	51·91	127	54·04	177	75·31	50	21·27	15	♀	VII
2	270	234	124	52·99	130	55·55	180	76·92	52	22·22	14	♂♀	VII
3	270	234	121	51·70	126	53·84	176	75·21	52	22·22	14	♂♀	VII
4	268	230	118	51·30	124	53·95	178	77·39	50	21·73	15	♂	VII
5	265	230	116	50·43	124	53·95	174	75·65	52	22·60	13	♂♀	VII
6	262	226	115	50·88	120	53·09	170	75·22	50	22·12	14	♂♀	VII
7	258	224	115	51·33	122	54·46	170	75·89	50	22·32	15	♂♀	VII
8	258	222	116	52·25	121	54·50	170	76·57	48	21·62	14	♂	VII
9	256	223	115	51·56	123	55·15	171	76·68	49	21·97	14	♂♀	VII
10	255	220	115	52·27	122	55·45	165	75·00	48	21·81	13	♂♀	VII
11	254	220	113	51·39	118	53·63	166	75·45	47	21·36	14	♂♀	VII
12	252	217	111	51·15	118	54·37	164	75·57	45	20·73	14	♂♀	VII
13	252	217	114	52·53	122	56·22	167	76·95	48	22·12	13	♂♀	VII
14	252	215	113	52·55	119	55·34	167	77·67	46	21·39	13	♂♀	VII
15	251	220	112	50·91	117	53·17	168	76·36	45	20·45	15	♂♀	VI
16	245	214	106	49·53	110	51·40	160	74·76	44	20·56	15	♂♀	VI
17	243	213	106	49·76	114	53·52	159	74·64	45	21·12	15	♂♀	III
18	234	198	102	51·51	108	54·54	152	76·76	43	21·71	15	♂♀	V
19	231	200	103	51·50	109	54·50	150	75·00	42	21·00	14	♂♀	I
20	230	199	101	50·75	109	54·77	151	75·87	42	21·10	13	♀	I
21	222	194	96	49·48	103	53·09	142	73·71	41	21·13	14	♂♀	II
22	219	189	97	51·32	102	53·97	144	76·13	42	22·22	14	♂♀	I
23	217	187	97	51·87	100	53·47	140	74·86	41	21·92	14	♀	II
24	210	183	94	51·36	99	54·09	138	75·40	39	21·31	13	♂	IV
(5) New Quay Bay, 4 in. mesh. Received November 19th, 1914.													
1	282	242	125	51·65	133	54·95	182	75·20	51	21·07	14	♂	IV
2	268	234	119	50·85	129	55·12	175	74·78	51	21·79	14	♂♀	IV
3	267	235	123	52·34	125	53·19	179	76·17	48	20·42	16	♂♀	V
4	267	230	120	52·13	125	54·34	176	76·52	48	20·87	14	♂♀	V
5	262	228	118	51·75	126	55·26	174	76·31	49	21·49	14	♀	IV
6	262	226	117	51·77	123	54·42	171	76·54	49	21·68	14	♂♀	V
7	262	223	116	52·01	118	52·91	171	76·68	51	22·87	15	♂♀	V
8	260	225	117	52·00	122	54·22	169	75·11	50	22·22	15	♂♀	V
9	260	221	115	52·03	121	54·75	171	77·37	47	21·26	15	♂♀	V
10	257	223	114	51·12	123	55·15	172	77·13	47	21·07	15	♂♀	V
11	254	222	113	50·90	121	54·50	172	77·47	44	19·82	16	♂♀	V
12	254	217	115	52·99	120	55·29	169	77·88	46	21·19	14	♂♀	IV
13	252	219	111	50·68	116	52·96	164	74·88	47	21·46	15	♀	V
14	251	217	112	51·61	118	54·37	163	75·06	46	21·19	15	♂♀	V
15	251	216	110	50·92	113	52·31	165	76·38	47	21·75	14	♀	IV
16	247	214	108	50·46	115	53·73	162	75·70	45	21·02	14	♂♀	V
17	246	214	109	50·93	113	52·80	161	75·23	44	20·56	14	♂♀	IV
18	243	211	106	50·23	112	53·08	160	75·83	43	20·38	14	♂	V
(6) Moelfre. Received November 23rd, 1914.													
1	259	223	116	52·01	122	54·70	172	77·13	47	21·07	14	♂	VII
2	255	220	112	50·91	120	54·54	166	75·45	46	20·90	15	♂♀	II
3	255	220	113	51·36	117	53·17	165	75·00	46	20·90	14	♂♀	VII
4	253	215	110	51·16	118	54·88	166	77·20	49	22·79	15	♂♀	VII
5	248	211	108	51·18	112	53·08	162	76·77	43	20·38	16	♀	V
6	246	213	109	51·17	115	53·99	163	76·52	46	21·69	14	♀	II

Table V—*Continued.*

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
7	241	207	107	51·69	113	54·59	160	77·29	45	21·73	15	♂	VII
8	235	201	106	52·73	108	53·73	154	76·61	44	21·88	14	♂♂	I
9	233	201	104	51·74	109	54·22	153	76·11	45	22·38	14	♀	VII
10	233	201	102	50·74	108	53·73	154	76·61	42	20·89	14	♂♂	V
11	230	199	102	51·25	111	55·77	150	75·37	43	21·60	14	♂♂	VII
12	229	198	99	50·00	106	53·53	145	73·23	42	21·21	13	♂♂	V
13	227	198	99	50·00	106	53·53	149	75·25	42	21·21	14	♂♂	I
14	225	195	98	50·25	104	53·33	150	76·92	43	22·05	14	♂	I
15	225	195	98	50·25	107	54·87	146	74·87	40	20·51	14	♂♂	I
16	224	196	100	51·02	106	54·08	148	75·51	41	20·91	13	♀♀	I
17	224	193	99	51·29	101	52·33	144	74·61	41	21·24	15	♀♀	I
18	223	191	98	51·30	106	55·49	146	76·43	42	21·99	13	♀	I
19	223	190	95	50·00	106	55·78	145	76·31	40	21·05	14	♂♂	I
20	222	191	98	51·30	102	53·40	144	75·39	40	20·94	14	♂♂	I
21	222	190	98	51·57	102	53·68	143	75·26	42	22·10	14	♀	I
22	221	190	97	51·05	101	53·15	142	74·73	41	21·58	14	♂♀	I
23	219	190	98	51·57	100	52·63	142	74·73	41	21·58	14	♂♀	I
24	218	190	95	50·00	103	54·21	141	74·21	41	21·58	13	♂♀	I
25	218	187	94	50·26	104	55·61	144	77·00	40	21·38	15	♀	I
26	217	188	98	52·12	102	54·25	142	75·53	42	22·34	15	♀	I
27	217	187	97	51·87	101	54·01	141	75·40	41	21·92	13	♀♀	I
28	217	185	92	49·72	101	54·59	139	75·13	40	21·62	13	♀♀	I
29	216	186	93	50·00	99	53·22	139	74·73	41	22·04	14	♀♀	I
30	216	185	95	51·35	102	55·13	142	76·75	42	22·70	13	♀♀	I
31	215	185	94	50·81	97	52·43	138	74·59	41	22·16	14	♂	I
32	215	184	95	51·63	98	53·26	137	75·54	40	21·73	15	♂♂	I
33	214	182	91	50·00	96	52·74	135	74·17	42	23·07	14	♂♂	I
34	211	181	91	50·27	100	55·24	136	75·13	41	22·65	12	♂♀	I
35	211	178	92	51·68	95	53·37	133	74·71	40	22·47	13	♂♀	I
36	207	177	89	50·28	96	54·23	134	75·70	40	22·59	14	♂	I
						(7) Pwllheli. Received November 23rd, 1914.							
1	269	234	122	52·13	128	54·70	178	76·06	51	21·79	15	♀	VII
2	269	233	121	51·93	126	54·07	179	76·82	51	21·88	15	♂♂	II
3	267	231	118	51·08	128	55·45	180	77·92	50	21·64	15	♂♂	VII
4	266	231	116	50·21	125	54·11	172	74·45	51	22·48	14	♀	II
5	265	229	115	50·21	120	52·40	169	73·80	48	20·96	15	♂♂	II
6	265	225	118	52·44	127	56·44	169	75·11	50	22·22	14	♂♂	II
7	263	231	117	50·64	124	53·68	178	77·05	48	20·77	16	♀♀	VII
8	263	227	119	52·42	121	53·30	173	76·21	49	21·58	15	♀	VII
9	263	227	117	51·54	126	55·50	174	76·65	51	22·46	15	♂♂	VII
10	263	226	115	50·88	123	54·42	172	76·10	48	21·23	14	♂♂	II
11	263	225	115	51·11	121	53·77	169	75·11	51	22·66	14	♀♀	II
12	262	228	119	52·19	122	53·50	172	75·43	51	22·36	14	♀	II
13	262	224	113	50·44	121	54·01	171	76·33	50	22·32	14	♂♀	VII
14	261	228	114	50·00	122	53·50	173	75·87	47	20·61	15	♀	II
15	261	225	118	52·44	125	55·55	174	77·33	49	21·77	15	♂♀	VII
16	261	225	118	52·44	121	53·77	174	77·33	48	21·33	14	♀	II
17	261	225	113	50·22	122	54·22	170	75·55	47	20·88	14	♀	VII
18	261	225	116	51·55	124	55·11	173	76·88	47	20·88	14	♂♂	II
19	259	223	114	51·12	123	55·15	171	76·68	49	21·97	14	♂♂	VII
20	259	222	112	50·45	121	54·50	169	76·12	47	21·17	14	♂♂	VII
21	258	220	113	51·36	123	55·91	168	76·36	47	21·36	14	♂♂	II
22	256	223	114	51·12	120	53·81	168	75·33	46	20·62	15	♂	V

Table V—*Continued.*

No.	T.	T.cd.	D.	% T.cd.	V.	% T.cd.	A.	% T.cd.	l.cp.l.	% T.cd.	K_2	s.	g.
23	255	221	114	51·58	120	54·29	168	76·01	49	22·17	14	♂♀♀	VII
24	255	220	115	52·27	120	54·54	168	76·36	46	20·90	14	♀♀	II
25	254	222	112	50·45	118	53·15	168	75·67	47	21·17	14	♀♂♂	VII
26	254	221	111	50·22	121	54·75	171	77·37	47	21·26	15	♂♂	VII
27	254	220	111	50·45	119	54·09	164	74·54	48	21·81	14	♂♂♀	II
28	254	220	111	50·45	115	52·27	165	75·00	47	21·36	15	♂♂♂	VII
29	252	216	109	50·46	119	55·09	167	77·31	46	21·29	15	♂♂♂	VII
30	250	218	112	51·37	115	52·75	164	75·22	47	21·56	15	♂♂♂	II
31	250	217	109	50·23	116	53·45	164	75·57	47	21·65	15	♂♂♂	VII
32	249	218	113	51·83	117	53·67	165	75·68	48	22·01	15	♂♂♂	II
33	249	210	110	52·38	116	55·24	159	75·71	46	21·90	14	♂♂	II
34	248	213	111	52·11	118	55·39	166	77·93	45	21·12	15	♂♀♂	II
35	247	217	114	52·53	114	52·53	162	74·65	46	21·19	14	♂♀♂	II
36	247	212	108	50·94	111	52·35	159	75·00	45	21·22	14	♂♀♂	II
37	246	213	109	51·17	117	54·93	164	76·99	46	21·59	15	♂♂♂	VII
38	246	210	107	50·95	115	54·76	160	76·19	46	21·90	14	♂♂	II
39	243	212	108	50·94	115	54·24	159	75·00	45	21·22	14	♂♂	II
40	241	206	103	50·00	108	52·42	154	74·75	44	21·36	13	♂♀♂	I
41	240	209	106	50·71	114	54·54	159	76·07	45	21·53	14	♂♂♂	II
42	237	207	104	50·24	110	53·14	155	74·88	43	20·77	14	♂♂	II
43	235	203	102	50·24	108	53·20	153	75·36	45	22·16	14	♂♂♀	II
44	231	203	103	50·73	108	53·20	152	74·87	44	21·67	14	♂	I
45	220	191	96	50·26	101	52·88	141	73·82	43	22·51	13	♂♀♂	I
46	210	182	90	49·78	93	51·09	136	74·72	38	20·87	14	♂	V
						(8) Moelfre. Received December 9th, 1914.							
1	271	233	120	51·50	126	54·07	180	77·25	47	20·17	15	♂	V
2	268	233	116	49·78	123	52·78	177	75·96	49	21·03	15	♂♂	V
3	268	233	120	51·50	127	54·50	178	76·39	49	21·03	16	♂♂♂	II
4	267	231	116	50·21	121	52·38	171	74·02	51	22·48	15	♂♂♂	II
5	265	230	119	51·73	125	54·34	178	77·39	47	20·43	16	♂♂♂	V
6	265	229	116	50·65	125	54·58	174	75·98	50	21·83	15	♂♂♂	II
7	262	226	115	50·88	121	53·53	172	76·10	50	22·12	15	♂♂♂	V
8	262	224	117	52·23	124	55·34	174	77·67	50	22·32	14	♂♂♂	II
9	261	225	113	50·22	123	54·66	168	74·66	47	20·88	14	♂♂♀	II
10	261	224	115	51·33	123	54·91	169	75·44	51	22·76	14	♂♂♂	II
11	260	226	116	51·32	124	54·86	172	76·10	47	20·79	14	♂♂♀	II
12	260	225	114	50·66	125	55·55	174	77·33	49	21·77	14	♂♂♀	II
13	260	224	114	50·89	124	55·34	173	77·23	49	21·87	15	♂♂	II
14	260	222	114	51·35	123	55·40	168	75·67	50	22·51	14	♂♀♂	II
15	259	225	114	50·66	120	53·33	172	76·44	48	21·33	15	♂♂♂	II
16	259	224	113	50·44	120	53·57	169	75·44	49	21·87	15	♂♂♂	II
17	258	222	110	49·54	123	55·40	170	76·57	48	21·62	16	♂♂♂	V
18	257	224	113	50·44	119	53·12	170	75·89	48	21·42	14	♂♀♀	II
19	257	220	111	50·45	120	54·54	168	76·36	50	22·72	14	♀♀	II
20	255	221	110	49·77	119	53·84	165	74·66	49	22·17	14	♂♂	II
21	255	221	114	51·58	122	55·20	173	78·28	48	21·71	14	♂♂	II
22	255	220	114	51·81	122	55·45	167	75·90	48	21·81	14	♂♂♂	II
23	254	221	114	51·58	120	54·29	169	76·47	46	20·81	14	♂♂♂	V
24	254	217	114	52·53	119	54·83	165	76·03	48	22·12	15	♂♂♀	II
25	252	219	113	51·59	120	54·79	167	76·25	48	21·91	14	♂♂♂	V
26	252	217	112	51·61	116	53·45	165	76·03	49	22·58	14	♂♂♂	II
27	249	213	109	51·17	118	55·39	164	76·99	47	22·06	15	♂♂	II
28	247	211	110	52·13	119	56·39	167	79·14	48	22·74	15	♂♂	II

Table V—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
29	241	205	106	51·70	113	55·12	158	77·07	46	22·43	14	♂♂	II
30	240	206	106	51·45	113	54·85	157	76·21	45	21·84	14	♂♂	II
31	239	206	104	50·48	114	55·34	162	78·64	45	21·84	14	♂♂	II
32	238	204	105	51·47	110	53·92	156	76·47	45	22·05	14	♂♂	II
33	237	206	103	50·00	114	55·34	156	75·72	46	22·33	14	♂♂	II
34	237	205	104	50·73	111	54·14	159	77·56	44	21·46	16	♂♂	II
35	237	202	106	52·47	111	54·95	157	77·72	46	22·76	13	♀♀	II
36	236	201	103	51·24	107	53·23	153	76·11	43	21·39	14	♀♀	IV
37	234	203	102	50·24	110	54·68	153	75·36	47	23·15	14	♀♀	II
38	230	200	102	51·00	108	54·00	153	76·50	43	21·50	13	♂♂	I
39	230	199	105	52·76	108	54·27	153	76·88	43	21·60	14	♂♂	I
40	226	194	99	51·03	106	54·63	146	75·25	43	22·16	14	♀♀	I
41	224	193	101	52·33	103	53·36	145	75·13	43	22·28	14	♀♀	I
42	224	192	100	52·08	103	53·64	145	75·52	44	22·91	14	♀♀	I
43	222	193	102	53·12	105	54·40	144	74·61	44	22·79	14	♂♂	I
44	222	187	95	50·80	104	55·61	142	75·93	41	21·92	13	♂♂	I
45	220	188	95	50·53	104	55·32	144	76·59	41	21·80	13	♀♀	II
46	219	189	96	50·79	103	54·49	142	75·13	40	21·16	14	♂♂	II
47	219	188	95	50·53	101	53·72	141	75·00	41	21·80	14	♂♂	I
48	217	187	95	50·80	100	53·47	143	76·47	41	21·92	15	♂♂	I
49	215	183	96	52·45	100	54·64	138	75·40	41	22·40	15	♂♂	I
50	205	175	89	50·85	96	54·85	136	77·71	40	22·85	14	♀	I
			(9) New Quay, 4 in. mesh. Received December 10th, 1914.										
1	270	232	123	53·01	127	54·74	178	76·72	50	21·55	14	♂♀	V
2	265	227	117	51·54	125	55·06	174	76·65	49	21·58	14	♂♂	V
3	263	227	119	52·42	127	55·94	175	77·09	49	21·58	14	♂♂	V
4	263	225	116	51·55	121	53·77	173	76·88	45	20·00	16	♂♂	V
5	262	225	118	52·44	123	54·66	171	76·00	48	21·33	14	♂♂	V
6	259	226	113	50·00	121	53·53	172	76·10	46	20·35	15	♂♂	V
7	259	225	111	49·33	120	53·33	173	76·88	45	20·00	15	♂♂	V
8	257	222	116	52·25	124	55·85	171	77·02	47	21·17	15	♂♂	V
9	256	219	113	51·59	118	53·88	168	76·71	48	21·91	15	♀♀	V
10	256	219	112	51·14	121	55·25	169	77·16	46	21·00	14	♂♂	V
11	255	221	113	51·13	116	52·48	166	75·11	46	20·81	14	♂♂	V
12	255	221	112	50·67	119	53·84	168	76·01	45	20·36	14	♂	V
			(10) Moelfre. Received December 17th, 1914.										
1	275	235	118	50·21	128	54·46	184	78·29	50	21·27	16	♂♂	VII
2	271	233	120	51·50	130	55·79	179	76·82	51	21·88	14	♂♂	VII
3	267	229	120	52·40	128	55·89	180	78·60	49	21·39	15	♂♂	VII
4	261	227	116	51·10	121	53·30	170	74·89	50	22·02	14	♀♀	VII
5	260	226	113	50·00	123	54·42	174	76·99	50	22·12	16	♂♂	VII
6	259	219	112	51·14	121	55·25	170	77·62	45	20·54	14	♂♂	VII
7	258	222	116	52·25	122	54·99	170	76·57	50	22·51	14	♂♂	VII
8	258	220	115	52·27	120	54·54	170	77·27	50	22·72	16	♀♀	VII
9	257	224	117	52·23	122	54·46	169	75·44	50	22·32	15	♂♂	VII
10	257	222	113	50·90	122	54·99	172	77·47	48	21·62	15	♂♂	VII
11	256	220	120	54·54	122	55·45	170	77·27	49	22·27	15	♂♂	VII
12	255	218	113	51·83	122	55·96	169	77·52	49	22·47	14	♂♂	VII
13	252	213	105	49·29	117	54·92	165	77·46	49	23·00	14	♂♂	VII
14	251	221	113	51·13	120	54·29	168	76·01	46	20·81	15	♂♂	VII

Table V—*Continued.*

No.	T.	T.cd.	D.		V.		A.		l.cp.l.		K_2	s.	g.
				% T.cd.		% T.cd.		% T.cd.		% T.cd.			
15	250	211	111	52·60	117	55·45	163	77·25	47	22·27	15	♀	VII
16	249	213	111	52·11	119	55·86	168	78·87	47	22·06	14	♀♂	VII
17	244	207	107	51·69	116	56·03	160	77·29	45	21·73	13	♂♂	VII
18	244	207	108	52·16	114	55·07	158	76·32	44	21·45	14	♂♂	VII
19	242	206	105	50·97	115	55·82	158	76·69	46	22·33	13	♂♂	VII
20	241	208	105	50·48	112	53·84	157	75·48	47	22·59	14	♂♂	VII
21	234	199	100	50·25	109	54·77	156	78·39	43	21·60	15	♀	I
22	232	203	105	51·72	112	55·17	157	77·34	45	22·16	16	♂♂	VII
23	232	199	102	51·25	107	53·76	151	75·87	44	22·11	14	♀♂	I
24	232	197	103	52·28	107	54·31	154	78·17	45	22·84	15	♀♂	I
25	231	199	100	50·25	108	54·27	150	75·37	44	22·11	14	♀♂	I
26	228	198	100	50·50	106	53·53	149	75·25	43	21·71	14	♀♂	I
27	227	196	99	50·51	105	53·57	146	74·49	44	22·44	14	♀♂	I
28	225	193	96	49·74	103	53·36	144	74·61	43	22·28	14	♀♂	I
29	225	193	98	50·77	105	54·40	147	76·16	42	21·76	14	♀♂	I
30	225	192	99	51·56	107	55·72	146	76·04	43	22·39	14	♀♂	I
31	224	193	99	51·29	104	53·88	149	77·20	43	22·28	14	♀♂	I
32	224	192	97	50·52	102	53·12	145	75·52	41	21·35	14	♂♂	I
33	224	191	99	51·83	105	54·97	145	75·91	44	23·03	14	♂♂	I
34	224	190	98	51·57	103	54·21	144	75·78	43	22·63	16	♀♂	III
35	223	191	97	50·78	106	55·49	147	76·96	42	21·99	14	♀♂	I
36	223	191	96	50·26	105	54·97	148	77·48	42	21·99	14	♂♂	I
37	223	191	98	51·30	103	53·92	142	74·34	41	21·46	14	♂♂	I
38	220	188	94	50·00	104	55·32	142	75·53	43	22·87	13	♂♂	I
39	220	187	98	52·40	103	55·08	143	76·47	43	22·99	15	♂♂	I
40	219	188	97	51·59	104	55·32	141	75·00	42	22·34	14	♂♂	I
41	219	186	96	51·61	104	55·91	142	76·34	43	23·11	12	♀♂	I
42	218	190	95	50·00	103	54·21	143	75·26	41	21·57	13	♀♂	I
43	218	188	99	52·66	104	55·32	145	77·12	41	21·80	14	♀♂	I
44	218	188	96	51·06	102	54·25	142	75·53	43	22·87	14	♀♂	I
45	216	185	96	51·89	101	54·59	142	76·75	41	22·16	15	♀♂	I
46	215	183	94	51·36	100	54·64	141	77·04	40	21·85	14	♀♂	I
47	214	183	99	54·09	107	58·47	139	75·95	41	22·40	14	♂♂	II
48	213	185	95	51·35	100	54·05	139	75·13	42	22·70	14	♂♂	I
49	213	182	93	51·09	100	54·94	139	76·37	40	21·97	15	♂♂	I
50	196	168	85	50·59	93	55·35	128	76·19	38	22·61	14	?	I
Mean				51·22		54·31		76·09		21·69	14·19		

ON "WHITEBAIT" COLLECTED IN MENAI STRAIT.

By Andrew Scott, A.L.S.

A number of samples of "whitebait" caught in the weir at Gorad Coch, near the Swillies in Menai Strait, between Anglesey and the mainland, were sent to me for examination in 1914 by Captain Robert Jones, the head fishery officer for that district. The collection lasted for seven months, from March to September, and samples were taken as the fish made their appearance. In some months two samples were taken, in others only one. Altogether ten samples were investigated. It was anticipated that some useful information would be obtained relating to the species of fish included under the general name "whitebait," and would settle the question whether young herring occur amongst the mixture. The results are satisfactory in so far that they show that the whitebait from Gorad Coch are young clupeoid fish, such as sprats and herring, and that young herring 35 to 67 millimetres in length are frequently present. In all probability the herring are hatched in spawning areas at the sea-bottom in some of the bays adjacent to the openings into the Strait. Many more samples will require to be dealt with to determine the frequency of their occurrence, and a careful investigation of the sea-bottom in Carnarvon and Beaumaris Bays would have to be made to find out if herring spawning takes place in these areas.

There is considerable difficulty in determining whether the smaller fish, of about 43 millimetres in length and under, are young herring or some other young clupeoid. They are scaleless and almost transparent. The position of the dorsal fin in relation to the pelvics is

no guide, as it has not taken up its final position. The relative differences in the beginning of the dorsal and pelvic fins in the adult sprat and herring are quite different from what is found in young stages under 43 millimetres. The beginning of the pelvic fins in adult sprats is usually distinctly in front of the dorsal fin. In the case of the herring, the dorsal fin starts in front of the pelvics. Ehrenbaum's figures in "Nordisches Plankton" show that young herring up to 34 millimetres in length have the pelvic fins well in front of the dorsal. It is evident, therefore, that the dorsal fin must change its position and gradually advance nearer the head as the fish grows and the scales make their appearance. This advance of the dorsal fin may not be completed till the young herring reaches a length of 47 millimetres, as shown by the table giving the measurements of the fish collected on May 28th. The only way to distinguish young herring from young sprats before the transformation is complete is by examining considerable numbers of fish. Young scaleless herring can be readily separated from young sprats when mixed up with them, by their bodies being much narrower and ribbon-like.

The following tables give the size of the fish, measured from the tip of the snout to the end of the caudal fin, the distance of the dorsal and pelvic fins from the snout, and the difference in the position of the beginning of the pelvic fins from the snout compared with the dorsal fin. The photographic illustrations 1-10 represent a typical fish from each sample, 11 and 12 the two extreme sizes from a sample collected on May 28th, 13-16 fish of nearly the same length but different character in a collection taken on July 17th.

The illustrations of fish of nearly the same size but different character are useful in showing the marked

distinctions that occur. The two lower fish are young sprats measuring 37·4 and 37·3 millimetres in length. The dorsal fin of the slightly larger one is 0·5 millimetre further away from the tip of the snout than the pelvics. The pelvic fins in the smaller one are 0·2 millimetre further away from the snout than the dorsal. The fish are moderately deep and the scales are developing. I regard the two upper fish as young herring. The first one is 34·6 millimetres long. The second is 38·9 millimetres in length. The dorsal fin of the former is 2·5 millimetres further away from the snout than the pelvics. In the latter it is 2·3 millimetres further away from the snout. Both fish are narrow and ribbon-like, and were probably almost transparent when alive. The scales are not developed. These characters agree well with the description of young herring of this length given by Ehrenbaum. If Ehrenbaum's identification of the young fish is correct, it is quite clear that considerable change takes place in the position of the dorsal fin in relation to the pelvics before the final characters are visible. The dorsal fin represents part of the remains of the embryonic fin which extended along the whole of the back, round the tail and along the ventral margin. The embryonic fin gradually disappears as the larva grows till only the persistent parts known as the dorsal, caudal and ventral fins are left. The pelvic fins are appendages developed after the transformation of the embryonic fin is completed, but before the dorsal part becomes a fixture, and are distinct outgrowths from the body. Their position is probably nearly permanent all through the life of the fish. As the fish grows in length and depth the dorsal fin is gradually pushed forward, and finally becomes attached to some of the dorsal spines of the backbone just above the pelvics.

If we compare the young herring (figs. 13 and 14) and the two young sprats (figs. 15 and 16) with the illustrations representing the first three samples, it can easily be seen that the fish captured on March 11th and 31st and on April 16th are young herring. The fish caught on March 11th measured 34·7 to 45·7 millimetres in length. The next lot taken on March 31st were probably part of the same shoal that visited the strait nearly a fortnight earlier, as the measurements are practically the same. Those caught on April 16th probably belonged to a later hatching than the first two samples, as they are distinctly smaller fish. Their size ranged from 33·7 to 40·7 millimetres. It is scarcely likely that they belonged to the same shoal represented on March 11th, as we would naturally expect larger fish after fully a month's longer time to grow. It will be noticed on looking over the column giving the difference in the pelvic fins that the variation is by no means uniform. Generally, however, the variation in the pelvic fins is greater in the smaller fish than in the larger ones. This ought to be the case if the dorsal fin moves forward as the fish grows. The sample taken on May 14th consisted of a mixture of young herring and sprats. Fig. 4 shows one of the herring. The whole of the fish captured on May 28th were young herring from 42·6 to 67·3 millimetres in length. Figs. 11 and 12 show the largest and smallest fish in this catch. The small one is nearly the same size as the largest fish caught in April, and it is evident that the collection contains at least two distinct generations of herring. The fish caught on June 29th were apparently all sprats, measuring from 23·6 to 43·6 millimetres in length. The sample taken on July 17th contained ninety-seven fish. Eight of them were young herring from 33·7 to 38·9 millimetres. The remainder were sprats from 37·3 to 53·6

millimetres long, with the keeled scales well developed. Only one herring was found in the sample taken on August 8th. The fish caught on August 24th were all sprats. The pelvic and dorsal fins in nearly all the sprats examined in August were exactly the same distance from the snout. The difference in the few exceptions was very slight. The sample taken on September 9th consisted wholly of sprats with well-developed keeled scales. The fish in this collection were typically sprats, but there is considerable variation in the relative positions of the dorsal and pelvic fins. This variation ranged from 0 to 2·3 millimetres, as shown in the table giving the results of the measurements of the September fish.

March 11th, 1914.

Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
45·7	22·2	21·5	0·7 −	40·6	21·1	19·0	2·1 −
44·9	22·1	21·7	0·4 −	40·4	21·2	20·3	0·9 −
44·5	22·0	20·8	1·2 −	40·1	20·5	19·3	1·2 −
44·2	22·1	20·7	1·4 −	40·1	21·1	19·3	1·8 −
43·7	22·1	20·9	1·2 −	40·0	20·9	19·0	1·9 −
43·4	21·7	20·5	1·2 −	39·7	20·4	18·4	2·0 −
43·1	21·9	20·5	1·4 −	39·4	20·1	18·4	1·7 −
43·1	21·4	19·6	1·8 −	39·3	20·3	19·2	1·1 −
42·7	21·9	20·5	1·4 −	39·2	20·2	18·6	1·6 −
42·7	21·9	20·2	1·7 −	39·2	20·4	18·6	1·8 −
42·7	21·9	20·2	1·7 −	38·8	19·9	18·2	1·7 −
42·5	21·5	19·8	1·7 −	38·4	19·9	17·3	2·6 −
42·5	20·9	19·3	1·6 −	38·4	19·9	17·7	2·2 −
42·2	21·3	19·7	1·6 −	38·2	20·1	18·1	2·0 −
41·8	21·4	20·1	1·3 −	38·0	19·6	17·6	2·0 −
41·6	21·3	19·4	1·9 −	38·0	20·3	17·8	2·5 −
41·6	21·1	19·7	1·4 −	37·9	20·0	17·6	2·4 −
41·6	21·3	19·7	1·6 −	37·8	19·5	17·3	2·2 −
41·5	20·9	18·9	2·0 −	37·8	20·1	17·7	2·4 −
41·5	21·0	19·9	1·1 −	37·5	19·6	17·9	1·7 −
41·4	21·3	19·6	1·7 −	37·2	19·8	17·6	2·2 −
41·3	21·5	20·1	1·4 −	37·1	19·5	17·2	2·3 −
40·9	21·4	19·2	2·2 −	36·9	19·6	17·8	1·8 −
40·9	21·0	19·2	1·8 −	36·9	19·8	17·1	2·7 −
40·7	21·2	19·3	1·9 −	36·6	19·5	16·9	2·6 −
40·7	21·0	19·7	1·3 −	35·6	19·1	16·5	2·6 −
40·6	21·6	19·7	1·9 −	34·7	18·7	16·2	2·5 −

March 31st, 1914.

Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
45·7	23·2	22·3	0·9 −	40·8	21·5	20·3	1·2 −
44·7	22·3	22·0	0·3 −	40·8	21·1	19·2	1·9 −
44·4	21·8	21·2	0·6 −	40·6	21·1	19·3	1·8 −
44·2	21·9	20·8	1·1 −	40·5	20·8	19·2	1·6 −
43·9	21·5	20·3	1·2 −	40·5	20·5	18·5	2·0 −
43·7	20·7	20·7	...	40·2	21·1	19·2	1·9 −
43·5	22·3	20·1	1·2 −	40·2	20·9	19·5	1·4 −
43·0	21·6	20·8	0·8 −	40·1	21·0	19·4	1·6 −
42·8	21·2	20·8	0·4 −	40·0	20·6	18·7	1·9 −
42·7	21·0	20·5	0·5 −	39·9	20·9	19·1	1·8 −
42·2	20·8	19·3	1·5 −	39·9	20·9	19·4	1·5 −
41·9	21·2	20·0	1·2 −	39·9	21·1	18·6	2·5 −
41·9	21·6	19·9	1·7 −	39·8	20·8	19·0	1·8 −
41·9	20·9	20·0	0·9 −	39·6	20·7	19·3	1·4 −
41·9	21·4	20·3	1·1 −	39·5	19·7	18·3	1·4 −
41·9	21·2	19·8	1·4 −	39·5	20·1	18·4	1·7 −
41·8	20·5	19·5	1·0 −	39·5	20·0	18·2	1·8 −
41·7	21·7	20·1	1·6 −	39·4	20·2	18·5	1·7 −
41·4	21·1	20·0	1·1 −	39·4	20·2	18·3	1·9 −
41·2	21·7	20·2	1·5 −	39·3	20·1	18·7	1·4 −
41·2	21·3	19·6	1·7 −	39·3	20·0	17·7	2·3 −
41·1	21·0	19·6	1·4 −	38·6	19·5	18·2	1·3 −
41·1	21·1	19·8	1·3 −	38·6	20·5	18·6	1·9 −
41·1	20·7	18·9	1·8 −	38·5	19·3	17·5	1·8 −
41·1	20·9	19·3	1·6 −	37·9	19·8	17·7	2·1 −
41·0	20·8	19·9	0·9 −	36·7	19·3	17·5	1·8 −
40·9	20·8	19·9	0·9 −	35·9	19·3	16·8	2·5 −
40·8	20·7	19·2	1·5 −				

April 16th, 1914.

Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
40·7	21·2	19·2	2·0 −	36·9	18·9	17·0	2·9 −
40·6	21·1	19·2	1·9 −	36·8	19·7	16·6	3·1 −
40·6	21·1	19·2	1·9 −	36·3	19·1	17·3	1·8 −
39·5	20·6	18·3	2·3 −	36·1	19·0	16·6	2·4 −
39·5	21·0	18·6	2·4 −	36·0	18·7	16·1	2·6 −
39·3	19·4	17·5	1·9 −	36·0	19·5	17·3	2·2 −
38·8	19·9	17·8	2·1 −	35·9	18·8	16·8	2·0 −
38·7	20·1	17·7	2·4 −	35·7	19·1	16·4	2·7 −
38·5	19·3	17·4	1·9 −	35·7	19·1	16·9	2·2 −
38·3	20·7	17·9	2·8 −	35·6	18·7	16·6	2·1 −
38·3	19·8	18·2	1·6 −	35·6	18·7	16·4	2·3 −
38·2	20·3	17·2	3·1 −	35·5	19·8	17·1	2·7 −
37·9	19·9	17·9	2·0 −	35·5	19·3	16·3	3·0 −
37·8	19·8	17·4	2·4 −	35·5	19·0	16·6	2·4 −
37·6	20·1	17·7	2·4 −	35·4	19·4	16·4	3·0 −
37·6	19·8	17·7	2·1 −	35·4	19·0	16·4	2·6 −
37·5	19·1	16·6	2·5 −	35·3	19·0	16·3	2·7 −
37·5	19·8	17·4	2·4 −	35·2	18·7	16·9	1·8 −
37·4	19·2	17·3	1·9 −	35·0	18·8	16·0	2·8 −
37·4	19·6	17·1	2·5 −	34·8	18·4	15·9	2·5 −
37·4	19·4	17·5	1·9 −	34·7	18·7	16·2	2·5 −
37·4	19·8	17·8	2·0 −	34·7	18·7	15·9	2·8 −
37·3	19·7	17·2	2·5 −	34·7	18·5	16·2	2·2 −
37·3	19·5	17·1	2·4 −	34·6	18·7	16·2	2·4 −
37·2	19·4	17·4	2·0 −	34·3	18·1	15·6	2·6 −
37·1	19·4	17·4	2·0 −	34·3	18·7	16·1	2·6 −
37·1	19·9	16·9	3·0 −	33·8	18·1	16·6	1·5 −
37·1	19·9	17·4	2·5 −	33·7	17·8	15·4	2·4 −
37·0	19·8	17·8	2·0 −				

May 14th, 1914.

Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
59·8	27·2	28·2	1·0 +	46·8	22·6	21·5	1·1 −
52·9	24·4	24·4	...	46·7	22·3	22·3	...
52·4	24·1	24·7	0·6 +	46·6	22·3	21·8	0·5 −
52·2	23·9	25·0	1·1 +	46·6	22·0	21·6	0·4 −
51·9	23·5	24·2	0·7 +	46·5	22·8	21·1	1·7 −
51·8	23·4	24·5	1·1 +	46·5	22·3	21·0	1·3 −
51·3	23·4	23·9	0·5 +	46·5	22·5	21·6	0·9 −
51·2	23·7	24·1	0·4 +	46·5	22·1	22·1	...
51·1	23·4	24·0	0·6 +	46·3	22·0	21·2	0·8 −
50·1	24·3	25·4	1·1 +	46·2	22·3	21·7	0·6 −
50·1	23·2	23·5	0·3 +	46·1	22·0	22·0	...
49·9	23·2	24·0	0·8 +	45·9	22·8	21·9	0·9 −
49·4	23·0	22·5	0·5 −	45·7	22·5	21·4	1·1 −
49·4	22·4	23·2	0·8 +	45·4	22·2	21·5	0·7 −
48·8	23·3	22·8	0·5 −	45·4	20·5	20·5	...
48·8	23·5	23·5	...	45·2	22·6	21·2	1·4 −
48·4	22·0	22·0	...	44·9	21·2	20·8	0·4 −
48·3	22·5	22·5	...	44·6	21·4	20·2	1·2 −
47·9	23·1	22·6	0·5 −	44·5	22·2	20·6	1·6 −
[illegible]·7	24·2	23·0	1·2 −	44·3	22·3	21·2	1·1 −
41·7	22·6	22·5	0·1 −	44·1	22·2	21·1	1·1 −
[illegible]·4	22·8	22·2	0·6 −	43·9	22·1	20·8	1·3 −
47·4	22·3	22·3	...	43·3	21·8	20·4	1·4 −
47·2	22·7	22·7	...	43·2	21·3	20·1	1·2 −
47·2	22·5	22·5	...	42·7	21·7	19·7	2·0 −
46·8	22·2	22·2	...				

May 28th, 1914.

Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
*67·3	29·7	32·2	2·5+	51·4	23·5	24·3	0·8+
65·7	30·7	32·2	1·5+	51·2	23·0	24·1	1·1+
64·5	28·7	30·6	1·9+	50·9	22·8	24·5	1·7+
58·6	27·2	28·6	1·4+	50·9	23·3	24·0	0·7+
57·9	25·9	27·6	1·7+	50·7	22·5	24·5	2·0+
56·6	25·5	26·6	1·1+	50·5	23·5	24·0	0·5+
56·6	25·9	26·8	0·9+	50·5	22·2	24·0	1·8+
56·5	25·1	26·7	1·6+	50·5	23·0	23·8	0·8+
56·5	24·3	26·2	1·9+	50·3	23·1	23·8	0·7+
56·4	26·0	27·6	1·6+	50·2	23·4	24·9	1·5+
56·3	25·2	26·0	0·8+	50·2	22·7	24·3	1·6+
56·1	25·7	26·7	1·0+	50·2	23·4	23·8	0·4+
55·7	25·5	26·3	0·8+	50·1	23·0	23·3	0·3+
55·7	25·6	27·0	1·4+	50·0	22·6	23·5	0·9+
54·8	24·6	25·5	0·9+	49·8	22·4	23·7	1·3+
53·4	24·4	25·4	1·0+	49·7	22·9	24·0	1·1+
53·2	24·1	25·7	1·6+	49·4	22·9	23·3	0·4+
52·8	24·2	26·0	1·8+	49·3	22·6	23·8	1·2+
52·5	24·7	25·1	0·4+	48·8	22·5	23·6	1·1+
52·0	24·4	25·8	1·4+	48·7	22·5	23·1	0·6+
51·8	23·7	24·8	1·1+	48·0	22·2	23·0	0·8+
51·8	24·0	24·5	0·5+	47·7	22·8	23·2	0·4+
51·7	24·0	25·5	1·5+	47·3	23·0	22·8	0·2−
51·6	23·4	25·3	1·9+	47·1	21·9	22·3	0·4+
51·6	23·3	24·4	1·1+	44·8	20·7	21·0	0·3+
51·6	23·8	25·3	1·5+	44·3	20·5	21·2	0·7+
51·6	23·3	24·8	1·5+	44·3	21·0	20·5	0·5−
51·5	23·6	24·3	0·7+	†42·6	18·7	17·2	1·5−

* Fig. 11, Plate III.

† Fig. 12, Plate III.

June 29th, 1914.

Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
43·6	20·5	20·5	...	37·0	17·1	16·5	0·6 −
43·2	19·8	19·7	0·1 −	37·0	17·5	16·8	0·7 −
41·9	19·5	19·5	...	36·9	18·0	16·8	1·2 −
41·3	18·3	18·3	...	36·6	16·6	15·7	0·9 −
40·7	18·7	18·4	0·3 −	36·6	17·0	17·0	...
40·1	18·6	18·0	0·6 −	36·3	16·8	16·2	0·6 −
39·9	19·0	17·8	1·2 −	36·3	17·5	16·5	1·0 −
39·5	18·5	18·3	0·2 −	36·2	17·6	16·8	0·8 −
39·5	17·9	17·9	...	36·0	17·8	16·6	1·2 −
39·3	18·4	17·5	0·9 −	36·0	17·4	16·5	0·9 −
39·2	18·2	17·3	0·9 −	35·9	17·4	16·6	0·8 −
39·0	19·0	17·6	1·4 −	34·9	16·8	15·9	0·9 −
39·0	18·2	17·8	0·4 −	34·2	17·1	16·9	0·2 −
38·9	18·5	17·6	0·9 −	33·7	16·5	15·5	1·0 −
38·8	18·3	18·3	...	33·1	16·5	15·2	1·3 −
38·7	18·3	17·9	0·4 −	32·9	15·8	14·7	1·1 −
38·6	18·5	17·6	0·9 −	31·8	15·8	14·7	1·1 −
38·4	17·8	17·8	...	31·3	15·4	14·0	1·4 −
38·2	18·4	17·5	0·9 −	31·0	14·3	13·0	1·3 −
38·1	17·0	17·0	...	30·8	15·7	14·2	1·5 −
38·0	17·4	17·4	...	30·6	15·9	13·0	2·9 −
38·0	18·0	16·9	1·1 −	29·7	15·0	13·4	1·6 −
37·7	17·8	16·8	1·0 −	29·2	14·8	12·8	2·0 −
37·5	17·8	16·9	0·9 −	23·9	15·5	13·3	2·2 −
37·5	18·3	17·7	0·6 −	23·6	14·9	13·3	1·6 −

July 17th, 1914.

Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
53·6	25·5	25·5	...	45·4	21·7	21·7	...
53·0	25·6	25·6	...	45·0	21·7	21·7	...
52·5	23·9	23·9	...	44·8	20·7	20·7	...
52·3	24·6	24·6	...	44·7	20·5	20·5	...
51·5	23·7	23·7	...	44·3	20·1	20·1	...
51·5	24·8	24·8	...	44·3	20·1	20·1	...
51·3	24·4	24·4	...	44·2	21·8	21·8	...
51·0	23·9	23·6	0·3 −	44·0	19·9	19·9	...
51·0	23·8	23·8	...	43·8	20·2	20·2	...
50·9	24·3	23·5	0·8 −	43·8	20·5	20·5	...
50·8	24·8	24·8	...	43·7	20·9	20·9	...
50·7	24·2	24·2	...	43·7	20·9	20·9	...
50·7	23·9	23·0	0·9 −	43·7	19·4	19·4	...
50·6	24·1	24·1	...	43·7	20·2	20·2	...
50·1	23·2	23·2	...	43·5	20·3	20·3	...
50·0	22·2	22·2	...	43·4	20·5	20·5	...
49·4	23·2	23·2	...	43·3	20·8	20·8	...
49·2	21·7	21·7	...	43·3	20·1	20·1	...
48·9	22·5	22·3	0·2 −	43·1	19·9	19·9	...
48·8	22·7	22·7	...	43·1	21·7	21·7	...
48·8	24·0	24·0	...	43·1	19·2	19·2	...
48·7	22·0	22·0	...	43·0	20·0	20·0	...
48·7	23·7	22·8	0·9 −	42·7	19·6	19·6	...
48·6	23·7	23·7	...	42·7	20·8	20·8	...
48·6	22·1	22·1	...	42·5	20·3	19·4	0·9 −
48·4	22·8	22·8	...	42·5	19·0	19·0	...
48·3	23·8	23·8	...	42·0	19·1	19·1	...
48·2	22·5	21·7	0·8 −	41·6	19·5	19·5	...
47·9	23·2	23·2	...	41·4	18·9	18·9	...
47·8	23·1	23·1	...	41·1	18·5	18·5	...
47·7	22·7	22·7	...	40·8	18·5	18·5	...
47·6	22·3	22·3	...	40·5	19·5	19·5	...
47·5	21·4	21·1	0·3 −	40·5	19·2	19·2	...
47·5	22·3	22·3	...	40·1	19·0	19·0	...
47·5	21·8	21·8	...	*38·9	18·2	15·9	2·3 −
47·5	21·7	21·7	...	38·7	18·4	18·4	...
47·5	22·9	22·9	...	38·3	17·0	17·0	...
47·3	21·5	21·5	...	38·3	18·0	18·0	...
47·2	22·0	22·0	...	37·5	17·0	17·0	...
47·0	22·4	22·4	...	†37·4	17·5	17·0	0·5 −
46·6	21·7	21·7	...	‡37·3	17·2	17·4	0·2 +
46·6	22·3	22·3	...	36·7	17·4	14·9	2·5 −
46·3	21·0	21·0	...	36·3	17·4	15·2	2·2 −
46·3	21·1	21·1	...	36·0	17·4	14·5	2·9 −
45·8	21·7	21·7	...	35·9	18·0	15·3	2·7 −
45·7	21·9	21·9	...	34·8	17·0	14·6	2·4 −
45·6	20·5	20·5	...	**34·6	17·1	14·6	2·5 −
45·5	21·5	21·5	...	33·7	16·2	13·8	2·4 −
45·5	21·3	21·3	...				

* Fig. 14, Plate III. † Fig. 16, Plate III. ‡ Fig. 15, Plate III. ** Fig. 13, Plate III.

August 8th, 1914.

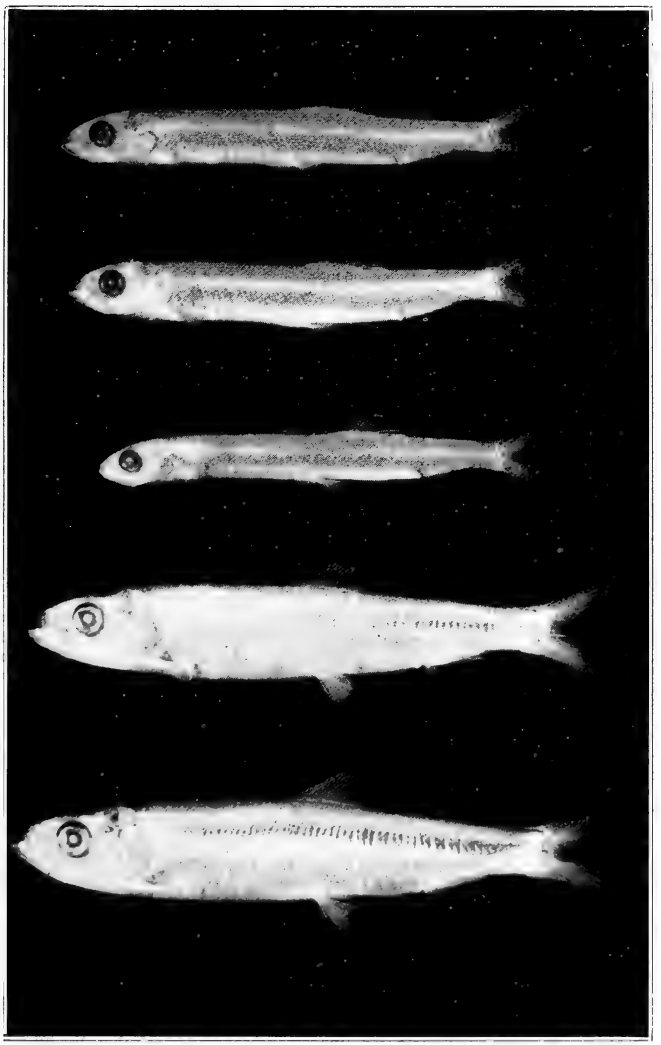
Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
52·1	23·2	23·2	...	46·1	20·3	20·3	...
51·5	23·1	23·1	...	45·9	20·2	20·2	...
50·9	25·7	25·7	...	45·8	20·7	20·7	...
50·7	22·8	22·8	...	45·8	21·5	21·5	...
50·6	25·0	25·0	...	45·4	20·2	20·2	...
50·3	21·9	21·9	...	45·4	21·0	21·0	...
50·2	24·4	24·4	...	45·3	18·4	18·4	...
49·9	22·4	22·4	...	45·2	19·9	19·9	...
48·8	22·3	22·1	0·2 −	45·2	20·5	20·5	...
48·8	22·5	22·5	...	45·1	20·3	20·3	...
48·8	21·7	21·7	...	44·9	20·8	20·8	...
48·5	21·9	21·9	...	44·8	20·0	20·0	...
48·5	22·2	22·2	...	43·9	20·5	20·5	...
48·1	21·4	21·4	...	43·3	19·2	19·2	...
48·1	21·8	21·8	...	42·3	18·8	18·8	...
48·0	20·8	20·8	...	42·0	18·7	18·7	...
47·4	22·3	22·3	...	41·2	19·4	19·4	...
47·3	21·9	21·9	...	40·7	15·7	17·7	2·0 +
47·0	21·5	21·5	...	39·1	19·4	19·4	...
46·8	21·5	21·5	...	38·7	16·6	16·6	...
46·7	21·0	21·0	...	38·5	17·6	17·6	...
46·5	21·5	21·5	...	38·3	16·7	16·7	...
46·3	21·4	21·4	...	36·0	16·2	16·0	0·2 −
46·2	20·7	20·7	...				

August 24th, 1914.

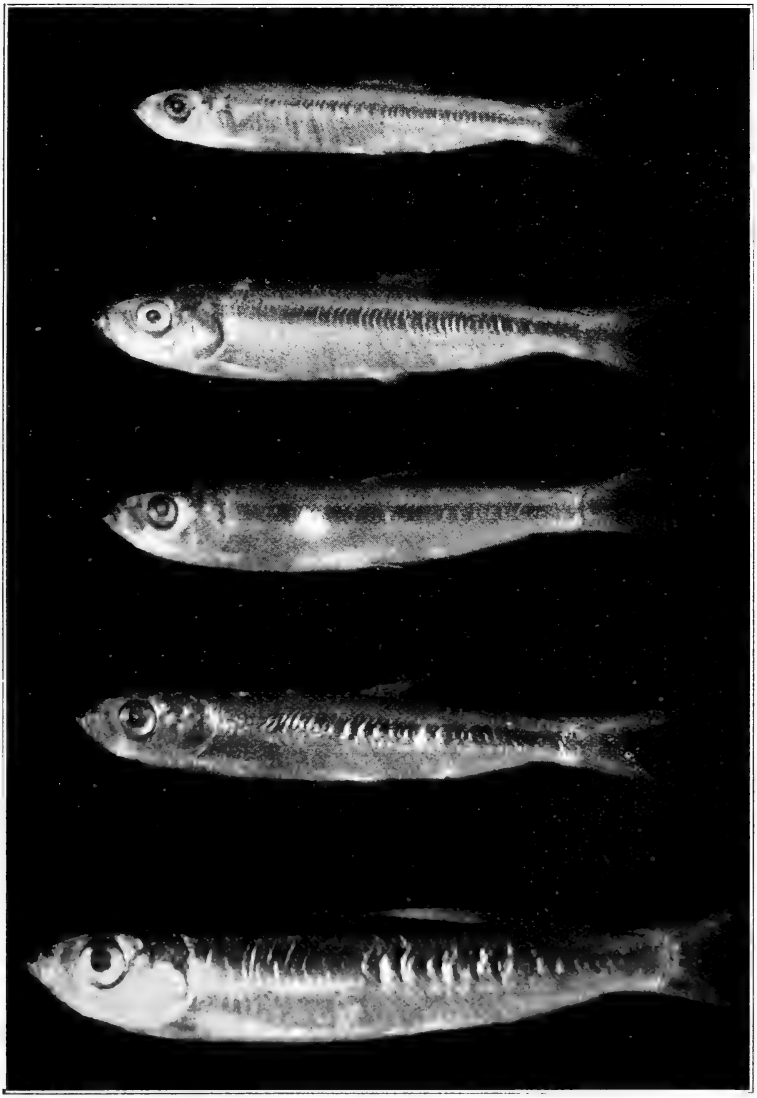
Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
58·2	25·9	25·7	0·2 −	48·8	21·8	21·8	...
57·2	25·8	25·8	...	48·8	22·0	22·0	...
56·5	26·3	26·3	...	48·7	22·0	21·8	0·2 −
56·1	25·5	25·5	...	48·3	22·6	22·6	...
55·7	25·5	25·5	...	47·8	20·7	20·7	...
55·4	25·8	25·8	...	47·2	22·0	21·8	0·2 −
55·0	25·6	25·6	...	46·7	21·3	21·3	...
54·5	24·8	24·6	0·2 −	46·4	21·0	21·0	...
53·7	24·9	24·9	...	46·3	21·3	21·3	...
53·0	25·0	24·2	0·8 −	45·8	21·2	21·2	...
52·7	23·7	23·7	...	45·8	22·7	21·1	1·6 −
52·7	25·0	24·8	0·2 −	44·8	22·8	22·8	...
52·6	23·4	23·4	...	44·8	21·7	21·7	...
52·6	23·9	23·9	...	44·7	20·2	20·2	...
52·5	24·0	24·0	...	44·5	20·5	20·5	...
52·5	23·0	23·0	...	44·4	21·0	21·0	...
52·4	24·4	24·4	...	44·3	20·3	20·3	...
52·3	24·0	24·0	...	44·1	19·4	19·4	...
51·7	24·0	24·0	...	44·0	20·0	19·8	0·2 −
51·1	23·7	23·6	0·1 −	44·0	21·0	21·0	...
50·3	23·0	23·0	...	43·5	19·5	19·5	...
50·3	23·1	23·1	...	43·5	20·3	20·3	...
50·3	22·4	22·4	...	42·5	19·3	19·3	...
49·8	22·3	22·3	...	42·3	19·1	19·1	...
49·4	22·5	22·5	...	41·5	20·5	20·5	...
49·3	23·4	23·4	...	41·2	18·3	18·3	...
49·2	22·4	22·4	...	40·5	18·9	18·9	...

September 7th, 1914.

Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −	Total length of fish.	Snout to beginning of dorsal fin.	Snout to beginning of pelvic fins.	Pelvic fins. + or −
mm.	mm.	mm.		mm.	mm.	mm.	
80·0	37·4	36·8	0·6 −	66·0	31·8	31·4	0·4 −
76·5	34·6	33·0	1·6 −	65·8	32·9	32·4	0·5 −
74·2	35·5	34·8	0·7 −	65·6	31·4	29·9	1·5 −
73·2	35·2	33·2	2·0 −	63·0	29·7	29·3	0·4 −
73·0	35·5	33·2	2·3 −	62·3	29·0	28·8	0·2 −
70·2	32·3	31·4	0·9 −	62·3	28·7	28·1	0·6 −
69·3	31·6	31·4	0·2 −	61·4	29·5	27·8	1·7 −
69·2	31·6	31·0	0·6 −	60·5	27·8	27·7	0·1 −
68·4	32·0	31·3	0·7 −	57·3	26·7	26·0	0·7 −
68·3	31·3	30·7	0·6 −	56·2	26·7	25·4	1·3 −
68·1	31·1	29·9	1·2 −	55·6	26·4	26·4	...
67·5	31·6	31·5	0·1 −	54·1	25·4	24·5	0·9 −
67·2	30·8	30·2	0·6 −	53·2	24·1	23·7	0·4 −
66·8	32·3	30·6	1·7 −	49·7	24·0	23·1	0·9 −



Typical Herring from each sample, March to May.



6.

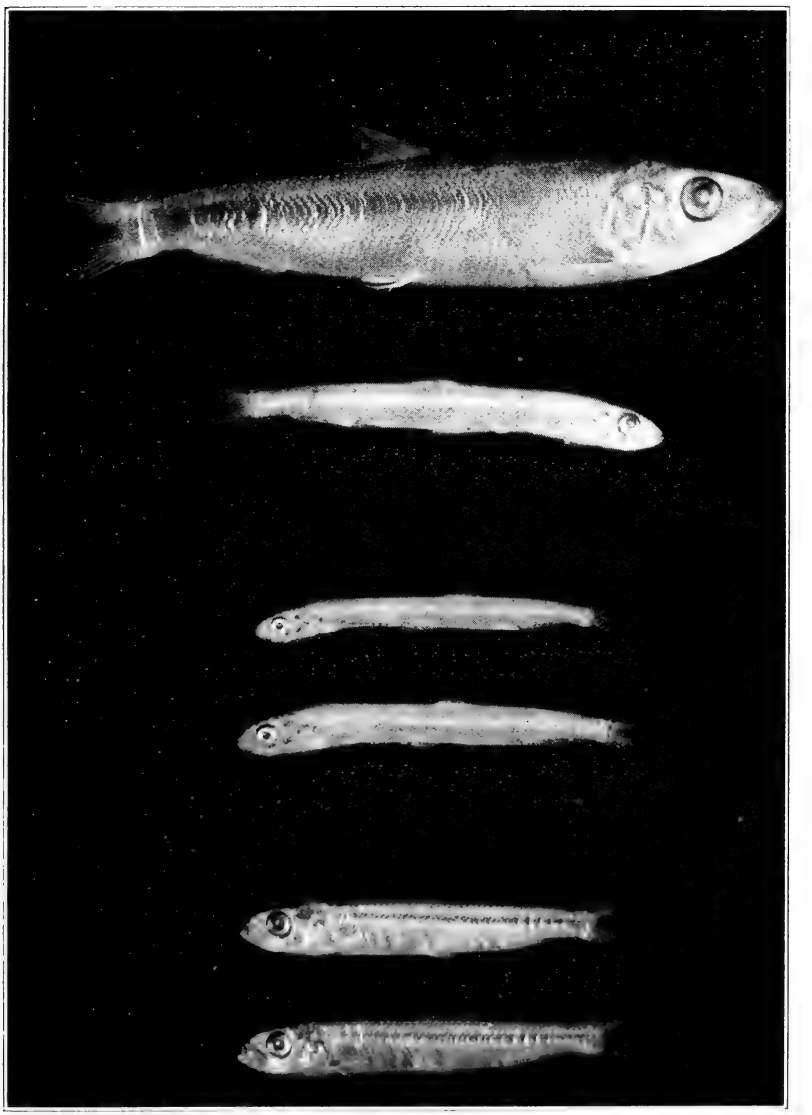
7.

8.

9.

10.

Typical Sprat from each sample, June to September.



FIGS. 11 and 12. Largest and smallest fish, May 28th.
,, 13 and 14. Young Herring, July 17th.
,, 15 and 16. Young Sprats, July 17th.

EXPLANATION OF PLATES.

Plate I.

Fig. 1. Young Herring. March 11th, 1914.
Fig. 2. ,, ,, ,, 31st, ,,
Fig. 3. ,, ,, April 16th, ,,
Fig. 4. ,, ,, May 14th, ,,
Fig. 5. ,, ,, ,, 28th, ,,

Plate II.

Fig. 6. Young Sprat. June 29th, 1914.
Fig. 7. ,, ,, July 17th, ,,
Fig. 8. ,, ,, Aug. 8th, ,,
Fig. 9. ,, ,, ,, 24th, ,,
Fig. 10. ,, ,, Sept. 7th, ,,

Plate III.

Figs. 11 and 12. Largest and smallest fish. May 28th.
Figs. 13 and 14. Young Herring. July 17th.
Figs. 15 and 16. Young Sprats. July 17th.

All the figures are magnified about one-third.

THE FAT-CONTENT OF IRISH SEA HERRING.

By Jas. Johnstone, D.Sc.

Throughout the period when biometric investigations on Irish Sea herrings were made by Mr. W. Riddell, fat-analyses of the muscle substance of the fish were also made. Samples of herring from the summer fishery near the south end of Isle of Man, from the winter fishery in Carnarvon and Cardigan Bays, and from the sprat fisheries in Morecambe Bay, and in the Estuary of the Mersey have been examined. Several interesting points suggesting further investigation have arisen in connection with this research, but so far time has not been available for their proper treatment. I hope, however, to return to this subject when the summer fishery of 1915 begins. The investigation may be of some importance in relation to methods of food preservation, a subject which indeed suggested it in the first instance.

The analyses were made by means of the ordinary Soxhlet apparatus, using carbon tetrachloride as a solvent instead of ether. The extreme oiliness of some of the fish made the sampling a matter demanding some care. The muscle substance could not easily be chopped up, because much oil would have been lost in the process. The herring to be sampled was, therefore, lightly scrubbed with a test-tube brush so as to remove the scales. The skin was then dried with a towel, and, by means of a sharp razor, a series of cuts were made through the flesh as close together as possible. A tangential cut was then made so as to free these thin sections of muscle substance, which were then lifted by clean forceps and put into a paper thimble contained in a weighing bottle. Both thimble and bottle had previously been dried in the water oven and weighed. The whole was then weighed and the weight of the sample of muscle substance obtained by

difference. The bottle with its contents was now dried in the water oven until the weight was constant, a matter usually of about 20 hours, or more. The fat was then extracted.

In the richest samples the oil oozed out through the thimble in the process of drying, and it was necessary to rinse out the weighing bottle, adding the solvent to the extraction flask. It was also necessary to plug the opening of the thimble lightly with dry cotton wool to prevent the breaking away of small fragments of the dried tissue and the entrance of these into the extraction flask through the siphon tube. In extraction the flask and Soxhlet were wrapped round with a towel so as to keep the temperature of the solvent as high as possible—it could almost be made to boil in the Soxhlet by this method. Extraction was usually carried on for two or three hours by which time all the fat was removed. Indeed after the third siphoning all the colour had usually disappeared from the solvent in the Soxhlet.

The flask containing the solution was then detached and the solvent distilled away. The flask was then dried in the steam oven till its weight was constant. This process was hastened considerably by frequent blowing into the flask by means of a small hand bellows. The heavy vapour of carbon tetrachloride was thus removed. As a rule constancy of weight was attained in about 8 to 10 hours. Checks on the accuracy of the analyses were made by drying and weighing the residues. The latter were also stored in order that Kjeldahl nitrogen analyses might be made, but no time was available for this latter work. The error in the analyses is, I think, fairly small; and when the individual differences of fish and fish are considered it may be concluded that further refinements in the methods of estimation would be futile.

The results are given in tables I to III. From the dates of the analyses the samples can be identified, and a comparison with Mr. Riddell's results (published in this report) can be made. The degree of ripeness of the fish and the relation of the sample

to the time of spawning can thus be ascertained. At the beginning of the series of analyses the herrings were mostly ripening fish in the condition denoted by Hjort's Nos. II and III in his scale of ripeness. At the end of the analyses the herrings were mostly spent fish.

Table I. Manx Herrings (Mature).

Date.	Sex, &c.	Weight of muscle substance.	Dry weight.	Weight of oil.	Percentage of oil.	Percentage of water.
1914						
June 3	Unripe ♂	7·195	...	0·383	5·3	...
,,	,, ♀	4·790	...	0·173	3·6	...
June 25	Unripe ♂	7·225	...	2·008	27·8	...
,,	,, ♀	8·264	...	2·205	26·8	...
July 2	Unripe ♂	7·793	3·309	1·901	24·4	57·5
,,	,, ♀	6·383	...	1·882	29·5	...
July 9	Unripe ♂	5·737	2·958	1·863	32·5	48·4
,,	,, ♀	6·688	3·644	2·339	34·9	46·6
July 31	Full ♂	6·140	2·380	1·038	16·9	61·2
,,	,, ♀	3·577	1·979	0·950	26·5	42·3
Aug. 22	Full ♂	5·258	2·442	1·246	23·7	53·5
,,	,, ♀	5·453	2·162	0·964	17·6	62·3
Sept. 4	Spawning ♂♀	4·869	1·782	0·791	16·3	63·4
,,	,,	4·972	1·980	0·902	18·1	60·2
Sept. 30	Spent ♂♀	5·669	2·043	0·512	9·0	63·9
,,	,,	5·635	1·897	0·503	8·9	66·3

Table II. Welsh Herrings (Mature).

Date.	Sex, &c.	Weight of muscle substance.	Dry weight.	Weight of oil.	Percentage of oil.	Percentage of water.
1914						
Oct. 27	Full ♂	4·846	1 755	0·901	18·6	63·8
,,	,, ♀	6·175	1·904	0·698	11·3	69·1
Nov. 19	Full ♂	4·823	1·911	1·086	22·5	60·4
,,	,, ♀	6·399	1·993	0·758	11·8	68·9
Nov. 20	Full ♂	5·506	2·015	1·054	19·1	63·4
,,	,, ♀	5·394	1·886	0·891	16·5	65·0
Dec. 9	Full ♂	5·472	1·978	1·019	18·6	63·8
,,	,, ♀	3·946	1·186	0·462	11·7	69·9
Dec. 18	Virgin ♂♀	4·038	1·252	0·416	10·3	68·9
,,	Spent ♀	4·517	1·296	0·349	7·7	71·3
1913						
Dec. 10	♂	2·866	...	0·117	4·1	...
,,	Full ♂	2·887	...	0·255	8·8	...
,,	♂	4·485	...	0·556	1·2	...
Dec. 19	Spent ♀	4·580	...	0·127	2·8	...
,,	,, ♀	3·594	...	0·313	8·7	...

Table III. Sprats and Immature Herrings.

Date.	Sex, &c.	Weight of muscle substance.	Dry weight.	Weight of oil.	Percentage of oil.	Percentage of water.
		Morecambe Immature Herrings.				
1914						
May 24	...	2·766	...	0·085	3·0	...
May 30	...	4·857	...	0·202	4·1	...
,,	...	3·108	...	0·101	3·2	...
		Morecambe Sprats, Immature.				
May 29	...	2·136	...	0·205	9·5	...
		Mersey Estuary Sprats, Mature.				
Jan. 27	...	5·230	1·566	0·624	11·9	70·0

Table I gives the results of analysis of the summer-caught herrings sent from Isle of Man. In the earlier analysis the tissue was dried so as to facilitate the fat extraction, but weighings were not made. Male and female fish were usually taken from each sample, and the separate analytical results are given. But, on looking at this and the other tables, one sees no significant sexual differences, so that the means of the separate estimations of male and female fish have been taken in making the graph of the results. Table II deals with winter-caught herrings from the Welsh Bays, and includes a few results obtained in December of 1913. Table III gives only a few results of analysis of small herrings and sprats from Morecambe Bay and the Mersey Estuary. The herrings were all immature. The Morecambe Bay sprats (almost "whitebait") were also very small. The Mersey sprats were large and mature fish with gonads fast approaching maturity. They would probably have spawned in the spring of this year.

The results of the tables, as regards the Manx and the Welsh fish, are represented in the graph which follows this.

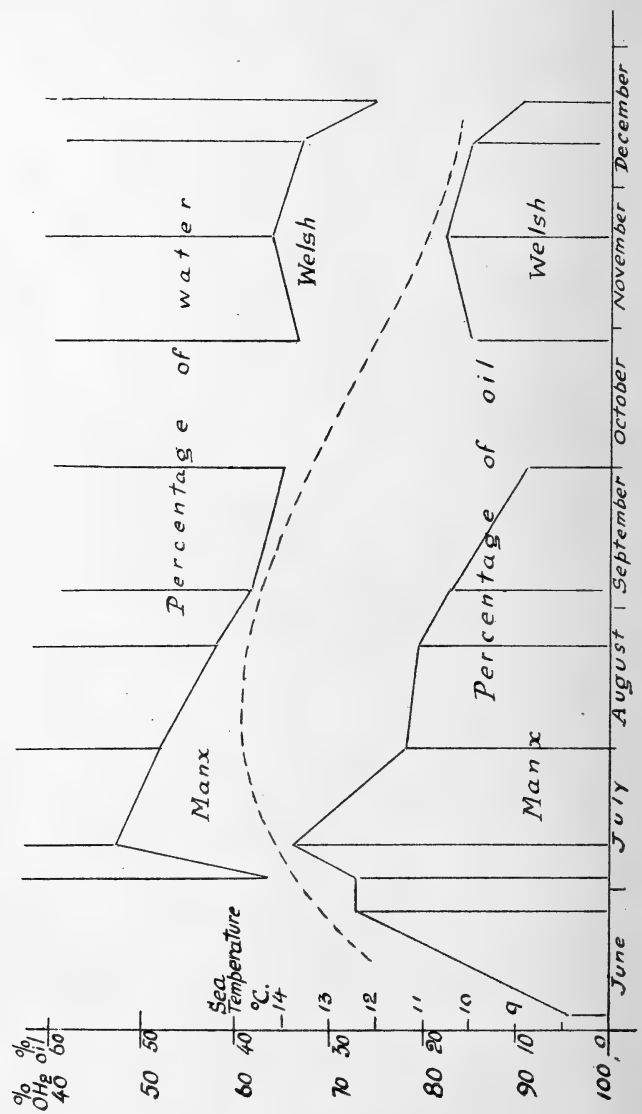


Fig. 1. Graphical representation of the variations in fat and water in the muscles of Manx and Welsh herrings. The broken line represents the variation in temperature of the sea at Carnarvon Bay Light Vessel.

We see, at once, some interesting relationships. It is clear, in the first place, that the variations of fat and water in the muscle substance of the fish are very closely complementary to each other.* The scale for the water contents has been reversed so as to show this relationship the more clearly. It is very apparent in the case of the Manx herrings, and much more so in the case of the Welsh herrings. As the fat-content increases, the water-content decreases, and *vice versa*. Now this is what one would expect. Carbohydrates are practically absent in most sea fishes; not even in the liver of well-fed herrings can these substances be detected.† The proportion of protein we may suppose, without evidence to the contrary, will tend to remain constant, so that it must be the water in the flesh which undergoes seasonal variations. This is not quite the case, as the graph shows, and there must be an appreciable variation in the percentage of protein in the muscle substance. Nevertheless the variation is slight compared with that of the water and fat.

To a certain extent the latter variation is similar, in its progress, to that of the sea-temperature. The latter cannot be exactly estimated, since the positions of the herring shoals were not ascertained when the samples were forwarded to us. Further these positions were, no doubt, variable, being nearer to the land at one time than at others, and this would make an appreciable difference in the temperature curve. The sea-temperature is, therefore, that at Carnarvon Bay Light Vessel, a position where the land hardly influences the annual variation of temperature, and which may be taken as generally representative of the water in the open sea round the south end of Isle of Man. There is a general relationship between

* This confirms Milroy's results of analysis of herrings made for the Scottish Fishery Board (see *24th Ann. Rept. Fishery Board for Scotland*, 1905 (1906), Pt. III, pp. 83-107. Also *25th Ann. Rept.*, 1906 (1908), Pt. III, pp. 197-208.

† See Stirling, *2nd Ann. Rept. Fishery Board for Scotland*. Appdx. F., No. I. pp. 31-46, Plates I and II, 1883 (1884).

fat-contents and sea-temperature, that is, metabolism is more intense the higher the temperature of the medium. Still the correspondence is not very close and we must find some other factor governing the variation in fat contents.

This latter factor is the reproductive cycle. The period preceding the final maturation of the genital products is marked by a large increase in fat-contents of the tissues of the fish, and the act of spawning is accompanied by a fairly rapid decrease in this fat-contents. We might generalise these facts by saying that the fish accumulates fat in its tissues in order that this stored nutritive matter might be drawn upon for the maturation of the eggs and spermatozoa (in the herring the masses of these products are about equal): the protein and fat present in the reproductive glands must come from somewhere. Now this generalisation cannot be quite true. The ratio of protein to fat in the ovaries increases greatly as the genital organs ripen, and the increase in mass of these organs is, therefore, not altogether due to the transference of fat from the muscular tissues to the genital organs. Further the greatest decrease in fat-contents occurs after spawning, when this stored nutritive material is no longer required in the reproductive process. Yet again there must be some relation between the spawning operation and the general metabolism of the animal in order that this change may occur. Evidently there are two independent factors which influence the metabolism of the fish (1) the variations of sea temperature, and (2) the reproductive cycle. It is certain that even virgin herrings would show as well marked a rise and fall of fat-contents as do sexually mature fish, thus virgin plaice do certainly show marked variations in "condition" which corresponded with seasonal changes in the sea. But, all the same, the maturation of the ovaries and testes and act of spawning do certainly influence the general metabolism of the animals. The rapid decrease of fat at the time of spawning

cannot be explained satisfactorily, for the final stage of maturation of the ova probably consists mainly in the imbibition of water from the general circulating fluids of the body.

We have also to notice the very high proportion of fat in these herrings. In July over one-third of the wet weight of muscle from a female fish consisted of fat. The percentage of fat was 34·9, that of water was 46·6, and if we take Milroy's average value of protein in the muscles of female herrings as 18 % these numbers will add up to 99·5 %. Comparing Table I with other published analyses of herrings we find the fat percentages very high, and this statement applies, to a less extent, also to the Welsh winter-caught herrings. Even the famous Loch Fyne summer herrings are less rich in fat than are these Manx ones, and one's own experience in merely handling the fish agrees with these analytical results.

The reason that the Loch Fyne herrings are less oily than the Manx ones is not that there is a corresponding difference in the planktonic food present in these sea areas. The summer crustacean plankton of Loch Fyne appears to be much denser than that round the south end of Isle of Man. The difference appears to depend on the fact that the spawning period of the Manx herrings occurs very shortly after the maximum of sea-temperature, so that the two factors, the increase of metabolism due to rise of temperature, and that due to the ripening genital products, work together in the same direction. Herring do spawn in Loch Fyne, but this appears to be rather exceptional—the shoals migrate out from the Loch in order to spawn. Only the temperature factor therefore operates in augmenting the fat-contents, and the temperature of Loch Fyne is, throughout the summer and autumn, lower than that of the sea to the south of Isle of Man.

Some other points of interest in relation to the morphology and bio-chemistry of the fat tissues are being investigated, but the opportunity has not yet occurred to complete this work.

REPORT ON THE PERIODIC SAMPLES OF SHRIMPS FROM THE MERSEY ESTUARY.

By T. Monaghan.
Assistant Naturalist in the Fisheries Laboratory.

These investigations were first carried out during 1912 and part of 1913, but a definite result could not be obtained on account of the insufficiency and irregularity of the samples sent to the laboratory. The result of the observations for 1912-13 were published in the Annual Report for 1913. This year the samples have been sent in with greater regularity so that the observations are more complete.

The samples were taken from the Rock Channel and Crosby Channel, both of which may be included in the term "Mersey Estuary." It will be noticed from the data (Table I) that, with a few exceptions, two samples were sent for examination each month, thus enabling me to compare the variation in numbers of the sexes more closely than in 1913. As each sample was sent the shrimps were first sorted out in their sexes and then counted. They were measured (from the tips of the antennules to the extreme end of the tail) in millimetres, and averages were determined for each group. At the same time a microscopic examination was made of the eggs on the berried females, and four different stages of development were recorded, viz. :—(1) Segmentation commencing. (2) The eye forming. (3) Eye fully developed and the appendages just showing. (4) Yellow and black pigment formed, the larvae ready to hatch. A number of eggs in each stage were then measured. These results are, however, not yet completed, and are consequently not given in this Report.

Table I shows the data as to samples obtained, and the average lengths of the berried females, non-berried females and males in each sample.

Table II gives the percentages of berried females, non-berried females and males for each month.

TABLE I.

Date, 1914.		Percentage of Females carrying eggs.	Number.	Average Length in mm.
January 9th	Berried Females	69·6	394	74·2
	Non-berried Females ...		172	65·5
			566	
	Males		234	53·7
			800	
February 3rd ...	Berried Females	50·8	230	63·0
	Non-berried Females ...		222	53·5
			452	
	Males		823	45·6
			1,275	
March 5th	Berried Females	50·2	204	65·3
	Non-berried Females ...		202	54·4
			406	
	Males		1,770	53·7
			2,176	
March 23rd	Berried Females	82·3	234	67·5
	Non-berried Females ...		50	63·5
			284	
	Males		253	52·0
			537	
April 15th	Berried Females	68·2	206	70·0
	Non-berried Females ...		96	64·2
			302	
	Males		752	52·7
			1,054	
April 30th	Berried Females	50·9	208	71·2
	Non-berried Females ...		200	58·0
			408	
	Males		1,150	54·5
			1,558	

TABLE I.—*Continued.*

Date, 1914.		Percentage of Females carrying eggs.	Number.	Average Length in mm.
May 12th	Berried Females	81·6	428	69·7
	Non-berried Females ...		95	61·7
			523	
	Males		384	53·0
			907	
May 27th	Berried Females	67·5	370	65·3
	Non-berried Females ...		178	66·0
			548	
	Males		1,152	55·7
			1,700	
June 12th	Berried Females	76·0	428	68·5
	Non-berried Females ...		135	58·0
			563	
	Males		131	52·5
			694	
June 26th	Berried Females	77·9	420	68·0
	Non-berried Females ...		119	59.5
			539	
	Males		47	51·0
			586	
July 9th	Berried Females	52·5	238	71·0
	Non-berried Females ...		215	66·7
			453	
	Males		170	56·5
			623	
July 27th	Berried Females	76·3	426	72·2
	Non-berried Females ...		132	65·2
			558	
	Males		30	50·2
			588	

TABLE I.—*Continued.*

Date, 1914.		Percentage of Females carrying eggs.	Number.		Average Length in mm.
August 7th	Berried Females	36·4	310		67·7
	Non-berried Females ...		540		59·0
				850	
	Males		1,000		48·75
			1,850		
September 9th ...	Berried Females	38·6	242		72·0
	Non-berried Females ...		384		61·7
				626	
	Males		700		52·0
			1,326		
September 30th...	Berried Females	28·7	121		73·5
	Non-berried Females ...		300		66·7
				421	
	Males		355		55·5
			776		
October 15th ...	Berried Females	24·1	74		77·0
	Non-berried Females ...		272		69·7
				346	
	Males		312		57·7
			658		
October 30th ...	Berried Females	5·2	14		74·6
	Non-berried Females ...		252		72·5
				266	
	Males		700		56·5
			966		
November 17th ...	Berried Females	1·8	8		76·2
	Non-berried Females ...		434		68·5
				442	
	Males		94		55·2
			536		

TABLE I.—*Continued.*

Date, 1914.		Percentage of Females carrying eggs.	Number.	Average Length in mm.
December 1st.....	Berried Females	7·5	78	69·2
	Non-berried Females ...		952	65·7
			1,030	
	Males		500	49·2
			1,530	
December 16th ...	Berried Females	24·6	75	70·5
	Non-berried Females ...		230	67·5
			305	
	Males		325	54·2
			630	
1915 January 26th ...	Berried Females	69·2	319	71·0
	Non-berried Females ...		142	68·0
			461	
	Males		291	55·0
			752	
February 9th.....	Berried Females	63·6	154	71·2
	Non-berried Females ...		88	62·0
			242	
	Males		678	54·2
			920	
February 23rd ...	Berried Females	73·9	256	67·7
	Non-berried Females ...		90	61·7
			346	
	Males		558	54·2

TABLE II.

1914.	Percentage of Males.	Percentage of Berried Females.	Percentage of Non-berried Females.
January (1)	29·2	49·2	21·5
February (1)	64·5	18·0	17·4
March (2)	74·5	16·1	9·2
April (2)	72·8	15·8	11·3
May (2)	58·9	30·6	10·4
June (2)	13·9	66·2	19·8
July (2)	16·5	54·8	28·6
August (1)	54·0	16·7	29·1
September (2)	50.1	17·2	32·5
October (2).....	62·3	5·4	32·2
November (1)	17·5	1·4	80·9
December (2)	38·1	7·0	54·7

Considering now the data given in the tables we may make some conclusions with respect to the spawning period of shrimps in the Mersey Estuary.

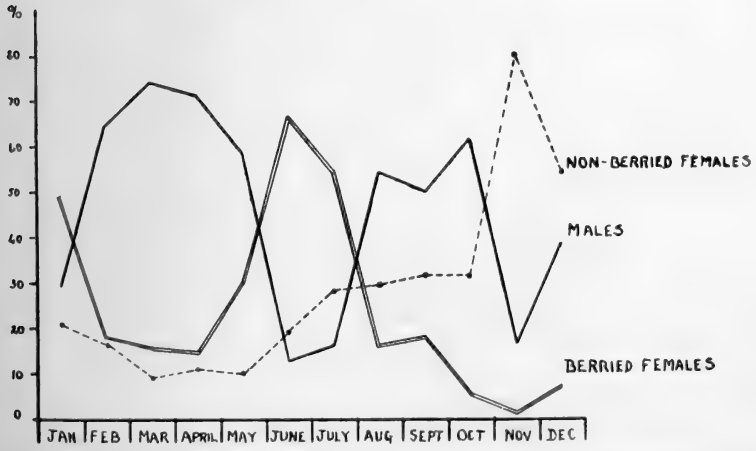


FIG. 1. Variations in the relative numbers of males, non-berried females, and berried females in the samples examined during 1914.

Fig. 1 is constructed from the data of Table II. The percentages plotted are calculated on the whole sample, and these figures may be necessary for a detailed consideration of the life-history of the shrimp in local waters. I do not consider it further in the meantime. It will be obvious, however, that the variation in the percentage of berried females in the whole catch may not give us accurate information as to the period of maximum spawning, for it is based on the males and females. We have, therefore, to calculate the percentage of females that carry eggs and regard this (provisionally) as giving information as to the period of the year during which spawning is in progress.

The data of 1913 and 1914 have therefore been combined, and some additional samples obtained in 1912 and 1915 are also included. The average percentages of female shrimps carrying abdominal eggs (berried females) have been calculated for each month. These new average percentages are as follows :—

Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May.	June.	July.	Aug.	Sept.	Oct.
3·2	23·4	66·3	62·7	70·7	68·9	74·9	69·0	73·4	41·9	33·7	7·6

These figures are represented by the following graph :—

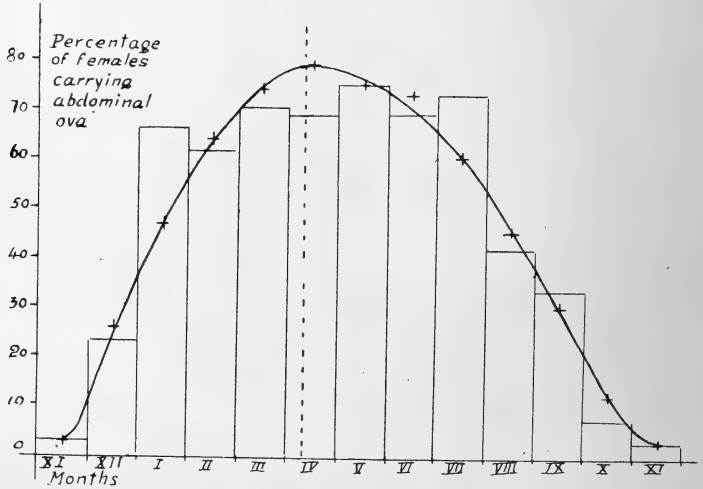


Fig. 2. Variation in the percentage of female shrimps carrying abdominal eggs during the years 1912-1915.

We may consider these data more closely. The columns represent the data of the above table. The +'s represent the rough data smoothed by the method described by Dr. Johnstone in last year's Report.* The mode, or time of maximal spawning, is about 10th April, when about 75 % of all the female shrimps' sample were found to be carrying abdominal eggs. The minimum proportion of berried females was observed in the samples taken during November, and this was the case in each of the years (1912, 1914 and 1915) in which samples were taken in that month. The proportion of berried female shrimps rises from the minimum towards the maximum rather more rapidly than it drops from the maximum towards the minimum.

The spawning period seems, therefore, to be a prolonged one, and indeed it is common experience everywhere off the Lancashire Coasts to find berried females throughout the year. But it does not follow that it is so prolonged as the figure indicates for, having extruded its eggs, the female carries them for a considerable time attached to her swimmerets. It is not known how long a period the eggs require for incubation, but according to Ehrenbaum* it may be four weeks (or even less) in summer, and four to five months in the winter. This is for the German North Sea Coast, and since the variation in the length of the incubation period depends on the temperature of the sea Ehrenbaum's estimates will probably be approximately true also for the Mersey Estuary shrimps.

During the spring and summer when the sea temperature is rising rapidly the proportion of female shrimps carrying abdominal eggs will, therefore, tend to be diminished by the hatching out of the larvae. This is, of course, also the case during the latter half of the year when the sea temperature

* *Rept. Lancashire Sea-Fish. Laby.* for 1913, p. 83.

* "Zur Naturgeschichte von *Crangon Vulgaris* Fabr." *Mitth. f. Sektion f. Küsten-und Hochsee fischerei, Deutschen Fischerei-Vereins*, Jahrgang, 1890. Berlin, 1890.

is falling, but the proportion will diminish far less rapidly than in the spring and summer, for the incubation period during the month of minimum sea-temperature is at least four times greater than during the month of maximum sea-temperature. If we could allow for this variability the curve of spawning of fig. 2 would fall more rapidly than it appears to do, that is to say, the spawning period would not be so prolonged as the figure indicates.

With the exception of Ehrenbaum's well-known work no good investigation of the life-history of the North European shrimp has been made, so that we do not know how long the incubation period is, nor how it varies with the temperature. Further, the stages of development are not so well-known that we can say exactly at what time, prior to the time of observation, an egg was spawned. By counting only new-laid ova we could, of course, make an estimate of the variations in the spawning time, and it was with this object that observations of the stages of embryonic development were made. But it will be necessary to carry on these observations, first of all, in an aquarium.

According to Ehrenbaum there are two spawning periods in the year with respect to the shrimps in the Heligoland Bight, the first from the middle of April to the beginning of June, and the second during October and November. It is not impossible that there may also be two chief spawning periods in Liverpool Bay, but the observations made so far do not appear to show that this is the case.

[Percy Sladen Memorial Trust Research.]

STUDIES OF CERTAIN PHOTO-SYNTHETIC PHENOMENA IN SEA-WATER.

I.—SEASONAL VARIATIONS IN THE REACTION OF SEA-WATER IN RELATION TO THE ACTIVITIES OF VEGETABLE AND ANIMAL PLANKTON.

II.—THE LIMITATIONS OF PHOTO-SYNTHESIS BY ALGAE IN SEA-WATER.

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Our attention was first drawn to the seasonal variations in the alkalinity of sea-water recorded in this paper, and to the remarkable degree to which green algae can reduce the hydrogen-ion concentration of sea-water in which they are grown, by preliminary observations made by Moore, Edie and Whitley at Port Erin in the Spring of 1912.

A severe epidemic of a disease, characterised by large areas or rounded spots of ulceration on the skin, had killed a considerable number of the plaice used for spawning purposes in the pond attached to the Fish Hatchery, although this pond had an abundant daily supply of fresh sea-water.

At the request of Professor Herdman the water of the pond was examined chemically, and the only important fact discovered was that it was much more alkaline than the water of the Bay. At the same time the pond-water was green in colour from the presence of floating mono-cellular algae, and a minute green flagellate Infusorian in great profusion, while,

as usual at this time of year, the sea-water in the Bay was almost free from green organisms. It was also observed that the water of the Bay was less alkaline to phenol-phthalëin than it had been on previous occasions when titrated in connection with other work.

For these reasons, it was determined to follow up the reaction of the sea-water at intervals throughout the round of the year, and also to make a series of observations as to the speed with which green organisms added to a confined volume of sea-water increased the alkalinity, and the limit to which the alkalinity could be so raised before photo-synthetic action ceased, or the organism perished.

I.—Seasonal Variations in the Alkalinity of Sea-Water.

Certain of the observations were carried out by Moore, Edie, and Whitley in August, 1912; the remainder were made by the present authors since that time.

Three types of observation were carried out :—

1. Titrations were made of a measured volume of sea-water to two coloured indicators, viz., phenol-phthalëin and methyl-orange using centi-normal solutions of hydrochloric acid. These titrations give the required amount of alkali to alter the hydrogen-ion concentration from approximately P_H, 10^{-8} to P_H, $10^{-4.5}$ and indicate quantitatively what has been termed qualitatively by Sörensen[2] the " Buffer " effect of the dissolved alkali-acid, or amphoteric, salts, and by Moore and Wilson[5] the " Reactivity " of the solution due to these same amphoteric solutes. This determination is one of great importance in estimating the characteristics of a physiological solution, since upon it depends the protective action against large variations in hydrogen-ion concentration.

2. The hydrogen-ion concentration was determined by the colorimetric method introduced by Friedenthal and Salm[1] and amplified and improved by Sörensen[2,3] and Palitzsch.[4]

3. The hydrogen-ion concentration was also determined electrometrically on several occasions, although this is a difficult estimation with sea-water on account of nearly all the "Reactivity" or "Buffer" action being due to bi-carbonate of magnesium. This was done to give a control to the colorimetric method, and on the whole the agreement was found to be excellent, when Sörensen's correction for the salt error had been applied.

A somewhat exclusive importance has been attached in recent years to a determination of the hydrogen-ion concentration of a physiological fluid such as blood-serum or sea-water. Important as this figure may be in indicating the actual reaction of the fluid, it gives no idea of the resistance of that reaction to change when alkali or acid is added, and from the point of view of the protective action of the physiological fluid to any living organisms of which it may form the environment or "external medium." This resistance to change is an all-important factor.

It is fashionable to-day to look askance at earlier methods of titration to coloured indicators, and to say that these only give the amount of acid or alkali necessary to bring the solution to an arbitrary level of hydrogen-ion concentration at which the indicator employed changes colour, and so titrations can furnish no indication of the hydrogen-ion concentration of the solution in its original condition. While this is true, and while we may now smile at the old-time results with coloured indicators which led to the same solution being written down as both acid and alkaline, and gave rise to such phrases as the "acidity" and "alkalinity" of the blood, it does not follow by any means that titration of physiological solutions is based on error and should be abandoned. In order to appreciate the properties as to alkalinity and acidity of a solution two things are essential, (1) to know the actual hydrogen-ion concentration and (2) the amount of alkali or acid required to be added to

shift the hydrogen-ion concentration between two definite values.

The first of these figures can be determined either electrometrically, or by the more rapid and convenient colorimetric method with indicators, which is based on the electrode method.

The second may be obtained by titrating to the change points of colour of two different indicators with a sufficient range between them.

The second figure is no less important than the first, for while the first gives the hydrogen-ion concentration with which the living organism is actually living in equilibrium, the second shows the amount of production of acid or alkali by the organism which is compatible with a given change in hydrogen-ion concentration, and hence demonstrates the degree of protection that the medium is capable of affording to the organism. All living organisms are extremely sensitive, both in their actual viability and also in the degree of their physiological activity to small changes in hydrogen-ion concentration, the growth and development and metabolic activity being all profoundly affected by comparatively slight changes.

The resistance to change in ionic concentration has been referred to as the "Buffer" effect by Sörensen, and was called the "Reactivity" of the solution by Moore and Wilson[6] to distinguish it from the "Reaction," which is quite a different thing.

Although the expression "Buffer Effect" has been used a great deal in late years, few attempts have been made to obtain a quantitative expression for the "Buffer Effect" or "Reactivity" although it was determined for blood-serum in 1906 by Moore and Wilson,[5] when the interesting result was obtained that it corresponds here to the total isotonicity figure of the serum as determined by the freezing-point method.

It was shown by these authors that the "Reactivity" can

be obtained by taking the algebraic differences in the titration figures to phenol-phthalëin, and to methyl-orange, or di-methyl-amido-azo-benzol.

When a solution containing acid-salts of carbonates or phosphates, or amphoteric electrolytes, such as proteins, has acid or alkali added to it there is a middle zone throughout which the hydrogen and hydroxyl-ionic concentrations vary very slowly, and outside this on either side there is abrupt rise and fall. By choosing two coloured indicators, one near each end of the range of such indicators, the breadth of this zone, or in other words, the "Buffer Effect" or "Reactivity," can be measured with fair accuracy. This is the figure which is determined by our titrations.

Another important effect which is given by measuring the variations in titration to a given indicator (suitably chosen so as to be sensitive to the varying factor) is the amount of variation in a metabolic product such as carbon-dioxide from one time or condition to another. Such an activity cannot be followed by determining hydrogen-ion concentration alone, although it is undoubtedly interesting to observe (as has been done below) the variation in the hydrogen-ion concentration corresponding to a given change in carbonic-acid concentration, arising from photo-synthesis or respiration.

But in a solution such as blood-serum or sea-water quite an appreciable variation in titration to phenol-phthalëin due to change in concentration of dissolved carbon-dioxide may occur, with scarcely a detectable change in hydrogen-ion concentration. A ten-thousandth normal solution of a caustic alkali is much more alkaline than blood serum or sea-water, but one c.c. of centi-normal acid added to 100 c.c. of this caustic alkali solution already suffices to remove the difference and makes the solution neutral, while such an addition scarcely appreciably affects the reaction of the serum or sea-water.

This important effect was pointed out by Moore, Roaf and

Whitley[5] in 1905, when investigating the effects of acid and alkali added to sea-water upon growth and development of Echinus eggs.

These authors state that "a solution of the mixed phosphates or carbonates in which there is an approximate balance between the concentration of hydrogen- and hydroxyl-ions such that these concentrations are nearly equal, cannot, however, be regarded as neutral in the same sense as distilled water is neutral, or as being acid or alkaline in the same sense as a solution containing only free acid or free alkali can be regarded as being acid or alkaline.

"Nor will such a solution of phosphates and carbonates, as is present in blood plasma, or sea-water, have a similar action upon living cells to either distilled water or a neutral solution of such salts as sodium chloride of equal osmotic concentration.

"Therefore blood plasma, and to a less extent sea-water, possess, on account of the mixed phosphates and carbonates which they contain, *a steadying action upon variations in the concentration of the hydrogen- and hydroxyl-ions.* When acid or alkali is added to the plasma, instead of there occurring a corresponding swing in the concentration of the hydrogen- and hydroxyl-ions, there takes place an alteration in the equilibrium of the ions of the phosphates and carbonates, which neutralises, in great part, the hydrogen- or hydroxyl-ions added, and prevents the plasma becoming markedly acid or alkaline. Without such a controlling action the life of the cells would be rendered impossible, for, as our experiments show, the living cell is most sensitive to even small variations in either hydrogen- or hydroxyl-ion."

The action of small variations in the hydrogen- or hydroxyl-ionic concentrations of plasma or other "external media" is only to-day gradually becoming recognised in the government of respiratory activity and other fundamental physiological functions, and hence attention may perhaps be drawn to the

fact that the above views were expressed in the Proceedings of the Royal Society nearly ten years ago, and that shortly thereafter the "Buffer" effect in the plasma was not only recognised, but estimated by Moore and Wilson, and published in 1906 in the first volume of the Bio-chemical Journal.

The following tabulated statement gives a record of the titration to phenol-phthalëin and to methyl-orange of the sea-water freshly drawn in the neighbourhood of Port Erin, Isle of Man, and in the Irish Sea, at various seasonal periods during the years 1912-14.

The titrations were made by adding four drops of 0·5 per cent. solution of phenol-phthalëin (by measure about 0·13 c.c.) to 100 c.c. of the sea-water, and then titrating with N/100 hydrochloric acid till the pink colour was just on the point of disappearing.

Two points are noteworthy, first that throughout the long series of observations the fresh sea-water was invariably alkaline to phenol-phthalëin indicating a value of hydrogen-ion concentration lying above P_H, 10^{-8}, or considerably higher than the values obtained by other recent observers such as Sörensen and Palitzsch[3,4] in sea-water from other regions, and, secondly, that there is a seasonal variation, the hydrogen-ion concentration being higher in winter, and decreasing in spring and summer. Although the latter variation is small, the differences in the amount of acid required to reduce the level to the neutral point of the phenol-phthalëin indicator show, when applied to the vast volumes of sea-water affected, great photo-synthetic activities of plants, and corresponding animal activities in oxidation. The results indicate a great crop, or annual variation in the water, corresponding to the seasonal variations on the land in the round of the year. The photo-synthetic crops in the sea-water, reckoned as carbohydrate produced, are similar to those on a land surface, and amount to several tons of carbohydrate per acre; this then forms the food for the floating animal-life of the sea.

Titration Values of Sea-Water at different Seasons of the Year.

Date.	Place of Collection of Sample.	No. of c.c. of N/100 HCl to neutralise 100 c.c. to phenolphthalëin.	"Buffer" or "Reactivity" Effect between P_H, $10^{-8.4}$ and P_H, $10^{-4} \times 10^{-4}$ N.	No. of c.c. of N/100 HCl required to neutralise 100 c.c. to methyl-orange.
		SUMMER 1912.		
Aug. 8—	Life-boat Slip	2·25 c.c.	—	—
„ 12—	Aquarium Tank	2·2	22·3	24·5 c.c.
„ 19—	Aquarium Tank	2·0	—	—
„ 24—	Aquarium Tank	2·2	—	—
„ 30—	Aquarium Tank	2·2	—	—
Sept. 3—	Aquarium Tank	1·8	—	—
„ 7—	Aquarium Tank	2·0	—	—
		WINTER AND SPRING 1912-13.		
Nov. 17—	Breakwater	0·8	—	—
„ 18—	Breakwater	0·75	—	—
„ 18—	Life-boat Slip	1·0	—	—
„ 21—	Irish Sea (by Fisheries Steamer from open sea)	1·0	—	—
Dec. 14—	Life-boat Slip	0·8	24·0	24·8
„ 15—	Life-boat Slip	1·2	24·8	26·0
1913				
Feb. 11—	Irish Sea (by Fisheries Steamer from open sea)	1·3	23·7	25·0
„ 13—	Breakwater (Small boat)	1·2	23·0	24·2
„ 14—				
„ 15—	Life-boat Slip	1·3	—	—
Apr. 8—	Life-boat Slip	1·65	23·85	25·5
„ 9—	Irish Sea (4 miles out, S.Y. Runa)	1·2	—	—
„ 10—	Life-boat Slip	1·6	22·1	23·7
„ 10—	Buoy at Breakwater (S.Y. Runa)	1·4	22·2	23·6
„ 11—	Irish Sea (5 miles out, S.Y. Runa)	1·5	23·3	24·8
„ 11—	Irish Sea (3 miles out, S.Y. Runa)	1·65	23·45	25·1
„ 12—	Irish Sea (3 miles out, S.Y. Runa)	1·75	22·25	24·0
„ 12—	Buoy at Breakwater (S.Y. Runa)	1·5	20·5	22·0
„ 12—	Life-boat Slip	2·2	22·3	24·5
„ 13—	Life-boat Slip	2·0	22·0	24·0
„ 14—	Port Erin Bay (Moorings S.Y. Runa)	2·1	22·5	24·6
„ 14—	Irish Sea (5 miles out, S.Y. Runa)	1·5	22·7	24·2
„ 14—	Life-boat Slip	2·25	22·95	25·2
„ 15—	Irish Sea (off Fleshwick Bay, S.Y. Runa)	2·75	21·65	24·4
„ 15—	Five Miles out (S.Y. Runa)	2·2	21·8	24·0
„ 15—	Life-boat Slip	2·75	21·25	24·0
„ 16—	Life-boat Slip	2·1	22·7	24·8
„ 17—	Life-boat Slip	2·5	21·9	24·4
„ 18—	Life-boat Slip	2·8	21·7	24·5
„ 19—	Life-boat Slip	2·3	22·4	24·7
„ 20—	Life-boat Slip	3·0	21·4	24·4
„ 21—	Irish Sea (5 miles out, S.Y. Runa)	1·8	—	—
„ 21—	Life-boat Slip	2·8	21·1	23·9

Titration Values of Sea-Water at different Seasons of the Year—*continued.*

Date.	Place of Collection of Sample.	No. of c.c. of N/100 HCl to neutralise 100 c.c. to phenol-phthalëin.	"Buffer" or "Reactivity" Effect between $P_H, 10^{-8.4}$ and $P_H, 10^{-4} \times 10^{-4}$ N.	No. of c.c. of N/100 HCl required to neutralise 100 c.c. to methyl orange.
	SUMMER 1913.			
June 14—	Life-boat Slip	3·2	21·3	24·5
„ 15—	Port Erin Bay (Middle of, Rowing-boat)	3·3	22·8	25·1
	SPRING 1914.			
Mar. 1—	Life-boat Slip	1·9	—	—
May 18—	Life-boat Slip	3·1	21·7	24·8
„ 18—	Outside Port Erin Bay (midway between Breakwater and Bradda Head)	2·5	22·8	25·3
„ 25—	Life-boat Slip	2·8	22·0	24·8
„ 25—	Breakwater	2·4	21·9	24·3
	SUMMER 1914.			
June 3—	Life-boat Slip	2·2	22·2	24·4
„ 3—	Breakwater	2·1	22·3	24·4
„ 12—	Life-boat Slip	1·9	23·0	24·9
„ 12—	Breakwater	1·8	23·1	24·9
„ 29—	Life-boat Slip	2·0	23·1	25·1
„ 29—	Breakwater	2·1	22·3	24·4
July 9—	Life-boat Slip	2·2	22·6	24·8
„ 9—	Breakwater	2·1	22·4	24·5
„ 15—	Life-boat Slip	2·6	21·8	24·4
„ 15—	Breakwater	1·9	22·6	24·5
„ 23—	Life-boat Slip	1·85	21·55	23·4
„ 23—	Breakwater	1·8	22·4	24·2
Aug. 1—	Life-boat Slip	1·8	22·7	24·5
„ 1—	Breakwater	1·75	22·65	24·4
	WINTER 1914-15.			
Jan. 1—	Life-boat Slip	0·3	—	—
„ 1—	Breakwater	0·8	—	—

The third column of figures in the table above is obtained by subtracting the "phenol-phthalëin" figure from the "methyl-orange" figure (since both titrations are in the same direction), and indicates the number of c.c. of N/100 acid or alkali necessary to bring 100 c.c. of sea-water *from the neutral point to phenol-phthalëin to the neutral point to methyl-orange or vice versa.* The neutral point to phenol-phthalëin corresponds

to a potential of hydrogen-ion concentration of approximately $P_H 10^{-8\cdot2}$, and the neutral point to methyl-orange to a hydrogen-ion potential of $P_H 10^{-4\cdot5}$. So that the figures indicate, expressed otherwise, the c.c. of N/100 acid required to produce this amount of change in hydrogen-ion concentration.

The limits are arbitrary, but they correspond closely to the extreme limits of a dilute solution containing, as the factors conferring its acidity or alkalinity, the carbonates or phosphates of the alkalies or of alkaline salts (such as calcium or magnesium).

The turning point of coloured indicators in such solutions is not a sharp line, but a band of some width. Roughly the "phenol-phthalëin neutral point" corresponds, in dilute solutions (such as are the physiological solutions) to the "bi-carbonate point" and to the "alkaline-phosphate point" respectively; while the "methyl-orange point" corresponds to the "complete carbonate neutralisation point" and the "acid-phosphate point" respectively.

These points accordingly give convenient limits for obtaining some quantitative measurement of that interesting property which has been called the "Reactivity" or "Buffer Effect" of a solution.

It is not claimed that they mark limits of *viability*, for, as will be seen in the next section of this paper, the green cells of marine algae and other marine plants, can live and photosynthesise up to the much more highly alkaline point (or lower hydrogen-ion potential point) marked by the conversion of all the carbonate or carbonic-acid, into "normal" carbonate.

However, these figures, representing the "Buffer Effect" or "Reactivity," do possess a substantial value, especially for the bio-chemist; for, a solution which possesses a low value of reactivity within this range will also possess a low value in the longer ranges such as that up to the normal carbonate on the alkaline side, or other defined limits.

The physiological interest of the figure so obtained lies in its relationship to the protective power of the medium against the invasion of the organism by alkaline or acidic products of metabolism, or such arising from other causes. Thus, sea-water is, while much more protective than fresh water, much less efficient than the secreted fluids of marine animals, and still less so than the blood-plasma or lymph of terrestrial animals. For, while mammalian serum has a reactivity (within the limits conventionally assigned above) of about N/5, that of sea-water, as given by the above table, is only about N/500.

To reduce the figures to fractions of "molar" concentrations, it is only necessary to remember that the titrations are made with N/100 acid, and expressed in c.c. required for 100 c.c. of sea-water. From this it follows that they must be divided by 10^{-4}. The figure for sea-water in our determinations as shown in the table accordingly varies around 21 N $\times$ 10^{-4} and 23 N $\times$ 10^{-4}.

The fact that sea-water freshly collected, at least in the region in which our samples have been taken, always gives a pink colour with phenol-phthalëin, and so is more alkaline than the body-media of both marine and terrestrial animals, is of some interest. The hydrogen-ion concentration of sea-water is accordingly lower than that of these media, the exponential value for the hydrogen-ion concentration lying between the limits of $P_H, 10^{-8\cdot1}$ and $P_H, 10^{-8\cdot4}$ (see p. 254).

It is of some interest to discuss the nature of the inorganic salt, or ion, to which this higher alkalinity of the sea-water is due. It is ascribed by Palitzsch[4] and by Brönsted and Wesenberg-Lund[15] to the calcium-ion, but our observations lead us to regard it as due to the accumulation of magnesium-ions in the sea-water. This accumulation arises from the higher relative solubility of the magnesium-ion in solutions of chlorides containing also carbonic acid-ions in excess. The

more rapid removal of calcium salts than of magnesium salts by marine organisms in the formation of shells also takes a part. As the result of the operation of these two causes, sea-water, as shown by analysis, contains magnesium-ions in much higher concentration than calcium-ions.

If a considerable volume of sea-water (one to two litres) be boiled for some hours in a Jena flask, adding from time to time boiled-out distilled water to make up for loss and so drive off excess of carbon-dioxide, a precipitate is obtained, which, on separation, is found to consist essentially of a mixture of oxide and carbonate of magnesium with very little calcium present.

If the water distilled off be collected, preferably in N/100 or N/10 alkali and the amount of dissolved carbonic acid be determined by titration to phenol-phthalëin with N/100 acid, the total amount which can be driven off is almost exactly half the figure given in the table for the titration to methyl-orange, while the residue of sea-water left behind in the flask now contains much more "normal" magnesium carbonate, and gives an intense pink with phenol-phthalëin. The fact that the carbon-dioxide which can be driven off by boiling the sea-water without addition of acid, or allowing great concentration of salts, is represented by approximately half the titration figure to methyl-orange of the sea-water in its natural condition, demonstrates clearly that the alkalinity is due to a bi-carbonate of an alkaline earth. Analysis of the precipitate formed shows that the alkaline earth chiefly concerned is magnesium.

This is shown by the two following experiments :—

Experiment 1. Took 1,000 c.c. of fresh sea-water, and distilled in Jena flask fitted to Liebig condenser, as in sea-water analysis operations for ammonia determinations. Eight successive distillates of 80 c.c. were taken off, the volume being restored in distilling flask at the end of each distillation. In the last distillation the carbon-dioxide coming off was very small. The total titration of the eight distillations was equivalent to 11·47 c.c. of N/10 acid; this for 100 c.c. of sea-water would be equivalent to 11·49

c.c. of N/100 acid, and this is very nearly one half of the average figure of 24 c.c. of N/100 acid obtained by direct titration of the sea-water to methyl-orange as given in the table above.

If the distillations be carried out to much higher concentrations of the sea-water, however, making up with boiled-out distilled water between each distillation, then practically all the carbon-dioxide can be driven off and the base causing the alkalinity of the sea-water left behind as a precipitate of magnesium oxide, only difficultly soluble in dilute hydrochloric acid.

This is shown in the following experiment :—

Experiment 2. One litre of fresh sea-water was placed in the distilling flask, and distilled, the distillate being caught in measured volumes of N/10 caustic soda, and back titrated at the end with N/100 hydrochloric acid.

In this case each distillation process was carried on until 600-750 c.c. of the water had distilled over, and four distillations in all were carried out. The titration figures in c.c. of N/10 caustic soda neutralised by the carbon-dioxide were as follows :— 14·14 − 2·67 − 2·07 − 1·72 = 20·56. It is seen that a little more than the amount of carbon-dioxide necessary to reduce from the bi-carbonate of magnesium to the normal carbonate comes off in the first distillation, then successively diminishing quantities as the carbonate with continued boiling passes towards the oxide.

As a result of the boiling a greyish-white precipitate forms, which on filtering off and taking up in hydrochloric acid proves to be a magnesium and not a calcium salt.

The alkalinity of sea-water is accordingly chiefly due to the bi-carbonate of magnesium, and the equilibrium lies at the point where there is such a proportion as is represented by a preponderance of magnesium bi-carbonate with a small excess of normal carbonate of magnesium. For the neutral point in dilute solution of magnesia and carbonic acid to phenol-phthalëin lies almost exactly at the bi-carbonate point, and a small excess of magnesium carbonate yields just such titration figures as have been shown above for sea-water.

Coming to the seasonal variations in alkalinity as indicated by the titration figures to phenol-phthalëin, a glance along the

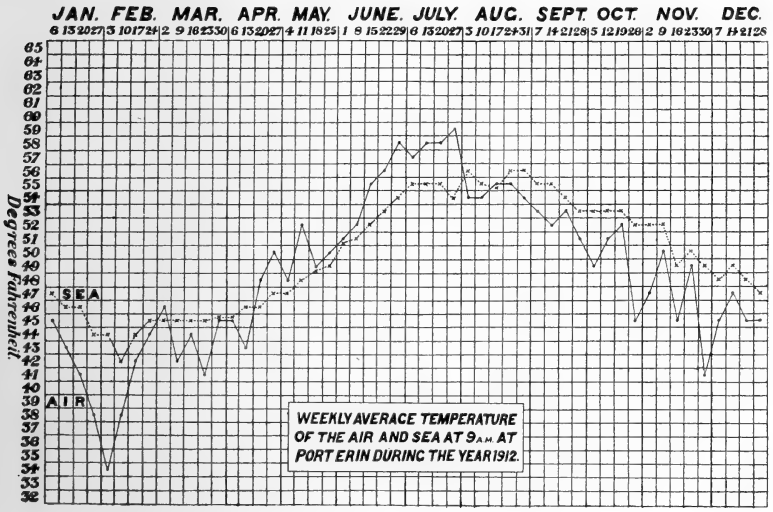
tables shows that there is a minimum in November, December, January, and February, increasing through the latter part of February and March to a maximum in May and June and then slowly falling off. Unfortunately measurements in the later half of September and October are lacking which might have shown the effects of the autumnal outburst of diatoms.

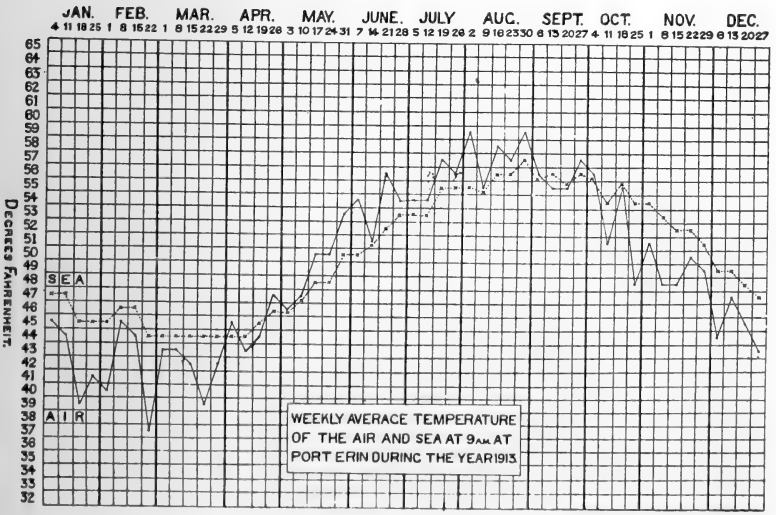
Taking the figures available, there is a variation from 0·3 to 1·2 in November and December up to 2·8 to 3·0 towards the end of April.

The difference of about 2 c.c. of N/100 alkali for 100 c.c. may not at first sight appear large. It may be taken as due to carbon-dioxide as bi-carbonate in the water, and 2 c.c. of M/100 carbon-dioxide per 100 c.c. is 2 c.c. of M/10 carbon-dioxide per litre, this is $4{\cdot}4 \times 2 = 8{\cdot}8$ milligrams per litre. Now the amount of carbon-dioxide which is photo-synthesised between December and April is probably much greater than this, because as the dissolved carbon-dioxide of the water is attacked by the diatoms in presence of light to form oxygen and build up organic carbon-compounds, the equilibrium between sea-water and the overlying atmosphere is upset and more carbon-dioxide is taken up from the air. To balance this, the absorption coefficient of carbon-dioxide for sea-water will decrease slightly at the somewhat higher temperature of the later Spring.

It might possibly be argued that the increased alkalinity of the Spring was due solely to this physical cause: that this is not the case is shown clearly by a comparison of the values of alkalinity with the temperatures of the sea-water in the years 1912 and 1913 as shown in the curves reproduced. It is observable that it is only in April that the temperature begins to rise, whereas the alkalinity commences to rise in February and March, and by the end of April is well on the way to its maximum.

Thus the table shows that in November, 1912, the value


JAN. FEB. MAR. APR. MAY. JUNE. JULY. AUG. SEPT. OCT. NOV. DEC.
Degrees Fahrenheit.
SEA
AIR
WEEKLY AVERAGE TEMPERATURE OF THE AIR AND SEA AT 9 A.M. AT PORT ERIN DURING THE YEAR 1912.


JAN. FEB. MAR. APR. MAY. JUNE. JULY AUG. SEPT. OCT. NOV. DEC.
DEGREES FAHRENHEIT.
SEA
AIR
WEEKLY AVERAGE TEMPERATURE OF THE AIR AND SEA AT 9 A.M. AT PORT ERIN DURING THE YEAR 1913.

lay below 1·0, while in February it had risen to 1·2 to 1·3, on April 8th it had reached 1·6, while the sea-water temperature had not moved till April 19th when it had only risen by 1° F. If the alkalinity at Life-boat Slip be taken from April 13th till April 20th, 1913, the average is 2·4. Thus, at the point where the temperature is only beginning to rise, the alkalinity has nearly reached a maximum. The alkalinity of April 20th is 3·0, compared with the figures 3·2 and 3·3 on June 14th and 15th, 1913. On the former day the temperature is only 1° F. above the average March temperature, on the latter days the temperature is 7° F. higher. The observations in June, July and August, 1914, show that there is no increase in alkalinity with temperature only.

It is also to be noted that the great Spring outburst of diatom activity appears before there would be time for any effect of rise of temperature. Although the first upward movement in temperature of the sea-water and the enormous increase in vegetable plankton lie close together in time, it is to be remembered that the physical cause must have a latent period ahead of the biological effect. The first movement of temperature is very slight, and when it is remembered that it must first stimulate growth, and then many succeeding generations arise before there is a massive increase in the vegetable plankton, it may probably be said that if the vegetable plankton (and the increased alkalinity) were due to the increased warmth of the water, then these would be found at least a week later in time than the first rise in temperature. There is, in fact, no such accordance observable, and it is far more probable that the increased length of day and altitude of sun, supplying a rapidly-increasing amount of solar light energy, are the potent causes in producing the diatom outburst and accompanying rise in alkalinity of sea-water.

If the actual variation in alkalinity observed be taken as a rough index of photo-synthetic activity, and as

an approximation the carbon be assumed to be all converted into carbohydrate, then some calculation may be given to demonstrate what such figures mean.

The amount of 8·8 milligrams per litre of carbon-dioxide changed corresponds to 2·4 milligrams of organic carbon, and this is 6 milligrams per litre of carbohydrate, synthesised, or 6 parts in a million of the sea-water—at first sight a ridiculously small quantity, but it is spread out in the sea. Six milligrams in a litre gives 6 grams in a cubic metre of sea-water. If the change by photo-synthesis occurred to *only* a depth of one metre, this would mean on a square kilometre of sea surface 6 millions of grams, or 6,000 kilograms of carbo-hydrate synthesised.

But the change occurs for a distance down in all probability of some hundreds of metres instead of one metre. Convection currents, winds and tides, mix the water thoroughly up for many metres deep.

The careful observations of Sven Palitzsch[4] have proved that the alkalinity of the sea-water does decrease as the depth from the surface increases, but there is scarcely any appreciable decrease at 100 metres, and even at 400 metres the decline is usually still small.

This decline in alkalinity at greater depths is of interest as an evidence of photo-synthetic activity and its relationship to alkalinity of the water. Its cause is probably three-fold, first the photo-synthetic activity decreases with depth as the intensity of the light diminishes in traversing the water; secondly, with increasing depth there is less admixture by currents with the more alkaline water due to photo-synthesis of the upper layers; and thirdly, organic débris of plant and animal in descending becomes oxidised, and these oxidations again set free carbon-dioxide which lowers in turn the alkalinity of the water.

If it be supposed, for the moment, that photo-synthesis

did not exist, then the whole depth of the sea in process of ages of time would come into complete equilibrium with the air, and the dissolved carbon-dioxide would be the same in the upper layers and in the depths, and accordingly there would be no drop in alkalinity in the deeper zones.

The slow drop in alkalinity demonstrates an effect of photo-synthesis, but it is so small within the upper portions that it may unquestionably be taken that photo-synthesis, aided by convection currents, has the full alkalising effect for at least 100 metres from the surface.

This supposition leads to a photo-synthetic effect of 300,000 kilograms of dry organic matter such as carbohydrate per square kilometre.

Expressed in English measurement this amounts to about two tons of *dry* organic matter per acre, and since vegetable crops do not contain on an average more than 20 per cent. of dry organic matter, this ocean crop corresponds to at least ten tons of moist plant organisms per acre.

The factors entering into such a computation, in the present fragmentary state of our knowledge, are of course vague, but the above is certainly a minimum, and it becomes obvious that the sea has annually a vast crop of green-plant organic matter comparable to that growing in the fields on land.

Another interesting point arising from these alkalinity determinations is that the degree of photo-synthesis and the corresponding weight of ocean crop is probably much more abundant nearer to the littoral. It is frequently observable in the table, that water taken along shore is more alkaline than that taken from on board a vessel three to five miles from shore.

Observations at great distances out at sea during the various seasons are most desirable but difficult to obtain.

Determinations of the Potential of Hydrogen-Ion Concentration.

This is the modern method of expressing the state of acidity or alkalinity of a solution at any given moment as distinct from the amount of added acid or alkali necessary to move the reaction to a given position. It is a static thing while the "Buffer Effect" or "Reactivity" is kinetic or dynamic. It might be termed the *reaction* of the fluid as distinct from its *reactivity*. The one expresses present position, the other ease or difficulty of moving into another determined position.

It has been known for some time that sea-water is an alkaline solution: this is stated by earlier observers such as V. Bibra,[6] Guignet and Telles[7] and Törnoe.[8] Later Natterer[9] observed that water from the eastern part of the Mediterranean was coloured red by phenol-phthalëin, and, unconsciously foreshadowing qualitatively Sörensen's later colorimetric method, was able to note by comparing the depths of shade of pink that the deep sea-water was less alkaline than the surface water.

Ruppin[10] was unable to note any pink coloration in water from the North Sea and Baltic, but as the water was drawn in 1909 from points near Kiel, the water may well have been contaminated sufficiently artificially to reduce slightly the alkalinity and throw it below the value necessary to give any colour reaction with phenol-phthalëin.

Loeb[11] found the water of the Atlantic Ocean at Wood's Hole to give a pink colour with phenol-phthalëin, while the water of the Pacific Ocean at Pacific Grove was less alkaline and gave no pink colour with this indicator.

Correlated with this, Loeb found the interesting fact that unfertilised ova of *Strongylocentrotus* developed parthenogenetically to a much greater extent in the more alkaline water

of the Atlantic than on the Pacific Coast. Addition of small amounts of alkali to the water of Pacific Grove, so as to bring its alkalinity up approximately to the level of the water of Wood's Hole, caused the parthenogenetic cleavage to increase. This is in agreement with the much earlier discovery of Loeb[12] that slight additions of alkali aided parthenogenesis and hastened cell-division. The same result on increased rate of cell-division was obtained by Moore, Roaf and Whitley[5] in fertilised eggs of *Echinus esculentus*.

This suggests an interesting thought as to the chemical causation of the outburst of animal life in the spring. It may be that it is the slow increase in alkalinity of the sea-water caused by the photo-synthetic activity of the increasing daylight upon the algae which reacts upon the animal life and causes increased cell division, as also that sexual activity (at least in the lower forms) which produces the shedding of ova and sperms. In any case it is quite clear that the increased alkalinity of the spring will be favourable to parthenogenesis, as also to cell-division after fertilisation.

The earliest observations by the hydrogen-electrode method upon the hydrogen-ion concentration of sea-water appear to be those made by Cottrell upon the suggestion of Loeb[12] in the water of San Francisco Bay. The experimental difficulties of such an observation as pointed out lately by Palitzsch are very great and were apparently not overcome by Cottrell, who found sea-water less alkaline than the neutral point (P_H, $10^{-7\cdot01}$).

The Danish observer, W. E. Ringer[13], was the first to overcome substantially most of the difficulties of the task lying in the way of obtaining hydrogen saturation of the electrodes and fluid without displacing carbon-dioxide, and so in the process of determination altering the alkalinity. This observer made a long series of observations of the water of the North Sea, the Zuidersee, and Bömmel-fiord, and found it to

vary slightly but to lie within the limits $P_H, 10^{-7.68}$ and $P_H, 10^{-8.24}$. The latter value lies well within the phenol-phthalëin range.

It is to the Carlsberg school of workers, and notably to Sörensen and Palitzsch[2,3,4], that we owe, first, extensive and careful determinations in various seas of the hydrogen-ion concentration of sea-water, secondly, a painstaking and elaborate investigation of the so-called " Salt Error " introduced into colorimetric observations by the action of the neutral salts of sea-water and chiefly the sodium chloride upon the coloured indicators.

Thanks to the labours of these observers, the deviation caused by the salt is now accurately known: it has been established that under given conditions this deflection in the colorimetric results is constant, and hence can be allowed for by deductions placed on record in the Sörensen-Palitzsch tables.

Accordingly, a rapid and accurate method is placed in the hands of observers for estimating the hydrogen-ion concentration of sea-water.

This colorimetric method of Sörensen has therefore been mainly used by us, but we have on certain occasions controlled it, by using the electrometric method with the electrodes. We have found concordance good when allowance is made for the " Salt Error " by use of the tables.

The Sörensen method has now become so well known that it need not be described in detail,* so its principle only will be pointed out. Mixtures in varying proportions of two solutions, one with a higher the other with a lower hydrogen-ion concentration, are prepared in a series of test-tubes. The hydrogen-ion concentrations of these mixtures have been directly determined once for all by Sörensen and tabulated so that they can be referred to, and have also been plotted in

* See Walpole, Bio-Chemical Journal, Vol. V, 1911, p. 207, and Vol. VIII, 1915, p. 628.

curves by Walpole,[16] which may be utilised instead of the tables. Minute directions are given by Sörensen for the preparation of the stock solutions from which the mixtures are made up at the time of each experiment. In making up the mixtures matters are so arranged that the volume of the comparison mixture is always 10 c.c. For example, calling the solutions A and B, a series of mixtures is made containing, say—(1) Nine of A and one of B, (2) Eight of A and two of B, (3) Seven of A and three of B, and so on. In an additional tube 10 c.c. of the sea-water is taken of which the hydrogen-ion concentration is to be determined. All the tubes are placed in a special test tube rack so that they slope at an angle of 45° to the horizontal and rest on a white surface such as milk-glass. An equal and definite volume of the coloured indicator to be used is now added to each tube and the tube containing the sea-water to be analysed for its hydrogen-ion concentration is carefully compared as to tint of colour with each tube, and that one selected with which it most closely matches. After a rough approximation a series of tubes lying closer in their steps of variation can be chosen. That one which matches closest is taken as having the same hydrogen-ion concentration as the sample to be tested. In the case of sea-water, after identification and establishment of the hydrogen-ion potential by table or chart, the salt error deduction must be made.

Naturally, different coloured indicators, and different comparison mixtures, are selected according to the level of hydrogen-ion concentration of the solutions to be tested. In our experiments on sea-water about Port Erin we have always found phenol-phthalëin appropriate as a coloured indicator, and the most useful comparative solution-mixture that known as "borate mixture" and hydrochloric acid. The "borate mixture" contains 12·404 grams of boric acid and 100 c.c. of normal caustic soda in one litre, and was carefully prepared according to Sörensen's directions. The hydrochloric acid

solution added in varying proportion to this is a deci-normal solution of hydrochloric acid. On one or two occasions the phosphate combination was utilised, one ingredient of this is one-fifteenth normal primary potassium phosphate (acid phosphate), the other one-fifteenth normal secondary sodium phosphate (alkaline phosphate), mixed as above described.

The following table gives the results of the observations :—

Seasonal Variations in Hydrogen Ion Concentration of Sea-Water.

Period of Year.	Potential of Hydrogen Ion Concentration colorimetrically determined with Borate and Hydrochloric Acid mixture, and corrected for Salt Effect (Sörensen).	Potential of Hydrogen Ion Concentration, electrometrically determined by hydrogen electrode method.
November, 1912	– 8·13	– 8·20
December, 1912.....	– 8·10	– 8·16
February, 1913	...	– 8·20
April, 1913	– 8·20	– 8·40
May, 1913	– 8·37	...
June, 1913	– 8·31	– 8·35
July, 1913	– 8·30	...

The results of the experiments demonstrate two things, first that the hydrogen-ion concentration for the Irish Sea in the neighbourhoods tested is fairly low indicating a correspondingly higher alkalinity ; secondly that the alkalinity is *increased* in the spring and summer months. The variations are too small to warrant any further deduction than this. Such observations are excessively difficult to carry out, but their trend is sufficient to confirm the results of the titration experiments, namely that the alkalinity of sea-water is lowest in winter and increases in the spring.

II.—The Limits of Photo-Synthesis by Algae as the Alkalinity Due to Their Action Increases.

As was stated at the outset, our attention was first drawn to the alkalinity question in relationship to photo-synthesis, by the plaice disease in the spawning tank, and the presence in this pond-water of an immense number of floating mono-cellular algae with which minute green flagellata were also present in great abundance. It was found then, in April 1912, that the alkalinity of the pond-water was very considerably higher than natural sea-water.

The diatom outburst of the Spring had not yet appeared and the alkalinity of the Bay water was low; on the other hand it was found that the alkalinity of one portion of the pond (which is separated into two parts by a wall and sluice) was such that it required 3·3 c.c. of centi-normal acid to neutralise 100 c.c. to phenol-phthalëin, and the other portion of the pond-water required 3·8 c.c. of centi-normal acid in a similar titration. As may be seen by comparing with the table given in Section I, these figures are above normal for fresh sea-water, and the pond-water was therefore in a pathological condition with regard to alkalinity.

The experiments of Loeb, and of Moore, Roaf and Whitley mentioned in Section I., indicate that such alkaline water would have a stimulating and in-co-ordinating action upon cell-division. In fact the amount of increased alkalinity would be just that which increases cellular activity. Given a provocative cause of any kind such as a bacterial infection, the conditions therefore were just those which would aid such an ulcerative disease as that from which the plaice were dying.

The pond-water was examined again for alkalinity and contrasted with the alkalinity of freshly taken water from Port Erin Bay in November, 1912; the results are shown in the following statement:—

November 17th, 1912. Took three samples of water (A) from Pond I. (large spawning pond), (B) from Pond II. (smaller pond at West end beyond sluice), (C) from open sea at Breakwater. A sample of 100 c.c. in each case was titrated with centi-normal hydrochloric acid, with phenol-phthalëin as indicator (4 drops of 0·5 per cent. phenol-phthalëin in the 100 c.c.) with the following results :—

(A) Pond I.	... ...	Required 1·9 c.c.
(B) Pond II.	... ...	,, 1·9 c.c.
(C) Breakwater	... ...	,, 0·9 c.c.

Thus, the water from the two ponds was approaching the spring alkalinity while the Bay water was at winter level.

The ponds were shortly afterwards emptied, the walls disinfected and algae removed as far as possible, and then refilled. Examined again in February, 1913, the alkalinity was found to be the same as that of the " Bay " water. Samples of " Pond " water and of " Bay " water taken then and titrated alongside each other as before, gave in each case an alkalinity represented by 1·3 c.c. of centi-normal acid.

It hence became obvious that the increased alkalinity was caused by the algal growth causing photo-synthesis and the conversion of bi-carbonate into alkaline normal carbonate.

The same effect was demonstrated in the determination of the hydrogen-ion concentrations in the " Pond " and " Bay " waters by Sörensen's method. At this earlier period the sea-waters were contrasted with mixtures of the two phosphatic solutions, because the alkalinity was not expected to run so high. The two pond-waters in November, 1912, matched at 9·8 c.c. of alkaline phosphate to 0·2 c.c. of acid phosphate, while the " Bay " water matched it at 9·7 c.c. alkaline phosphate to 0·3 c.c. of acid phosphate. After correction for salt effect these values correspond to $P_H\ 10^{-8.3}$ for the " Pond " water and $P_H\ 10^{-8.1}$ for the " Bay " water.

It thus became of interest to determine the limits to which, under the most favourable circumstances of a smaller volume of sea-water exposed to light in presence of green algae, such increase of alkalinity and depression of hydrogen-ion concentration could be carried.

The fact that sea-water is usually alkaline to phenolphthalëin is sufficient to show that, save for an infinitesimal amount of dissolved carbon-dioxide requisite to keep up potentially the partial pressure of carbonic acid anions in the water, all the carbonic acid is present as a bi-carbonate along with a small fraction as normal carbonate.

When photo-synthesis commences this small potential amount of carbon-dioxide is synthesised into organic carbon compounds, this change upsets the equilibrium and so the bi-carbonate (of magnesium) breaks up yielding a supply which restores the tension in solution of carbonic acid. The question is, how far can this process go before the rising alkalinity destroys the algae, and the answer is beautifully given by the following experiments.

It has already been shown by Nathansohn[14] that aquatic plants can assimilate perfectly in water containing bi-carbonates, but not in water containing only normal carbonates; but as far as we are aware, the exact point of stoppage in a natural mixture of carbonates and bi-carbonates such as is present in sea-water, or the value of the hydrogen-ion concentration at the end of the process, have not hitherto been determined.

The result is interesting—the photo-synthetic action stops at the precise point where all the available bi-carbonate has been converted into carbonate, and the hydrogen-ion concentration has then reached the surprisingly low value indicated by the potential $P_H, 10^{-9.1}$.

Experiment 1. August 29th, 1912. A sample of sea-water was taken at the "Life-boat Slip" and this was carefully neutralised by adding the calculated amount of centi-normal

hydrochloric acid as estimated from a titration of its alkalinity. A volume of 2,000 c.c. was measured off and to this a quantity of green algal confervae found growing in a vessel in the laboratory was added. The amount of moist algal matter added was not estimated, but it certainly did not exceed half a gramme. The algae were added at 12.15 p.m. on August 29th and the whole was placed in a wide-mouthed bottle, which the mixture just filled. The bottle was stoppered and left in the open air exposed to daylight. A sample was taken off and tested as to alkalinity on August 30th at 9.15 p.m.; the alkalinity had risen so that 3·8 c.c. of centi-normal acid were required to neutralise 100 c.c. to phenol-phthalëin. Thus the alkalinity as a result of photo-synthesis in such a restricted volume had already risen above anything naturally found in sea-water.

A second titration was carried out on September 1st at 10.20 p.m., that is about 82 hours from the commencement of the experiment, when the alkalinity was found to have increased enormously, 9·7 c.c. of N/100 acid being required to neutralise 100 c.c. of the solution to phenol-phthalëin. A third titration was made at 6 p.m. on September 2nd, the value of the alkalinity to phenol-phthalëin had now reached 11·1 c.c. A fourth titration at 9·20 p.m. on September 3rd gave 11·7 c.c.; a fifth, on September 4th at 10.20 p.m. gave 12·3 c.c.; a sixth, on September 5th at 8.40 p.m. gave 12·2 c.c.; and a seventh, at 6 p.m. on September 6th gave 11·4 c.c.

A naked-eye examination during the experiment showed that the algae remained green, and apparently the growth was healthy until September 4th when the alkalinity had reached its maximum. From this point onward the growth commenced to turn brown and die, and on September 6th, when the alkalinity had commenced to fall off again, the green organisms were evidently dead. The drop in the figure was hence probably due to bacterial decomposition.

The figure obtained at the point of maximum alkalinity in the above experiment bears an interesting relationship to the figure for the total alkalinity of sea-water, as shown by the titrations to methyl-orange in the first table of the preceding section of the paper. Since the turning point in colour of methyl-orange lies above the hydrogen-ion concentration of carbonate mixtures, it gives the total content of the sea-water in all bases, including magnesium and calcium oxides, and the value for such bases lies between 24 and 25 c.c. of centi-normal acid for 100 c.c. of sea-water. Now the maximum value of the alkalinity at which the algae cease to photo-synthesise, and die, corresponds to 12·3 c.c. of centi-normal acid or exactly one half of the total alkaline bases. The algal cells behave like a sensitive colour-indicator and cease to functionate precisely at the point where all the bi-carbonates have been converted into normal carbonate. Up to this point, they have, in presence of light actively converted carbon-dioxide into organic carbon compounds and have flourished; at this point the gradient of alkalinity begins more rapidly to rise and they are killed off.

Experiment 2. At 3 p.m. on November 17th, 1912, a wide-mouthed bottle holding approximately two litres was filled with sea-water at the Life-boat Slip, about three grams moist weight of *Ulva enteroides* was added, and the bottle, stoppered, was exposed to the daylight on the wall of the Fish-spawning pond. A sample of the water was titrated at 10.45 a.m. on November 18th, and required 2·4 c.c. of centi-normal acid to neutralise to phenol-phthalëin, the neutralisation figure of the "Bay" water being at the same date 0·75 c.c. Thus, in a winter day when the exposure to daylight could not have exceeded four hours of diffuse daylight, the photo-synthetic activity had brought the alkalinity of this confined volume of sea-water from the mid-winter to the spring level. Titrated again at 4 p.m. the alkalinity had increased to 5·9 c.c.;

the day was bright but without much sunshine, so that on a winter day in about five hours interval the alkalinity had increased, as a result of photo-synthesis, from 2·4 c.c. to 5·9 c.c. The bottle and the contained algae was carried over to Liverpool, being exposed on board the steamer, and later on the roof of the laboratory, to such illumination as was available. It was titrated for a third time on November 22nd at noon. The alkalinity had reached 7·4 c.c. of centi-normal acid per 100 c.c. or more than double the maximum alkalinity in late spring or summer in the sea.

The hydrogen-ion concentration was also determined in this experiment after about four hours' exposure in November daylight; it matched at 9·9 c.c. of alkaline phosphate in the phosphatic mixture, the normal sea-water standing at 9·7. Next day at 4 p.m. it was more alkaline than the full strength of the alkaline phosphate, so that the hydrogen-ion concentration was below P_H, $10^{-9.1}$.

Experiment 3. February 13th, 1913. A sample of sea-water was taken at 12·40 p.m. and a small quantity of green seaweed (*Ulva enteroides*) was placed in it. A titration of the water as then taken gave 1·3 c.c. of centi-normal acid per 100 c.c. The bottle containing about 2,000 c.c. of sea-water, and being quite full and stoppered, was exposed to the daylight. February 14th, at 11.30 a.m., the seaweed was floating at the top and showed bubbles of gas entangled. Unstoppered, stirred up, allowed to come to rest, and took 100 c.c. for titration. The sample required 5·9 c.c. of centi-normal acid to neutralise it to phenol-phthalëin. A Sörensen determination gives a match at 7·6 Borate to 2·4 HCl, equivalent to P_H, $10^{-8.6}$. The water removed for the determinations was replaced by fresh sea-water so as to fill the bottle, which was re-stoppered and left exposed to the light. On February 15th at 11.30 a.m. the water was titrated again, the weather having been bright in the interval, but with little direct sunlight. The seaweed

had floated to the top and contained a good many bubbles of gas. The contents were well stirred up and a sample taken for titration; the result was 8·5 c.c. A Sörensen determination gave a match at 9·1 Borate and 0·9 c.c. HCl, equivalent to $P_H 10^{-8.9}$.

The next titration made in this experiment was carried out at 11.30 a.m. on February 16th. There had been a little bright sunlight on the afternoon of February 15th, and a dullish morning on the 16th. The titration gave 11·1 c.c. The Sörensen determination gave an alkalinity exceeding the full strength of the Borate solution, that is to say above $P_H 10^{-9.1}$.

This shows that in an interval of three days in February, the small quantity of seaweed used in the experiment had been able to increase the alkalinity of two litres of sea-water almost to the maximum point.

The bottle containing the sea-water and green seaweed was taken over to Liverpool and exposed on the roof of the Bio-chemical Laboratory. The weather was fairly bright, and the bottle was exposed to diffuse light during the day, but was not in any direct sunlight. Analysed on February 17th, that is, four days from the commencement of the experiment, it gave an alkalinity corresponding to 12·4 c.c. per 100 c.c. of centi-normal acid. A titration against methyl-orange for total alkali gave 24·0 c.c. of centi-normal acid.

Here again the full point of alkalinity is reached at almost exactly one half of the total alkali available.

The quantity of seaweed in the water was separated and analysed. Dried from adherent moisture and weighed in the moist condition, it amounted to 3·05 grams. After drying at 105°C, this yielded 0·568 gram of dried matter. Incinerated this left 0·180 gram of ash, or about 30 per cent. of the dried weight. The ash contained Na, Mg, Fe, Cl, and SO_4, and P_2O_5. There was a considerable amount of iron in the ash.

Experiment 4. This experiment was made to test the rapidity of action in an early summer period and was not carried to the end as the others. June 14th, 1913, at 10 a.m., seaweed placed in water, about 2,000 c.c., in fully-stoppered bottle. Titration value of water, at commencement, 3·7 c.c. alkaline. Again titrated at 10 a.m. next day, after bright sunshine meanwhile, titration had increased to 8·7 alkaline. Sörensen had gone up from 6·1 : 3·9, Borate and Hydrochloric acid to 9·4 : 0·6, equivalent to P_H, $10^{-9.0}$.

Summary and Conclusions.

1. Photo-synthesis by green algae causes a marked diminution in hydrogen-ion concentration in sea-water, and in confined volumes of water this variation in the direction of increased alkalinity may act as the inducing or favouring cause for pathological conditions and disease.

2. Certain salts of the sea-water (notably carbonate and bi-carbonate of magnesium) act as a steadying agency in preventing too rapid variations in the ionic concentrations of hydrogen and hydroxyl, and so safeguard life in the ocean. Within viable limits, physiological activity may be stimulated at certain seasons in which the alkalinity of the sea-water is increased, or, in other words its hydrogen-ion concentration diminished. Thus a rise in alkalinity in Spring would aid, along with temperature and sunlight, in producing increased cell-division.

3. The "Buffer" effect or "Reactivity" of sea-water has been estimated between two fixed points, viz., the turning point to methyl-orange and the turning point to phenol-phthalëin, and found to correspond to about 22×10^{-4} N. This range of "Reactivity" does not show seasonal variation, and the hydrogen and hydroxyl ionic concentrations simply vary within its limits.

4. The "Reactivity" effect is mainly produced by dissolved magnesium bi-carbonate, and not calcium bi-carbonate as usually stated.

5. In all cases the normal fresh sea-water gave a pink colour with phenol-phthalëin indicating a potential of hydrogen-ion concentration lying below P_H, 10^{-8}, the average being about P_H, $10^{-8.2}$.

6. The seasonal variations in P_H have been followed out by the colorimetric method of Sörensen and by the Hydrogen Electrode method, and a small but distinct increase in alkalinity in Spring has been detected.

7. This vernal increase in alkalinity is not due to increasing temperature disturbing the equilibrium between the carbon-dioxide of sea-water and atmosphere, for the rise in alkalinity clearly precedes in time the rise in temperature. It is caused by photo-synthesis as is shown by its coincidence in its occurrence with the rapid lengthening of the day in March and the increasing sun's altitude, as also by the great changes in alkalinity which may be produced by exposure of sea-water containing algae to sunlight.

8. Algae continue abstracting carbon-dioxide and so increasing alkalinity until all the bi-carbonates have become changed into normal carbonates, and then definitely cease to functionate and rapidly die at this latter point. The potential of hydrogen-ion concentration falls to P_H, 10^{-9} before synthesis ceases. At this point the sea-water gives an intense pink to phenol-phthalëin, and titration gives a figure almost exactly half that of the total reactivity effect.

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LITERATURE.

1. Friedenthal, and Friedenthal and Salm. Arch. f. Anat. u. Physiol., 1903, S. 550. Zeitsch. f. Electrochem. Bd. X, 1904, S. 113, S. 341; Ibid., Bd. XIII, 1907, S. 125.

2. Sörensen. Biochem. Zeitsch. Bd. XXI, 1909, S. 131, and other papers in Comptes rendus du Lab. de Carlsberg, notably T. VIII, pp. 1, 396.

3. Sörensen and Palitzsch. Comptes rendus du Lab. de Carlsberg, T. IX, 1910. Biochem. Zeitsch. Bd. XXIV, 1910, S. 387.

4. Palitzsch. Report on the Danish Oceanographical Expeditions, 1908-10. Carlsberg Fund, ed. by Joh. Schmidt, Copenhagen, 1912. Vol. I, p. 239. Biochem. Zeitsch. Bd. XXXVII, 1911, S. 116. Comptes rendus du Lab. de Carlsberg, T. X, 78, 1911.

5. Moore, Roaf and Whitley. Proc. Roy. Soc. B. Vol. 77, 1905, p. 102.

Moore and Wilson. Biochemical Journal, Vol. I, p. 297, 1906.

6. V. Bibra. Liebig's Annalen, Bd. 77, 1851, S. 90.

7. Guignet et Telles. Comptes rendus de l'Acad. des Sciences, T. 83, 1876, p. 920.

8. Tornöe. Den norske Nordhavsekspedition, 1876-8, 27, 1880.

9. Natterer. Denksch. de kais. Akad. d. Wissensch. math-naturwissen. Klasse, Bd. 59. Ber. d. Kom. f. Erforschung d. östl. Mittelmers, 83, 1892, und 60, 54, 1893.

10. Ruppin. Wissensch. Meeresuntersuch. Kiel, neue Folge, 11, 281, 1909.

11. Loeb. Arch. f. d. ges. Physiol. Bd. 118, 186, 1907.

12. Loeb. Arch. f. Entwickelungsmechanik d. Organismen, 1898, Bd. VII, 631. Arch. f. d. ges. Physiol, 1903, Bd. 99, 637. Ibid., 1904, Bd. 101, 340. Ibid., Bd. 103, 1904, 506. University of California Publications, Physiology, 1903-4, Vol. 39, 139.

13. Ringer. Verhandlingen u. h. Rijksinst. v. h. ondersook. der zee, 1908.

14. Nathansohn. Ber. u. d. Verhandl. d. k. Sachs. Gesellsch. d. Wissensch. zu Leipzig. math-naturwissen. Klasse, Bd. 59, 1907.

15. Brönsted und Wesenberg-Lund. Internationale Revue d. ges. Hydrobiologie u. Hydrographie, 1912, S. 251.

16. Walpole. Biochemical Journal, Vol. V, 1911, p. 207, and Vol. VIII, 1915, p. 628.

HYDROGRAPHIC OBSERVATIONS MADE IN THE IRISH SEA DURING 1914.

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The hydrographic observations in the eastern portion of the Irish Sea which were begun in 1907 were continued during 1914, but owing to various circumstances they were very incomplete during the past year.

A certain amount of data has, however, been collected, and it has been thought worth while to record it. Observations were made during the months of February, April, May, June, July, and September.

Various causes had prevented hydrographic cruises being made during January, March, and August, and at the end of September the observation steamer was taken over by the Admiralty.

The 24 stations at which water samples were collected were the same as during 1913 and are indicated on the chart given in last year's Report on p. 191.

February 3 to 7, 1914.

Stations I to IV, 3/2/14. Surface observations only.

Station.		Time.	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
I	54°N. ; 3°30′W.	9.25 a.m.	5·1	17·82	32·20	25·47
II	54°N. ; 3°47′W.	10.30 a.m.	6·3	18·63	33·66	26·47
III	54°N. ; 4°4′W.	11·40 a.m.	6·65	18·74	33·86	26·58
IV	54°N. ; 4°20′W.	12.55 p.m.	7·9	18·98	34·29	26·75

Station V, 53°53′N. ; 4°46′W. 5/2/14 (9.35 a.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	8·35	18·99	34·31	26·71
30	8·15	18·98	34·29	26·71
60	8·15	18·98	34·29	26·71

Station VI, 53°43′N. ; 4°44′W. 5/2/14 (10.55 a.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	8·65	18·97	34·27	26·62
30	8·5	18·96	34·25	26·63
70	8·45	18·97	34·27	26·64

Station VII, 53°33′N. ; 4°41′W. 5/2/14 (12.15 p.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	8·1	18·96	34·25	26·69
30	7·95	18·95	34·23	26·70
83	7·95	18·95	34·23	26·70

Stations VIII to XIX. Surface observations only.

Stations XX to XXIV were not visited owing to the very bad weather experienced during this cruise.

Station.		Date and Time.		T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
			p.m.				
VIII	53°27′N. ; 4°5′W.	5.2.14	3.0	6 8	18·69	33·77	26·49
			a.m.				
IX	53°31′N. ; 3°31′W.	6.2.14	9.25	6·2	18·47	33·37	26·26
X	53°37′N. ; 3°45′W.	6.2.14	10.25	6·7	18·70	33·78	26·53
XI	53°43′N. ; 3°58′W.	6.2.14	11.20	7·15	18·88	34·11	26·72
			p.m.				
XII	53°48′N. ; 4°12′W.	6.2.14	12.15	7·4	18·92	34·18	26·74
XIII	53°54′N. ; 4°27′W.	6.2.14	1.10	7·6	18·96	34·25	26·76
XIV	54°32′N. ; 4°37′W.	6.2.14	5.10	8·3	18·91	34·16	26·59
XV	54°37′N. ; 4°45′W.	6.2.14	6.10	8·3	19·00	34·33	26·72
			a.m.				
XVI	54°35′N. ; 4°27′W.	7.2.14	8.0	6·6	18·70	33·78	26·54
XVII	54°34′N. ; 4°12′W.	7.2.14	9.0	6·2	18·59	33·58	26·43
XVIII	54°32′N. ; 3°55′W.	7.2.14	10.0	6·3	18·67	33·73	26·53
XIX	54°29′N. ; 3°43′W.	7.2.14	11.0	5·3	18·04	32·59	25·76

April 8, 1914.

Stations I to VII. Owing to bad weather only surface observations were made.

Station.		Time.	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
I	54°N.; 3°30′W.	10.10 a.m.	7·3	18·10	32·70	25·59
II	54°N.; 3°47′W.	11.15 a.m.	7·55	18·75	33·87	26·48
III	54°N.; 4°4′W.	12.20 p.m.	7·8	18·96	34·25	26·73
IV	54°N.; 4°20′W.	1.25 p.m.	8·0	19·04	34·40	26·82
V	53°53′N.; 4°46′W.	3.0 p.m.	7·9	19·07	34·45	26·87
VI	53°43′N.; 4°44′W.	4.0 p.m.	7·9	19·06	34·43	26·86
VII	53°33′N.; 4°41′W.	5.0 p.m.	7·8	18·96	34·25	26·73

May 5 to 7, 1914.

Stations I to IV, 5/5/14. Surface observations only.

Station.		Time.	T°	Cl°/$_{oo}$	S°/^{oo}	σ_t
I	54°N.; 3°30′W.	8.40 a.m.	9·35	18·19	32·86	25·41
II	54°N.; 3°47′W.	9.45 a.m.	8·95	18·75	33·87	26·26
III	54°N.; 4°4′W.	10.45 a.m.	9·05	18·82	34·00	26·35
IV	54°N.; 4°20′W.	11.45 a.m.	9·15	19·02	34·36	26·61

Station V, 53°53′N.; 4°46′W. 5/5/14 (1.30 p.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	9·05	19·06	34·43	26·69
30	8·9	19·06	34·43	26·71
60	8·85	—*	—	—

* Bottle broken in transit.

Station VI, 53°43′N.; 4°44′W. 5/5/14 (2.45 p.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	9·1	19·10	34·51	26·74
30	8·95	19·09	34·49	26·74
65	8·9	19·09	34·49	26·75

Station VII, 53°33′N.; 4°41′W. 5/5/14 (4 p.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	9·3	19·06	34·43	26·65
30	9·2	19·06	34·43	26·66
70	9·05	19·05	34·42	26·67

Stations VIII to XXIV. Surface observations only.

Station.		Date and Time.		T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
VIII	53°27′N.; 4°5′W.	6.5.14	a.m. 11.15	9·15	18·94	34·22	26·50
IX	53°31′N.; 3°31′W.	6.5.14	p.m. 3.0	9·80	18·50	33·42	25·78
X	53°37′N.; 3°45′W.	6.5.14	4.0	9·45	18·71	33·80	26·13
XI	53°43′N.; 3°58′W.	6.5.14	5.0	9·15	18·91	34·16	26·46
XII	53°48′N.; 4°12′W.	6.5.14	5.55	8·95	19·04	34·40	26·67
XIII	53°54′N.; 4°27′W.	6.5.14	6.55	8·95	19·05	34·42	26·68
XIV	54°32′N.; 4°37′W.	7.5.14	5.35	9·45	18·78	33·93	26·22
XV	54°37′N.; 4°45′W.	7.5.14	4.55	9·30	18·41	33·26	25·72
XVI	54°35′N.; 4°27′W.	7.5.14	3.45	9·45	18·26	32·99	25·50
XVII	54°34′N.; 4°12′W.	7.5.14	2.50	9·45	18·49	33·40	25·82
XVIII	54°32′N.; 3°55′W.	7.5.14	1.55	9·55	18·21	32·90	25·40
XIX	54°29′N.; 3°43′W.	7.5.14	12.45	9·85	17·92	32·38	24·95
XX	54°24′N.; 3°57′W.	7.5.14	a.m. 9.20	9·20	18·15	32·79	25·38
XXI	54°20′N.; 4°13′W.	7.5.14	8.25	9·25	18·58	33·57	25·97
XXII	54°15′N.; 3°57′W.	7.5.14	p.m. 8.0	9·55	18·42	33·28	25·70
XXIII	54°10′N.; 3°42′W.	7.5.14	8.50	8·95	18·43	33·30	25·81
XXIV	54°5′N.; 3°27′W.	7.5.14	9.45	9·70	18·10	32·70	25·23

June 2 and 3, 1914.

Stations I to IV, 2/6/14. Surface observations only.

Station.		Time.	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
I	54°N.; 3°30′W.	5.55 p.m.	11·6	18·43	33·30	25·36
II	54°N.; 3°47′W.	6.55 p.m.	11·9	18·32	33·10	25·15
III	54°N.; 4°4′W.	9.20 p.m.	11·8	18·42	33·28	25·31
IV	54°N.; 4°20′W.	10·20 p.m.	11·1	18·79	33·95	25·96

Station V, 53°53′N.; 4°46′W. 3/6/14 (9.40 a.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	10·2	19·01	34·34	26·43
30	9·9	19·08	34·47	26·58
65	9·9	19·07	34·45	26·56

Station VI, 53°43′N.; 4°44′W. 3/6/14 (10.50 a.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	10·25	19·09	34·49	26·53
30	10·1	19·09	34·49	26·55
56	10·15	19·10	34·51	26·56

Station VII, 53°33′N.; 4°41′W. 3/6/14 (12 noon).

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	10·5	19·04	34·40	26·41
30	10·28	19·05	34·42	26·46
62	10·24	19·06	34·43	26·49

July 9 to 10, 1914.

Stations I to IV, 9/7/14. Surface observations only.

Station.		Time.	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
I	54°N.; 3°30′W.	11.0 a.m.	15·5	18·48	33·39	24·64
II	54°N.; 3°47′W.	1.30 p.m.	13·6	18·58	33·57	25·18
III	54°N.; 4°4′W.	2.35 p.m.	13·6	18·72	33·82	25·38
IV	54°N.; 4°20′W.	3.30 p.m.	13·2	18·86	34·07	25·66

Station V, 53°53′N.; 4°46′W. 10/7/14 (11.50 a.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	13·5*	18·98	34·29	25·76
30	12·1	19·02	34·36	26·09
83	11·95	19·02	34·36	26·11

* Possibly this should have been 12·5°.

Station VI, 53°43′N.; 4°44′W. 10/7/14 (1 p.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	12·65	19·05	34·42	26·02
30	12·4	19·05	34·42	26·07
68	12·35	19·05	34·42	26·08

Station VII, 53°33′N.; 4°41′W. 10/7/14 (2.5 p.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	13·4	18·91	34·16	25·68
30	13·23	18·91	34·16	25·76
58	13·2	18·91	34·16	25·76

September 7 to 8, 1914.

Stations I to IV, 7/9/14. Surface observations only.

Station.		Time.	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
I	54°N. ; 3°30′W.	2.25 p.m.	16·9	18·41	33·26	24·22
II	54°N. ; 3°47′W.	3.20 p.m.	16·2	18·68	33·75	24·76
III	54°N. ; 4°4′W.	4.15 p.m.	15·4	18·94	34·22	25·29
IV	54°N. ; 4°20′W.	5.15 p.m.	15·2	18·96	34·25	25·37

Station V, 53°53′N. ; 4°46′W. 8/9/14 (9 a.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	14·6	19·02	34·36	25·58
30	14·5	19·00	34·33	25·58
91	14·4	19·00	34·33	25·60

Station VI, 53°43′N. ; 4°44′W. 8/9/14 (10.10 a.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	14·8	19·01	34·34	25·53
30	14·72	19·01	34·34	25·54
62	14·7	19·01	34·34	25·55

Station VII, 53°33′N, ; 4°41′W. 8/9/14 (11.25 a.m.)

Depth (metres)	T°	Cl°/$_{oo}$	S°/$_{oo}$	σ_t
0	15·6	18·95	34·23	25·27
30	15·55	18·94	34·22	25·26
50	15·45	18·95	34·23	25·30

During the past year an important paper on the seasonal changes of salinity in the Irish Sea has been published by D. J. Matthews (*Fisheries, Ireland, Sci. Invest.*, 1913, IV [1914]). It is based on the hydrographic work carried out by the Irish Department of Agriculture and Fisheries.

Matthews shows that there is a general northward flow of water through the Irish Sea, thus confirming the conclusions drawn by myself in a paper on " The flow of water through the Irish Sea," published in the *Lancs. Sea-Fish. Lab. Report* for 1909 (also *Trans. Biological Soc. Liverpool*, Vol. 24, 1910).

As regards the hydrographic conditions in the mouth of the Bristol Channel, Matthews holds different views from those expressed by myself in the above-mentioned paper. Perhaps he is right, but, as he practically admits himself, further work is necessary to settle the point.

I must, however, protest against the manner in which Matthews has referred to a paper by Nielsen (*Meddelelser fra Kommissionen for Havundersögelser: Hydrografi*, Vol. 1, No. 9, Copenhagen, 1907), in such a way that it appears to give support to his own assumption of an eddy-like circulation of the water in the mouth of the Bristol Channel.

Nielsen originally concluded that " all around Ireland there flows a current in an anticyclonic direction." He says also (loc. cit., p. 25) that " the north-going current, west of Ireland, sends a branch around the north coast of the island and down through the Irish Channel, so that this island has a coast current in an anticyclonic direction like Iceland, Scotland, etc." He came to this conclusion largely owing to a paper by Matthews (*Report on the Physical Conditions in the English Channel*, 1903. *First Report of the North Sea Fisheries Investigation Committee* [*southern area*] London, 1905), in which hydrographic results obtained off the south of Ireland and in the English Channel were interpreted as indicating a southerly flow of water from the Irish Sea.

Now that this southward flow of water can no longer be assumed, Matthews, forgetting apparently why Nielsen postulated the anticyclonic circulation *all around* Ireland, quotes him as postulating an anticyclonic circulation of the water *only off the south of Ireland* (an eddy, that is to say) and uses this as some sort of support for his own assumption of an eddy in a cyclonic direction in this same Bristol Channel area.

The diagram I published, in the paper on the flow of water through the Irish Sea already referred to, indicated an anticyclonic eddy in the Bristol Channel which carried some less saline water southward.

I would like to point out that in the charts on which it was based, which were published in the same paper, all the results obtained by the Plymouth observers were included and the actual salinities marked on the charts. The course of the isohalines depended almost entirely on these results, and not in any essential manner upon the few " liner " observations which were also indicated on the charts. Matthews, no doubt correctly, objects to the inclusion of these latter results as being less trustworthy ; but if they are left out the course of the isotherms is not affected and Matthews' criticism of the manner in which the charts have been drawn is not really justified.

ON THE PELAGIC FISH-EGGS COLLECTED OFF THE SOUTH-WEST OF THE ISLE OF MAN IN 1914.

By Andrew Scott, A.L.S.

I hoped it would have been possible to continue the report, commenced last year, on the pelagic fish-eggs taken in the plankton collected by the Lancashire and Western Sea-Fisheries steamer, but this has not been possible. An accident to the ship in February laid her up for repairs during the most important month of the spring. By the time the repairs were completed, and the ship ready for sea again, the maximum spawning period in 1914 was past. Later on in the year the ship was taken over on Government Service, and the sea work brought to an abrupt conclusion for the time being. Only thirteen of the collections taken while the vessel was carrying on investigations contained pelagic fish-eggs, compared with eighty-three in 1913.

The samples of plankton taken throughout the year in Port Erin Bay, and on occasions outside, around the south-west of the Isle of Man in connection with the "Intensive Study" investigations, have been the only means of obtaining continuous information regarding the occurrence of pelagic fish-eggs in 1914. This in-shore area, especially Port Erin Bay, is more liable to be influenced by the action of winds and currents than the open sea. The appearance of eggs in the plankton taken in the Bay, then, will probably only give an approximate idea of the spawning periods compared with the results that might be obtained in the open water of the central area. The central area may be regarded as the portion of the Irish Sea extending from between Cumberland

and the Isle of Man to off the coast of North Wales. This includes one or two fairly well defined spawning grounds. The investigations carried on at the South-West of the Isle of Man in 1914 do not alter the approximate duration of the spawning period that is given in the XXI Annual Report for the species dealt with below. There is a well marked difference in the number of eggs of the valuable food fishes present in the Bay compared with the area outside, as shown by the tables given. It is evident, therefore, that spawning takes place outside, and the eggs are carried into the Bay by winds and currents.

The arrangement here is the same as that adopted in the XXI Annual Report, page 233.

Clupea sprattus, Linn.—Sprat.

Eggs of the sprat were observed in the plankton collected in Port Erin Bay on May 25th. They appeared to be continually floating about this area during June and July, but none were found later than July 30th. A collection taken off Kilan Head in Cardigan Bay, by the Fisheries steamer on April 5th, was estimated to contain 805 sprat eggs. Two were found in a haul 8 miles S.E. of Point Lynus on May 6th. Our records extending over eight years show that sprat eggs may occur in the plankton from the beginning of April until the middle of September. It is possible that the actual spawning period in some parts of the Irish Sea may be even earlier than is indicated by the presence of the eggs in the plankton. The reproductive organs of sprats sent to me from Morecambe for investigation on February 5th, 1915, were nearly all well advanced towards maturity. In one or two cases the ovaries and testes were quite mature.

Gadus callarius, Linn.—Cod.

The pelagic eggs of the cod are generally to be found in small numbers in the plankton taken inside the breakwater at Port Erin between the end of February and April. They are no doubt carried into the Bay by winds and currents. It is scarcely likely that spawning will take place in the shallow water of the Bay. Cod eggs are moderately abundant at times outside the Bay from a few miles West of Bradda Head to South of Calf Island. Some of the collections taken at Station III have been estimated to contain from 865 to 1,590 cod eggs. The first cod eggs appeared in the Bay on February 26th. Throughout the next two months very few of the bi-weekly hauls in the Bay, and outside of it, were without the eggs of this fish. Plankton collected 10 miles off Point Lynus on February 5th contained five cod eggs. Another sample from 11 miles N. by E. of the Liverpool North-West Lightship, on February 17th, contained three eggs. The hauls made in the central area of the Irish Sea and in Carnarvon Bay in April showed cod eggs to be generally distributed.

The following is a list of the cod eggs observed at the South-West of the Isle of Man in 1914:—

		No. of Eggs.			No. of Eggs.
Feb.	26—Port Erin Bay ...	31	Apr.	4—Station I	974
Mar.	4—Port Erin Bay ...	19	„	4—Station III	1,590
„	11—Port Erin Bay ...	4	„	8—Off Spanish Head	44
„	18—Port Erin Bay ...	19	„	9—Port Erin Bay ...	5
„	21—Port Erin Bay ...	2	„	11—Off Spanish Head	321
„	24—Port Erin Bay ...	15	„	11—Port Erin Bay ...	7
„	27—Port Erin Bay ...	6	„	14—Off Spanish Head..	17
„	30—Port Erin Bay ...	1	„	14—Port Erin Bay ...	8
Apr.	1—2 miles W. of Bradda Head ...	282	„	15—Station III	280
„	1—2 miles off Perwick	16	„	16—Station III	753
„	1—Off Spanish Head	32	„	17—Station III	6
„	2—Station I	32	„	17—Off Bradda Head...	162
„	2—Station III	160	„	18—Station III	2
„	2—S. of Calf Island ...	5	„	18—Off Bradda Head...	10
„	2—Port Erin Bay ...	4	„	18—Off Calf Island ...	1
„	3—Station I	533	„	20—Station III	4
„	3—Station III	865	„	20—N. of Calf Island...	3
			„	20—S.W. of Calf Island	6

Station I lies 5 miles West of Bradda Head.

Station III lies 3 miles North-West of Bradda Head.

Gadus aeglefinus, Linn.—Haddock.

There has been a marked scarcity of this fish in the Irish Sea during the past five or six years. No haddock eggs were obtained in 1911 and 1912. Only two were seen in 1913. They were from a haul taken five miles N.W. from Peel, Isle of Man, on May 7th. The investigations carried on in 1914 give no indication that there will be an early recovery of this fishery. A migration of the adults into the Irish Sea, however, may take place quite unexpectedly at any time. Herdman and Dawson, in the Lancashire Sea-Fisheries Memoir No. II, "Fish and Fisheries of the Irish Sea," page 47, state that the value of the Irish Sea fishery for haddock in 1900 was £35,000. The only eggs observed during 1914 were obtained from plankton collected in Port Erin Bay on May 12th and 18th. The haul on May 12th contained two eggs, and that on the 18th one only.

Gadus merlangus, Linn.—Whiting.

The eggs of the whiting appeared in the Bay plankton on March 4th, and were generally distributed there and in the open area outside during March and April. The open sea collections taken in connection with the "Intensive Study" investigations usually contained much larger numbers of eggs than those taken in the Bay. It was estimated that fully eight thousand were present in a haul from Station III on March 3rd. The collections taken in the central area of the Irish Sea in April showed the eggs to be fairly plentiful everywhere. A haul taken on April 14th, 8 miles N. $\frac{1}{2}$ W. from Liverpool North-West Lightship, was estimated to contain 4,900 whiting eggs.

The following list gives the distribution of the eggs off the South-West of the Isle of Man in 1914:—

		No. of Eggs.
Mar.	4—Port Erin Bay ...	8
„	7—Port Erin Bay ...	23
„	11—Port Erin Bay ...	16
„	18—Port Erin Bay ...	21
„	21—Port Erin Bay ...	4
„	24—Port Erin Bay ...	24
„	27—Port Erin Bay ...	6
„	30—Port Erin Bay ...	2
„	31—3-4 miles W. of Bradda Head ...	32
Apr.	1—2 miles W. of Bradda Head ...	773
„	1—2 miles off Perwick	26
„	1—Off Spanish Head..	41
„	2—Station I	218
„	2—Station III	1,040
„	2—S. of Calf Island...	22
„	2—Port Erin Bay ...	69
„	3—Station I	2,113
„	3—Station III	8,145
Apr.	4—Station I	1,369
„	4—Station III	1,120
„	8—Off Spanish Head...	134
„	9—Port Erin Bay ...	291
„	11—Off Spanish Head..	485
„	11—Port Erin Bay ...	93
„	14—Off Spanish Head..	178
„	14—Port Erin Bay ...	51
„	15—Station III	850
„	16—Station III	1,451
„	17—Station III	84
„	17—Off Bradda Head...	85
„	18—Station III	36
„	18—Off Bradda Head...	67
„	18—Off Calf Island.....	51
„	20—Station III	94
„	20—N. of Calf Island...	4
„	20—S.W. of Calf Island	20
„	24—Port Erin Bay ...	10
May	4—Port Erin Bay ...	1

Gadus virens, Linn.—Green Cod.

Eggs which were probably identical with those of the green cod were first noticed in the Bay plankton in 1914 on January 28th. They occurred throughout the whole of February, and once in March, the 31st. A haul taken on February 26th contained 114 eggs identified as *Gadus virens*. Green cod eggs were present in only one of the collections taken outside the Bay, but this is probably due to the spawning period of the fish being over before the "Intensive Study" investigation of the outside area commenced. None were found in the plankton from the central area of the Irish Sea.

Gadus luscus, Will.—Bib.

Gadus minutus, Linn.—Poor Cod.

Eggs of both these species of fish occur in the plankton of the Irish Sea, but the difference in size is so very slight that it is almost impossible to separate

them with certainty. They were observed in the central area nearly a fortnight earlier than in Port Erin Bay in 1914. The eggs were found for the first time in the Bay plankton on March 4th, and were of frequent occurrence both inside the Bay and at the observation stations outside during March, April and May. As usual, the largest numbers were taken at the open sea stations. It was estimated that 2,660 bib or poor cod eggs, probably both species, were present in a haul taken at Station III on April 3rd.

The maximum spawning period of the valuable gadoids frequenting the area at the South-West of the Isle of Man in 1914 appears to have been at the beginning of April. The largest numbers of eggs were found on April 3rd and 4th.

Onos spp., Risso.—The Rocklings.

The eggs of one or more species of rockling are amongst the first of the pelagic fish eggs to make their appearance in the plankton of the Irish Sea, and are usually the last to disappear. Their occurrence in the Bay in 1914 extended from January 19th right on through the spring, summer and autumn until October 9th. This is almost identical with the distribution found in 1910. There is a distinct variation to be found amongst the eggs, both in the size of the egg itself and of the oil-globule. It is almost certain that two species of rocklings are occasionally represented in the same haul. Rockling eggs collected in September measured 0·832 mm. in diameter with an oil-globule 0·176 mm., and 0·67 mm. in diameter with an oil-globule 0·15 mm.

Scomber scomber, Linn.—Mackerel.

Eggs identified as those of the mackerel only occurred twice during 1914. Two were found in Port

Erin Bay plankton collected on July 27th and again on July 30th.

Drepanopsetta platessoides, Fabr.—Long Rough Dab.

The well-defined eggs of the long rough dab, after being apparently absent from the area round the South-West of the Isle of Man for three years, occurred in thirteen hauls taken in the spring of 1914. All the records except one are from outside Port Erin Bay, between Bradda Head and Calf Island. They were present in numbers varying from one to nine in nearly every sample of plankton taken between April 2nd and 20th. The fish no doubt spawns in the soft muddy area in the vicinity of Bradda Head. The eggs were not observed in any of the collections from the central area of the Irish Sea. The following is a list of the positions where the plankton containing these eggs was collected:—

		No. of Eggs.			No. of Eggs.
Apr.	2—Station I	1	Apr.	11—Off Spanish Head..	9
„	2—Station III	3	„	11—Port Erin Bay ...	1
„	3—Station I	2	„	15—Station III	1
„	3—Station III	3	„	16—Station III	5
„	4—Station I	2	„	17—Station III	1
„	4—Station III	2	„	20—S.W. of Calf Island	1
„	8—Off Spanish Head..	1			

Zeugopterus punctatus, Bl.—Muller's Top-knot.

The eggs of a species of top-knot, which is probably the above, appeared to be generally distributed in the Bay from April 24th to the end of June in 1914, and in the adjoining area outside from March 31st to April 20th. There does not appear to be the same marked difference in the numbers generally present in the Bay compared with those found in the area outside. That is clearly the case with the valuable food fishes such as the cod, whiting, plaice, &c. The following table of distribution is given for the sake of comparison with the food fishes:—

	No. of Eggs.
Mar. 31—3-4 miles W. of Bradda Head ...	2
Apr. 4—Station III	6
„ 8—Off Spanish Head	1
„ 11—Off Spanish Head	2
„ 14—Off Spanish Head	3
„ 15—Station III	2
„ 16—Station III	9
„ 18—Station III	1
„ 20—Station III	2
„ 20—N. of Calf Island...	4
„ 20—S.W. of Calf Island	57
„ 24—Port Erin Bay ...	16
„ 27—Port Erin Bay ...	12
„ 30—Port Erin Bay ...	8
May 4—Port Erin Bay ...	10
„ 12—Port Erin Bay ...	6
„ 14—Port Erin Bay ...	9
„ 18—Port Erin Bay ...	5
May 21—Port Erin Bay ...	52
„ 25—Port Erin Bay ...	3
„ 28—Port Erin Bay ...	40
June 1—Port Erin Bay ...	11
„ 11—Port Erin Bay ...	5
„ 15—Port Erin Bay ...	3
„ 18—Port Erin Bay ...	12
„ 22—Port Erin Bay ...	3
„ 25—Port Erin Bay ...	1
„ 29—Port Erin Bay ...	4

Lepidorhombus megastoma, Donov.—Megrim or Sail Fluke.

The pelagic eggs of the megrim or sail fluke occur frequently outside Port Erin Bay, but are only captured irregularly inside the breakwater. They first made their appearance in 1914 in a haul taken in the Bay on March 21st, and again on the 30th. The eggs were present in nearly every haul taken at the observation stations outside the Bay from April 1st to 20th. In one of these hauls from outside the Bay it was estimated that there were 494 megrim eggs. They were generally distributed in the central area of the Irish Sea throughout the month of April. The following table gives the distribution of megrim eggs off the South-West of the Isle of Man in the plankton collected in 1914:—

	No. of Eggs.
Mar. 21—Port Erin Bay ...	1
„ 30—Port Erin Bay ...	1
Apr. 1—2 miles off Perwick	1
„ 1—Off Spanish Head..	1
„ 2—Station I	24
„ 2—Station III	70
„ 2—S. of Calf Island...	4
„ 3—Station I	494
„ 3—Station III	189
„ 4—Station I	207
„ 4—Station III	76
„ 8—Off Spanish Head..	18
„ 9—Port Erin Bay ...	9
Apr. 11—Off Spanish Head..	104
„ 11—Port Erin Bay ...	37
„ 14—Off Spanish Head..	52
„ 14—Port Erin Bay ...	35
„ 15—Station III	42
„ 16—Station III	244
„ 17—Station III	5
„ 17—Off Bradda Head...	8
„ 18—Station III	7
„ 18—Off Bradda Head...	7
„ 18—Off Calf Island.....	11
„ 20—Station III	26
„ 20—S.W. of Calf Island	3

Pleuronectes platessa, Linn.—Plaice.

The pelagic eggs of this important food fish were observed in the Bay collections as early as February 26th, 1914. They appeared to be fairly plentiful at times in the area extending from Bradda Head to Calf Island during the first two weeks of April while the "Intensive Study" investigations were being conducted. One of the hauls taken at Station III was estimated to contain 461 plaice eggs. The egg is easily recognised by its large size, corrugated shell, and absence of oil-globule. The following table gives the records of plaice eggs from the Bay plankton and from the open sea:—

	No. of Eggs.		No. of Eggs.
Feb. 26—Port Erin Bay ...	3	Apr. 11—Off Spanish Head..	3
Mar. 7—Port Erin Bay ...	1	„ 11—Port Erin Bay ...	6
„ 30—Port Erin Bay ...	1	„ 14—Off Spanish Head	3
Apr. 2—Station I	5	„ 15—Station III	21
„ 2—Station III	15	„ 16—Station III	93
„ 2—S. of Calf Island...	2	„ 17—Off Bradda Head ..	2
„ 3—Station III	41	„ 18—Off Bradda Head ..	4
„ 4—Station I	58	„ 20—Station III	1
„ 4—Station III	461	„ 20—N. of Calf Island...	1
„ 8—Off Spanish Head..	3	„ 20—S.W. of Calf Island	2

Pleuronectes limanda, Linn.—Dab.

The eggs of the dab were only observed three times in the area at the South-West of the Isle of Man in 1914. Four were found in a haul taken at Station III on April 4th, one off Spanish Head on April 8th, and three at Station III on April 17th. They were generally distributed, and at times fairly plentiful, in the central area of the Irish Sea during the whole of April.

Pleuronectes microcephalus, Donov.—Lemon Sole.

Pelagic eggs identified as those of the lemon sole do not appear to occur very often in the plankton collected at the South-West of the Isle of Man. None are recorded during the first six years of the "Intensive Study"

investigations. They were only observed once in the Bay in 1914, when three were found in a haul taken inside the breakwater on April 14th. Lemon sole eggs were also identified once in the plankton collected in the central area. A gathering taken ten miles S.W. from Morecambe Bay Light Vessel on April 5th contained 150 eggs of this fish.

Callionymus lyra, Linn.—Dragonet.

The very characteristic eggs of this fish were first taken in the Bay plankton in 1914 on February 10th. They appeared to be generally distributed in the South-West area until the end of June. Dragonet eggs are not so abundant there as in the central area of the Irish Sea, where a single haul may contain as many as 10,000. The following table gives the distribution of dragonet eggs at the South-West of the Isle of Man in 1914:—

	No. of Eggs.		No. of Eggs.
Feb. 10—Port Erin Bay ...	1	Apr. 11—Port Erin Bay ...	13
„ 17—Port Erin Bay ...	1	„ 14—Off Spanish Head	92
„ 20—Port Erin Bay ...	1	„ 14—Port Erin Bay ...	8
„ 26—Port Erin Bay ...	5	„ 15—Station III	88
Mar. 4—Port Erin Bay ...	4	„ 16—Station III	96
„ 7—Port Erin Bay ...	4	„ 17—Station III	5
„ 11—Port Erin Bay ...	4	„ 17—Off Bradda Head..	14
„ 18—Port Erin Bay ...	4	„ 17—Port Erin Bay ...	5
„ 21—Port Erin Bay ...	1	„ 18—Station III	7
„ 24—Port Erin Bay ...	2	„ 18—Off Bradda Head..	12
„ 27—Port Erin Bay ...	4	„ 18—Off Calf	2
„ 30—Port Erin Bay ...	2	„ 20—Station III	34
„ 31—3-4 miles W. of Bradda Head ...	3	„ 20—N. of Calf Island...	1
Apr. 1—2 miles W. of Bradda Head ...	8	„ 20—S.W. of Calf Island	5
„ 1—2 miles off Perwick	2	„ 24—Port Erin Bay ...	7
„ 1—Off Spanish Head..	8	„ 27—Port Erin Bay ...	3
„ 2—S. of Calf Island...	4	May 10—Port Erin Bay ...	1
„ 2—Port Erin Bay ...	1	„ 14—Port Erin Bay ...	9
„ 3—Station III	6	„ 18—Port Erin Bay ...	2
„ 4—Station I	8	„ 21—Port Erin Bay ...	20
„ 4—Station III	27	„ 25—Port Erin Bay ...	3
„ 8—Off Spanish Head..	4	„ 28—Port Erin Bay ...	9
„ 11—Off Spanish Head..	62	June 11—Port Erin Bay ...	10
		„ 15—Port Erin Bay ...	1
		„ 29—Port Erin Bay ...	3

AN INTENSIVE STUDY OF THE MARINE PLANKTON AROUND THE SOUTH END OF THE ISLE OF MAN.—PART VIII.

By W. A. Herdman, F.R.S., Andrew Scott, A.L.S., and H. Mabel Lewis, B.A.

[Introductory Note.—This is now the eighth year (1914) of our detailed analysis of the plankton collected week by week at Port Erin. Whether we shall now be able to complete our contemplated ten years of continuous observations seems a little doubtful. The taking of periodic samples in Port Erin Bay is still going on, and will probably continue to go on as before, but the observations in the open sea at three and five miles west of Bradda Head, which have been carried out every year during the time of the great vernal maximum, will not be possible during the present ninth year of the work.

The plan of work in collecting the samples, in working them up and in preparing this report, has been practically the same as in previous years. Mr. H. G. Jackson, M.Sc., again acted as my very efficient scientific assistant on the yacht "Runa" during the work at sea in April, 1914, and carried out the preservation of the material and the preliminary examination under my direction at the Port Erin Biological Station; while the six weekly Bay samples throughout the year were taken by Mr. T. N. Cregeen, and were carefully preserved by Mr. Chadwick.

Furthermore, the three joint authors have divided between them the rest of the work on the usual plan. Mr. Scott has carried on the further and more detailed examination of the samples; Miss Lewis has done the statistics, calculating the totals and averages, and drawing curves for all the groups and many of the individual organisms; while I have been responsible for supervising

the whole, and for the form in which the report is now presented. I feel that Mr. Scott and Miss Lewis have done the major part of the hard work, and that I deserve little credit except for planning the work, enabling it to be carried out, and expressing my opinion on details of the investigation and on the general conclusions.

As in the case of the last few years, we do not consider the present report to be an exhaustive statement of the results to be obtained from a study of the collections. It is again only an interim report to record the progress of the investigation. We look forward to giving a fuller discussion of the ten years' material when the series of observations is completed. We have now in hand a considerable bulk of unpublished figures, curves and other data.

We may refer readers to the previous parts of this report (from 1907 onwards) for any desired details as to the apparatus and methods of work and the results so far obtained.—W. A. Herdman.]

Material Available.

The collections made during 1914 have amounted to 393—all taken within the same limited sea-area off the Isle of Man as in former years. Our table for the whole series of samples taken during the eight years of the investigation is now as follows:—

Year.	At Sea, from Yacht.		In Bay throughout Year.	Totals.
	Spring.	Autumn.		
1907	218	279	138	635
1908	156	242	157	555
1909	329	147	231+49	756
1910	107	249	296	652
1911	120	84	314	518
1912	87	0	299	386
1913	82	41	282	405
1914	102	0	291	393
Totals	1,201	1,042	2,057	4,300

On account of the meeting of the British Association in Australia in the summer of 1914, the yacht "Runa" was not put in commission that autumn, and consequently no samples were obtained outside Port Erin Bay at that time. Nor have we any supplementary material this year from any other parts of the district. As Mr. Riddell has stated on an earlier page of this report, the Sea-Fisheries steamer "James Fletcher" ceased to make her periodic cruises after September, and the plankton samples, although worked up, are not reported on, as they form an incomplete series. They remain available, if it seems desirable, for inclusion with those of the following year.

No change has been made in the nets employed, or in the method of using them (see former reports).

Plankton of Port Erin Bay in 1914.

As before, the plan of work within the Bay has been to take two horizontal hauls (coarse and fine nets) and one vertical haul twice each week throughout the year—that is, about 24 hauls per month. The twelve months of 1914 are represented by these hauls as follows:—

Months	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII
No. of Hauls	24	21	24	27	24	27	27	27	24	27	18	21

A glance shows that only in one month (November) has weather interfered much with the work. As a rule the collections were made with regularity, and the monthly average is high. We are much indebted to Mr. Cregeen, of the Biological Station, for his successful endeavours to keep us supplied with the necessary statistical data.

On the whole, the agreement between the evidence given by the smaller vertical hauls at the mouth of the Bay and that of the much larger surface hauls obtained on the same occasions is satisfactory, but naturally the vertical hauls sometimes yield additional information.

Our total plankton curve for the Bay did not this year rise to quite such a high point in the vernal maximum as that reached in the spring of 1913, but on the other hand the autumnal maximum was greater in 1914, and, in fact, the curve remains at a higher level from August onwards to the end of the year (fig. 1).

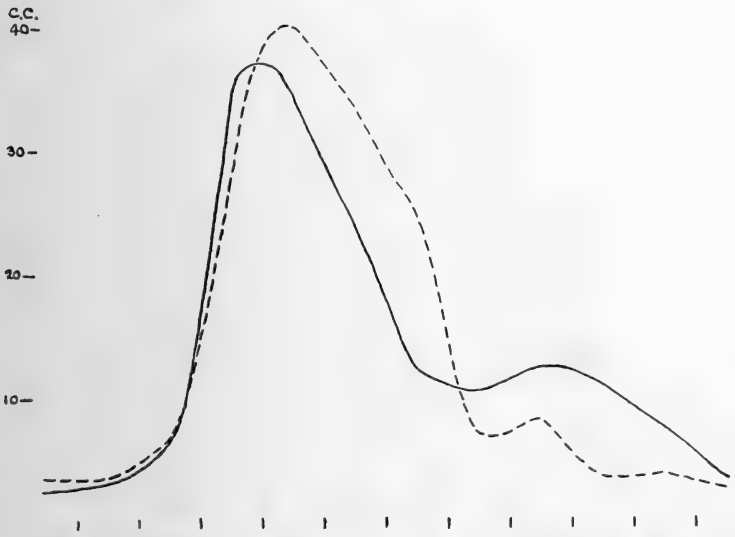


FIG. 1.—Curves for the total plankton in the years 1914 (whole line) and 1913 (dotted line).

The following table shows the monthly averages of the total catch, and of the chief groups of the plankton per haul of the standard net (coarse and fine nets together forming one standard "double haul"):—

T

1914.	Double hauls.	Average catch.	Diatoms.	Dinoflag-ellates.	Copepoda.	Copepod. nauplii.
		c.c.				
January ...	8	2·5	29,725	2,179	4,923	1,524
February...	7	3·2	70,486	1,973	7,032	3,380
March	8	6·5	759,072	5,701	7,276	7,385
April	9	34·7	3,257,761	15,278	26,121	66,601
May	8	34·8	22,370,831	37,365	12,042	40,920
June	9	24·5	2,364,992	13,911	27,281	43,978
July	9	12·7	1,259,516	5,654	40,949	44,089
August ...	9	10·8	380	2,443	37,889	52,372
September	8	12·7	206,177	1,375	69,702	32,711
October ...	9	11·3	908,827	20,658	27,411	51,022
November	6	7·9	569,727	22,503	54,549	18,905
December	7	3·7	77,220	16,343	18,546	6,984

From this table we see that the spring maximum was again in May, agreeing in this respect with that of 1911 and of 1913—though really earlier in the month than in the latter year. The actual largest haul was 88·5 c.c. on May 4th. On the whole, the curve of the monthly averages of the total catch agrees fairly well with that for 1913, but while the averages for May, June, and July are lower, those from August to the end of the year are higher. We noticed last year (1913) that the autumn maximum was an unusually low one; this year it is somewhat higher, but not so high as we have sometimes recorded.

It will be noticed from the table that although the Copepoda, as usual, attain their highest numbers much later in the year than the Diatom maximum, the Dinoflagellates this year attain to their maximum in the same month, May, with the Diatoms. Last year (1913) the Dinoflagellate maximum—a much greater one than this year—was as late as July. We consider that, however, to be unusually late.

We have drawn curves for these various groups in the two years, but consider it unnecessary to reproduce them, as the changes from month to month can be readily

followed with the eye on the table above and the corresponding one in our last report.

The summer minimum of Diatoms was unusually well marked in 1914, when the average standard haul fell in August to less than 400. In 1913 it was over 94,000. The autumnal increase, in 1914, to over 900,000 in October was much more marked than in the previous year, when the maximum was 187,000, in September. But, in fact, all the groups show larger numbers in the autumn and winter months of 1914 than in the corresponding months of 1913.

There was a rather unusual temporary increase of Copepoda in April (26,000, while May showed only 12,000), and apparently a sudden change from this zooplankton to the phytoplankton took place at the end of April. There was a large zooplankton haul (62·5 c.c.) on April 27th, consisting chiefly of Copepoda, and an equally large (70 c.c.) haul on April 30th, which was almost a pure phytoplankton (*Chaetoceras*, *Lauderia*, and *Eucampia*).

Diatoms.

There is nothing very remarkable to record in connection with the occurrence of Diatoms in the Bay plankton of 1914. The numbers first rose to over a million on March 24th, had increased to over 15 millions by April 30th, attained their maximum of 155,288,000 on May 4th, then dropped to $11\frac{1}{2}$ millions on May 7th, and remained fairly high till the second week of July. Then came the summer minimum. They began to increase again at the end of September, and reached to between one and two millions on October 9th, 15th, 18th, and November 3rd. The maximum of over 155 millions on May 4th is mainly composed of a single species, namely,

Chaetoceras debile, of which there were 148 millions present in the double haul. It may be recalled that the previous year (1913) it was another species, *Asterionella japonica*, that was present in great abundance, running up to nearly 200 millions per haul on May 16th. In 1913 the vernal phytoplankton at Port Erin was an "Asterionella-plankton," in 1914 it was a "Chaetoceras-plankton." We pointed out last year the probability that amongst the competing common spring Diatoms some slight advantage enables sometimes one form and sometimes another to gain the mastery and become for a short time enormously abundant.

The More Important Genera of Diatoms.

We give here our usual short summary of the distribution throughout the year of the more important Diatoms.

Biddulphia.—The spring maximum (78,100 on March 30th) was again low, as in the two previous years, but the autumn figures resemble closely those of 1911 when there was the unusually high maximum of 660,600 on November 24th; this year we have 800,400 on November 3rd. These are the only two years of our investigation in which the autumn maximum has been higher than the spring maximum. This includes the two forms *B. mobiliensis* and *B. sinensis*, of which sometimes the one and sometimes the other is the more abundant.

Chaetoceras.—This was again by far the most abundant Diatom in the plankton, and is represented in our gatherings throughout the year. The spring increase began early in March, the numbers reaching to over a million before the end of the month. On April 30th we had 10,443,500, and on May 7th 10,207,200, while the maximum occurred between those dates with the

enormous haul of 151,220,000 on May 4th. As pointed out above, 148 millions of this total belong to the species *C. debile*. After the middle of May the numbers rapidly fell off to a few tens or hundreds in August. They rose again in September, and reached 1,792,200 on October 9th.

Coscinodiscus.—This genus was more abundant both at the time of the spring and the autumn maximum than in any previous recorded year. The maximum was on April 30th, with 930,000 in the standard haul, and in October we had 102,000, on the 18th, and on November 3rd 83,000. The genus was practically unrepresented in our nets from the middle of June till the beginning of October.

Rhizosolenia.—The highest monthly average (2,013,916) was, as usual, in June (in 1913 it was in July, but in every other year in June), but the actual maximum was on July 2nd with 6,726,000. The genus was entirely absent from our gatherings after July 16th until October 12th, from which date till the end of the year the largest haul was only 6,000 (October 18th). Only in 1908 have the autumn numbers for *Rhizosolenia* been lower than in this year.

Thalassiosira.—This genus was less abundant in 1914 than in the previous four years, but the numbers were higher than in 1907-9. It appeared in our nets for the first time in the middle of March (with the exception of 20 specimens in one haul in January), and the numbers increased to the maximum of 750,600 on May 7th, and then fell to zero by the end of the month. It was represented again from October 9th to 23rd (15,000 on October 15th), and on two occasions in November (1,000 on the 9th, 300 on the 23rd), and was then absent for the rest of the year.

Guinardia.—The numbers for 1914 are lower than usual, the maximum being only 1,740,000 on June 11th. This year agrees with every other recorded year in having the *Guinardia* maximum in June. The highest numbers for the autumn were only 8,400 on October 12th, and 8,500 on the 18th.

Lauderia.—The maximum of this genus has been sometimes in April and sometimes in May. This year it was in the latter month, with 3,600,000 on May 4th, and the high monthly average of 831,800. Absent after June 1st until September 28th, the numbers then rose again to 88,000 on October 15th, and fell to zero by November 19th.

We give here, for comparison with those published in other years, the table showing the monthly averages of the above seven genera of Diatoms.

1914.	Biddul-phia.	Chaeto-ceras.	Coscino-discus.	Rhizoso-lenia.	Thalassi-osira.	Guin-ardia.	Laud-eria.
Jan. ...	10,075	8,195	7,397	55	2	132	69
Feb. ...	22,233	36,124	8,257	7	0	560	9
Mar. ...	45,100	625,606	49,839	2,642	2,969	4,331	162
April ...	9,972	2,355,146	329,406	93,167	8,114	55,100	373,193
May ...	2,652	20,832,575	17,285	417,112	108,131	89,762	831,800
June ...	26	15,731	430	2,013,916	0	333,356	378
July ...	26	15,085	42	1,240,334	0	3,996	0
Aug. ...	6	374	0	0	0	0	0
Sept. ...	85	206,032	25	0	0	0	12
Oct. ...	26,140	820,602	38,002	1,638	3,329	3,113	12,579
Nov. ...	406,100	110,187	37,292	420	217	3,462	275
Dec. ...	33,000	29,500	8,584	17	0	10	0

The increase and diminution of the several forms, month by month, is shown almost as clearly as it would be by a curve.

Dinoflagellata.

The monthly averages for *Ceratium* and *Peridinium* in 1914 were as follows:—

[1914.	Ceratium tripos.	Peridinium spp.	1914.	Ceratium tripos.	Peridinium spp.
January	2,102	2	July	3,987	1,582
February	1,734	0	August	1,003	1,422
March	3,835	84	September ...	1,335	20
April	7,993	3,519	October	19,522	0
May	12,947	17,322	November ...	21,917	333
June	5,744	8,027	December ...	15,571	0

This year agrees with 1912 in having the spring maximum of both groups in May, but the distribution in 1914 was more regular than in the former year. It will be seen from the figures given above that the curve of the monthly averages is very regular indeed, rising in the case of *Ceratium* from February to May, then falling to a minimum in August and rising again to the autumn (and this year higher) maximum in November. A similar regularity is to be noticed in the case of *Peridinium*. The actual maximal hauls are, for *Ceratium*, 30,000 on May 12th and 32,000 on October 15th, and, for *Peridinium*, 48,000 on May 14th, and 2,000 on November 3rd, this last figure being the sole record we have for the last three months of the year, and evidently, therefore, quite an exceptional occurrence.

Noctiluca.

Noctiluca miliaris was rather more abundant in 1914 than in the previous year, and the maximum appears to have been in May (10,200 on the 21st, 10,800 on the 28th). This is unlike 1912, when we stated that, "on the whole, *Noctiluca* seems to be least plentiful in spring and early summer, and to become more abundant in autumn." Last year we recorded as characteristic "the constant presence of the organism in small quantity." But on the whole, as we then pointed out, it is more usual for "*Noctiluca* to be either totally absent, or present in great profusion."

Copepoda.

We give here, as usual, a short summary of the occurrence of the commonest species of Copepoda.

Calanus finmarchicus.—In previous years the summer maximum has usually been in July, with a second climax in October (September in 1913); but in 1914 the maximum was in August (7,320 on the 6th), and the numbers were then low till the end of the year.

Pseudocalanus elongatus.—The maximum haul this year was 41,000 on June 22nd, but the monthly average for April was higher than that for June. In the curve of the monthly averages we have three high peaks, in April, June and August, with depressions in May and July.

Oithona similis.—The maxima this year were in July and November, with a smaller one in September. In July the largest hauls were 54,180 on the 2nd, and 73,600 on the 24th; while in November there was an unusually large haul of 199,300 on the 9th. This late autumn maximum was much higher than in any previous recorded year.

Temora longicornis.—The highest monthly average was this year in June, but the two largest individual hauls were one in April (23,200 on the 24th), and one in July (18,190 on the 27th). From this summer maximum the numbers fall off gradually to the end of the year.

Paracalanus parvus.—The maximum was, as usual, in September, following the summer minimum, and this year it was very much higher than we have had to record before, the largest haul being 138,300 on September 10th.

Acartia clausi.—The numbers were, on the whole, lower than in most recent years, the largest haul being only 17,400, on July 13th. There were only six hauls with numbers exceeding 10,000, two in June, one in July,

and three in August, so that the maximum, though low, appears to be spread over these three months.

Anomalocera patersoni.—This species was present in eleven hauls taken during April, May and June, the largest being 4,800 on April 24th, and the others ranging from 340 to 2. The large April haul probably represents an unusual invasion of this oceanic species.

Centropages hamatus.—*Centropages* is only entirely absent from our nets in two months during 1914, namely, January and December, but the numbers are, as usual, not very high. The maximum was 500, on August 20th.

We need give no further records of *Microcalanus pusillus*. It is apparently no longer of importance in our district, and there are several other forms, such as *Isias clavipes*, which are quantitatively more worthy of record.

The monthly average hauls for the eight more important species of Copepoda in 1914 are as follows:—

1914.	Calanus.	Pseudo-calanus.	Temora.	Centro-pages.	Anomal-ocera.	Acartia.	Oithona.	Para-calanus.
January ...	13	1,175	6	0	0	131	3,302	257
February ...	3	855	4	1	0	50	5,520	555
March	2	1,091	197	6	0	35	5,755	91
April	613	13,332	3,369	15	611	643	7,392	67
May	144	2,119	2,160	73	28	2,192	5,200	48
June	610	11,003	4,229	21	4	5,501	5,891	0
July	1,540	4,998	3,722	87	0	4,720	23,860	2,019
August	1,687	10,679	2,018	103	0	5,444	11,932	5,857
September	74	3,731	853	98	0	2,707	16,937	44,207
October ...	47	4,020	92	69	0	4,409	8,987	9,746
November...	19	1,664	12	1	0	921	45,845	6,013
December	2	1,456	0	0	0	303	14,874	1,817

Again we find there are slight differences in detail between this and other years, but on the whole the curves are in general much the same, and the history throughout the year can be readily followed by the eye. For example, *Temora* is obviously a summer species with its maximum in June and a minimum in mid-winter, while

Paracalanus shows a minimum in spring and early summer and a maximum later in the autumn.

Cladocera.

The numbers for this group were again low, as they were in 1913. It is obvious from the following histories that our two species of Cladocera maintain their character as a summer group.

Podon intermedium first appeared in the Bay gatherings on April 27th, and was usually present from that time onwards to October 18th. It was most abundant at the end of May and beginning of June (850 on May 28th was the maximum), and again in August (e.g., 560 on the 13th).

Evadne nordmanni made its first appearance in the Bay on April 9th, reached its maximum of 1,370 on May 28th, was present in varying quantities up to August 6th, and again from September 3rd to 19th, and on October 23rd.

Sagitta.

Sagitta bipunctata was again represented in practically every haul of our nets, but the numbers were lower than usual. On only twelve occasions throughout the year in our "official" hauls did the numbers rise to over 100, and the maximum (on May 28th) was only 400.

It ought to be noted, however, that while the ordinary surface gatherings taken across Port Erin Bay in spring contained only small numbers of *Sagitta*, we found that by using weighted nets in a deeper zone of water outside larger hauls of *Sagitta* were obtained. We had found the same on previous occasions, and it is evident that sometimes most of the *Sagitta* are below the surface.

Oikopleura.

Oikopleura dioica was, as usual, present in the Bay gatherings throughout the year. The maximum haul was 35,060 on June 1st, but the highest monthly average was in April (10,403 as against 10,099 in June), while there was a depression between these two peaks in May, when the monthly average was only 2,387. The numbers fell in July and August, but were fairly high again in September and October (10,910 on September 28th; 9,500 on October 18th; 8,550 on November 3rd). On the whole this record is more like that of 1912 than of 1913; but the differences between the three are not very great.

Various Larvae.

Echinoderm larvae were again, as in 1913, most abundant in February and in March, but the numbers reached were not quite so great—20,000 on February 26th being the maximum haul at that time of year. A few were present in other months, and there was a quite exceptional haul of over 25,000 on June 22nd, 1914.

Polychaet larvae were present throughout most of the year, and the record was very much like that of 1913, the maximum being again in spring. The largest haul in 1913 was 115,000 on March 3rd, and that of 1914 was 150,200 on March 7th.

The "Mitraria" Polychaet larva is again seen by this year's record to be a cold water form, having its maximum in spring and being rare or absent in the warm months of summer. The largest hauls were 2,580 on January 13th, 8,200 on February 26th, 4,100 on March 18th, and 10,000 on April 9th. Although February and April show the greatest individual hauls, March has the highest monthly average (1,875 against 1,431 in February

and 1,444 in April). The lowest monthly average is 9, in July.

Crab Zoeas were present in very small quantities in 1914, in most months the numbers recorded being only two or three in a haul. The largest haul, and, in fact, the only one which runs to three figures, is 320 on April 24th.

A considerable haul of the "Mysis" stage of *Crangon*, amounting to about 560 individuals, was obtained on April 8th, at a mile off Spanish Head, with a net weighted so as to tow at about 5 fathoms. Crangon larvae, although frequently present, usually occur in much smaller numbers per haul.

The Nauplii of Balanus ranged in 1914 from January to May inclusive, and during these months the average haul and the greatest single haul were as follows:—

1914.	Nauplius Stage.		Cypris Stage.	
	Average.	Greatest haul.	Average.	Greatest haul.
January ...	17	90	—	—
February...	6,504	39,800, on 26th	—	—
March	20,330	126,800, on 24th	—	—
April	14,509	76,600, on 24th	16	140, on 30th
May	3,600	28,000, on 4th	343	1,920, on 21st
June	—	—	7	61, on 1st

After this large haul early in May the numbers fell off rapidly, and not a single Nauplius was captured in June. In the previous year the range was from the end of January to late in June, and the largest haul was over 450,000 on March 26th, a good deal larger than anything obtained in 1914.

The Cypris stages made their appearance (two individuals) on April 20th, and increased slowly up to 1,920 on May 21st, and then, after a haul of 61 on June 1st, rapidly disappeared, the last captured being a single specimen on June 25th. The great reduction in

numbers in passing from the Nauplius to the Cypris stages is always interesting.

Gastropod larvae were again present in considerable quantity in every month of the year. The largest individual hauls were on February 26th (5,200), October 28th (5,800), and November 9th (13,800). In the previous year the largest hauls were in March and November. It looks as if there were two different sets of Gastropods, the one reproducing in early spring, and the other in late autumn. These larvae obviously require closer study and identification.

Lamellibranch larvae were also present in quantity throughout the year, and were more abundant than the Gastropods in every month. The highest records were on February 26th (8,800), May 12th (6,050), September 15th (15,200), October 31st (16,000), November 9th (13,000), and December 9th (15,800). These numbers, though not quite so high, correspond fairly well with those for 1913.

Medusae and Ctenophora.

Although Medusae were present in small numbers throughout the year, they only reached a hundred or over per haul in April, May, June, and October (500, the maximum, on October 9th). The numbers are a good deal smaller than those present in June, July, and September of 1913.

Ctenophora do not seem to have ever been present in any quantity in 1914. Nothing corresponding to the vast swarms of *Pleurobrachia* which we have occasionally met with in the past occurred during this year.

Fish Eggs.

Mr. Andrew Scott has dealt with the fish eggs in more detail in a separate article* in this report, so we

* See p. 274.

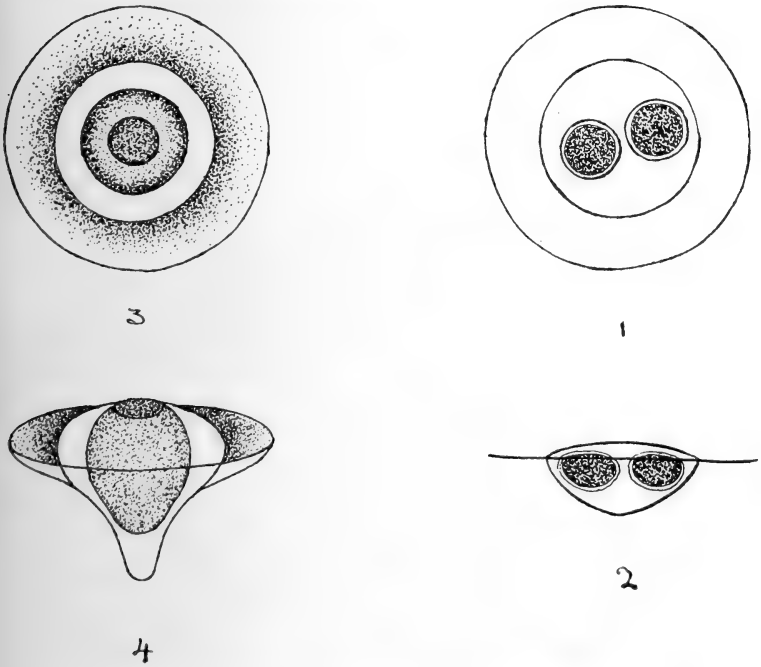
need only note here that the Rockling eggs ranged in occurrence from January 19th to October 9th, and attained their highest monthly average (80 per haul) in May; while all other fish eggs together ranged from the end of January to the end of July, and the highest monthly average was 106, in April. The greatest haul of Rockling eggs was 301 on May 21st, and of all other fish eggs taken together 445 on April 9th. It must be remembered that these numbers are for Port Erin Bay, and may be greatly exceeded by hauls in the open sea outside. Eggs belonging to as many as eight species of food-fishes occurred in some of the hauls in the open sea during April.

Further Remarks.

We have no special occurrences of rare or oceanic forms to record this year; but we may note the presence of two interesting organisms which have appeared frequently in the plankton from the Lancashire coast (Barrow Channel), and which we figure, from living specimens, as a help to the identification.

Figures 1 and 2 show an organism which is not uncommon in our local plankton. Seen from above (fig. 1) it is quite circular in outline, and with the exception of the protoplasmic bodies in the centre, it is perfectly transparent, so that the periphery is difficult to make out. The central area inside the outer rim seems curved so as to have a biconvex shape in profile view (fig. 2). It always contains one, frequently two, and rarely three or four, of the circular protoplasmic bodies. This seems to be the organism named by Hensen (who recorded it from the Western Baltic) the "Barbierbeckenstatoblast."

Figures 3 and 4 show an organism which is less commonly met with here. In this form the outer rim curves upwards, and we have never seen more than one protoplasmic body in the centre. The central cavity, containing the oval-shaped protoplasmic body, is somewhat like a spinning-top with a bluntly rounded point. This form appears to be the organism described by Cleve as *Pacillina* (*Fungella*) *arctica*, from plankton collected

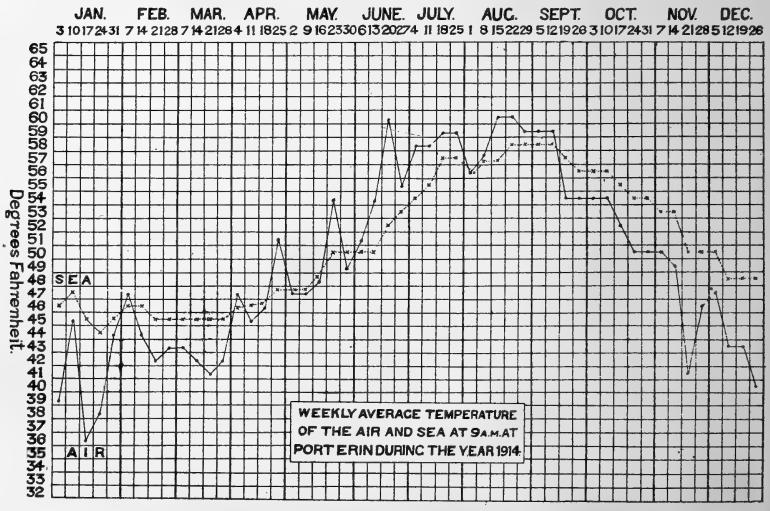


by the Swedish Expedition to Spitzbergen in 1898. It has also been recorded from the North Sea, the Skagerrak and the Kattegat. A form described by Vanhöffen under the name "Chinesenhut" closely resembles Cleve's *Pacillina*, but differs in the outer rim being turned downwards and in having the produced part of the capsule containing the protoplasmic body quite pointed.

Both these forms, or very closely related organisms, are figured by Lohmann in his "Eier und Cysten des Nordischen Planktons,"* and he is of opinion that the two are closely related and that Bergh's view that they are Molluscan egg-capsules is correct. Two or three other observers have expressed similar opinions since.

We are certainly inclined to regard both of these as Gastropod eggs, and the form shown in figs. 1 and 2 has a close resemblance to the egg-capsule of the common *Littorina littorea*. Dr. W. M. Tattersall has kindly sent a sketch of the egg-capsule of *L. littorea*, on which he has been making observations,† and this shows the closest resemblance to our fig. 1 above.

We give here the usual temperature chart of Port Erin Bay, drawn from Mr. Chadwick's daily records.



* Nordisches Plankton, Bd. I., Lief. 10, p. 17.

† Which will be shortly published in Irish Fishery Bd. Sci. Investig.

L.M.B.C. MEMOIRS.

No. XXIII. TUBIFEX

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INTRODUCTION.

I. Methods.

My investigations have been carried out on the living worm by teasing out certain parts of the body, and by means of a large number of series of transverse and longitudinal sections.

I have found the method of teasing out certain parts of the worm, either in water or salt solution, quite invaluable. By this means all the reproductive organs can be liberated and examined in the living condition, and very pretty staining effects have been obtained by the addition of methylene blue. Certain parts of the nephridia can also be obtained in this way, especially the vesicle referred to later. The vesicle is always removed with portions of the nephridial tubes, but it is difficult to remove all the nephridium because the tubes are much coiled, and the nephrostome lies in the next segment in front as in all other Oligochaeta.

In preparing specimens for section cutting, various preserving fluids and staining reagents were employed. The killing process is a difficult one, as the contortions of the worm on the addition of any chemicals are very violent. The worm, if left to itself during the process, becomes so bent and shrunken that it is useless for sectioning. It is advisable, therefore, to hold the worm at both ends while it is being immersed in the killing fluid, and thus any serious contraction of the internal organs may be prevented. The great disadvantage of this method is that the brain and anterior part of the nerve cord are usually pressed out of shape, but it is easy to hold a few specimens nearer the middle, while killing them, when the anterior end can be obtained in fairly good condition.

For killing and preserving, Tellyeoniczky's aceto-bichromate was first used, but the tissues shrank badly in this reagent. I then tried Perenyi's fluid and Bles' fluid,* and these, while killing the worm very quickly, do not cause the tissues to shrink. Although, later on, other killing reagents were used, I found none so generally satisfactory as Bles' and Perenyi's fluids.

Zenker's fluid was used, especially for nephridia, with good results.

The staining reagents most commonly used were borax carmine with picro-indigo-carmine as a counter stain, and brazilin,† which is a very delicate and effective stain. Iron haematoxylin was also used, and this rendered the muscle fibres very distinct.

II. Historical Survey.

Before entering upon a description of the minute anatomy of *Tubifex rivulorum*, which forms the subject of this Memoir, it will be interesting to notice to whom we are indebted for the present condition of our knowledge of this form.

As early as 1745 Bonnet referred to it, but was content with describing certain peculiarities in the habits and general form of the worm, and with referring to its method of regeneration after artificial fission. He did not attempt to give any details of its structure. Schoeffer in 1764 gave a figure and description of *Tubifex rivulorum*, which he called "Kleinen Wasseraal."

* Bles' fluid ... ... 90 parts 70% alcohol.
7 ,, strong Formol.
3 ,, Glacial Acetic Acid.

† Brazilin ... ... 1% Iron Alum in 70% alcohol.
0·5 % Brazilin;

O. F. Müller in 1773 classed it under Lumbricus as *L. tubifex*, but his description is very imperfect, and is largely corrected by D'Udekem. Lamarck (1816) separated *Lumbricus tubifex* and *L. lineatus* of Müller's Lumbricus and formed a new genus, Tubifex: the first he called *Tubifex rivulorum*, the second *Tubifex marinus*. Owing to apparent similarities in the structure of these worms and that of the Naiads, Lamarck united them in a class Hespides. In 1842 Hoffmeister recalled the attention of naturalists to the old "genus *Lombric*" of Müller, which he divided into three new genera, Lumbricus, Enchytræus and Sænurus, and he placed *Tubifex rivulorum* in his new genus Sænurus. It differed from the "genus *Lombric*" by the absence of a gizzard and by the fact that the setae are of unequal length. His observations on *Tubifex rivulorum* are very incomplete and inaccurate.

Grube in 1851 classed Tubifex among the Naiads under the name given to it by Hoffmeister—(*Sænurus variegatus*), and he assigned to it the following characters: "Ohne Kiemen, Borsten Bundelchen zweizelich, obere Borsten haar und hakenformig, selten obere und untere hakenformig, Blut lebhast roth oder roth gelb."

Budge (1850-51) has provided us with descriptions of the genital and respiratory organs. These descriptions are more accurate than those of Hoffmeister, but still very incomplete.

D'Udekem in 1855 published an account of the anatomy of the worm, dealing with all the systems in order, but studying them only in the living animal and in preparations made by teasing out parts of the body and then subjecting them to examination. His observations are certainly more detailed and accurate than those of

earlier observers, but there are several inaccuracies, most of which have been noticed and corrected by still more recent observers. Many of the terms then in common use have since been discarded, and to some of these Beddard in his Monograph of the Oligochaeta refers, viz., the capsulogenous gland is now known as the spermatheca, the cloaca as the spermiducal gland, the vesicula seminalis as the sperm sac and the matrice as the egg sac.

In 1871 McIntosh published a paper in which he gives short accounts of certain of the organs, but no complete description of any system except the circulatory, as he concentrated attention on certain minor points such as the perivisceral fluid and corpuscles and the granular glands which form a complete investment around the intestine and dorsal vessel. His results will be considered in detail later.

Vejdovsky in 1884 published his well-known work "System und Morphologie der Oligochaeten." In this he makes frequent reference to Tubifex, and increases a good deal our knowledge of the structure of this worm. But in a work which embraces the whole of a large group it is impossible to describe in any great detail the structure of one member of that group. The same may be said of Beddard's Monograph of the Oligochaeta, published in 1895.

Several other papers have been inserted in the Bibliography at the end of this work. Some of these deal only with one small part of the subject, and reference will be made to them in the right places. Others have been mentioned in the list because they have some more distant bearing on the work, but yet are sufficiently allied to it to be of interest.

It has seemed advisable to revise all this scattered work, and, by a careful and detailed investigation of the

worm, to build up a Memoir which shall give, as far as possible, a full account of the structure of this form, together with any other points of interest which may arise during the investigation.

This paper, previous to its publication, was presented as a thesis for the D.Sc. degree of the University of London, and in this connection I wish now to express my gratitude to Professor Jackson who kindly photographed the drawings for me. It is with pleasure also that I acknowledge my indebtedness to Professor Dendy for valuable suggestions made while the work was in progress and for careful criticism of the finished work, which was carried out under his supervision in the Zoological Laboratory at King's College.

III. General Remarks.

Tubifex rivulorum is found in large numbers in the mud of the estuarine Thames, where, at low water, they may often be seen as bright red masses. In every consignment of mud which was examined this species was always found associated with another worm—*Limnodrilus udekemianus*—both belonging to the family Tubificidae. The two worms can easily be distinguished from one another if they are examined under a low power of the microscope; but, with practice, it is also possible to separate them when in the mass. One notices that the anterior segments of all of them are reddish in colour, but there is a marked difference in the posterior segments of the two forms. In the case of *Tubifex rivulorum* the posterior segments also are red in colour, but in *Limnodrilus udekemianus* the red colour is masked by the presence of pigment in the body wall. The pigment is yellow or orange in colour, and is confined to a narrow

band in each segment, forming an incomplete ring around it and giving the worm a striped appearance.

If specimens of both worms are examined under the microscope, it will be seen that the setae are different, Tubifex showing two kinds of setae, capilliform and sigmoid, while Limnodrilus has only sigmoid.

If the receptacle in which the worms are being kept is suddenly jarred with a sharp knock, they all simultaneously contract, hiding themselves almost completely in the mud. *Tubifex rivulorum* is also sensitive to light, for, if a bright beam from an arc lamp be projected through the water on to the mass, all the worms belonging to this species contract instantly and disappear. *Limnodrilus udekemianus* appears to be unaffected by the light, for the posterior end of the body still continues to wave about in the water.

If individuals of both species be placed on a slide, it is possible to distinguish them by their movements. *Tubifex rivulorum* is much more rapid in its movements, and retains its activity much longer than *Limnodrilus udekemianus* after being deprived of the mud in which it lives.

EXTERNAL CHARACTERS.

Tubifex rivulorum is a small worm varying from 30-50 mm. in length, and is of a bright red colour. The colour is due to the presence of red blood, which is clearly visible through the transparent body wall. The worm is bluntly pointed at both ends, the widest part of the body being in the anterior third. The body is divided throughout its length into a number of segments, all quite clearly marked off from one another, and decreasing in size towards the posterior end. The number of segments

in a fully-developed worm varies from 112 to 130. The worm never exhibits any "secondary" annulation of the segments, such as occurs so generally in Limnodrilus.

The Prostomium:

This appears as an outgrowth or process of the first segment. It is conical in shape with a slightly blunted apex and overhangs the mouth on the dorsal surface. It is separated from the first segment by a transverse furrow. It is extremely sensitive, and is undoubtedly used as an organ of touch, and consequently is plentifully supplied with nerves which arise from the brain.

Setae:

The setae, as in all Oligochaeta, are the organs of locomotion. They consist of chitinous rods derived from specialised cells of the epidermis. Part of each seta is buried in the body wall, and may project into the body cavity, but the rest protrudes beyond the surface of the body.

The development of these organs has been carefully observed and described by earlier writers, especially Vejdovsky, who first established the fact that all setae are ectodermal in origin. A varying number of setae are implanted in "setigerous sacs or follicles" which arise as invaginations of the epidermis. The arrangement of the setae in a sac is like that of the sticks of an open fan (Pl. II, fig. 5). Each seta has its origin at the base of the sac, but it is lodged in a separate cavity and divided from its fellows by a small piece of the body wall. The cuticle, which forms the outermost layer of the body wall, is continued into each of the cavities, and forms a lining to it for the greater part of its length. Near the blind end of the sac the cuticle is absent, and is replaced by a large

group of ectodermal cells whose boundaries are indistinct, but whose nuclei are large, round and nucleolated. According to Vejdovsky, any of these cells are capable of giving rise to new setae to replace those which may have been lost. The free ends of the setae are seen constantly to change their position as the worm rapidly coils and uncoils itself. Sometimes they are directed forwards, then backwards, and often, especially when the worm is quiet for a moment, they are placed almost at right angles to the body. When the worm is crawling forwards the setae are always pointing somewhat backwards, but a sudden twist of the body is sufficient to change their position completely.

There are two sets of muscles by which the setae are moved :—

(*a*) The parieto-vaginal muscles, which are attached to the base of the setigerous follicle and to the body wall on all sides of it. These muscles regulate the movements of the whole follicle. If those lying on the posterior side of the sac contract, the inner ends of the setae are drawn back, and the free ends are pushed forward, whereas, if those muscles on the anterior side contract, the setae are directed backwards (Pl. II, fig. 5, *m.p.*).

(*b*) The intrafollicular muscles (fig. 5, *m.f.*). These are attached to the body wall and to the individual setae, and thus the movements of the setae can be regulated.

The development of the setae in this worm resembles, in all essentials, that of the setae of other Oligochaeta, and therefore it is only necessary to refer to it briefly. The setae appear as small cones of chitinous substance at the bottom of the sac. The apex of the seta is first developed and then growth in length proceeds from the inner end until the seta has pierced the body wall and attained its full length.

The setae are arranged in four bundles in every segment of the body except the first and the last. Two of these bundles are ventral in position, and lie one on either side of the mid-ventral line. These are spoken of as the ventral bundles (Pl. III, fig. 11, *v.bl.*). The other two are much more dorsal in position, and are referred to as the dorsal bundles (fig. 11, *d.bl.*).

Three kinds of setae are present, viz., capilliform, uncinate, and pectinate (Pl. II, fig. 6). The distribution of the capilliform setae is limited, for they are confined to the dorsal bundles, and do not usually extend further back than the 30th segment. The uncinate setae are common to both dorsal and ventral bundles throughout the body, while the pectinate form is limited to the dorsal bundles of the anterior segments, and is absent from some of these.

The capilliform setae are long, straight, hair-like in form, and narrowing to an extremely fine point at the free end (Pl. II, fig. 6, *d*). By far the greater part of the seta is exposed, only about a quarter of it being embedded in the setigerous sac. These setae are quite smooth and devoid of barbs, such as are described by Beddard (1895) as occurring in *Lophochaeta ignota*. The longest ones occur in segments 6 to 9; behind the latter segment they become smaller and smaller, until, at last, they completely disappear. It is very unusual for there to be more than three capilliform setae in a bundle, and very commonly only one or two are present. In those cases where three setae are present, they vary considerably in length, and as a rule two of them are long while one is much shorter.

Beddard (1895), in his classification of the setae of Oligochaeta, divides them into two main groups:—

(*a*) Long, slender setae gradually diminishing in diameter towards the pointed extremity—capilliform.

(*b*) Shorter setae of a curved form, something like an elongated S with a thickening at about the middle—sigmoid setae.

The uncinate and pectinate setae of *Tubifex rivulorum* belong to this latter class. Of these, the uncinate form is much more common than the pectinate. They are both of the same general shape, but the thickening referred to by Beddard is not very strongly developed. Neither does it occur at the middle of the seta, but is always nearer the outer free end and approximately divides the seta into a distal third and a proximal two-thirds (Pl. II, fig. 6, *a*) In the uncinate setae the distal end is bifid, but the two prongs vary somewhat in size and shape in different setae. The uncinate setae in the dorsal bundles have the two prongs equal in size and of the same shape, that is, somewhat narrow throughout and tapering gradually to a very fine point at the distal end (Pl. II, fig. 6, *b*). In the ventral bundles, where the uncinate setae only are present, the two prongs are of slightly different size and shape. The dorsal one is somewhat slender, and comes to a sharp point at the tip. The ventral one is shorter than the dorsal, and of a blunter nature, its apex being somewhat more rounded (Pl. II, fig. 6, *a*). There is never, however, such a marked difference in the size of the prongs as in the case of Limnodrilus, where the ventral one is always much shorter than the dorsal.

The number of uncinate setae which may be found in a bundle differs somewhat in different regions of the body. For instance, in both dorsal and ventral bundles of the first two setae-bearing segments there are never more than two setae, and these are very small compared with those which come close behind. In segments 3 to 9 or 10 they reach their maximum size, and there are often four or five present in the ventral bundles, but rarely more than

three in the dorsal ones, where they are accompanied by capilliform and sometimes by pectinate setae. More posteriorly they decrease in size and number, soon dwindling down to two in a bundle, and near the posterior end of the body it is usual to find only one small seta in each bundle.

The pectinate setae are, as already mentioned, less numerous than the uncinate type, and are confined to the dorsal bundles of the anterior segments. There, therefore, it is possible to find in the same bundle setae of three different kinds: capilliform, uncinate, and pectinate, the total number rarely exceeding six. The pectinate setae resemble the uncinate of the dorsal bundles in general form. The only difference between them is the presence, in the pectinate form, of subsidiary prongs between the two main divisions of the seta (Pl. II, fig. 6, *c*). These subsidiary prongs are figured and described by McIntosh, Lankester and Beddard. Their number varies from 1 to 4, the latter number occurring most rarely.

I have not yet been able to identify the webbed setae described by Lankester (1871). He discusses at some length the form of the sigmoid setae in this worm. He states that he has often seen sigmoid setae, in the dorsal bundles of the first ten segments, which are provided with a web between the two prongs. Benham, who also investigated this species, could not identify the web. During my observations on the setae I have particularly looked for the appearance of this web, but have always failed to find it. A possible explanation of this difference of opinion is that Lankester was looking at the pectinate setae which do occur in the dorsal bundles of these anterior segments only, and mistook the subsidiary prongs for a continuous web stretching across between the two main prongs. Again, Lankester recounts how he has seen

a number of hairs (6 or 7) surrounding certain of the setae near the apex. He states that they result from the splitting up of the horny substance of the seta. He goes on to describe how a number of small dark particles are placed at intervals along the hairs. It is quite easy to recognise the condition that he describes, but it seems to me that his observations may bear another interpretation. When the worms are kept in the laboratory for a time, even though they are placed under running water, they become infested with fungus growths. These have the form usually of long, delicate filaments which appear to have their origin between the prongs of the setae, and present exactly the appearance described by Lankester.

THE BODY WALL.

As in most Oligochaeta the body wall consists of the following layers:—(1) Cuticle, (2) Epidermis, (3) Circular Muscles, (4) Longitudinal Muscles, and (5) Peritoneal Epithelium.

1. *The Cuticle* is a delicate layer of non-cellular nature which lies outside the epidermis, and is formed as a secretion from the epidermal cells. It completely invests the body, and is about 3 μ thick.

2. *The Epidermis* consists of a single layer of cells throughout, but varying somewhat in thickness in different parts of the body. For purposes of description we may divide the epidermis into:—(*a*) Ordinary or extra-clitellar epidermis; and (*b*) Clitellar epidermis.

(*a*) *Ordinary or Extra-Clitellar Epidermis.* The ordinary epidermis consists of two well-marked types of cells, viz., gland cells and columnar supporting cells, and is from 6 μ to 8 μ thick. The gland cells are large, somewhat irregular in outline, and with the nucleus, as a rule, situated in the lower half of the cell. The

cytoplasm of these cells is densely granular. The nuclei are rounded or slightly oval in shape, and each contains a nucleolus (Pl. II, fig. 5, *hyp.*).

The gland cells are separated from one another by the columnar epidermal cells, or "supporting cells" as Atheston (1898) calls them. They are narrower than the gland cells, generally columnar in outline with the broader end lying next to the cuticle. The nuclei are oval in shape, quite conspicuous, and situated near the middle of the cell.

(*b*) *Clitellar Epidermis.* The clitellum does not develop until the worm has nearly reached sexual maturity. It then becomes differentiated from the ordinary epidermal cells, and is confined to the principal reproductive segments, viz., segments 11 and 12. It has the form of a complete girdle, but is less well developed on the ventral than dorsal surface. It merges gradually into the ordinary epidermis anteriorly and posteriorly. It is discontinuous, of course, at the openings of the setigerous follicles, the spermathecal pore and penis.

It is composed of a single layer of cells throughout. At first, the cells are little different from the ordinary gland cells of the epidermis. The nucleus is a conspicuous rounded body, situated at the base of the cell, and exhibits a very well-marked reticulum with a nucleolus. The cytoplasmic contents of the cell are finely granular.

This is the usual structure of the clitellum, even in individuals which appear to be quite mature, and, consequently, this is the condition which has been described by most observers. Atheston (1898), for example, says of the gland cells of the clitellum that they are smaller and more numerous than those of the ordinary epidermis, otherwise they are similar.

Ekitaro Nomura (1913), in his description of *Limnodrilus gotoi*, gives a short account of the clitellar gland cells as they occur in that worm. He says that the gland cells are 20-23 μ long, that is, four times the length of the ordinary clitellar cells, and 8-10 μ across. "Three well-marked stages can generally be observed in mature specimens: a highly vacuolated condition, a more or less granulated condition, and one in which the cells contain many globules."

In most of the specimens of *T. rivulorum* that I examined the clitellar gland cells were only a little larger than the ordinary gland cells, and the contents were granular. In a few cases, however, the clitellum was enormously enlarged, and I can only suppose that the maximum development of the clitellum is not reached until a very short time previous to the formation of the cocoon. The cells at this time may be as much as 40 μ to 45 μ long, and 11 μ to 14 μ across (Pl. III, fig. 12). They are almost oblong in shape, tapering just a little at the inner end. The nuclei are still visible, but are much less distinct. They remain near the inner end of the cell. The increase in size of the cells at this time is accompanied by the deposition in them of a large number of globules of the secretion which forms the cocoon. Many of the cells are quite full of the secretion, which masks, almost completely, the cytoplasmic contents. In others, where the secretion is present in smaller quantities, it is arranged fairly regularly in the form of rounded masses, 2 μ in diameter, which congregate principally near the lateral walls of the cell, where they are arranged in longitudinal rows (Pl. III, fig. 12, *se. c.*). The cytoplasm is somewhat vacuolated. Between these enlarged and modified gland cells are supporting cells, which are as long as, but much

narrower than, the gland cells. The nuclei of these cells are oval in shape, and may be situated in any part of the cell (fig. 12, *in. c.*). Certain fibres from the longitudinal muscular layer may turn outwards and come into close contact with the inner ends of the gland cells.

3. *The Circular Muscle Layer* lies just within the epidermis. It is an extremely delicate layer, and is with difficulty recognised in transverse and longitudinal sections. Occasionally it can be fairly clearly seen in an oblique section, when the individual fibres appear as incomplete bands surrounding the body (fig. 5, *c. m.*).

4. *The Longitudinal Muscles* are better developed than the circular, and can be seen in longitudinal sections of the body wall as elongate fibres. Seen in transverse section they have the appearance of flat plates or lamellae embedded in a granular substance. There is no axial core, as is so characteristic of the longitudinal muscles of Lumbricus, neither are they divided into dorsal and ventral regions by lateral lines, as is described by Nomura (1913) for *Limnodrilus gotoi* (fig. 5, *l. m.*).

5. *The Peritoneal Epithelium* consists of a single layer of somewhat flattened cells with prominent rounded nuclei.

COELOM AND COELOMIC FLUID.

The coelom of *Tubifex rivulorum* is, as is usual in the Oligochaeta, well marked and spacious, and divided into a series of compartments by septa. The number and arrangement of the septa correspond to the external segmentation of the body. The septa between segments 1 to 4 are incomplete. The coelom only communicates indirectly with the exterior—by means of the nephridia and genital ducts. There are no dorsal pores such as are

found in earthworms. The coelom shows no division into different cavities, other than that due to segmentation, with the exception of the egg sac and sperm sac, which are simply portions of the coelom bounded by special walls and arising as simple outgrowths of the coelom of certain segments. These organs will be described in detail with the rest of the reproductive system.

The coelom is lined throughout by the delicate peritoneal epithelium which is also reflected over the other organs in the body cavity. The shape and size of the cells forming this epithelium vary considerably in different parts of the body, but for the present these modifications in its structure will only be mentioned briefly. The parietal and visceral layers are those which form the innermost layer of the body wall and the outermost layer of the intestinal wall respectively. The cells of the parietal layer have already been described. The visceral layer is formed chiefly of much enlarged pear-shaped cells—the "chloragogen cells" of Claparède. This applies to the visceral layer around the intestine. In the region of the buccal cavity, pharynx and oesophagus, the cells of this layer very nearly resemble those of the parietal layer. In certain regions of the nephridial tubes the peritoneum is composed of very large, vesicular cells which are very easily detached from the tube and from each other. In other parts of the tube, chiefly those nearer the nephrostome and nephridiopore, the ordinary, flattened epithelial cells are present.

The coelom is always filled with a colourless coelomic or perivisceral fluid, by which all the organs of the body are constantly bathed. Suspended in the fluid are a number of colourless corpuscles which may be described

as coelomic or perivisceral corpuscles. The number of these corpuscles in different individuals varies enormously, but two kinds are usually recognisable—amoeboid and spherical. In some individuals the spherical corpuscles are much more numerous than the amoeboid, while in others they are more evenly distributed. It is often rather difficult to decide whether a particular corpuscle is in the amoeboid or spherical condition. It is not really actively amoeboid, yet it is irregular in outline and with no clearly defined contour. It would seem, therefore, that transitional stages may occur at such times when an amoeboid corpuscle, having, as Beddard suggests, become loaded with granules of an excretory nature, ceases to be amoeboid and gradually becomes spherical in outline. Both kinds of corpuscles are granular, the granules sometimes being of a yellowish hue.

THE ALIMENTARY CANAL.

The general arrangement of the various regions of the alimentary canal can be clearly seen if the living worm be examined under the microscope, owing to the transparency of the body wall. If examined in this way, it will be seen that the wall of the alimentary canal in the first five segments is almost colourless, or only slightly yellowish in colour. In segment 6, however, the appearance changes very suddenly, for the wall has now a blackish or brownish hue, due to the presence of specialised cells of a glandular nature known as chloragogen cells, which will be described in detail later. More posteriorly the colour again changes, becoming gradually lighter and lighter until in the region of the anus the wall of the alimentary canal has again a pale yellow colour. As in all Oligochaeta, the alimentary canal has

the form of a straight tube extending from the mouth, situated on the ventral surface of the first segment, to the anus, which is surrounded by the last segment of the body, or the anal segment.

The alimentary canal can be divided into the following regions:—(1) mouth, (2) buccal cavity, (3) pharynx, (4) oesophagus, (5) intestine, and (6) anus.

1. *The Mouth.* The mouth is ventral in position, surrounded by the first segment of the body or peristomium, and overhung by the prostomium. When closed it appears as a transverse slit bounded by a slightly puckered wall, but when it is open the aperture is rounded (Pl. II, fig. 4, *mo.*).

2. *Buccal Cavity.* The mouth leads into the buccal cavity, which is short—only extending through the first segment of the body. It is partly covered by the cerebral ganglia, dorsally. The buccal cavity is capable of extrusion, but this does not seem to take place very often under ordinary conditions. I have, however, frequently observed it when ether has been added to the water in order to quiet the worm when it is under observation. The buccal cavity can then be seen as a somewhat frilled organ protruding from the mouth. When once the cavity has been extruded, it is not readily drawn in again.

A transverse section of the buccal cavity, when not extruded, shows it to be of wide calibre, with a straight, unfolded wall. The latter is very thin, and is composed of a single layer of epithelial cells, somewhat cubical in shape, with well-marked nuclei and numerous short cilia. Outside this layer of epithelial cells a few muscle fibres are scattered, but they are not sufficiently numerous to form definite muscular layers (Pl. II, figs. 4 and 7, *bu.c.*).

3. *The Pharynx.* Leading out of the buccal cavity is the pharynx, which extends through segments 2 and 3.

It is surrounded by the peripharyngeal connectives which connect the cerebral ganglia, lying dorsally, with the sub-oesophageal ganglion or anterior end of the ventral nerve cord. The pharynx can never be everted, and is provided with exceedingly muscular walls, the musculature being chiefly dorsal in position. The lumen is folded and ciliated, the cilia being considerably longer than those of the buccal cavity (Pl. II, fig. 4, *ph.*).

4. *The Oesophagus.* The pharynx opens into the oesophagus, which also extends through two segments (4 and 5), and communicates at its posterior end with the intestine. The oesophagus is a narrow tube, its lumen somewhat folded and ciliated. The wall is thin, consisting of a ciliated epithelium internally and a few muscle fibres externally, arranged in circular and longitudinal directions, the circular muscles inside (fig. 4, *oe.*).

Situated in those segments occupied by the oesophagus, and, to a less extent, those occupied by the pharynx, are organs of a glandular nature known as septal glands. These organs occur in many Oligochaeta, but are specially well developed in aquatic forms, although they have been described by Vejdovsky as being very well marked in *Allolobophora foetida.* These glands are attached primarily to the septa, but may extend on to the walls of the oesophagus, and to a large extent lie freely in the body cavity. Each gland is a mass of pear-shaped cells, the narrower part of each being prolonged into a duct which opens into the oesophageal region of the alimentary canal. The cells possess very distinct nuclei, situated in the broader part of the cell (Pl. II, fig. 4, *s.gl.*).

5. *The Intestine.* The intestine commences in segment 6, and extends throughout the entire remaining length of the body to the anus, which, as already stated,

opens to the exterior on the last segment (Pl. II, fig. 4, *in.*). It is overlaid in the anterior segments by the supra-intestinal blood vessel, and more posteriorly by the dorsal vessel, and itself covers the ventral vessel which lies freely in the body cavity. The intestine is kept in place by the septa, which constrict it slightly at intervals, and by special muscles which pass from it to the body wall. Its lumen is larger than that of the oesophagus, and is unfolded. It is ciliated throughout, the cilia being specially long and abundant in the posterior segments of the body, especially the last six or seven segments.

The structure of the intestinal wall from within outwards is as follows:—

(*a*) A single layer of somewhat elongated cells forming the ciliated epithelium. The cilia are not very conspicuous except at the anterior and posterior parts of the intestine (Pl. III, fig. 13, *ct.ep.*).

(*b*) A very thin muscular layer composed of a number of isolated circular (on the inside) and longitudinal (on the outside) muscle fibres (fig. 13, *c.m.; l.m.*).

The wall of the intestine is very vascular, and the blood vessels are situated between the ciliated epithelium and the layer of circular muscles (fig. 13, *b.v.i.*). These vessels form what is known as the intestinal network (see circulatory system).

(*c*) A glandular layer composed of a very large number of unicellular glands which cover the entire surface of the anterior half of the intestine, and also cover the dorsal blood vessel (fig. 13, *c.c.*). It is to the presence of these glands that the dark colour of the intestinal wall is due. They were called chloragogen cells by Claparède, and this name is still applied to them. When examined in section, they are seen to be somewhat

club-shaped cells with a fine external membrane enclosing a large number of granules of two kinds, some being much larger than the others, and of a dark brownish hue. Both kinds of granules are very inert, and stains and other reagents have little or no effect upon them.

If the body wall of the worm be ruptured, these cells, which are very easily dislodged from the wall of the intestine, can be liberated into the water. In the water they swell considerably, and the two kinds of granules which they contain can be clearly seen (Pl. III, fig. 14). The larger ones tend to congregate near the centre of the cell, the outline of which becomes rather indistinct and shadowy when the cell is immersed in water. The smaller granules can be more clearly seen near the periphery of the cell where the larger granules are not so abundant. All the granules of a cell which has been freshly liberated from the body of a living worm exhibit active molecular movements, and this movement is kept up for some time if the wall of the cell is not broken. Gradually, however, this molecular movement becomes slower and slower until it finally ceases. The granules then appear to congregate at one part of the cell, sometimes at the side, sometimes nearer the middle, the rest of the cell being quite clear and transparent. Probably this massing together of the granules heralds the breaking down of the cell.

All these points in the structure of the chloragogen cells can be made out in the fresh material. This amount of investigation was carried out with considerable care by McIntosh, and his results were published in his paper entitled "On some points in the structure of Tubifex" (1871). In quoting Dr. Buchholz' paper "Beiträge zur Anat: der Gattung Enchytræus" (1862), McIntosh mentions that this author has put a distinct nucleus in

all his figures of the chloragogen cells of Enchytræus. McIntosh appears to have made efforts to demonstrate the presence of a nucleus in the fresh, unstained chloragogen cells of Tubifex, but owing, as he says, to the large number of granules always present in the cell, he was not able to do this. It certainly does need considerable care in observation in order to see the nucleus, but it is comparatively easy to do so if the cells are floated on to a large quantity of water on a slide and a cover-glass put on. The cells then roll about in the water, and during their movements it is often possible to see the nucleus. McIntosh also states that even after careful examination of transverse and longitudinal sections of the worm he arrived at the same result. I find, however, that in any section of the intestinal part of the worm, which has been appropriately stained, viz., with borax-carmine, the nuclei can be very plainly seen, as they are large and each has a distinct nucleolus (Pl. III, fig. 13).

The true shape of the cells is better seen in sections than by examining them in the fresh condition. In sections they are seen to be truly pear-shaped, the broader end lying freely in the body cavity. In such preparations the nucleus, which is oval in shape, is usually situated in the narrower part of the cell. It is quite useful to make permanent smears of these cells and stain them on the slide. A stain which differentiates the nucleus particularly well is Brazilin. In these smears the granules stain very slightly: not nearly so heavily as does the nucleus (Pl. III, fig. 15).

Rice (1902) has published a paper on the chloragogen cells of *Lumbricus herculeus*, and he enters in some detail into their origin, growth and structure, and gives some suggestions as to their function. It is evident from his descriptions that their growth and structure are very

similar to those of the chloragogen cells of Tubifex. Rice carried out a series of feeding experiments, and found that neither excess of food, nor starvation, nor a varied diet, had any effect upon these cells. He also pointed out that the granular contents are apparently lifeless, for no stain appreciably affects them, nor are they altered in any way by the addition of strong acids. The lifeless condition of the adult cell suggested to Rice the possibility that these cells have performed their function in the young worm—presumably they would have some connection with the elaboration of food, as they cover the dorsal vessel as well as the intestine—and have become functionless in the adult. This theory is supported by the fact that these cells in a very young worm exactly resemble those of the ordinary peritoneum. In the larger worms a gradual development into typical chloragogen cells can be traced. The cells increase in length, the characteristic granules appear, and they become less and less responsive to stains, with the exception of the nucleus which stains throughout. Earlier observers believed these cells to be secretory in function, but, judging from their lifeless condition in the adult, their function, if any, would surely be one of excretion rather than secretion.

CIRCULATORY SYSTEM.

The circulatory system, as in all other Oligochaeta, is a closed one, having no communication with the coelom. It consists of a series of main vessels having a longitudinal direction, and united with one another by lateral vessels in each segment. The blood vessels are well-developed tubular structures, and their walls are very delicate. In none of the vessels does there seem to be an epithelial

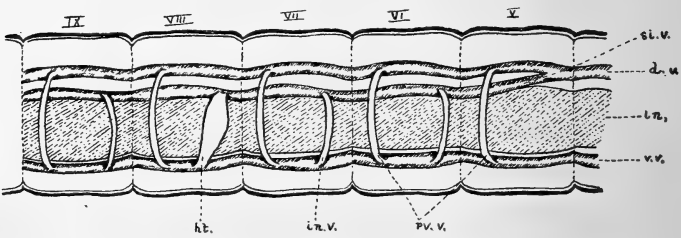
lining. Its place in these lower forms is taken by a delicate membrane, destitute, apparently, of any cellular structure. Beddard described the same condition in other aquatic worms, viz., Naiads and Enchytræidae. It is only in the true earthworms that this membrane is replaced by an epithelium consisting of large and conspicuous cells. In the smaller vessels, such as the periviscerals and intestinals, the wall consists of nothing more than this membrane covered externally by a single layer of flattened, peritoneal cells. Such branches must necessarily be non-contractile. The walls of the main blood-vessels show a rather more complicated structure, but still remain very thin and delicate. The membrane, mentioned above, is supported externally by a few muscle fibres. The circular muscles are very few, and are situated just outside the membrane. The longitudinal fibres are better developed, and lie just underneath the peritoneal epithelium, which forms the outermost covering of the wall.

Owing to the rapid movements of the worm, the dorsal and ventral vessels are constantly thrown into a number of zig-zags or S-shaped portions, which are rendered much more apparent by the addition of chloroform or ether to the water in which the worm is placed, when the movements of the worm become very violent.

The main trunks which have a longitudinal direction are the dorsal, supra-intestinal, and ventral vessels.

1. **The Dorsal Vessel** extends through the entire length of the body from the anal segment to the prostomium (Pl. I, fig. 1, *d.v.*). It lies dorsal to the alimentary canal for the greater part of its length, but in the region of the reproductive organs it changes its position and comes to lie nearer the ventral vessel. Further back it reassumes its original position.

2. **The Supra-intestinal Vessel** is always attached to the dorsal wall of the intestine, and is invested by a layer of chloragogen cells continuous with those which form the outermost layer of the intestinal wall (Pl. III, fig. 9, *si.v.*). It originates in segment 5 as an offshoot of the dorsal vessel which lies above it, and it extends through the body to a short distance behind the segments containing the reproductive organs. Nomura (1913) states that in *Limnodrilus gotoi* this vessel opens into the dorsal vessel again at the posterior part of the body. Although I have carefully examined many serial sections, I have not been able to find this second opening of the supra-intestinal into the dorsal vessel in *Tubifex rivulorum*. The vessel seems rather to diminish gradually in size, and finally to disappear. In those segments of the mature worm which contain the reproductive organs, the supra-intestinal vessel is slightly displaced and lies a little to one side of the mid-dorsal line. This is probably due to the pressure exerted by the reproductive organs on the other organs in the body.



TEXT-FIG. 1. Diagram of segments *V—IX* showing the positions of the principal blood vessels with their connections in these segments. *d.v.* dorsal vessel, *si.v.* supra-intestinal vessel, *in.* intestine, *v.v.* ventral vessel, *pv.v.* perivisceral vessel, *in.v.* intestinal vessel, *ht.* heart.

3. **The Ventral Vessel** extends through the whole length of the body, and lies beneath the alimentary canal between it and the nerve cord (Pl. I, fig. 1, *v.v.*). It

commences in segment 1, where it is paired, its two parts uniting with the two branches into which the dorsal vessel divides, also in segment 1. These two parts of the ventral vessel pass backwards as two converging trunks as far as segment 3, where they unite. It is attached to the intestine throughout the greater part of its length, but occasionally appears to lie freely in the body cavity. It should be noticed that the nephridia of the posterior segments of the body are situated close to the ventral vessel with the walls of the nephridial tubes closely pressed against it. In the last segment of the body the dorsal and ventral vessels unite and are slightly coiled.

These longitudinal trunks are connected with one another by a series of commissural vessels. These are of two kinds: (*a*) intestinal, (*b*) perivisceral or coelomic.

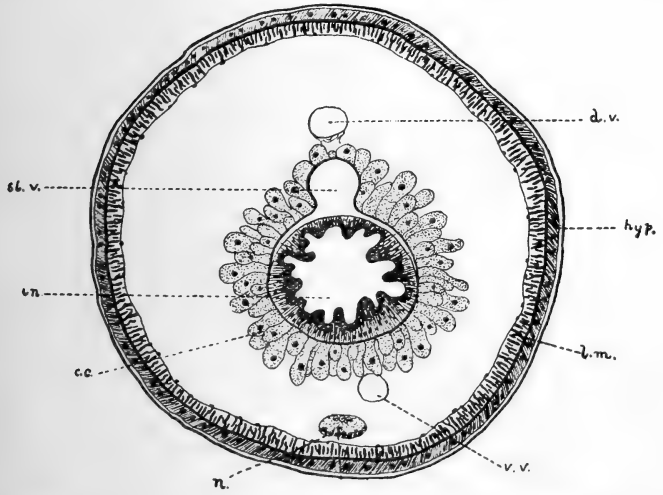
(*a*) *Intestinal Vessels.* The principal intestinal vessels, of which there is a pair to each segment, connect the supra-intestinal with the ventral vessel and are well developed in all segments behind the fifth. They have their origin from the supra-intestinal vessel near the middle of the segment and well in front of the perivisceral trunks (Pl. VI, fig. 42, *in. v.*). These intestinal vessels do not lie freely in the body cavity but pass beneath the layer of chloragogen cells, and thus encircle the intestinal wall. In the posterior segments they are connected with the dorsal instead of the supra-intestinal vessel.

In addition to this principal intestinal trunk, there are accessory vessels, which are, however, much less conspicuous (fig. 42, *in.v.*[1]). These are best seen by treating the living worm with ether on a slide. At first, this has the effect of causing strong contractions of the intestine and blood vessels, which rather hinder than aid the observer. But if the worm is allowed to remain on the slide for

some time these contractions become less violent, as the animal becomes exhausted, until, finally, all movement ceases except the occasional contraction of the intestine. The body of the worm becomes somewhat flattened, and the chloragogen cells, which at first completely hide the underlying tissues, become clearer. It is then possible to investigate the blood vessels which pass to the intestine wall. These branches become filled with blood, and, if the worm is lying on its side, are comparatively easy to see. The number and arrangement of these smaller intestinal vessels vary in different parts of the body. The largest vessel, which has already been described, is constant. The variable ones are the accessory intestinal branches which lie between the intestinal proper and the perivisceral trunk (Pl. VI, fig. 42). In the anterior segments of the intestinal region of the body there may be as many as five pairs of accessory intestinal vessels, all branched and forming a vascular network over the intestinal wall, but they do not appear to have any connection with the ventral vessel. In the posterior segments these vessels gradually decrease in size and number, and become very difficult to follow.

(*b*) *The Perivisceral Vessels* are present in all segments of the body except segment 8. They arise in pairs from the dorsal vessel near the posterior border of each segment, and are connected with the ventral vessel below. These are quite large trunks, passing out almost at right angles to the dorsal vessel, and form in most segments a series of complex coils, often extending from one end of the segment to the other (Pl. VI, fig. 42, *pv. v.*). This coiling of the perivisceral vessels allows of ample freedom of motion for the worm—a very necessary precaution in view of its rapid and sudden movements. They do not branch, and, therefore, there is no integu-

mental network in this worm, such as is described by Beddard as occurring in Ilyodrilus and Bothrioneuron. In the anterior segments of the body the perivisceral trunks lie freely in the body cavity. More posteriorly, particularly in that part of the body which is waving about in the water, these vessels, while not branching, are always pressed closely against the body wall for a good part of their length, and remain in this position



TEXT-FIG. 2. Diagrammatic tranverse section through segment *IX* to show the relative positions of the principal blood vessels. *hyp.* epidermis, *l.m.* longitudinal muscles, *c.c.* chloragogen cells, *in.* intestine, *si.v.* supra-intestinal vessel, *d.v.* dorsal vessel, *v.v.* ventral vessel, *n.* nerve cord.

permanently. This would suggest that aeration of the blood takes place through the body wall, which is very thin near the posterior end of the body. At its anterior end the dorsal vessel divides into two branches, which become slightly coiled and then dip down ventrally to become continuous with the anterior ends of the ventral vessels. In segment 2 the perivisceral vessels are given off from the dorsal vessel near the posterior border of the segment.

Instead of passing out at right angles to the dorsal vessel, as is the rule, they have a somewhat more forward direction and may extend into the segment in front, the septum between segments 1 and 2 being absent (Pl. VI, fig. 41, *pv. v.* 2). They become slightly coiled, and then dip down to open into the ventral vessel a short distance behind their origin from the dorsal vessel.

The perivisceral vessels of segments 3-7 have a similar distribution to that described above for segment 2, but in segments 4-7 they lie entirely in the segment to which they properly belong, owing to the presence of septa between adjacent segments, thus preventing the passage of the vessels from one segment to the other. It should be noted also that the right and left perivisceral of segments 2 and 3 open into the right and left branches of the ventral vessel respectively.

In segment 8 the perivisceral vessels are absent. In segments 9 and 10 they become coiled around the anterior and posterior sperm sacs respectively. In segments 11-17 of a mature worm which contain the ovisac the perivisceral are always coiled around this organ. In all the segments behind those containing the reproductive organs these vessels pass simply from the dorsal to the ventral vessel.

The "hearts" are situated in segment 8. They originate from the supra-intestinal vessel above, in the posterior part of the segment, and open into the ventral vessel below (Pl. I, fig. 2, *ht.*). They are much enlarged, especially near their origin from the supra-intestinal trunk, and are contractile.

The blood is a red, non-corpusculated fluid, the haemoglobin being dissolved in the blood plasma.

THE NERVOUS SYSTEM.

The nervous system of *Tubifex rivulorum* is formed upon the same plan as that of all Oligochaeta. It consists of cerebral ganglia united to a ventral chain of ganglia or nerve cord by peripharyngeal connectives. As in most other Oligochaeta the whole nervous system lies completely in the body cavity. It is possible to examine the arrangement of the ganglionated chain and peripheral nerves in the living worm, especially after the addition of ether to the water in which the worm is lying. The ether increases the transparency of the body wall so that the internal organs can be seen more easily, and in some cases the alimentary canal becomes so contorted that the ventral nerve cord is left quite freely exposed for a considerable distance. This is a very useful method of investigation, as it enables one to compare the form and size of the ganglia, and the proportion of connective to ganglion in the segment in different parts of the body, and these proportions vary a good deal. It is, however, of very little use for a detailed examination of the brain. In the living worm it is usually very difficult to define the outlines of the brain, even after the addition of ether. It has happened, however, very occasionally, that I have been able to see the brain quite clearly by this method. The difficulty which is usually experienced in examining the brain in the living condition is due partly to the greater transparency of the brain compared with that of the body wall, and partly to the contractions of the pharynx over which the brain is situated. The great drawback to the use of ether is that the blood vessels in the anterior region of the body become somewhat distended and quite full of blood. This, of course, only serves to obscure the brain more.

Even though one may be fortunate enough occasionally to get a good idea of the shape of the brain, it is better to complete the investigation by means of serial sections, which are, of course, necessary for a proper examination of histological details, both in the brain and ventral nerve cord. Many attempts were made to make use of *intra-vitam* methods of staining, as it was hoped that the nervous system would be rendered more distinct when the tissues were stained in the living condition. These attempts, however, were not successful, as the stain (methylene blue) penetrated the body wall very slowly and incompletely, and did not reach the nervous system at all.

The Cerebral Ganglia, as in most aquatic Oligochaeta, lie far forwards, just behind the prostomium. Their posterior border lies at the level of the boundary between the buccal cavity and pharynx. They are dorsal in position, and form a comparatively large brain, the structure of which is complicated by the presence of several lobes. The brain is concave behind and in front, and consists of a solid mass of nerve fibres and nerve cells. Anteriorly it is produced into three horns or lobes, two lateral and one median: the former may be described as the antero-lateral lobes, and the latter as the anterior median lobe (Pl. I, fig. 1, *an.l.*; *m.l.*). From these, nerves arise which will be considered later. The anterior median lobe is characteristic of the family Tubificidæ, but is not always present as a simple lobe. In a few more highly specialised forms, *e.g.*, Bothrioneuron, it consists of a median nerve communicating with a small ganglion placed a little way in front of the brain (Beddard).

The antero-lateral lobes described above are not figured by Vejdovsky (1884) in his drawing of the brain

of *Tubifex rivulorum*, and he definitely states that the brain in this region is devoid of processes or lobes. He figures, however, postero-lateral lobes from which well-developed muscles pass to the body wall, and these lobes can be clearly seen (Pl. I, fig. 1, *ps.l.*), but I have never been able to find the muscles. Beddard described, in his definition of this species, a median, smaller, posterior lobe corresponding to the anterior one; but this I have never been able to see. It is particularly easy to see the outline of the brain just at this point, as the dorsal vessel runs under it here, and the red colour of the blood in this vessel causes the brain to show sharply outlined against it. In addition to the lobes already mentioned, there are yet two more to be noted, and these are very important, as it is from them that the peripharyngeal connectives arise. They are situated between the antero-lateral lobes and the postero-lateral lobes, and are slightly more ventral in position than either of these. They pass obliquely outwards, downwards and backwards, and finally bend sharply inwards, terminating in the nervous bands known as peripharyngeal connectives which finally unite, in the median line ventrally, with the first ganglion of the nerve cord known as the sub-pharyngeal ganglion (Pl. II, fig. 8 *sp.g.*).

Cerebral Nerves. I have been able to recognise three pairs of these, two pairs arising from the brain proper and one pair from the peripharyngeal connectives. Of these three pairs of cerebral nerves, one pair arises from the median anterior lobe of the cerebral ganglia. They really originate as one nerve forming a continuation of the median lobe, but this condition only obtains for a short distance, as the single nerve soon divides into two branches which diverge from one another. Both branches, however, pass forwards towards the tip of

the prostomium, with which they ultimately become connected. Each nerve branches several times, and gives rise to a large number of extremely fine fibres, which terminate in the sensory cells with which the prostomium is amply provided. It is the presence of these sensory cells which renders the prostomium such an important tactile organ.

The second pair of nerves arising from the cerebral ganglia form continuations of the antero-lateral lobes of the brain, which have already been described. They are short, and pass directly to the lateral walls of the prostomium and first segment. Here they branch, forming a number of fine fibres which spread themselves over and into the body wall of this region. The third pair arise as branches of the peripharyngeal commissure, and pass to the body wall of the first segment, where they also branch (Pl. II, figs. 7 and 8).

The Ventral Nerve Cord originates at that point where the two connectives arising from the cerebral ganglia unite below the gut, having first encircled it. The nerve cord is a ganglionated chain extending from the second segment to the last segment of the body, and it lies freely through all its length in the body cavity. It is surrounded by a connective tissue sheath, from which, at intervals, branches arise and pass to the body wall. By this means the nerve cord is kept in position in the body cavity. It is necessary to exercise care in identifying nerves, for these branches of the connective tissue sheath closely resemble the nerves in appearance, though not, of course, in structure.

The cord is also enclosed in a muscular sheath, which, however, does not completely encircle it, but is confined principally to the dorsal surface. The sheath is very delicate, and is composed of a few muscle fibres

placed longitudinally. There is a single ganglion situated near the posterior border of each segment, and these are connected into a continuous chain by a series of connectives which unite adjacent ganglia. In the anterior segments the ganglion and connective are about equal in length, the connective, if anything, being a little longer. More posteriorly, however, where the segments become shorter, the proportions of these two parts of the nerve cord also change. The connectives become shorter and shorter, but the ganglia change very little in size, so that, finally, the connectives are extremely short, and the ganglion occupies almost the whole length of the segment. But throughout the length of the nerve cord the two parts are sharply marked off from one another. This is very clearly seen if one examines the living worm, but when the nerve cord is seen in section the difference between the two is even more apparent. The connective is of about the same diameter throughout its length, but the diameter of the ganglion varies in different parts. In most cases it can clearly be divided into three lobes placed end to end and separated from one another by constrictions. Of these lobes, the first is the largest, while the other two are of nearly the same size, the posterior one being very slightly smaller.

The ventral nerve cord gives off branches or peripheral nerves in each segment. There has been a considerable difference of opinion amongst earlier authorities as to the number of these branches in each segment. Vejdovsky (1884) figures no fewer than five pairs; D'Udekem (1855) states that there are three pairs, but Nasse (1882) was only able to find two. After careful examination of the living worm and sections, I have come to the conclusion that there are three pairs of these lateral nerves in each segment. Their places of

origin from the nerve cord are approximately the same in all the segments. There is always one pair arising from the connective and two pairs from the ganglion. The first pair comes off from the connective just behind the septum, while of the two pairs originating from the ganglion one springs from the most anterior of the three lobes, just at that point where the connective merges into the ganglion. The third pair is to be found more posteriorly, and usually originates at or near the constriction which separates the first and second lobes of the ganglion.

These three pairs of nerves all have a similar distribution. They are true lateral nerves, and do not, as in some forms, *e.g.*, most of the Lumbriculidae, arise from the mid-ventral line as a single nerve, and after entering the body wall divide into two branches. They arise, on the contrary, from the lateral part of the cord, but slightly nearer the ventral than the dorsal surface. They pass out at right angles to it, and extend for some distance in the body cavity before plunging into the body wall.

Histology of the Nervous System.

In order to get a clear idea of the histological details of the structure of the brain and nerve cord, it is necessary to examine transverse and longitudinal sections of these organs. If the sections are appropriately preserved and stained, the various elements become fairly well differentiated. The two outermost coverings of the nerve cord have already been mentioned, but in sections it can be seen that both the connective tissue sheath and the muscular layer are extremely delicate. The most conspicuous part of the former are the nuclei of the connective tissue cells, and, indeed, in places it is very difficult to identify any other structural details. The

nuclei are oval in shape, and the cells to which they belong are considerably flattened. The muscular layer has already been sufficiently described.

The brain is composed of both nerve cells and nerve fibres, the cells being disposed dorsally and laterally, while the fibres occupy the ventral and central parts of the brain. The nerve cells, in section, appear as almost rounded bodies with clearly marked rounded nuclei. The nerve fibres are embedded in a transparent matrix, and when cut transversely give the brain, in the region in which they are, a finely granular appearance.

The nerve cord, as already mentioned, can be divided into connectives and ganglia, the ganglia being represented on the surface of the cord as swellings between the connectives. In sections it can be seen that the difference in size of the cord in different parts is due to the presence or absence of nerve cells. The connectives are formed only of nerve fibres, whereas the ganglia possess in addition a large number of nerve cells, their lateral and ventral position being different from that which they occupy in the brain (Pl. VII, fig. 46, *n.c.; f.*). The mass of nerve fibres, particularly in the ganglia, is divided up into different regions by the interposition between the nerve fibres of a delicate fibrous layer, which appears to be a continuation of the connective tissue sheath which completely surrounds the cord (fig. 46).

As in most Oligochaeta, giant fibres are to be seen in the nerve cord throughout its length. These occupy the same position in the cord as do those of Lumbricus, that is, they lie close to its dorsal surface. The number of fibres present, however, varies in different parts of the body. Anteriorly, just behind the brain and for several segments, the cord contains only one giant fibre, and this lies in the mid-dorsal line. More posteriorly there are

three giant fibres, the middle one being the largest. They are still situated dorsally, and lie side by side.

The structure and function, and the history of our knowledge, of the giant fibres in certain Polychaet worms, especially *Halla parthenopeia,* has been dealt with in great detail by Ashworth (1908), and it therefore seems unnecessary to refer to the subject in detail here. It may, however, be advisable to include a brief account of the structure of a giant fibre as described by Ashworth, as follows:—

Each giant fibre consists of a sheath composed of interlacing glia fibres of different diameters and embedded in a finely granular protoplasm. The centre of each fibre is occupied by a bundle of fibrillae, also embedded in a matrix known as the interfibrillar substance. The number of these fibrillae in a fibre differs in different Annelids. The space which is always present between the bundle of fibrillae and the sheath of the fibre is filled with a semi-fluid perifibrillar substance, which is colourless, hyaline and contains very fine granules.

In my preparations of the nerve cord of *Tubifex rivulorum* only the sheath of the giant fibres has been clearly marked, the fibrillae and the perifibrillar substance not staining at all well, thus giving the fibres the appearance of empty tubes. This is due to the extreme difficulty which is always experienced in attempting to differentiate clearly the fibrillae of the giant fibres. All ordinary methods of preserving and staining the material, with which I was familiar, were unsuccessful, and as Ashworth's paper did not come into my hands until this work was ready for publication, I have not yet had an opportunity of testing whether his methods of preserving and staining *Halla parthenopeia* are equally successful in the case of *Tubifex rivulorum.*

THE EXCRETORY ORGANS.

The excretory organs or nephridia consist of a system of paired tubes, which are present in most segments of the body. They are absent from a few of the anterior segments, but as in most aquatic Oligochaeta they begin well before the genital segments, from which, however, they are absent. I have been able to trace these nephridia as far forward as segment 7, but not in front of that. Behind the genital segments they are to be found through all the remaining segments of the body.

The nephridia are coiled tubes occupying two segments, and, typically, two only, but in a few cases I have seen the coils of a nephridium of one segment lying in the segment behind, but this must be considered as an abnormal condition. Each nephridium is provided with an internal and an external aperture, the former opening into the body cavity, the latter to the exterior. The nephridia lie nearer the ventral surface of the body than the dorsal, and are situated one on either side of the ventral vessel and very close to it. In the posterior segments of the body certain of the coils of the tube appear to be very intimately connected with this vessel, so that even though the movements of the worm may be extremely violent, these tubes do not become displaced.

Nasse (1882), in his work on the family Tubificidae, described the presence of branches of the ventral vessel which arise near the nephridia and pass into close connection with these organs. That is, he claims that there are special blood vessels conveying blood from the ventral vessel to the nephridia, in which organs, presumably, the blood is purified. Nasse does not explain how the blood is returned to the main circulation, but he suggests, at any rate, an arrangement of the vessels and

nephridia which is closely allied to that found in higher Oligochaeta and typically in Lumbricus. Vejdovsky (1884) contradicts this, and definitely states that there is no connection between the ventral vessel and the nephridia. I am disposed to agree with Vejdovsky that there are no special branches passing from the ventral vessel to the nephridia, but at the same time we ought not to lose sight of the fact that, as has already been stated, the nephridia are always in close connection with the ventral vessel. This suggests the possibility that the excretory products which collect in the blood during its passage round the body are passed out from it, while in the ventral vessel, to the nephridia, the walls of which are very thin and would form no barrier to the diffusion through them of these waste products.

Since the nephridium occupies two segments, it follows that the tube must pierce the intervening septum. We may, therefore, divide the whole nephridium into pre-septal and post-septal portions. The pre-septal part consists of the nephrostome, or funnel, and a very short, uncoiled portion of the nephridial tube. By far the greater part of the nephridium, therefore, lies behind the septum, and consists of a much coiled tube which can be divided into certain regions according to the structure of its walls. This tube terminates in the external aperture or nephridiopore, which is situated at the apex of an enlarged vesicle forming the terminal portion of the nephridial tube. It is possible by careful teasing out of part of the intestinal region of the living worm to separate portions, at any rate, of the nephridium. It is extremely difficult, however, to obtain a good view of the nephrostome and nephridiopore by this means. In some cases the nephridium can be examined *in situ* if the body wall be sufficiently transparent, and in this way the cilia can

often be seen in motion. If the tube be removed from the body, the cilia very quickly cease to move, and it is impossible then to decide whether any particular portion of the tube is ciliated or not (Pl. VI, fig. 43).

The nephrostome is small and very simple in structure. Its diameter in the widest place is only slightly greater than that of the nephridial tube. Its lumen is a little larger than that of the tube, and its walls are somewhat thicker, making it funnel-shaped. It is composed of a very few cells, the nuclei of which are large, round and nucleolated (Pl. VII, fig. 47). The inner borders of the cells, those abutting on the lumen, are ciliated, the cilia being long and pointing chiefly in one direction, namely, from the free end of the funnel down the tube. Some of the cilia, however, fringe the free edge of the funnel, and these are particularly long and very active. By their rapid movements they create a current in the direction of the funnel, into which the excretory products are drawn. The pre-septal part of the tube is extremely short and uncoiled, and its cavity is directly continuous with that of the funnel. Its walls are thin, and the nuclei of the cells are flattened in a direction parallel to that of the tube. The tube is ciliated in this part.

The post-septal portion of the nephridium can be sub-divided into three regions:—(1) A delicate, much-coiled tube with extremely thin walls. (2) This passes into a slightly thicker walled tube of a yellowish colour, and decked with specially modified peritoneal cells. (3) This again passes into a somewhat thinner walled tube, which is covered with specialised peritoneal cells for part of its length, and which finally opens into a small vesicle communicating with the exterior at the nephridiopore (Pl. VI, fig. 43, *t.*).

(1) The first portion of the tube has thin, transparent walls, and is profusely coiled, the coils being quite irregular in their arrangement and forming no definite loops such as are so characteristic of the nephridia of Lumbricus and other types. Its cavity is intracellular and ciliated throughout. It is invested by an extremely delicate peritoneal covering, the cells of which are much flattened and possess oval nuclei.

The first portion of the tube is separated from the second by a curious structure, which, as far as I can tell, has never been described by any other observer as occurring in Tubifex. It has been figured by Eisen (1885), however, in the nephridia of *Spirosperma ferox*. He calls it an ampulla, and this name will be used for the corresponding structure as found in Tubifex. Eisen describes the ampulla as an enlargement of the tube of the nephridium, but he does not attempt to give any account of its structure. He does not even venture to express an opinion as to whether it is a permanent or only a temporary structure. Beddard (1895) in his monograph refers to the ampulla in his account of the characters of the family Tubificidae, as occurring in Limnodrilus, *Spirosperma ferox* and many earthworms. He gives, however, no account of its structure.

In Tubifex the ampulla is undoubtedly a constant organ, for I have never failed to find it in any of the specimens that I have examined, whether in sections or by teasing out the nephridium from the living worm. Further, it is present in the nephridium of all segments from the anterior to the posterior end of the body. The ampulla always occurs between the first portion of the nephridial tube and the second, which is characterised by walls of a yellowish colour. The ampulla is usually pear-shaped, although in a few cases it is somewhat more

circular in outline. The first part of the tube opens directly into it at its broader end, while the second portion of the tube leaves it at the opposite or pointed end (Pl. VII, fig. 48). The structure of the ampulla is always the same. It has the form of a swollen bladder bounded by a delicate wall composed of a single layer of cells with prominent oval nuclei. The junction of the first part of the tube with the bladder is marked by a circlet of specially large rounded cells, with prominent nuclei, which by their arrangement form a sort of collar round the tube. In the cavity of each ampulla there is, as a rule, a brownish, granular mass of irregular shape lying somewhat nearer to its broader end (Pl. VII, fig. 48, *gr. m.*). In many cases this mass appears to have no connection with the wall of the bladder, but in others I have been able to see exceedingly fine processes extending from it to the wall. In an ampulla removed from the living worm this mass appears to be solid, but in section it is seen to be hollow, its cavity being continuous with that of the first part of the nephridial tube. It is very difficult to decide whether this central canal of the granular mass ends blindly, or whether it communicates with the cavity of the ampulla. I am inclined to think that the latter is the case. There is no suggestion of branching of this canal, nor the formation of fine nephridial tubules within the mass itself. In fact, the latter seems to be composed of a large number of inert granules which are affected by neither killing reagents, preserving fluids nor stains. As already mentioned, the nephridial tube is ciliated throughout, but just at the point where it enters the ampulla it is provided internally with cilia which are particularly long and which project into the ampulla, or more accurately into the canal of the granular mass

(Pl. VII, fig. 48, *ci.*[1]). If one liberates an ampulla with a portion of the tube from the living worm, these cilia exhibit lively movements even for some time after those in the nephridial tubes have become motionless. The wall of the ampulla is not ciliated.

(2) The second portion of the nephridial tube opens out from the pointed end of the ampulla, and directly after its origin it is bent back sharply so that it runs for a short distance parallel to the ampulla and the first part of the tube. This part of the tube is characterised by the fact that its walls are somewhat thicker than those of the first part, that they are yellowish in colour, and the cells of which they are formed are very granular. The tube is not much coiled, but is bent on itself two or three times to form well-marked loops which lie approximately parallel to each other and appear to be bound together in a common investing membrane. For the first part of its length this tube is provided with a layer of peritoneum, composed of flattened cells similar to those investing the first part of the tube. These gradually give place to specially modified cells known as vesicular peritoneal cells (Pl. VI, fig. 44, *v. pt.*). These are large, rounded, bladder-like cells with very thin walls. These cells are particularly well seen in the fresh material, but are liable to undergo considerable shrinkage during the processes of killing and fixing. When examined in the living condition, these cells may be clear and devoid of any special solid contents, or they may be filled with minute brownish granules which exhibit active molecular movements even after the cell has been dislodged from the wall of the nephridium.

(3) The third part of the tube is ciliated, is of somewhat wider calibre than the first part, and has thin walls. The proximal portion is covered with vesicular peritoneal

cells similar to those described above, but distally the peritoneum is again composed of flattened cells. The distal end of this tube expands rather suddenly to form a small, contractile vesicle, which opens to the exterior at the nephridiopore. The tube is ciliated to its distal end, and a tuft of specially long cilia projects into the vesicle. The latter is somewhat pear-shaped when fully expanded, narrowing considerably as it approaches the nephridiopore, which is situated on the ventral surface of the body, a short distance in front of the ventral setae on either side.

REPRODUCTIVE ORGANS.

Tubifex rivulorum is hermaphrodite, the ova and spermatozoa maturing at the same time. The worms exhibit fully-developed reproductive organs during the autumn and early winter, that is, from October to December. The first cocoons are laid about the beginning of November, and can be found in large numbers in the mud for the next two months, after which time their formation ceases. The mature worms show well marked differences in external appearance from those in which the reproductive organs are not well developed, especially in the anterior segments. The sexual organs occupy segments 9-16 or 17 in quite mature worms, and these segments are considerably swollen and of a dull whitish colour, due mainly to the large size of the sperm sac and to the large ova which are present. During May, June and July it is comparatively common to find worms which present somewhat this appearance, and it would be easy to make an error and suppose that such worms were mature if they were not more minutely examined. Further investigation shows that this whiteness and

opacity are due to the presence of parasites belonging to the species *Urospora saenuridis*, which will be described later.

The reproductive system is a complex affair, as is usually the case in hermaphrodite forms, and it will be well, first of all, to enumerate the various organs which assist in the process of reproduction and then enter into their structure and arrangement in detail afterwards. First, then, there are the *ovaries* (Pl. I, fig. 2, *ov.*) and *testes* in which the ova and spermatozoa respectively undergo the earlier stages of their development. They are not, however, permitted to come to maturity in these organs, but are transferred quite soon to special sacs known as the *egg sacs* (Pl. I, fig. 3, *ovs.*) and *sperm sacs* (Pl. I, fig. 2, *sp. s.*), which, at first, occupy only one segment each, but in the mature worm may extend through several segments. When the ova and spermatozoa are fully developed, they are transferred to the cocoon by means of special ducts, which are provided with external apertures perforating the body wall. These ducts are known as the *oviducts* (female) and the *sperm ducts* or *vasa deferentia* (male) (Pl. I, fig. 2, *v. d.*). The spermatozoa do not reach the cocoon directly, by means of the vasa deferentia, but are transferred, during copulation, to special organs set apart for their reception. These organs are known as the *spermathecae* (Pl. I, fig. 2, *sp.*). The terminal portion of each vas deferens is dilated to form an elongated chamber known as the *spermiducal gland* (Pl. I, fig. 2, *at.*), which opens to the exterior by the *penis*, a chitinous organ, capable of protrusion (Pl. I, fig. 2, *pe.*). The penis aids in the transference of the sperm from the sperm sac of one worm to the spermathecae of the other during copulation. In connection with the spermiducal gland, and formed as a

proliferation of some of its cells, is an irregularly-shaped mass known as the *prostate gland*. The cells of this gland secrete a cementing substance which is passed into the spermatheca with the sperm, and is used for moulding the spermatozoa into a solid mass, of characteristic shape, known as a *spermatophore*. The position which these organs occupy in the body is constant, and will be noticed in the description of the various organs.

I. The Gonads.

Both ovaries and testes are present in the same worm, there being only a single pair of each. As is usual in the Oligochaeta, the testes lie in front of the ovaries, and in Tubifex they are situated in adjacent segments, the testes in segment 10 and the ovaries in segment 11. In their relative segments they occupy exactly the same position in relation to the other organs present in that segment; that is, they lie one on either side of the intestine and slightly nearer the ventral than the dorsal surface of the body.

Not only do they correspond in their position in the segment, but, in the young condition at any rate, they are exactly similar in appearance and structure. Both ovaries and testes are derived from peritoneal epithelium, and appear in the earliest stages of their development as small masses of a few undifferentiated cells forming the germinal epithelium. In such a condition as this it is only by noticing the segment which they occupy that one can distinguish between the gonads. They are attached to the septum forming the anterior boundary of the segment which they occupy and hang freely into the body cavity. At a little later stage, but when the cells of the germinal epithelium are still undifferentiated, the

shape of the mature gonad is indicated. They are both somewhat pear-shaped, the broader end being attached to the septum (Pl. I, fig. 2, *ov.*). In a mature worm, however, the ovaries often attain so great a size that they are somewhat bent round in the segment and the original shape is lost: this is also partly due to the presence of the oldest ova at one side of the ovary. The ovaries persist throughout the reproductive season, and only attain their full size when the rest of the reproductive organs are developed.

If, however, in a mature worm one seeks for the testes, one will not be able to find them. This is due to the fact that they have been completely enclosed in the sperm sac, which is of large size and in which the spermatozoa complete their development. It is necessary, therefore, to examine much younger worms in order to find the testes. In fact, they are quite well developed in those individuals in which there is no trace of any other part of the reproductive system except the ovaries. The testes develop somewhat earlier than the ovaries, and, therefore, in a young worm it will be easy to identify them without verifying their position, as they will be larger than the ovaries. When the testis has attained its full size it consists of a mass of rounded cells with clearly marked nuclei, but without any specially characteristic features. These may be called the spermatogonia, or sperm mother cells, and here the development of the spermatozoa in the testis ceases. The spermatogonia must be transferred to the sperm sac before further development can take place.

Many of the earlier writers confused the testis with the sperm sac or with part of it. D'Udekem (1855), for example, speaks of the testis as occupying segment 8 where it appears as an unpaired organ below the intestine

and having the form of a voluminous gland, greyish in colour. What he has described as the testis is really a portion of the sperm sac, for the latter in a mature worm not only extends backwards through several segments, but may pass forwards in front of that segment which originally contained the testis. McIntosh (1871), too, has confused the sperm sac with the testis in stating that the testis of one side remains in segment 9, while its fellow extends back as far as segment 16.

The ovaries are still small when the testes have attained their full size, but while the latter soon disappear the ovaries gradually increase in size until they occupy a large proportion of segment 11. In a fully developed ovary it is easy to recognise ova in several stages of development.

II. Development and Structure of the Spermatozoa.

The germinal epithelium in the testis gives rise by ordinary cell division to a number of spermatogonia, which, at an early stage in their development, are separated from the testis and pass into the sperm sac, which, at first, is a simple, undivided sac. The spermatogonia, when they leave the testis, are uninucleate, but very soon the nucleus divides several times, and, as its division is not at once accompanied by division of the cytoplasm, each spermatogonium or sperm morula, as it is called, becomes multinucleate (Pl. V, figs. 28 and 29). Calkins (1895a), who has described in detail the spermatogenesis of Lumbricus, states that the spermatogonia, while still in the testis, are multinucleate. Careful examination of many preparations of the spermatogonia of Tubifex has led me to decide that in this form, at any rate, the spermatogonia only become multinucleate on leaving the testis.

The nuclei of the sperm morula, which, when first formed, are scattered irregularly through the cell, gradually become arranged more regularly around the periphery of the cell, the central portion of which has no nucleus and is, therefore, entirely cytoplasmic in nature. The cytoplasm surrounding the nuclei at the periphery of the sperm morula exhibits slight cleavage marks around each nucleus, and these cleavages deepen until the nuclei are completely constricted off from one another, while still remaining in connection with the cytoplasmic mass occupying the centre of the morula, which remains undivided throughout and is known as the sperm blastophore (Pl. V, figs. 30 and 31). During this protoplasmic cleavage the sperm morula or sperm polyplast increases considerably in size, and the nuclei are large, rounded, and possess nucleoli. Each of the elements making up the fully formed sperm morula is called a spermatocyte, and these are at first large and comparatively few in number (Pl. V, fig. 31). The spermatocytes are arranged extremely regularly around the sperm blastophore, but this arrangement can only be fully appreciated when the sperm morula is viewed in section (Pl. V, fig. 32).

The nucleus of each of the spermatocytes now undergoes karyokinetic division, probably twice, and the cells then divide so that the number of elements comprising a sperm morula in its later stages of development is very much increased. The cells thus formed are much smaller than the spermatocytes, as there is no appreciable increase in the size of the whole morula at this stage (Pl. V, fig. 33). The cells resulting from the division of the spermatocytes are known as spermatids, and from these the mature spermatozoa are derived by a simple metamorphosis. The spermatids are at first rounded in shape, and the nucleus forms the greater part of the cell, the

cytoplasm being much reduced in quantity. Gradually, however, the spermatids become oval in shape. The nucleus of the spermatid then elongates considerably to form the filiform head of the spermatozoon so characteristic of the group (Pl. V, fig. 34). The middle piece of the spermatozoon is not very conspicuous, but appears as a direct continuation of the head. The distal extremity of the spermatid now becomes much drawn out, forming a long tail which is, in the normal spermatozoon, considerably longer and thinner than the head. It stains but faintly with nuclear stains, as it is composed of cytoplasm only, derived in all probability from the delicate layer of protoplasm surrounding the nucleus of the spermatid. At first, the fully formed spermatozoa are arranged very regularly around the blastophore, as regularly, in fact, as were the spermatids from which they were derived, each one with the anterior end of the head buried in the substance of the blastophore. The tails of the spermatozoa are at first quite straight, and while still in this condition become motile (Pl. V, fig. 35). When the spermatozoon is quite matured and ready to leave the blastophore, the tail usually exhibits a simple coil at the free end. The spermatozoa now become much less regularly arranged on the blastophore and by degrees they separate completely from it. When they are set free they remain for a time in the cavities of the sperm sac, but finally they find their way to the ciliated funnel of the vas deferens, around which, in the mature worm, they congregate in great numbers.

In most of the individuals I have examined, I have found developing in the sperm sac, side by side with the normal spermatozoa, other structures which resemble them to a certain extent, but which can easily be distinguished from them. At first I was inclined to look

upon these structures as parasites, but a closer examination of their development has led me to suggest that two kinds of spermatozoa are present. On this understanding the same terminology will be adopted in the following description as was used above in describing the development of the normal spermatozoa.

The central blastophore from which the spermatocytes are constricted off is larger, and more irregular in outline, than that on which the normal spermatozoa are formed. The spermatocytes themselves, too, are less regularly arranged, often tending to be formed in groups of four or five together (Pl. V, fig. 36). The nuclei of the spermatocytes stain very deeply. The changes taking place during metamorphosis can be easily followed. The spermatocytes, which are at first rounded, become oval in shape, the outer free end being considerably more pointed than the opposite end, which is embedded in the blastophore (Pl. V, fig. 37). The spermatids formed by division of the spermatocytes are much larger and less numerous than those developed from a normal sperm polyplast, and their arrangement on the blastophore is as irregular as that of the spermatocytes from which they are derived (Pl. VI, fig. 38). In the earlier stage of metamorphosis there is no semblance of a true tail, but when the head has lost its oval form and becomes much elongated, the tail is gradually differentiated. At first it is quite short, but it increases in length as the head of the spermatozoon becomes fully developed, and in the final stages the tail is as long as, or a little longer than, the head (Pl. VI, fig. 39). The mature spermatozoa are quite irregularly arranged, but they tend to lie together on the blastophore in bundles of five or six, just as the spermatocytes have been described as doing, and from it they finally separate. In some cases I have been able to

find in the same sperm smear most of the stages in the development of these spermatozoa, but in other cases they seem to occur much more rarely.

At first I tried to incorporate the stages of development described above in my description of the development of the normal spermatozoa, but when I was able to get such a complete series of each I gave up the attempt. I now suggest that this worm possesses dimorphic spermatozoa, a condition which has been described as occurring in Paludina amongst the Mollusca and in certain Amphibia, Birds and Mammals. In Paludina the two kinds of spermatozoa are different in shape, the normal one being divisible into a spirally coiled head and an extremely long tail. The larger kind is vermiform, and bears a tuft of cilia at one end. In Birds and Amphibia the two kinds of spermatozoa are of the same form, but differ in size, the smaller one being functional.

In *Tubifex rivulorum* both kinds of spermatozoa are elongated structures, and each is divisible into head, middle piece, and tail, but the proportions of these parts to each other in the two forms are rather different (Pl. VI, fig. 40). The head of the normal spermatozoon is small, forming about one-sixth of the total length (Pl. VI, fig. 40A); it is oval in shape with a sharply pointed anterior extremity, and measures about 4μ in length. It is composed almost entirely of nuclear substance, there being such a small quantity of cytoplasm present as to be almost negligible. The middle piece, which is shorter than the head, lies behind that structure, and gives attachment to the tail. It is composed of cytoplasm and stains only slightly with nuclear stains, such as haematoxylin. The tail is very long and delicate, and is also composed of cytoplasm. The other spermatozoa are much larger, and the head forms about half the whole

structure. The head is elongated, but less sharply pointed at the anterior end than is that of the normal spermatozoon (Pl. VI, fig. 40B). The middle piece is less distinctly marked off from the head than in the first case, and the tail is straight throughout.

III. The Sperm-sac.

The sperm-sac is developed, as in all Oligochaeta, for the reception of the spermatogonia, which are early removed from the testis and transferred to this organ. In it the mature spermatozoa are formed.

In a worm in which the reproductive organs as a whole are immature, the sperm-sac can be seen in its simplest condition. It is an unpaired structure, and arises as a pouch-like outgrowth of the septum between segments 10 and 11. This outgrowth is directed backwards and projects into segment 11. The cavity thus formed is at first quite simple, and is bounded by a layer of flattened peritoneal epithelium which is derived from the tissue of the septum. As the number of spermatozoa increases, the sperm-sac also enlarges, and it soon becomes too big for the segment in which it developed, and projects still more posteriorly into segment 12. In a mature worm the number of spermatozoa is very large, and these are present in the sperm-sac in all stages of development. The sperm-sac increases tremendously in size, extending further and further back until it may occupy as many as seven or eight segments (Pl. I, figs. 2 and 3, *sp. s.*). It is the presence of this large sperm-sac which gives the mature worm a swollen and opaque appearance in the region of the reproductive segments. It is very common also to find that the sperm-sac has encroached upon the segments in front of that occupied by the testis, and

segment 9 at least, and occasionally segment 8, also enclose a portion of the sac. The development of this sac both anteriorly and posteriorly to the segment in which it first appears may be due to simple pressure exerted by the developing spermatozoa.

It has already been said that the sac arises as a simple outgrowth of the septum between segments 10 and 11. As it increases in size, however, its structure becomes more complicated. The cavity becomes divided up into a series of much smaller spaces by the growth inwards of its walls. These small spaces remain in communication with one another, and are filled with spermatozoa in all stages of development. It is difficult to believe that the tremendous number of sperm morulae present in the sac have all been derived from the small and inconspicuous testes. Some writers have suggested that the peritoneal epithelium lining these coelomic spaces of the sperm-sac is capable of forming germinal tissue. If all the spermatozoa were derived from the spermatogonia of the testes which are transferred, as they are formed, to the sperm-sac, one would expect to find the youngest sperm morulae nearest the segment in which the testes lie. But this is not the case, for it is common to find spermatogonia and fully developed spermatozoa lying side by side in any part of the sperm-sac, and there is no suggestion of regularity in their arrangement. This irregular arrangement is what one would expect if the epithelium of any part of the sperm-sac were capable of forming germinal tissue.

Throughout the life of the worm the ciliated funnel continues to hang freely into the cavity of segment 10, and never becomes enclosed in the sperm-sac as is the case in Lumbricus. It does not seem quite clear, perhaps, at first sight, how the mature spermatozoa from the sperm-sac reach the funnel. If, however, we bear in mind the

development of the sperm-sac, there is little difficulty in understanding this. The sperm-sac extends forwards into segment 9, and backwards as far as segment 16, in both cases ending blindly. There remains, however, an opening from the sperm-sac into segment 10, from which it was derived as simple, pouch-like outgrowths of the anterior and posterior septa, respectively. Although the structure of the sperm-sac becomes much more complicated as the worm matures, the sac retains its connection with segment 10, and so the spermatozoa can pass into the coelom, and thence to the surface of the expanded funnel of the vas deferens which lies in the same segment.

IV. The Sperm Ducts or Vasa Deferentia.

The spermatozoa when mature are conveyed to the exterior by special ducts known as the sperm ducts or vasa deferentia. As is always the case in the Oligochaeta, the number of these ducts corresponds to the number of the testes, therefore in *Tubifex rivulorum* there is a single pair. Each duct consists of a much coiled tube with its origin in segment 10—the segment which contains the testes; and its external aperture in segment 11. The whole duct, however, does not lie in these two segments—its length is so great and it exhibits such complex coiling that it extends posteriorly through several segments, sometimes reaching as far back as segment 15.

For the purpose of description we can divide the sperm duct into the following regions:—(1) The ciliated funnel (Pl. I, fig. 2, *ci. f.*), (2) the coiled tube (Pl. I, fig. 2, *v.d.* 1, *v.d.* 2), (3) the spermiducal gland with the prostate (Pl. I, fig. 2, *at.*), and (4) the penis (Pl. I, fig. 2, *pe.*).

These can all be studied in detail by means of transverse and longitudinal sections, but it is interesting and instructive to liberate the sperm duct from the body in the living condition. This is possible by appropriate and careful teasing out of the reproductive segments of a mature worm. During this teasing-out process, the duct can be recognised as a small, whitish, shining mass, formed of a number of coils, which, with care, can be unravelled on the slide.

It is comparatively easy to get the tube with the spermiducal gland and penis still attached, but the structure of the penis cannot be seen well by this means, the only feature which is clearly brought out being the chitinous nature of the penis sheath. It is a much more delicate operation, however, to get the extreme anterior portion of the tubular part with the ciliated funnel still attached. The difficulty is due to the fact that the funnel is situated in a different segment from that occupied by the tube, and the latter is usually broken off at the septum. If one is fortunate enough to get the funnel also, the shape of the latter can be fairly clearly seen, and its appearance will be described later. Although this method of investigation is invaluable for obtaining a correct idea of the relation of the various parts of the sperm-duct to one another, it is of no use for histological details.

1. **The Ciliated Funnels** of the sperm ducts are situated in the segment in front of that which contains the tubular part of the duct, that is, they lie in segment 10, or the segment that contains the testes, and they open directly into the body cavity. When seen in the living condition the funnel has the form of a flat, plate-like expansion, but when sections are examined it will be seen that its shape and appearance vary a good

deal in worms of different ages. In a very young worm in which both testes and ovaries are present, but before the development, to any great extent, of the sperm sac, the ciliated funnel is visible. It is the first part of the vas deferens to be formed, and appears as a local proliferation of the cells of the septum between segments 10 and 11. It rests at first directly on the septum, and it is only at a later stage, after the formation of the tube itself, that it hangs freely into the body cavity. Even at such an early stage as this, it is slightly hollowed out, with its convex side attached to the septum and its concavity opening into segment 10. As the vasa deferentia develop the ciliated funnels increase in size, lose their direct connection with the septum 10/11, and take up their normal position in the segment, being situated one on either side of the intestine and a little ventral in position. Each funnel forms a markedly cup-shaped expansion of the anterior end of the vas deferens. It has a circular outline in transverse section, and opens into the body cavity by a wide aperture (Pl. IV, fig. 18B).

The funnel is composed of a single layer of cells which, when seen in section, are quadrangular in shape, those nearer the centre of the funnel being squarish in outline, while those nearer the edge are somewhat longer than broad (Pl. IV, fig. 17, *ct. ep.*). They are all provided with large nuclei, those in the squarish cells being almost spherical, while the others are oval in shape, but all possess a very distinct nucleolus. This epithelium is ciliated, the cilia being very conspicuous both in stained preparations and in the living specimen. In the latter, the cilia do not seem to be disposed equally over the inner surface of the funnel, but rather to be confined to certain tracts. There is nothing in the sections, however, to suggest this arrangement, and it is very likely that the

cilia are not all equally active at the same time, but that some cease entirely to vibrate for a time, and then resume their action again. Outside the ciliated epithelium is a single layer of very much flattened peritoneal cells, which is continuous with the peritoneum, completely investing the rest of the vas deferens. This layer is extremely delicate, but is rendered more conspicuous by the somewhat swollen, oval nuclei which the cells contain (Pl. IV, fig. 17, *pt.*).

The ciliated funnel of a mature worm has a very characteristic appearance. It is no longer cup-shaped, but the edge is so sharply recurved towards the vas deferens that the funnel becomes much more shallow and is almost turned inside out (Pl. IV, fig. 17). The surface of the funnel which bears the cilia is thus exposed as fully as possible. There is not the least doubt that this interesting change in the shape of the funnel is intimately connected with the transference of mature spermatozoa from the sperm sac to the vas deferens. But it is difficult to say whether this change in shape is completed before the spermatozoa actually reach the funnel, in order to expose as much surface as possible for their reception, or whether it is due entirely to the pressure exerted by the enormous number of spermatozoa which congregate upon it. In a mature worm the whole of the ciliated surface of the funnel is covered with a dense mass of spermatozoa, not arranged irregularly, but with their heads entangled amongst the cilia of the funnel, and their tails lying parallel to the cilia (Pl. IV, fig. 17, *sm. t.*). At first sight, the funnel appears to be provided with enormously long cilia, but a closer examination of the stained preparations reveals the true state of things. Just outside the boundary of the cells composing the funnel is a narrow zone which is clearly differentiated

from the rest of the apparent cilia. This differentiation is due to the staining properties of this region, for the stain which is absorbed is a nuclear stain such as haematoxylin or borax carmine, whereas the cilia stain only with a plasma stain. This deeply-stained zone just outside the cells of the funnel is caused by the presence of the heads of the spermatozoa, which always stain heavily with a nuclear dye (Pl. IV, fig. 17, *sm. h.*). In many of the preparations which I examined the cilia of the funnel were not visible at all owing to the large number of spermatozoa present, but in a few cases the spermatozoa have slightly shrunk away from the funnel, and in these the true cilia can be seen clearly. Their length is not more than one-third or one-fourth that of the spermatozoon tails. It seems to me that we have here an example of the phenomenon of stereotropism, so well known already in the case of spermatozoa, which appear to be always attracted by any surface. If one examines a funnel in the living condition one often sees a very large number of spermatozoa hovering around and on the funnel, and it is very possible that in the processes of killing and fixing the worm they are maintained in this position.

2. **The Coiled Tube.** The ciliated funnel opens into the tubular part of the sperm-duct, just in front of the septum between segments 10 and 11. The tube, therefore, perforates the septum. It belongs typically to segment 11, but owing to its great length it extends posteriorly through several segments, although its external aperture remains in segment 11. If the tube be examined under the microscope directly it has been removed from the body, it will be seen as a long, delicate, transparent structure with a very clearly marked cavity. If one watches it for a short time it will be seen that the

cavity of one part of the tube is surrounded on all sides by a wall of equal thickness. This part is nearer to the ciliated funnel, and its walls are thin. On the other hand, the part nearer to the spermiducal gland has much thicker walls, and, moreover, these walls contract a little and the cavity of the tube is thrown out of position so that it no longer lies centrally, but takes on the form of a very open spiral, the whole tube meanwhile becoming slightly shorter. If transverse sections of the sperm duct were cut after its removal from the body, the canal would be seen to be lying nearer the outer wall, first on one side, then on the other. In sections of the sperm duct cut when it is in position in the body, however, the canal lies centrally for all its length, which seems to indicate that the spiral curve of the canal in the first case was due to contraction on its removal from the body. Beddard states in his Monograph on the Oligochaeta that the coiled tube in all the members of the Tubificidae is ciliated throughout, but this statement is not entirely correct. In the immature form of *Tubifex rivulorum* cilia are present throughout the whole length of the duct, but the coiled tube of the mature worm can be divided into two parts, one ciliated, the other non-ciliated. The former lies nearer to and in connection with the ciliated funnel, while the latter opens into the spermiducal gland. These ciliated and non-ciliated parts of the tube can be easily recognised, both in the living condition and in sections. We will first confine our attention to the vas deferens of the mature worm, though it will be necessary later on to refer to the immature form for the purpose of comparison.

(*a*) The ciliated portion of the tube occupies about half its total length, and follows immediately upon the ciliated funnel (Pl. I, fig. 2,

v.d. 1). Ciliary action does not occur by any means regularly in this part, for it is often completely in abeyance, but the cilia are so long that they can be easily seen, even when they are motionless.

In the wall of the ciliated portion of the tube we can recognise the following layers:—(1) Ciliated epithelium, (2) Muscular layer, and (3) Peritoneum.

The ciliated epithelium forms the innermost layer of the wall of the duct and is composed of a large number of flat, annular or ring-like cells piled very regularly on one another. Each cell is perforated in the centre by a large, rounded lumen, about 42 μ in diameter, the wall around it being comparatively thin. A continuous tube is formed by the regular arrangement of the cells and by the fact that the cavity is pierced in exactly the same position in all the cells (Pl. IV, fig. 20A). Each cell possesses a single, elongated, somewhat spindle-shaped nucleus which may extend almost half way round the cell (Pl. IV, fig. 19A). It is pointed at both ends and somewhat broader near the middle, where it exhibits a well-marked, rounded nucleolus. The inner edge of each cell is plentifully provided with cilia which are rather longer than the radius of the cavity, and have a somewhat spiral arrangement (Pl. IV, fig. 19A, *ci*). The cells themselves appear to be embedded in a structureless matrix which stains with plasma stains and forms an extremely delicate layer right round this part of the duct. The centre of the tube is usually occupied by a dense mass of spermatozoa, which in the living condition can be seen travelling down the tube. The muscular layer is but feebly developed in this part of the tube. It consists of a single layer of longitudinal muscle fibres which are disposed at equal distances around the tube (Pl. IV, fig. 20A; *l.m.*). They do not pass straight down the wall

of the tube, but have a somewhat spiral arrangement. The peritoneum, which completely invests the tube, is composed of a single layer of very much flattened cells with rather large, oval, nucleolated nuclei (Pl. IV, fig. 20, *pt.*).

(*b*) The non-ciliated portion. The ciliated part of the tube gives place gradually to the second part, which is characterised by the absence of cilia and the presence of a much thicker wall. Consequently, this part of the canal has a diameter almost twice as great as that of the first part (Pl. I, fig. 2, *v.d.* 2.). The lumen of the tube is of exactly the same diameter throughout, so its increase in size is due entirely to the greater thickness of its wall.

The epithelial cells which form the innermost layer of the wall of this second part of the tube have the same general form as those described above. That is, they are ring-shaped, piled one on top of the other extremely regularly, and each one is perforated in the centre by a rounded lumen. Each cell has its greatest thickness around the lumen of the tube, and tapers off considerably at its outer edge (Pl. V, fig. 21). The nuclei of these cells resemble very closely those of the first part of the tube, and are situated near the inner edges of the cells (Pl. IV, fig. 19B). The greater thickness of the wall of this part of the tube is due to a considerable increase in the quantity of the matrix in which the cells are embedded, and by which they are surrounded (Pl. V, fig. 21, *ma.*). This matrix still stains only with plasma stains, but it is no longer structureless, for it is characterised by the formation in it of a large number of exceedingly fine fibrillae. These are not visible in the fresh material, but are rendered very distinct by the action upon them of fixing reagents and by the use of appropriate stains.

They are arranged very regularly, originating at the inner edge of the cell and radiating out to the periphery. These fibrillae are so numerous that there is but little of the normal cytoplasm remaining. Owing to the large number of these fibrillae which are present, and to the consequent disappearance of the cytoplasm of the cells, the cell boundaries become very indistinct, and are barely recognisable. The nuclei, however, persist in their original position, and indicate, sufficiently clearly, what was the arrangement of the cells (Pl. IV, fig. 20B).

In the mature worm these cells are not ciliated, but the inner edge of each is considerably thickened and strengthened by the deposition of a secretion resembling cuticle in appearance and staining reactions. Since the inner edge of each cell is thus thickened, the secretion has the form of a series of circular bands arranged extremely regularly throughout the length of the tube (Pl. IV, fig. 20B, *an.r.*). It is extremely difficult to decide what is the exact nature of the substance composing these rings. Its position in relation to the cell and to the lumen of the duct suggests a chitinous or cuticular secretion, but there is no doubt that it is capable of contraction, which suggests that it is muscular in nature. These circular bands convert this part of the vas deferens into a much more solid structure than it would otherwise be, and also help to keep the lumen open. Outside the epithelial layer is a single layer of longitudinal muscle fibres arranged similarly to those of the first part of the tube. They are continuous, at the end of the vas deferens, with those which surround the spermiducal gland. A peritoneal covering invests the vas deferens in this part also, and its cells are flattened, forming a single layer, and provided with oval nuclei.

The change from the narrower to the wider part of

the vas deferens is a gradual one, and this transitional part of the tube is worth further examination. As has been already noticed, the lumen remains of the same size throughout. The wall, however, thickens gradually. The cells of which it is composed become somewhat larger, and are a little further apart from one another, but their boundaries are quite distinct (Pl. V, fig. 21). The matrix increases in quantity, it is developed between adjacent cells and also in a thick layer outside them. The fibrillae are quite distinct, and very numerous even in this region. For a short distance the cells of the second part of the tube are ciliated, but there are no annular rings. As soon as the rings appear the cilia are lost, and at this stage also the cell boundaries become less distinct.

The vas deferens of quite a young worm is ciliated throughout, and the wall of the second part of the tube is only very slightly thicker than that of the first. That is, the matrix is only present in small quantities at first, and there are but slight indications of the annular rings.

It seems strange that so little notice has been taken of the complicated structure of the vas deferens in this worm, and, possibly, in many others. Beddard, as well as stating that the tube is ciliated throughout, adds in another place that the wall in Oligochaeta as a whole is usually composed of an inner ciliated epithelium and an outer peritoneal covering. He seems to consider it a very exceptional condition for there to be any muscular elements at all, and he quotes Eudrilus (an earthworm) as his only example. Nasse (1882) seems to have been the first and only observer to note the essential points in the structure of the vas deferens of Tubifex, but his description gives one the impression that he was uncertain on some points, such as the actual form of the cells, whether they were annular or only spindle-shaped, and whether an

external peritoneal covering is present or not. In a paper recently published in the *Zoologischer Anzeiger*, Dr. Bohumil Cejka (1913) gives a short account of the structure of *Litorea krumbachi*, in which he deals with the vas deferens. From his description of the histological structure of this tube, I am inclined to think he has seen something of the structure which I have attempted to describe. He says, for instance, that two regions are recognisable. In a transverse section of the first part of the duct he describes the tube as circular and the lumen as being provided with strong cilia. At the periphery he has seen black fibrils, which he believes play an important part in contraction. The second part of the tube, he says, is not ciliated, and has thicker walls which are composed entirely of gland cells—a structure which is quite different from that just described for *Tubifex rivulorum*.

3. **The Spermiducal Gland with the Prostate.** This gland has the form of an elongated, somewhat pear-shaped and curved sac, into the broader end of which opens the second part of the vas deferens, while the narrower end communicates with the exterior by the penis, which will be described later. The cavity of the spermiducal gland is a direct continuation of the cavity of the vas deferens, and is also continuous with that of the penis (Pl. VII, fig. 45). We may, therefore, consider the gland and penis as being modified portions of the sperm duct. Attached to the gland at one side, and near its swollen extremity, is a glandular, irregularly-shaped, lobate mass forming what is known as the prostate (Pl. VII, fig. 45, *pr.*). The cells of which the prostate is composed are large and pear-shaped, with prominent rounded nuclei situated usually in the swollen part of the cell. The narrower part of each cell forms its

duct, and in sections these are all seen to converge towards one point, that point being where the prostate is in communication with the spermiducal gland. At this point, the muscular and peritoneal layers of the wall of the gland are interrupted so that the cells of the prostate and those of the innermost layer of the gland are intimately connected with one another. In fact, it has been said by those observers, *e.g.*, Vejdovsky, who have studied the development of the spermiducal gland and the prostate, that the latter is simply formed as a proliferation of the cells of the lining epithelium of the gland.

The wall of the spermiducal gland is composed of the following layers:—(*a*) An inner epithelium, (*b*) A muscular layer, and (*c*) Peritoneum.

Beddard states that the lining epithelium of the spermiducal gland in Limnodrilus is ciliated, and this suggests the possibility that a similar condition obtains in the other members of the family Tubificidæ. Vejdovsky certainly believed it to be ciliated in Tubifex, as he states that the prostate is derived from cells of the ciliated lining epithelium. In the mature worm, at any rate, I have never been able to identify cilia in the gland, and as the second portion of the vas deferens is not ciliated this is not surprising. The cells of which this epithelium is composed differ a good deal in structure and appearance at different stages of their development. In a young worm, whose reproductive organs are all developed but not yet matured, the lining epithelium of the spermiducal gland is composed of a single layer of rather low cells, cubical in shape, with oval nuclei which are large and conspicuous and situated near the outer end of the cell. The cell is filled with a dense, granular cytoplasm, which stains deeply with nuclear stains such as borax carmine. Just at the

junction of the prostate with the gland, this single layer of cells gives place to a mass of irregularly shaped, somewhat elongated cells which encroach a good deal upon the cavity of the gland in this region. In a mature worm the appearance of these cells is very different. They become much enlarged, so that they tend to obliterate the cavity of the gland. Their outlines become less distinct, and the nuclei, which remain near the outer end of the cells, are very difficult to identify. This increase in the size of the cells is accompanied by a change in their contents. The cytoplasm becomes less granular and very vacuolated, and its appearance suggests the presence of some secretion which stains readily with "plasma" stains, such as picro-indigo-carmine (Pl. VII, fig. 45). Outside the epithelium is a layer of longitudinal muscle fibres placed somewhat obliquely, and outside this again a delicate peritoneal covering of flattened cells.

4. **The Penis.** The terminal portion of the spermiducal gland is modified to form a protrusible penis which opens to the exterior on the ventral surface of segment 11. The structure of the penis is somewhat complicated. Its cavity is continuous with that of the gland, and it is surrounded by a penis sheath, which, when the penis is retracted, is double, the outer layer being continuous with the epidermis of the body wall (Pl. VII, fig. 45, *pe.s.*). We may, therefore, speak of the outer and inner penial sheaths, both consisting of a single layer of epithelial cells, somewhat cubical in shape and provided with conspicuous, rounded, nucleolated nuclei (Pl. V, fig. 22, *pe.s.* and *pe.s.*[1]). The cuticular secretion, which forms a delicate layer outside the cells of the epidermis, is continued over the cells of the penial sheath, and in the retracted condition of the penis may completely fill the space between the outer and inner penial sheaths. The

outer sheath is provided with muscles externally, these being disposed mainly in two ways (Pl. V, fig. 22). There is a comparatively delicate layer of circular muscles completely investing the sheath, and, from this layer, oblique muscles pass, in a somewhat irregular fashion, to the body wall. Certain of these fibres branch, and the branches anastomose with the longitudinal muscles of the body wall.

The cavity of the penis proper is surrounded by a single layer of elongated cells (Pl. VII, fig. 45, *le.*) which encroach considerably on the lumen. Near the apex of the penis they are so large that they almost obliterate the lumen altogether.

V. The Development and Structure of the Ova.

The ovary of a young worm consists of a solid mass of undifferentiated cells, of which those which are nearest to the septum to which the ovary is attached are the youngest. The nuclei of these cells are often to be seen undergoing mitotic division, and by this means the number of the cells making up the ovary is increased. The first formed cells are gradually pushed further away from the septum, and these give rise to potential ova or oogonia, which can be recognised by the large size of their nuclei as compared with the amount of cytoplasm surrounding them.

Vejdovsky suggests that in such a form as Rhynchelmis the oocytes are at first amoeboid, and that their subsequent rapid increase in size is due to the ingestion of the ova around them. I have never been able to identify amoeboid oocytes in Tubifex, but some sections have revealed what appears to be an equally effective mode of nutrition. The young ovary is, as has already

been stated, composed of a solid mass of cells, all of which seem to have equal chances of developing into mature ova. In many cases, however, development only continues in those cells around the periphery of the ovary. The central cells do not develop any further, but gradually lose their individuality, the cell membranes disappear, and the nuclei are absorbed. These changes in the central cells result in the formation of a central, non-nucleated, cytoplasmic core surrounded by developing oocytes. It is interesting to notice that these oocytes are distinctly pear-shaped: the narrower part of the cell being embedded in the central mass, while the nucleus is lodged in the broad part of the cell (Pl. V, fig. 26, *oc.*). Certain of these cells develop much more rapidly than the others, and increase so much in size that they protrude considerably beyond the surface of the ovary, and are only enclosed by a fine membrane composed of a single layer of much-flattened cells. It is very easy to conceive of these oocytes being dislodged from the ovary, and this is what actually does happen, for when they have reached a certain size they are transferred to the egg sac, while still immature, and here they complete their growth.

In the course of my investigations of the living worm, I have several times succeeded in teasing out and freeing from the body the ovary, in a more or less complete condition. In most cases, its appearance very nearly resembles that seen in sections, and although the oocytes in this condition are very transparent, it is possible, by the addition of a little methylene blue, to stain them sufficiently to examine them before disintegration sets in, which, by the way, takes place very rapidly. As a rule, there is no special blood supply to the ovaries. It has already been mentioned that the perivisceral vessels of segments 10, 11 and 12 are much enlarged in the mature

worms, and that these vessels give off important branches which are directed backwards over the reproductive organs. There are, however, no special branches from these vessels to the ovaries. In one case I found quite a different state of affairs on teasing out the ovary from the living worm (Pl. V, fig. 25). Only a portion of the ovary was obtained on this occasion, but the oocytes present were situated on all sides of a central "stem" or rod in which a blood vessel was lying—this could easily be identified by the red colour of the blood which was still present in it (Pl. V, fig. 25, *st.*). The oocytes were arranged quite irregularly on the stem. Large and small ones lay side by side, sometimes grouped together into clusters, sometimes occurring singly. Each oocyte was attached to a short branch of the main stem terminating in a tiny swollen head, the oocyte itself being spherical in outline, or nearly so. Opposite to each stalk a short branch was given off from the blood vessel—this branch entered the stalk and terminated in the swollen head already mentioned. It was not possible, of course, to decide what was the connection of this "ovarian vessel" with the rest of the circulatory system, as the ovary was completely isolated. Although I examined many more ovaries I never saw this interesting condition again.

As the oocytes contain no yolk-granules as long as they remain part of the ovary, they are then most suitable for purposes of examination, as the yolk-granules tend to obscure the other structures in the egg (Pl. V, fig. 27). The structure of such an oocyte of Rhynchelmis has been described in detail by Vejdovsky (1884), but the description does not entirely agree with the conditions obtaining in an oocyte of Tubifex, at the same age.

The oocyte is spherical in outline, and surrounded by a delicate investing membrane. The cytoplasm forms

a fine network, the density of which is only slightly greater at the periphery than nearer the centre of the cell. In Rhynchelmis, Vejdovsky figures a much denser portion of the cytoplasm around the periphery with a more delicate network in the interior. The nucleus is large and rounded, and at this stage is situated near the centre of the cell, but as the ovum matures it comes to lie nearer the periphery. The nucleus is surrounded by a well-marked nuclear membrane, but I have not been able to identify any perforations through which the nucleoplasm is in communication with the cytoplasm, such as are figured by Vejdovsky as occurring in Rhynchelmis. The nucleus is composed of a large number of chromatin granules embedded in the usual linin network—the linin stains only faintly, but the chromatin stains much more deeply, and, therefore, shows up well in good preparations.

The nucleus contains one or more nucleoli, which appear to have a different structure in different ova. In all cases there is one large nucleolus which is spherical in outline, and which we may describe as the "principal" nucleolus.* In some cases this nucleolus has a vacuolated appearance, showing one or two fairly large vacuoles, in other cases it stains uniformly and appears to be an undifferentiated mass of chromatin, while in yet other ova it has a granular appearance (Pl. V, fig. 27, *p.nul.*). In addition to this principal nucleolus there is usually a number of smaller masses which we may call "accessory" nucleoli.* These are not always present, but in those oocytes in which they do occur their number varies from 2 to 5. These accessory nucleoli very often have no connection with the principal one, but sometimes one large accessory nucleolus is in conjunction with the principal one, forming a compound body. In this case

* Compare Wilson's "The Cell in Development and Inheritance," p. 127.

the accessory nucleolus usually stains more deeply with a nuclear stain than does its companion (Pl. V, fig. 27, *ac. nul.*).

Very soon after the oocyte has been transferred to the egg sac, yolk granules appear in the spaces of the cytoplasmic network, the cytoplasm meanwhile shrinking away from the wall a little. Later on the yolk granules become so numerous that the protoplasm cannot be seen at all. The formation of the granules seems to have no connection with the position of the nucleus, that is, they are scattered irregularly through the cytoplasm. The granules are spherical bodies of comparatively large size, varying from 2 μ to 4 μ in diameter. They stain very deeply with both nuclear stains such as haematoxylin and plasma stains, such as picro-indigo-carmine (Pl. V, fig. 24, *y. gr.*).

It is interesting to notice that maturation of the egg-cell actually begins while the latter is still enclosed in the egg sac, but I have not been able to decide exactly how nearly the process is completed before the egg is extruded, as all the specimens I have obtained which have been undergoing maturation have been at about the same stage. The oldest oocytes within the egg sac often exhibit a fully-formed nuclear spindle, the nuclear membrane having already disappeared (Pl. V, fig. 24). The spindle is somewhat dumbbell-shaped, and exhibits the normal structure. The two centrosomes are far apart from one another, but are connected by a large number of spindle threads. The star or aster (Pl. V, fig. 24, *as.*) is formed by a number of delicate threads which, surrounding the centrosome, radiate out into the adjacent cytoplasm. The chromatin has grouped itself into a number, about 24, of small rounded masses, which are arranged on the equator of the spindle (fig. 24, *chr.*). The nucleus at this time is situated near the periphery of the cell.

VI. The Egg Sac.

The egg sac is set apart for the reception of immature ova which have become separated from the ovary. In this organ the ova complete their growth and undergo the first stage of their maturation process. The egg sac is an unpaired structure, and arises as a single, pouch-like outgrowth of the septum between segments 11 and 12. It is directed backwards, and in its simplest condition occupies only segment 12. But it soon enlarges considerably, and extends into the segments behind the twelfth. The sperm sac in the course of its development becomes pushed into the egg sac, so that we have here the curious condition of one sac lying actually within the other (Pl. III, fig. 11, *sp.s.*, *ovs.*).

The cavity is, of course, coelomic in origin, and remains undivided throughout, that is, there is no division of its cavity into smaller ones, such as occurs in the sperm sac. The wall of the egg sac is thin and composed of a single layer of flattened peritoneal cells, with no muscular elements at all (Pl. I, fig. 3, *ovs.*).

Egg-cells of different ages can be found in the egg sac, and their position in this organ suggests very forcibly that they have all been derived from the ovary. The youngest oocytes are situated in the most anterior segments: that is, in those nearest the ovary, and many of these have few or no yolk granules. The oocytes, when liberated from the ovary, fall into the cavity of segment 11, in which the ovary lies. As the egg sac is an outgrowth of the posterior septum of this segment, it is a very simple matter for the oocytes to find their way from the segment into the egg sac.

VII. The Oviducts.

There has been a considerable difference of opinion amongst earlier observers as to the position of the oviducts in most members of the Tubificidæ. Without going into the whole history of the question, we may notice that D'Udekem (1855) believed that in *Tubifex rivulorum* the oviducts were connected with the male ducts, indeed that they actually surrounded the terminal portion of the vasa deferentia. His views were supported by later observers, such as Claparède (1861) and Eisen (1885). Vejdovsky at first was inclined to agree with D'Udekem's description of the relations between these two ducts; but later he changed his opinions, and came to the conclusion that the oviducts are situated between segments 11 and 12. This conclusion was based upon certain experiments which he performed. For example, he kept worms in certain chemical reagents, and observed the extrusion of the eggs between these two segments.

For some time I was unable to identify the oviducts in my sections, but Mr. E. S. Goodrich, F.R.S., was kind enough to lend me some of his slides in which they were shown quite plainly. Since then, on a re-examination of my own preparations, I have been able to distinguish these structures, though much less clearly than in those of Mr. Goodrich—to whom I wish to express my indebtedness.

There can be no doubt that, while the oviducts are small and comparatively inconspicuous, they are normally present, at any rate in the fully mature worm. There is a single pair lying in the intersegmental line 11, 12 (Pl. I, fig. 2, *ovi.*). The duct is very short, and opens internally by a wide, funnel-shaped opening into the coelomic cavity of segment 11, while the external opening, which is small and inconspicuous, lies in the same longitudinal line as the male openings.

VIII. The Spermathecae.

During copulation the spermatozoa of one worm are transferred to the other, and stored in special organs known as the spermathecae (Pl. I, fig. 2, *sp.*). In *Tubifex rivulorum* there is one pair of these organs situated in the same segment as the testes, viz., in segment 10. Their general form is best seen by liberating them from the living worm. This can easily be done if a mature worm be placed on a slide and a cover-glass put upon it. The slightest pressure on the cover-glass is usually sufficient to rupture the body wall in the region of the reproductive segments, which are much distended owing to the large number of spermatozoa and ova developed. If the body wall be ruptured, the spermathecae, which for the greater part of their length lie freely in the body cavity, are usually forced out, but remain attached to the worm at their external apertures (Pl. VII, fig. 49). The ease with which this operation can be performed is due to the fact that the spermathecae are of considerable length and are bent round in the segment. They are also resistant to pressure, to some extent, and as soon as the body wall of the worm is ruptured they spring free. They are visible to the naked eye in this condition as small, pear-shaped, opaque, but glistening bodies, but it is necessary, of course, to examine them under the microscope in order accurately to describe their form. When examined thus, the whole organ can be divided into two regions:—(*a*) a pouch or sac-like portion (Pl. VII, fig. 49, *sp.* p^1.) which narrows considerably to form (*b*) the duct opening to the exterior near the ventral setae of segment 10 (Pl. VII, fig. 49, *sp. d.*).

The proportions of these two parts to one another

vary considerably according to the condition of the worm—in a mature worm the duct is usually somewhat longer than the pouch, but in the younger condition they are more nearly equal. Typically, the spermatheca lies entirely in segment 10, but about the time of copulation, and certainly when the spermatozoa are to be found in these organs, they enlarge considerably and may be either coiled round in segment 10 or extend into segment 11, or even more posteriorly into segment 12 (Pl. IV, fig. 16, *sp.*). In the latter case there is a sharp bend in the spermatheca, and the duct runs forwards again parallel to the pouch in order to open to the exterior on segment 10. The pouch is quite simple, although it is so voluminous, and is devoid of diverticula; neither has it any connection with the alimentary canal, as is the case in certain Enchytræidae.

The structure of the wall of the spermatheca differs considerably in different parts. The wall of the pouch is thin, but three layers are recognisable. Internally there is a single layer of epithelial cells which seem to be of two kinds. Certain of the cells are squarish in shape and have very large nuclei, which are oval or rounded in shape, and usually include one principal and one or more accessory nucleoli. These cells are probably glandular in nature. Between these are smaller, narrow cells, whose nuclei are much less conspicuous, and which we may term interstitial cells. The wall of the pouch is strengthened by the presence of a few scattered muscle fibres placed parallel to the longitudinal axis of the spermatheca and outside the layer of epithelial cells. Outside this again, the wall is covered by a single layer of flattened peritoneal cells, the nuclei of which are oval in shape and form the most conspicuous part of the cells.

The duct when seen in transverse section is circular

in outline, and its wall shows the same layers as are to be seen in the wall of the pouch, but their structure and arrangement are somewhat different (Pl. V, fig. 23). There is a very marked change in the character of the cells forming the inner epithelium. There has been considerable discussion amongst the earlier observers as to whether the epithelium of the duct is ciliated or not in the spermathecae of species belonging to the family Tubificidae. Beddard, although he mentions the views of different authors upon the subject, does not venture any opinion himself. He states in his general account of the spermathecae of the Oligochaeta that ciliation does not, as a rule, occur, but that it has been described in a few forms, for example, Tubifex. As no reference was given, I have been unable definitely to trace the paper from which he obtained this information. It seems likely, however, that he is referring to a paper by Nasse (1882), which he mentions in his general description of the family Tubificidae. Nasse definitely states that there are cilia in the duct of the spermathecae of Tubifex, "In Ausführungsgange trägt das Epithel eine Cuticula und flimmert." Vejdovsky (1884), careful observer though he was, has denied the presence of cilia, and Stolc in his monograph on the Tubificidae (1888) does not figure them. I am now in a position to confirm the statement made by Nasse, for in sections of the spermathecal duct, both transverse and longitudinal, the cilia are particularly obvious (Pl. V, fig. 23, *ci.*). They are not visible when one examines the spermatheca in the living condition, for the wall of the duct is rendered opaque by a thick, muscular covering. The epithelium of the duct is composed of a single layer of cells almost columnar in shape, but considerably longer than broad, and with large oval nuclei. The cilia are rather short, shorter

considerably than the radius of the cavity of the duct, so that they do not obstruct the passage much.

The muscular layer is much more powerfully developed around the duct than on the pouch. The muscles form a double layer, those on the inside being arranged circularly, while outside this is a very definite sheath of longitudinal muscles. These appear to be continuous with those on the pouch, the circular muscles being interposed between them and the ciliated epithelium. The cells of the peritoneum covering the duct are somewhat different from those investing the pouch. They are no longer flattened, but squarish in outline, and form a rather more conspicuous layer than that of the pouch (Pl. V, fig. 23, *pt.*). The nuclei are usually situated near the middle of the cell, the contents of which are very granular.

The terminal portion of the duct of the spermatheca is protrusible, and when it is protruded the duct narrows considerably near the end and leads to the exterior by a very narrow but straight passage. Near the spermathecal pore (Pl. IV, fig. 16, *sp. p.*) the ciliated epithelium ceases, and that which lines the narrow passage leading to the exterior is similar in structure to that forming the epidermis of the body wall, with which it is directly continuous.

The muscular layer extends to the end of the duct, where the muscles become connected with those of the body wall. When the terminal portion of the duct is retracted, the passage to the exterior is no longer straight, but its wall is folded back in the form of a cone or arrow-head.

IX. The Spermatophores.

A spermatophore consists of a large number of spermatozoa cemented together by a substance derived from the prostate, the cells of which open into the spermiducal gland as already described. These spermatophores are comparatively solid and resistant structures, and can always be found in the spermathecae of a mature worm. When properly formed, they have a very characteristic shape and structure, but it is quite common to find ill-formed ones which are not at all typical in shape or structure. This seems to be due to a lack of the cementing substance.

The number of these structures which may be found in one spermatheca varies considerably. I have often found only one, more commonly two or three, and occasionally as many as five or six. They vary, too, very much in size, but, as a rule, the perfectly formed ones are nearly or quite as long as the spermatheca in which they are found. They are arranged quite irregularly in the spermatheca, sometimes lying coiled up entirely in the pouch, and in other cases occupying the duct and whole length of the pouch, and yet lying coiled up in it. Their arrangement can best be seen when the spermathecae are liberated from the living worm, as described above (Pl. VII, fig. 49). They can then be examined within the spermatheca, or, with care, it is possible to liberate them by rupturing the wall of this organ, when those which are coiled up in it break free and may be removed in a perfect condition. They stain well in a very dilute solution of methylene blue, but permanent preparations cannot be made in this way. They can, however, be fixed to the slide, and stained first with borax-carmine and counter-stained with picro-indigo-carmine, when the

various parts will be well differentiated, the heads of the spermatozoa staining well with the borax-carmine, and the tails with the picro-indigo-carmine.

The spermatophores are visible to the naked eye, and when first liberated from the spermatheca appear as small, fine, white, glistening bodies, the largest being about 1-2 mm. long. When examined under the microscope they will be seen to have quite a complicated structure, but before describing this it will be well, perhaps, to say a word or two about their formation. During copulation, the spermatozoa, together with the secretion of the gland cells of the prostate, are transferred from one worm to the spermathecae of the other. It seems certain that the moulding, which must take place before the spermatophores attain their final shape, is carried out entirely in the spermatheca, and moreover in one part only of the spermatheca, viz., its duct. The duct leading from the external aperture to the pouch is so narrow, and the quantity of sperm forced in so great, that the mass must necessarily conform very closely to the shape of the duct. And this we find is the case even to the minutest detail. We will first describe the general form and minute structure of the spermatophore, and then show how this agrees with the general shape of the spermathecal duct.

The fully-formed spermatophore is a narrow, much elongated structure, many times longer than broad, and somewhat worm-like, its widest part being near the middle and tapering off at both ends. The anterior end is always simple in structure, but the posterior end may be similar to it or may terminate in a beautifully moulded and curved conical head (Pl. VII, fig. 50). It has been suggested that the presence or absence of a conical head is characteristic of two different species, but this is

impossible, as I have found spermatophores with and without the head in the same spermatheca or in the two spermathecae of the same individual. I shall refer to this again later.

The minute structure of these spermatophores has been described by Vejdovsky (1884), and by Lankester (1871, A). When examined in transverse section, the spermatophore is seen to be circular in outline. In the centre is a cavity which we will call the axial cylinder (Pl. VII, figs. 51 and 52, *ax.c.*). It extends from end to end of the spermatophore, and even into the conical head. It is widest in the centre, and tapers off at both ends. This cavity is filled with a substance which, when the spermatophore has just been liberated from the worm, is granular in appearance. This central mass stains deeply with borax-carmine, and in permanent preparations has the appearance of a mass of longitudinal fibres, which usually become somewhat shrunken by the action on them of reagents. Outside the axial cylinder is a very narrow dark band, which stains much more deeply than the substance of the axial cylinder (Pl. VII, figs. 51 and 52, *sm. h.*). For this reason it is very conspicuous. When highly magnified it is seen to consist of a very large number of minute oval masses, which, owing to their staining properties, I conclude are the heads of the spermatozoa. The rest of the spermatophore is made up of a cementing material in which the tails of the spermatozoa are embedded (Pl. VII, figs. 51 and 52, *sm. t.*). As was pointed out both by Vejdovsky and Lankester, these are placed parallel to one another, but obliquely to the axis of the spermatophore. The tails of the spermatozoa appear as striations in the homogeneous substance in which they are embedded, and when viewed from the surface are seen to pass obliquely round from left to right.

Both Vejdovsky and Lankester state that not all the tail is embedded in the cementing material, but that a small portion lies outside, so that the whole spermatophore is surrounded by the free ends of the spermatozoa. The spermatophores which I have examined show a certain amount of variation in this respect. In many cases the tails are undoubtedly partly free from the cementing material, and the free ends are placed at right angles to the longitudinal axis of the spermatophore, and not obliquely to it. In some cases, however, the layer of cementing material is thicker than in others, and when that is so there is no portion of the spermatozoon lying free. Whether this is an abnormal condition, or merely due to the amount of cementing material present, is very difficult to decide.

Lankester's description differs from this in several details. He states that there is a narrow, highly refringent band outside the axial cylinder and another of similar nature outside the layer of cementing material and spermatozoa, between it and the free ends of the tails of the spermatozoa, the latter being in constant vibratile motion. He also says that the whole spermatophore exhibits movement when liberated from the spermatheca into a dilute salt solution. Although I have performed this operation many times, I have never succeeded in persuading the spermatophore to exhibit the active movements which have been described. Further, Lankester believes that the head of the spermatozoon is much elongated, of almost the same length as the tail, and that the head as well as part of the tail is buried in the cementing material towards the outside of the spermatophore. I think there is no doubt that the heads of the spermatozoa lie just outside the axial cylinder, but very close to it, and are confined to the deeply staining zone

described above, and that the oblique striations already referred to are caused by the tails of the spermatozoa alone.

It is very evident that the very characteristic form of the spermatophores is due entirely to the shape of the spermathecal duct and its external aperture. The duct is circular in section, so also is the spermatophore. During the rapid movements of the worm it is very natural that the spermatophore should be forced through the duct into the wider pouch by a slightly spiral motion, and this explains the oblique arrangement of the tails. The muscular walls of the duct may also assist in forcing the spermatophore into the pouch. As a rule, the first-formed spermatophores, those which are forced into the pouch soon after or during copulation, are pointed at both ends or without the arrow-shaped head described above. On the other hand, it is very usual to find that the spermatophores which are formed last, that is, those which lie partly in the duct and partly in the pouch, are provided with the conical extremity. There can be little doubt that the arrow-shaped head is moulded in that part of the duct following directly on the external aperture, for the shape of the two are exactly similar when the spermathecal duct is retracted. I conclude, therefore, that the spermatophores, moulded first in the spermatheca, are forced through the duct and into the pouch quite rapidly—partly by pressure from behind exerted by other masses of spermatozoa and cementing material, and partly by the contractions of the worm. At any rate, the process would seem to be such a rapid one that the spermatophore does not remain any length of time in the duct. On the other hand, towards the close of copulation, the formation of the spermatophores would take place more slowly, and the posterior ends of the last

one or two could remain in the terminal portion of the spermathecal duct a sufficient time to allow of their being moulded to its shape. It is true that I have never observed the posterior end of the spermatophore lying in this position, but I have found it several times only a short way along the duct and with the conical head perfectly moulded.

X. The Cocoon.

It is common amongst all Oligochaeta for the ova and spermatozoa to be deposited in cocoons, and *Tubifex rivulorum* is no exception to the rule. The worms are sexually mature in the autumn, and the cocoons are first seen in November. They are deposited on the mud in which the worms live, and very soon are completely buried in it. The cocoons are made of a fibrous substance secreted by the glandular cells of the clitellum, and have a characteristic form. They are usually whitish or greyish in colour and semi-transparent, but when viewed with the naked eye they appear opaque, but this is due to the eggs which they contain. They are usually oval in shape, but sometimes become more nearly spherical, and at either end they are drawn out into a short neck through which, when the young worms are ready to hatch out, they emerge (Pl. VII, fig. 54). The neck is filled with a plug of the same substance of which the cocoon is formed, but it is probable that it is of a less resistant nature. The number of eggs which may be present in a cocoon is very variable. Sometimes there is only one, more commonly from four to nine, but occasionally the number is greater, and in a very few cases I have seen as many as thirteen or fourteen. Although I have not been able to find any spermatozoa even in a freshly-laid

cocoon, there can be no doubt that in *Tubifex*, as in other Oligochaeta, the spermatophores are passed into the cocoon with the eggs, and are there dissolved to set the spermatozoa free. The rest of the cocoon is filled with a clear, colourless fluid, probably albuminous in nature. It seems to depend largely on the number of eggs in a cocoon as to whether they all develop into embryos or not. I have been able to count as many as nine young worms in a cocoon, all of which hatched out, but never more than that number; so it is probable that, as a rule, all the eggs in a cocoon develop, but when the number is unusually large certain only of them develop, the others breaking down. The young worm when hatched exactly resembles the adult in external form, and, with the exception of the reproductive organs, all the organs of the body are well represented. The newly-hatched worm is about a quarter of an inch long, and consists of 30 to 35 segments. The alimentary canal is complete with the exception of the anus, which does not develop until later. The whole canal contains a large number of yolk granules, the remains of those found in the egg, and there are some also in the coelomic cavity. The dorsal and ventral blood vessels are visible, the former already slightly contractile. The setae are perfectly developed, but small in size, and there are not more than two or three in each bundle in the anterior segments, and one more posteriorly.

PARASITES.

Tubifex rivulorum is a host for several internal parasites, and in addition to these it often has attached to the body wall externally Vorticellae and Fungi.

The Vorticellae are not true parasites, for they all possess an active circlet of cilia at the distal end and a

mouth. The worm is simply made use of in this case as a substratum to which the Vorticellid becomes attached. These Protozoa are very abundant on some individuals, and completely absent from others, but, as a rule, when present, they are confined to the posterior segments of the body, those which wave about freely in the water.

The fact that Fungi often attack these worms was recognised by McIntosh (1871), who speaks of Fungi growing on the dis-organised anterior segments, while the posterior ones are in full activity. I have often observed fully active worms which are infected with Fungus growths, and it seems very probable that the Fungus increases so much in quantity that it actually causes the disintegration of the segments which are attacked—the anterior segments are usually affected first, but the growth may spread throughout the entire length of the body. These Fungi appear, as a rule, to attach themselves in or near to the setigerous sacs, for they are usually to be seen emerging from between the two prongs of the sigmoid setae, forming long, delicate filaments, several of which may originate from the same seta-bundle.

The internal parasites may be found in the alimentary canal, sperm sac and body cavity. McIntosh states (1871) that he has found numerous examples of Opalina amongst the sandy mud in the intestinal canal. He gives figures of some of these specimens, which have very diverse forms. These worms were captured from the margin of the River Tay. It is interesting to note that, although I have examined numerous worms from the Thames both in sections and in the living condition, I have never once found Opalina in the alimentary canal. I have been more successful, however, in finding parasites in the sperm sac and body cavity. During the summer months of 1912, when the worm was really immature,

several specimens were found which at first sight had the appearance of maturity, for, in the region of the reproductive organs, the body was white, swollen and opaque. When the body wall in this region was punctured, parasites escaped in large numbers. At first they appeared as small, rounded, bodies, white and glistening, each of which proved to be a cyst bounded by a fairly thick wall (Pl. VII, fig. 53 A). If a cover-glass be put on, or the cyst be subjected to slight pressure, the wall bursts and its contents are liberated. These consist of an enormous number of extremely minute spores, each one somewhat awl-shaped, and provided with a caudal filament at one end. At the opposite end is a rounded, clear space, and between the two what may be described as the body of the spore, consisting of granular protoplasm and embedded in it an elongated nucleus, apparently broken into two parts (Pl. VII, fig. 53 B, *nu.*). This parasite was first described by Kolli, and was named by him *Urospora saenuridis*. It is a Gregarine of the sub-order Eugregarinae, and tribe Acephalina.*

McIntosh (1871) described these parasites as awl-shaped bodies, but he did not appreciate their real significance, as he interpreted them "as stages in the development of the spermatozoa." Nasse also saw them (1882), and interpreted them correctly as parasites occurring in the sperm sac. Minchin* mentions another Gregarine parasite (*Synactinomyxon tubificis*) as occurring in the sperm sac of *Tubifex rivulorum*, but I have not been fortunate enough to find it.

The last parasite noticed as occurring in Tubifex during my observation of the worm was Caryophyllaeus, a Cestode belonging to the family Caryophyllacea of the

* *Vide* "A Treatise on Zoology."—Ed. by E. Ray Lankester. Part I, pp. 194 and 298.

Cestoidea Monozoa. The parasite occupies the coelomic cavity, and has only been found during the summer months, when the reproductive organs are undeveloped. The mature form is to be found in the intestine of certain fishes, and the young in the anterior segments of *Tubifex rivulorum*, usually in segments 10 to 17. Each one is provided at the anterior end with a characteristic mobile organ capable of being thrown into a series of undulating folds. At the opposite end the cylindrical body is produced into a tail provided with three pairs of hooklets.

BIBLIOGRAPHY.

1908. Ashworth, J. H. The Giant nerve cells and fibres of Halla parthenopeia. Philos. Trans. Royal Society of London, Vol. 200, p. 427.

1898. Atheston, L. The epidermis of T. rivulorum with especial reference to its nervous structure. Anat. Anz., XVI, pp. 497-509.

1893. Beddard, F. E. On the Atrium and Prostate in the Oligochaeta, P.Z.S., pp. 475-487.

1895. ——— A Monograph of the Order Oligochaeta, Oxford.

1888. ——— The Nephridia of Earthworms. Nature, Vol. XXXVIII, p. 221.

1911. ——— Spermatophores of Earthworms. P.Z.S., p. 412-420.

1890. Benham, W. B. "Atrium" or "Prostate." Zool. Anz., XIII, pp. 368-372.

1880. Bloomfield, J. The general features of the Development of Spermatozoa in the Vermes. Zool. Anz., III, pp. 65-67.

1745. Bonnet, C. Traité d'Insectologie, ou Observations sur quelque espèces de Vers d'Eau douce, qui, coupés par morceaux, devinnent autant d'animaux complèts. Paris.

1850. Budge, J. Ueber die Geschlechtsorgane von Tubifex rivulorum. Arch. f. Nat., XVI, pp. 1-8, Pl. 1.

1851. ——— Ueber Respirationsorgane von Tubifex rivulorum. Verh. Nat. Ver. Preus. Rheinl., VII, pp. 258-9, Pl. V, Fig. 5.

1862. Buchholz, W. H. Beiträge zur Anat. der Gattung Enchytraeus. Schriften der Physik. ökonom. Gesellschaft in Konigsberg, p. 108.

1881. Bütschli, O. Beiträge zur kenntniss der Fisch-Psorospermien. Zeit. f. wiss. Zool., Vol. 35, pp. 629-651.

1895. Calkins, G. Observations on the Yolk nucleus of Lumbricus. XIV. Trans. New York Acad. Sciences.

1895a. ——— Spermatogenesis of Lumbricus. Journ. of Morph., Vol. XI.

1913. Cejka Bohumil. Litorea krumbachi—Ein Beitrag zur Systematik der Enchytraeiden. Zool. Anz., XIII, Band Nr. 4.

1861. Claparède, E. Recherches Anatomique sur les Oligochaetes. Mem. Soc. Phys. Geneve, XVI, p. 217-291.

1905. Depdolla, Ph. Untersuchungen uber die Spermatogenese von Lumbricus terrestris. Zool. Anz., Bd. XXVIII.

1886. Dewitz, J. Pfluger's Archiv, Vol. 37, p. 219, 1885; Vol. 38, p. 358, 1886.

1886. Diffenbach, O. Anatomische und Systematische Studien an Oligochaeta limicolae. Inaugural Dissertation, Giessen. From Ber. Oberhess. Ges., XXIV, pp. 65-109.

1856. Doyère, M. P. Essai sur l'Anatomie d. l. Nais sanguinea. Mem. Soc. Linn. Normandie, Vol. X, pp. 306-321.

1855. D'Udekem, J. Histoire naturelle du Tubifex des ruisseaux. Mem. Cour. et des Sav. Etr. Belg., XXVI, 38 pp.

1879. Eisen, G. Preliminary Report on Genera and Species of Tubificidae. Bih. K. Vet. Ak. Handl., V, No. 16, 26 pp.

1885. ——— Oligochaetological researches. Ann. Report Commissioner of Fish and Fisheries, Washington, pp. 879-964.

1894. Foot, K. Preliminary note on the Maturation and Fertilisation of the egg of Allolobophora foetida. Journ. of Morph., IX, No. 3.

1879. Fraisse, P. Ueber Spermatophoren bei Regenwurmern. Arbeit. Zool. Inst. of Wuerzburg, Bd. V, pp. 38-54, Pl. 4.

1895. Goodrich, E. S. On the structure of Vermiculus pilosus. Q.J.M.S., XXXVII (*n.s.*), p. 253.

1851. Grube, A. E. Die Familien der Anneliden mit Angabe ihrer Gattungen und Arten. Berlin, 164 pp. (Also in Arch. f. Nat., XVI.)

1884. Hammond, A. Some further researches on Tubifex. Journ. Micr. and Nat. Sci., III, pp. 147-155, Pl. XVI.

1906. Hargitt, Chas. W. Experiments on the behaviour of tubicolous Annelids. Jour. Exp. Zool., Vol. 3, pp. 295-320.

1899. Hatai, S. On Limnodrilus gotoi. Annot. Z. Japon, III, pp. 5-11.

1889. Hermann, F. Beiträge zur Histologie des Hodens. Arch. f. mikr. Anat., XXXIV.

1909. Hesse, Edmond. Quelques particularités de la spermatogenèse chez Oligochaeta. Arch. Zool. exper. (4), T. 10, pp. 441-446.

1842. Hoffmeister, W. De Vermibus quibusdam ad Genus Lumbricorum pertinentibus Dissertatio Berolini, 28 pp., 2 plates.

1909. Joseph, H. Die Amöbocyten von Lumbricus. Ein Beitrag zur Naturgeschichte der cellulären centren. Arb. Z. Inst. Wien, XVIII, pp. 1-60.

1906. Kammerer, P. Der Bachrohrenwurm Tubifex rivulorum (Lamarck). Wochenschr. Aquar. Terrar. Kunde. Jahrg. 3, pp. 467-468, 480-482, 491-492.

1816. Lamarck, J. Histoire naturelle des animaux sans vertèbres. 1st edition, Vol. III, p. 225.

1871. Lankester, E. R. Outlines of observations on the organisation of Oligochaetous Annelids. Ann. and Mag. Nat. Hist., Ser. IV, Vol. VII, pp. 90-101.

1869. ——— A Contribution to the knowledge of the lower Annelids. Trans. Linn. Soc., XXVI, pp. 631-646.

1871a. ——— On the structure and origin of the Spermatophores or Sperm ropes of two species of Tubifex. Q.J.M.S. (*n.s.*), XI, pp. 180-187, Pl. X and XI.

1906. Loeb, J. Dynamics of living matter. New York, p. 156.

1895. Matthews, A. Maturation, fertilisation and polarity in the Echinoderm egg. Journ. Morph., X.

1871. McIntosh, W. C. On some points in the structure of Tubifex. Tr. Roy. Soc. Edin., XXVI, pp. 253-267.

1908. Michaelsen, W. Zur Kenntniss der Tubificiden. Arch. Nat. Jahrg., 74 Bd., 129-162.

1908. Müller, C. Regenerationsversuche an Lumbriculus variegatus und Tubifex rivulorum. Arch. Entw. Meck. Bd. 26, pp. 209-277.

1773. Müller, O. F. Vermium Terrestrum et Fluviatilium acc. Historia Hafn et Lips.

1882. Nasse, D. Beiträge zur Anatomie der Tubificiden. Diss. Inaug. Bonn, 32 pp., 2 plates.

1913. Nomura, Ekitaro. On two species of aquatic Oligochaeta (Limnodrilus gotoi and L. willeyi). Journ. of Coll. of Science, Imperial Univ. of Tokyo, Vol. XXXV, Art. 4.

1875. Perrier, E. Sur le Tubifex umbellifer. Arch. Zool. Exp., IV, Notes pp. VI-VIII.

1903. Pierantoni, Umberto. L'Ovidutto e la emissione delle uova nei Tubificidi (Contributo alta biologia degli Oligochaete marini). Arch. Zool. Napoli, Vol. 1, pp. 108-119.

1893. Randolph, H. Ein Beitrag zur Kenntniss der Tubificiden. Vierteljahrschr. Nat. Ges. Zurich, XXXVII, pp. 145-147.

1869. Ratzel, F. Beiträge zur anat. und systematisch Kenntniss der Oligochaeten. Zeitschrift f. wiss. Zoologie, Bd. XVIII, pp. 563-591, Pl. 42.

1867. ——— Beiträge zur anat. von Enchytraeus vermicularis. Zeit. f. wiss. Zool., Bd. XVIII, pp. 105-109, Taf. 6, 7.

1902. Rice, Wm. J. Studies in Earthworm chloragogues. Biol. Bull. Woods Holl, III, pp. 88-94.

1878. Rolleston, G. The blood corpuscles of the Annelids. Journ. of Anat. and Physiol., Vol. XII, Pl. III, pp. 401-418.

1764. Schoeffer, J. Abhandlungen von Insecten Regenberg.

1906. Southern, R. Notes on the genus Enchytraeus with description of new species. Irish Naturalist, Vol. 15, pp. 179-185.

1909. ——— Contributions towards a Monograph of the British and Irish Oligochaeta. Proc. Roy. Irish Acad., Vol. XXVII, p. 119.

1913. Stephenson, J. On Intestinal Respiration in Annelids, with considerations on the origin and evolution of the Vascular System in that Group. Trans. Roy. Soc., Edin., Vol. XLIX, Part III (No. 14).

1909. ——— Report on a Collection of the smaller Oligochaeta made by Capt. F. H. Stewart, I.M.S., in Tibet. Rec. Ind. Mus., Calcutta, III, pp. 105-114.

1910. ——— On some littoral Oligochaeta of the Clyde. Trans. Roy. Soc. Ed., Vol. XLVII, Pt. 1 (No. 2).

1910a. ——— Studies on the aquatic Oligochaeta of the Punjab. Rec. Indian Mus., Vol. V, Part I.

1912. ——— On a collection of Oligochaeta mainly from Ceylon. Spolia Zeylanica, Vol. VIII, Pt. XXXII, p. 251.

1888. Stolc, A. Monographie Ceskych Tubificidü Morfologicka a systematicka studie. Abk. Bohm. Ges., Series VII, Vol. 2, No. 11, 45 pp.

1907. Szüto, Andor. Adatok az édesvízi csövágó féreg. (Tubifex tubifex). Mull kivalasztó szerveinck ismeretéhez. Állatt Kozlem. Kot. 6, pp. 31-36.

1884. Vejdovsky, F. System u. Morphologie der Oligochaeten. Prag., 166 pp., 16 plates.

1891. ——— Note sur une Tubifex d'Algerie. Mem. Soc. Zool. Fr., IV, pp. 596-603, Pl. XV.

1907. Watson, A. T. The habits of tube-building worms. Brit. Assoc. Rep., 76th meeting, p. 599.

1906. Wagner, F. Zur Oecologie des Tubifex und Lumbriculus. Zool. Jahrb. Abt. Syst., Bd. 23, p. 295.

1852. Williams, T. Blood proper and chylaqueous Fluid of Invertebrate animals. Philos. Trans., Pt. II.

1904. Wilson, E. The cell in Development and Inheritance.

EXPLANATION OF PLATES.

List of Reference Letters.

a. = Uncinate setae from ventral bundle.

ac. nul. = Accessory nucleolus attached to principal one.

ac. nul[1]. = Accessory nucleolus.

am. = Ampulla.

an. l. = Antero-lateral lobe.

an. r. = Annular ring of vas deferens.

ar. h. = "Arrow-head" extremity of spermatophore.

as. = Aster.

at. = Spermiducal gland.

ax. c. = Axial cylinder.

b. = Uncinate setae from dorsal bundle.

bl. = Blastophore.

br. = Brain.

bu. c. = Buccal cavity.

b. v. = Blood vessel.

b. v. i. = Blood vessel in intestinal wall.

b. w. = Body wall.

c. = Pectinate seta.

ca. f. = Caudal filament.

ca. s. = Capilliform setae.

c. c. = Chloragogen cells.

ce. = Cells of vas deferens.

chr. = Chromosomes.

cht. = Chromatin.

ci. = Cilia.

ci. f. = Ciliated funnel.

cl. = Clitellum.

cle. = Cleavage marks.

c. m. = Circular muscles.

co. s. = Connective tissue sheath.

ct. ep. = Ciliated epithelium.

cu. = Cuticle.

cu[1]. = Prolongation of cuticle into setigerous follicle.

cy. = Cytoplasm.

cy. m. = Undivided cytoplasmic mass to which oocytes are attached.

d. = Capilliform setae.

d. bl. = Dorsal seta bundle.

d. t. = Dorsal tubes or neurochord.

d. v. = Dorsal vessel.

e. = Subsidiary prongs.

eg. = Eggs.

e. m. = Opening of prostate into spermiducal gland.

epi. = Epispore.

f. = Fibres of nerve cord.

fb. s. = Fibrillar substance.

fl. = Flagellum.

g. c. = Gland cells of epidermis.

gr. m. = Granular mass.

h. = Large blackish-brown granules.

ht. = Heart.

hyp. = Epidermis.

i. = Smaller granules of chloragogen cells.

in. = Intestine.

in. c. = Interstitial cell of epidermis.

in. v. and *in. v*[1]. = Intestinal vessels.

le. = Large cells surrounding the lumen of the penis.

l. m. = Longitudinal muscles.

m. = Muscles from pharynx to body wall.

ma. = Matrix.

m. f. = Intrafollicular muscles.

mi. = Middle piece.

m. l. = Anterior median lobe.

mo. = Mouth.

m. p. = Parieto-vaginal muscles.

mu. = Muscular layer.

mu. l. = Muscular layer, circular inside, longitudinal outside.

n. = Nerve cord.

n. b. = Nerves to body wall.

n. c. = Nerve cells.

n. cd. = Non-ganglionated part of nerve cord.

n. cd^{1}. = Ganglionated part of nerve cord.

ne. = Nephridium.

n. f. c. = Nuclei of young follicle cells.

np. = Nephridiopore.

n. pr. = Nerve to prostomium.

ns. = Nephrostome.

n. s. f. = Nuclei of setigerous follicle.

nu. = Nucleus.

nu^{1}. = Nuclei in mitotic division.

nul. = Nucleolus.

nu. mem. = Nuclear membrane.

nu. sp. = Nuclear spindle.

oa. = Ovum.

ob. m. = Oblique muscles.

oc. = Oocyte (pear-shaped).

oc. y., oc. y^{1}. = Young and older oocytes respectively.

oe. = Oesophagus.

ov. = Ovary.

ovi. = Position of oviduct.

ovs. = Ovisac.

pe. = Penis.

pe. s. = Outer penis sheath.

pe. s^{1}. = Inner penis sheath.

ph. = Pharynx.

p. nul. = Principal nucleolus.

pp. c. = Peripharyngeal commissure.

pr. = Prostate.

pro. = Prostomium.

pr. t. = Preseptal part of nephridial tube.

ps. l. = Postero-lateral lobe.

pt. = Peritoneum.

pv. v. = Perivisceral vessel.

pv. v. 2 = Perivisceral vessel of segment 2.

pv. v. 3 = Perivisceral vessel of segment 3.

r. = Wall of nephrostome.

s. = Setae.

se. = Septum.

se. c. = Secretion of gland cells.

s. gl. = Septal glands.

s. f. = Setigerous follicle.

si. v. = Supra-intestinal vessel.

sm. t. = Tails of spermatozoa.

sm. h. = Heads of spermatozoa.

sp. = Spermatheca.

spc. = Spermatocyte.

sp. d. = Spermathecal duct.

sp. g. = Sub-pharyngeal ganglion.

sph. = Spermatophore.

spm. = Spermatids.

spo. = Spores.

sp. p. = Spermathecal pore.

sp. p^{1}. = Spermathecal pouch.

sp. s. = Sperm sac.

s. s. = Setigerous sac.

st. = Stalk.

t. = Contractile enlargement of nephridial tube.

te. = Position of the testes in the immature worm before the development of the sperm sac.

u. = Wall of ampulla.

va. ep. = Vacuolated epithelium.

v. bl. = Ventral seta bundle.

v. d. = Vas deferens.

v. d. 1 = Ciliated part of vas deferens.

v. d. 2 = Non-ciliated part of vas deferens.

v. pt. = Vesicular peritoneal cells.

v. v. = Ventral vessel.

w. cy. = Wall of cyst.

x. = Thin-walled coiled part of tube.

y. = Wider part of tube with vesicular peritoneal cells.

y. gr. = Yolk granules.

z. = Thick-walled tube leading to nephridiopore.

Plate I.

Figs. 1 to 3. Diagrammatic representation of the first eighteen segments of the body with the principal organs in each segment, as they appear in the living worm.

Plate II.

Fig. 4. Longitudinal section through the prostomium and first six segments of the body to show the anterior part of the alimentary canal and nervous system. × 160.

Fig. 5. Section through one of the dorsal setigerous sacs. × 510.

Fig. 6. Isolated setae from dorsal and ventral bundles. × 300.

Fig. 7. Transverse section through the first segment of the body, showing the buccal cavity and brain. × 200.

Fig. 8. Transverse section through the first segment, showing brain, peripharyngeal connectives and sub-pharyngeal ganglion. × 200.

Plate III.

Fig. 9. Transverse section through segment 8 in the region of the hearts. × 160.

Fig. 10. Transverse section through segment 11, showing the ovaries and one spermatheca. × 100.

Fig. 11. Transverse section through segment 14, showing the ovisac and sperm sac. × 90.

Fig. 12. Longitudinal section through the body wall of a mature worm in the region of the clitellum. × 520.

Fig. 13. Longitudinal section through the wall of the intestine, showing the details of its structure. × 370.

Fig. 14. Chloragogen cells in the living condition, drawn directly after their separation from the wall of the intestine. The nucleus is not shown, as it is very difficult to see in the fresh cells. × 1000.

Fig. 15. Chloragogen cells: the cells were separated from the wall of the intestine, fixed in Perenyi's fluid and stained with Brazilin on the slide. The irregularity in shape is due to the shrinkage. × 1000.

Plate IV.

Fig. 16. Longitudinal section through the reproductive segments 10-13 of a mature worm. One of the spermathecae (*sp.*) only is shown cut twice. It is much enlarged and extends into segment 12. The septa between the segments were not visible in the preparation. × 120.

Fig. 17. Longitudinal section through the ciliated funnel of a mature worm covered with spermatozoa, showing its structure and connection with the vas deferens. × 230.

Fig. 18. (A) Longitudinal section through the ciliated funnel of an immature worm. × 250.
(B) Transverse section of the same. × 250.

Fig. 19. (A) Transverse section of ciliated portion of vas deferens. × 800.
(B) Transverse section of non-ciliated portion of vas deferens. × 800.

Fig. 20. (A) Longitudinal section through ciliated portion of vas deferens. × 800.
(B) Longitudinal section through non-ciliated portion of vas deferens. × 800.

PLATE V.

Fig. 21. Longitudinal section of vas deferens. The section shows the transition from the ciliated to the non-ciliated part of the duct. × 800.

Fig. 22. Transverse section of the retracted penis. × 570.

Fig. 23. Transverse section of the spermathecal duct. × 360.

Fig. 24. An oocyte from the ovisac, undergoing maturation. × 160.

Fig. 25. An unusual form of the ovary which was liberated from the body and drawn in the living condition.

Fig. 26. Longitudinal section through a portion of the ovary, showing pear-shaped oocytes. × 480.

Fig. 27. Longitudinal section through a portion of the ovary, showing typical young and older oocytes, the latter just before their separation from the ovary. × 280.

Figs. 28-35. Stages in the development of the normal spermatozoa.

Fig. 28. Uninucleate spermatogonium from testis. × 820.

Fig. 29. Multinucleate spermatogonium from sperm sac. × 1720.

Fig. 30. Early stage in the formation of the spermatosphere, showing the first cleavage of the cytoplasm. × 1720.

Fig. 31. Formation of spermatosphere complete. Surface view of spermatosphere with spermatocytes. × 470.

Fig. 32. Section of spermatosphere with spermatocytes. × 470.

Fig. 33. A rather later stage. × 670.

Fig. 34. Formation of spermatids which are just beginning to elongate. × 610.

Fig. 35. Spermatozoa fully formed, but still attached to the blastophore. × 350.

Figs. 36-40. Stages in the development of "giant" spermatozoa.

Fig. 36. Formation of spermatocytes. × 660.

Fig. 37. Spermatids just beginning to elongate. × 660.

Plate VI.

Fig. 38. Later stage in the elongation of the spermatids. × 660.

Fig. 39. Immature "giant" spermatozoa. × 660.

Fig. 40. (A) Ordinary form of the spermatozoa. × 1710.

(B) "Giant" form of the spermatozoa. × 780.

Fig. 41. A figure drawn from a living specimen to show the arrangement of the blood vessels in the prostomium and first five segments of the body. The coiling of the perivisceral vessels is slightly simplified. × about 220.

Fig. 42. A figure drawn from a living specimen to show the arrangement of the blood vessels in an intestinal segment. × 180.

Fig. 43. A diagrammatic representation of a complete nephridium.

Fig. 44. A small portion of the nephridial tube covered with vesicular, peritoneal cells—drawn in the living condition.

Plate VII.

Fig. 45. Longitudinal section through the spermiducal gland, prostate and penis. × 110.

Fig. 46. Transverse section of the nerve cord. × 630.

Fig. 47. Longitudinal section of a nephrostome with the preseptal part of the nephridial tube, part of the body wall and septum. × 520.

Fig. 48. Longitudinal section of an ampulla of a nephridium. × 790.

Fig. 49. Spermathecae liberated from the body through a rupture in the body wall caused by pressure. × 50.

Fig. 50. External form of a very large spermatophore. × 55.

Fig. 51. Longitudinal section of part of a spermatophore. × 620.

Fig. 52. Transverse section of a spermatophore. × 620.

Fig. 53. (A) Section of a complete cyst of *Urospora saenuridis*. × 390.
(B) Detailed drawings of two of the spores. × 1900.

Fig. 54. General view of a cocoon with ripe eggs.

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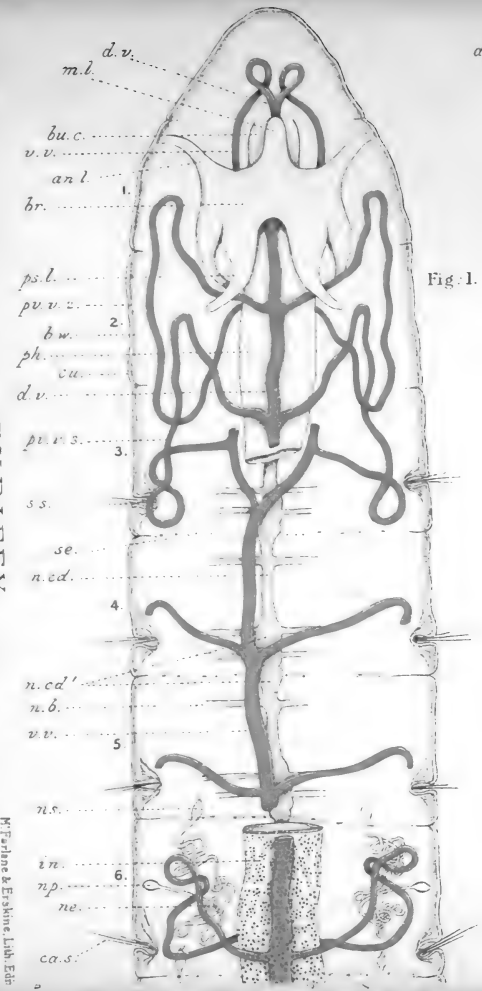

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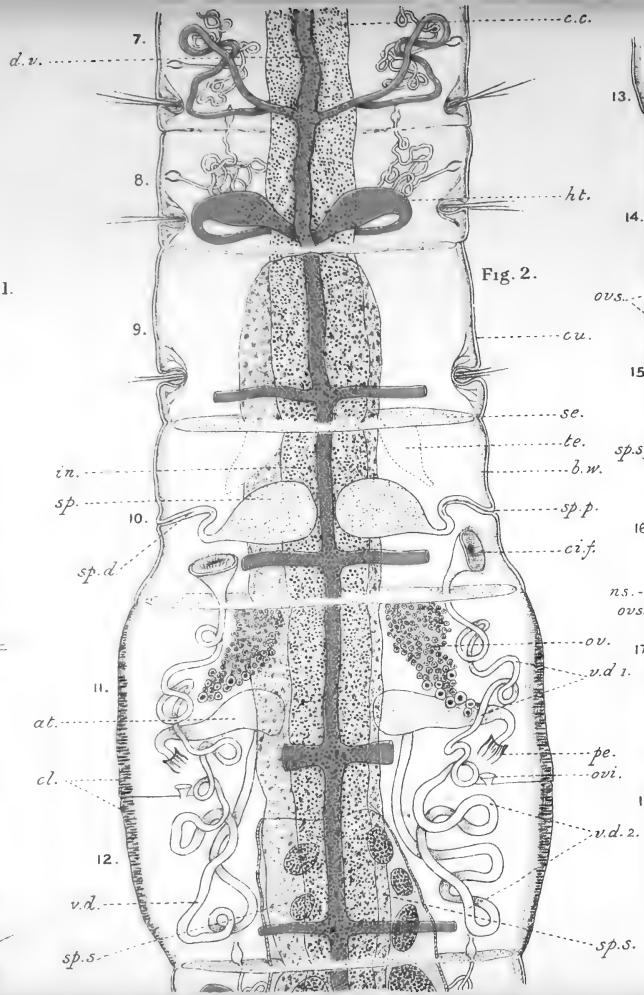

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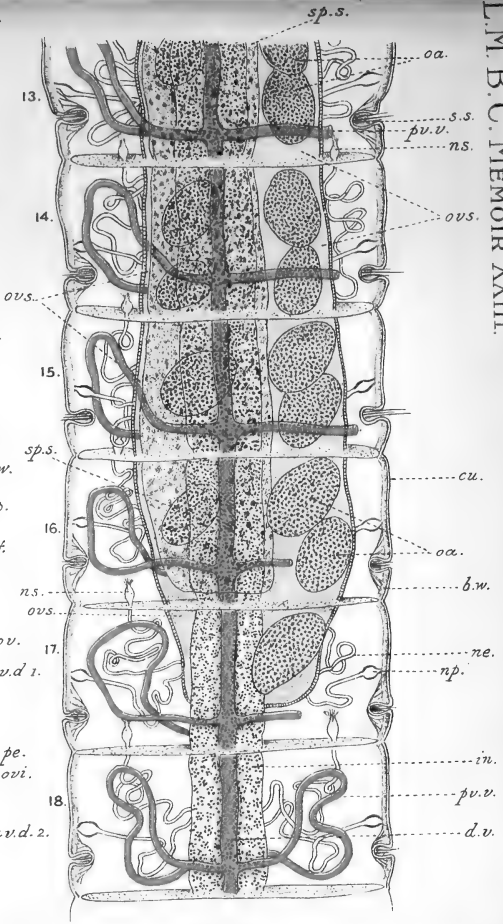

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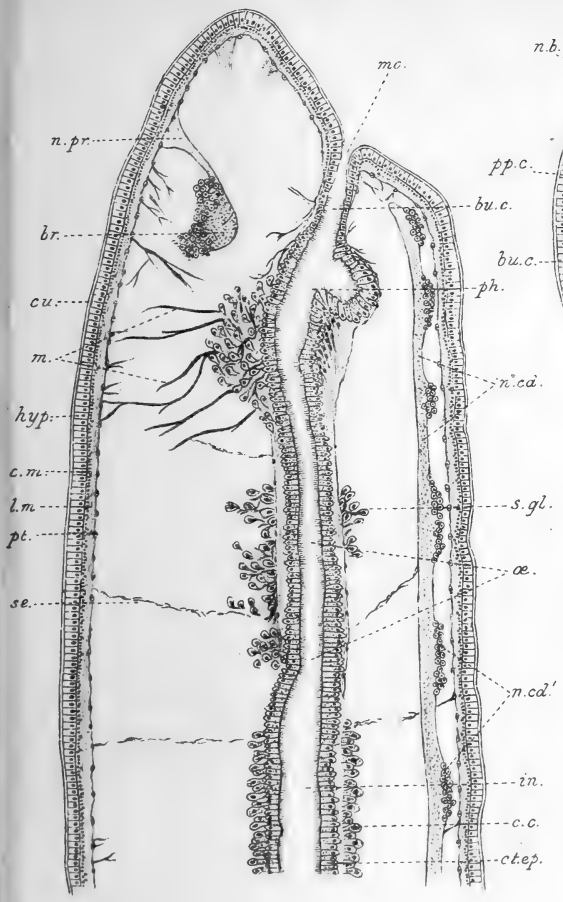

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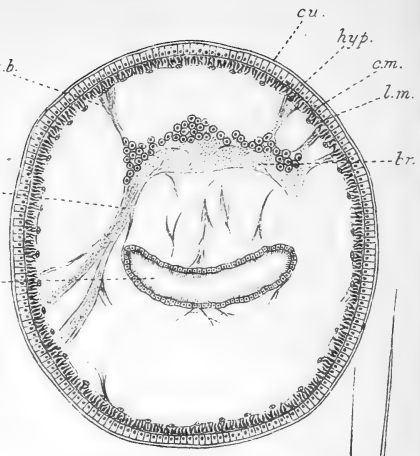

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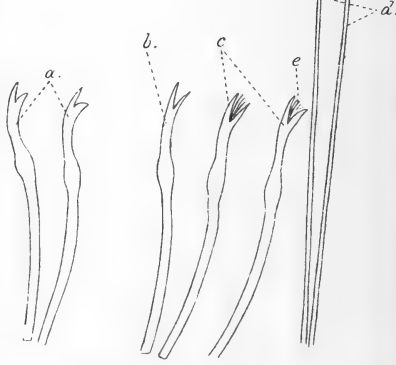

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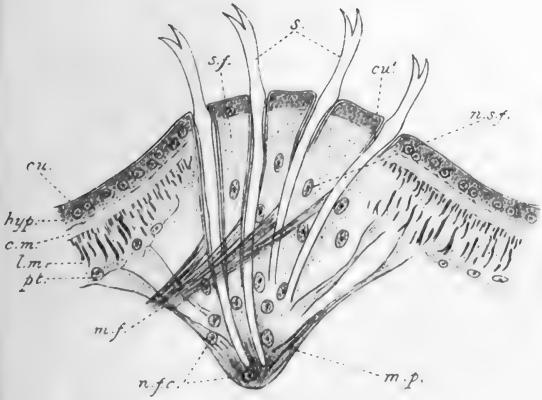

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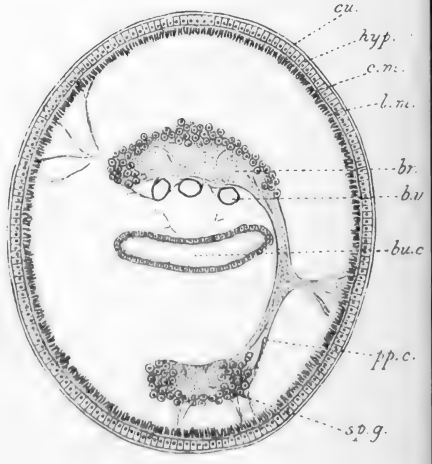

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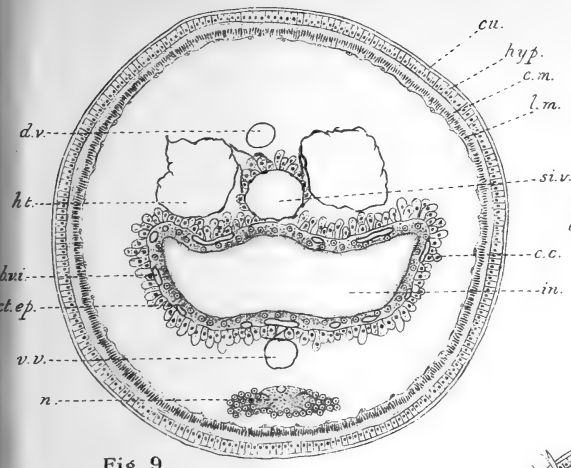

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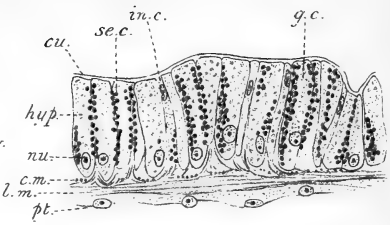

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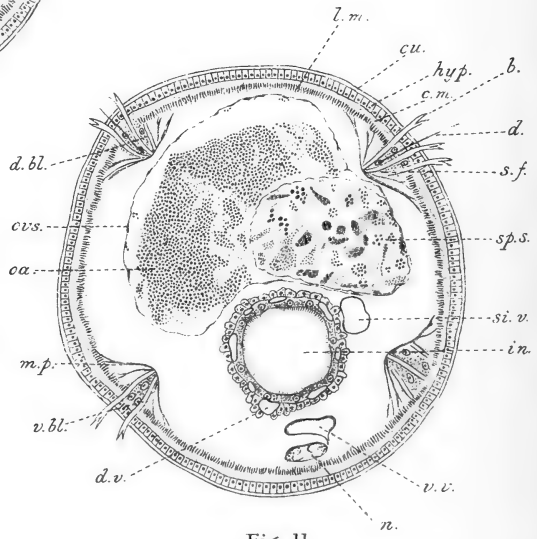

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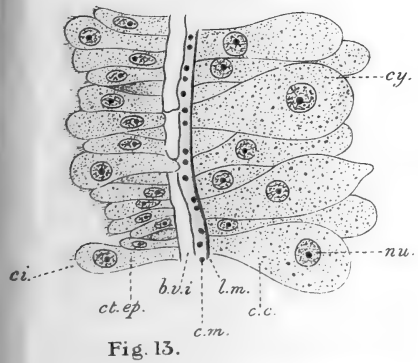

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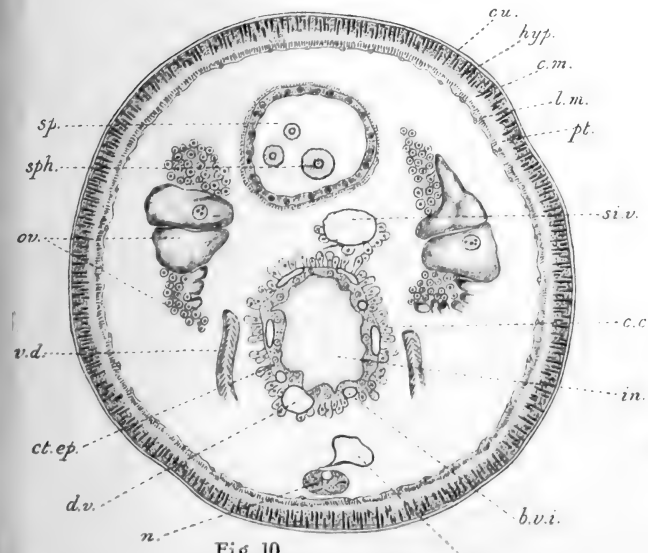

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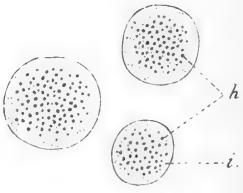

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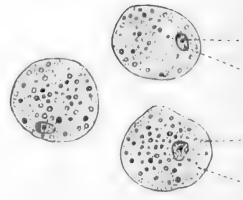

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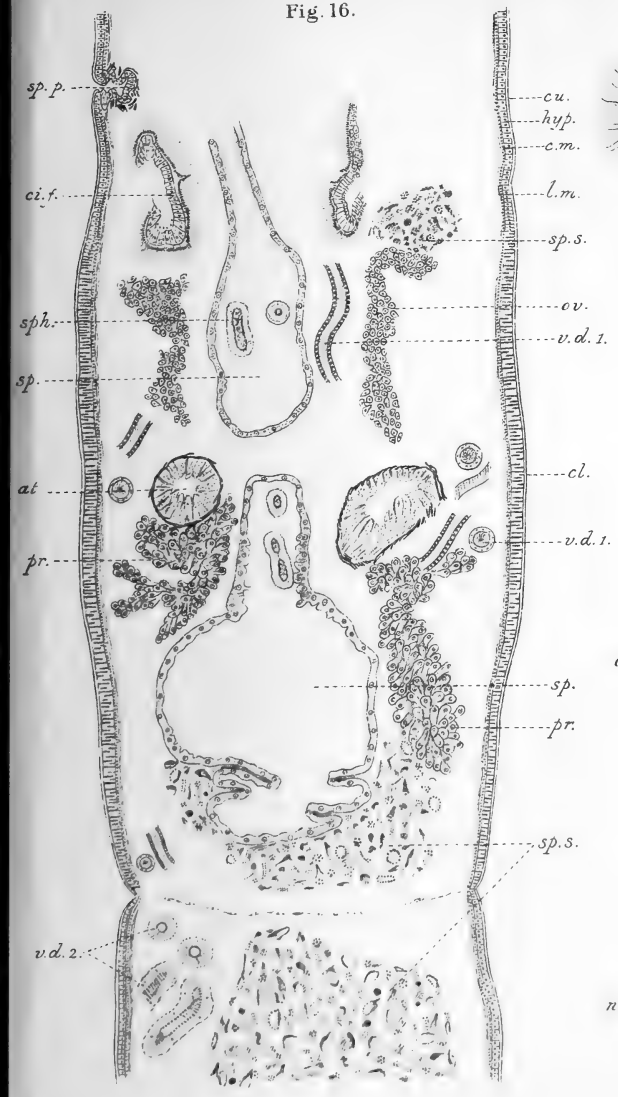

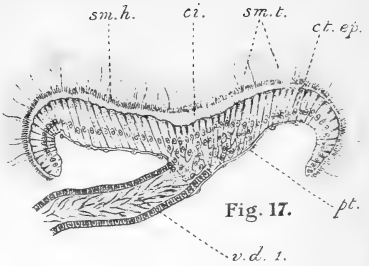

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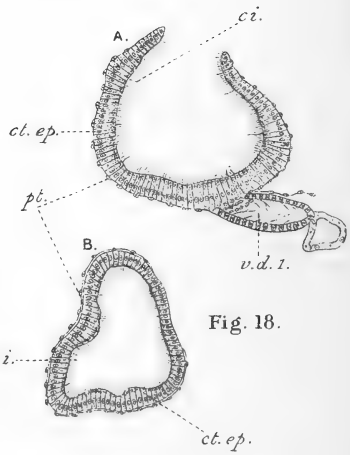

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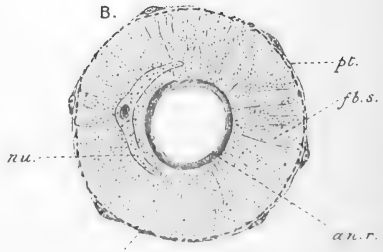

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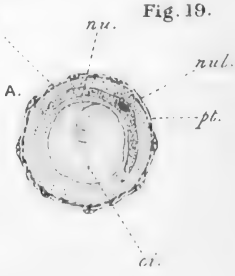

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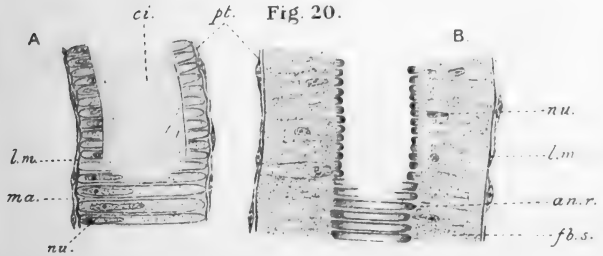

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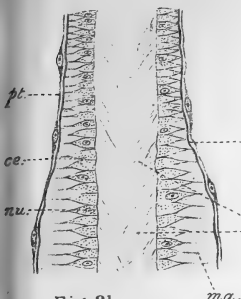

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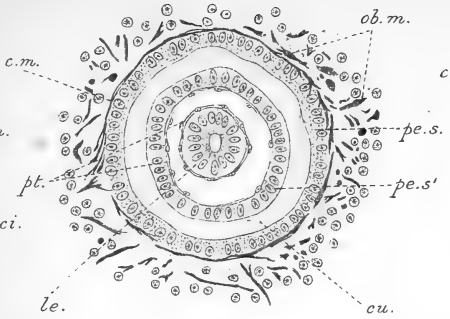

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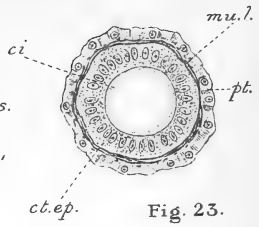

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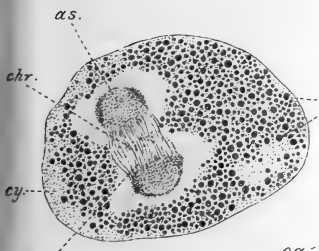

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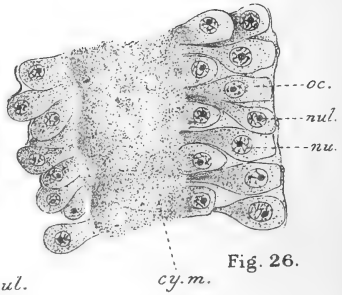

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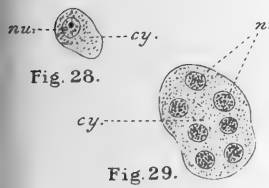

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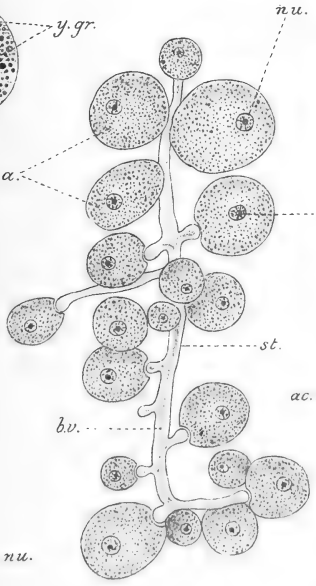

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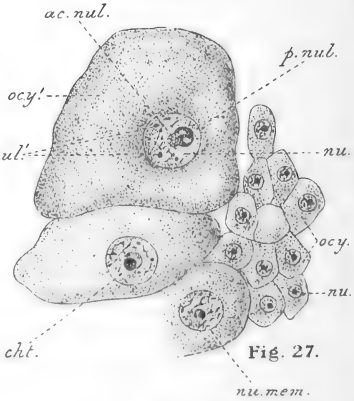
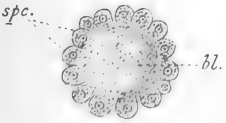


Fig. 27.

Fig. 30.

Fig. 31.



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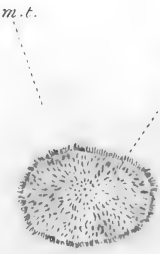

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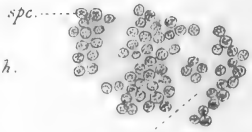

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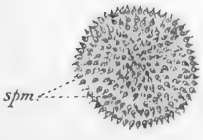

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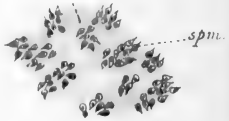

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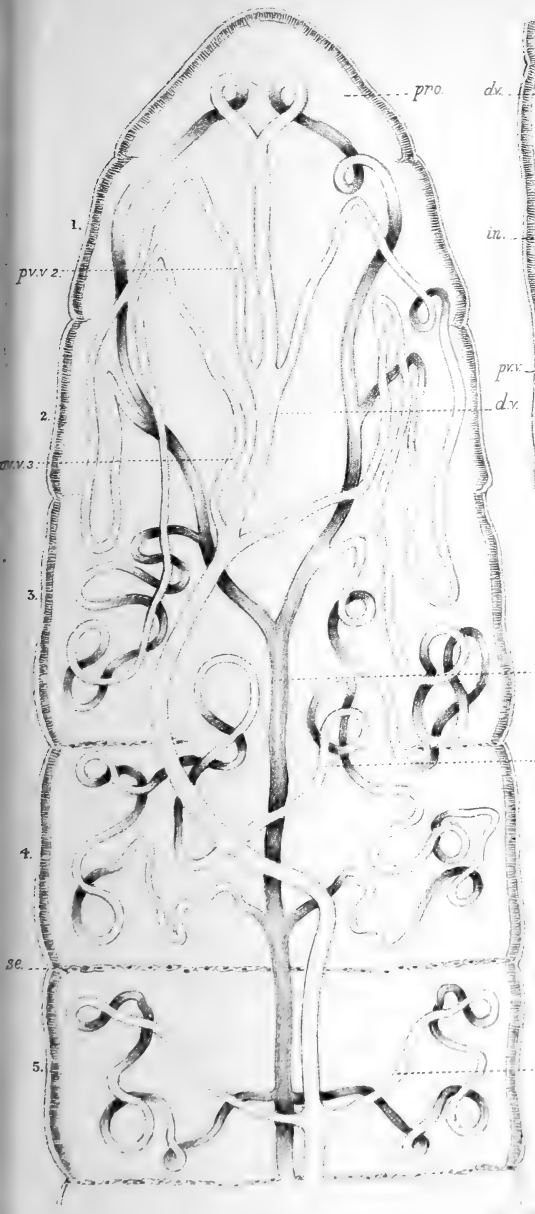

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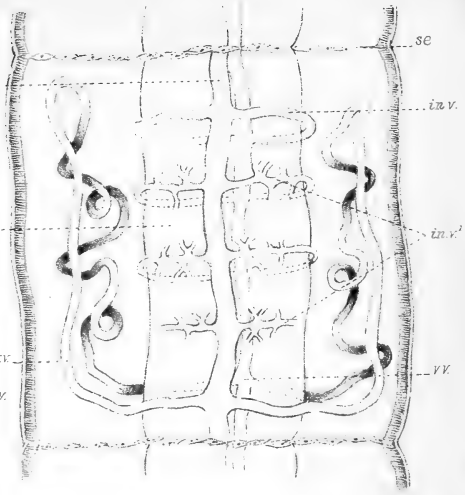

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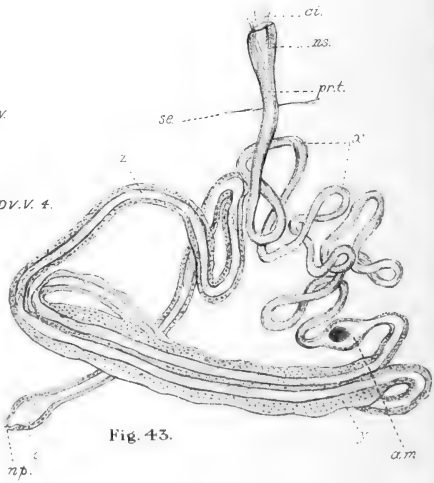

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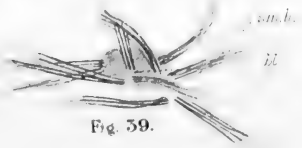

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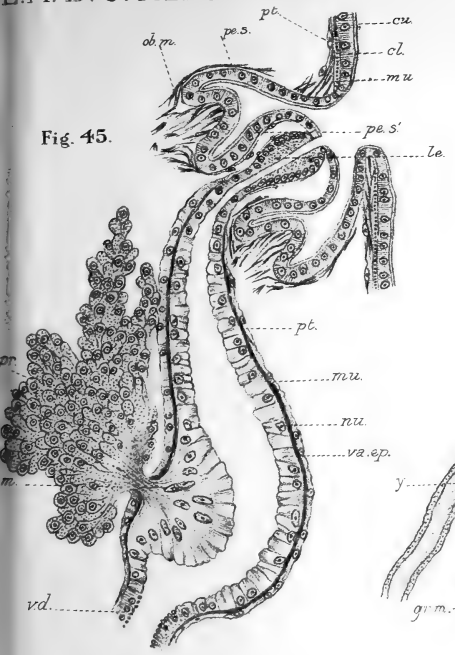

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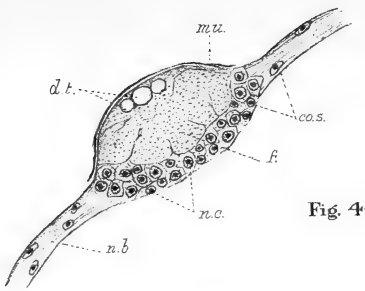

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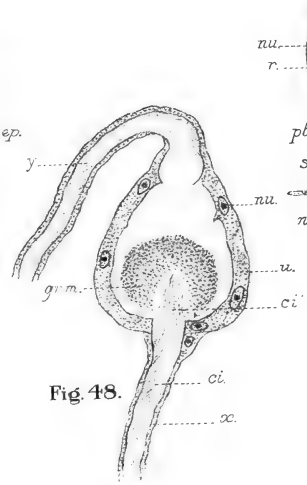

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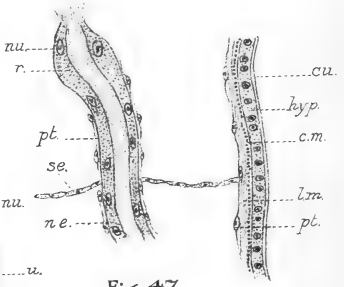


Fig. 47.

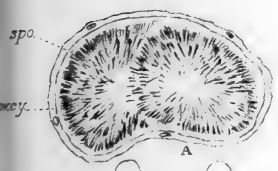
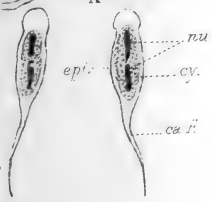


Fig. 53



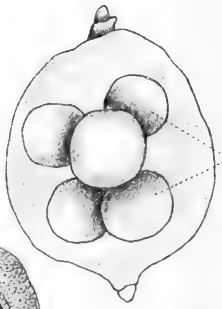


Fig. 54.

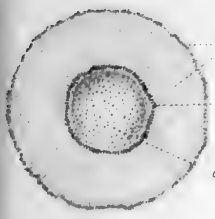


Fig. 52

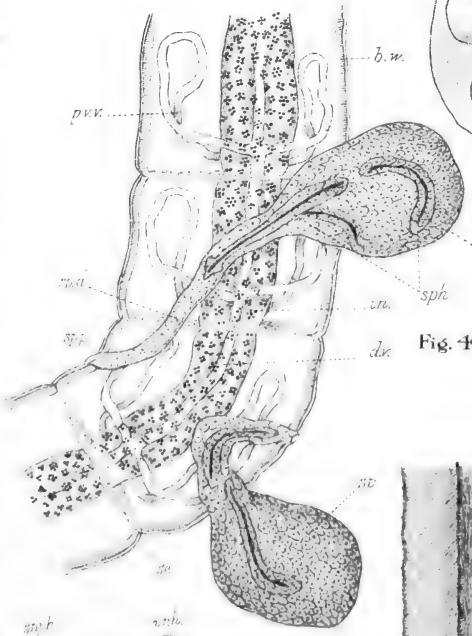


Fig. 49.

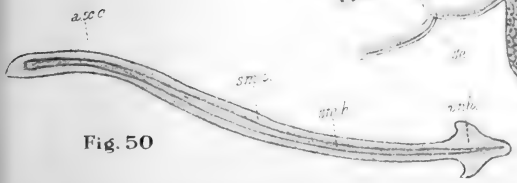


Fig. 50

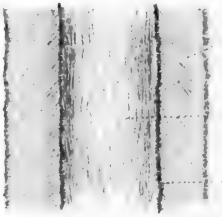


Fig. 51.