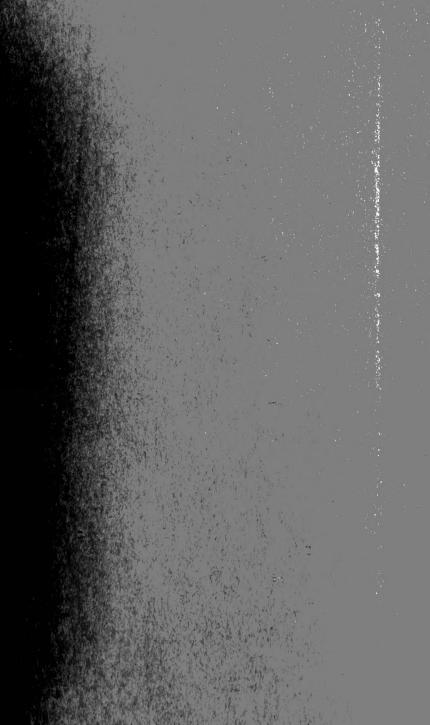
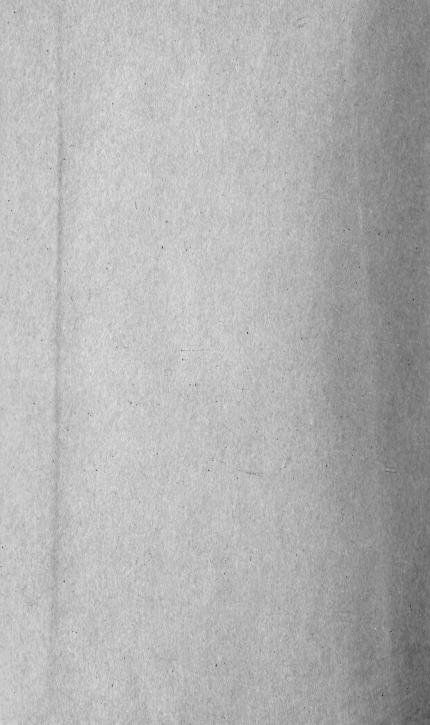
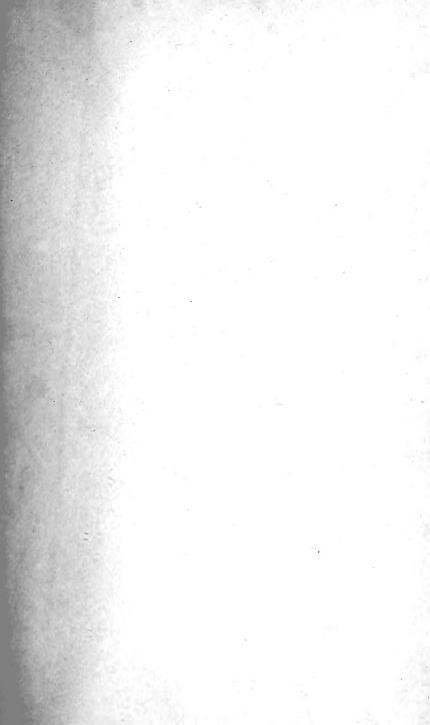
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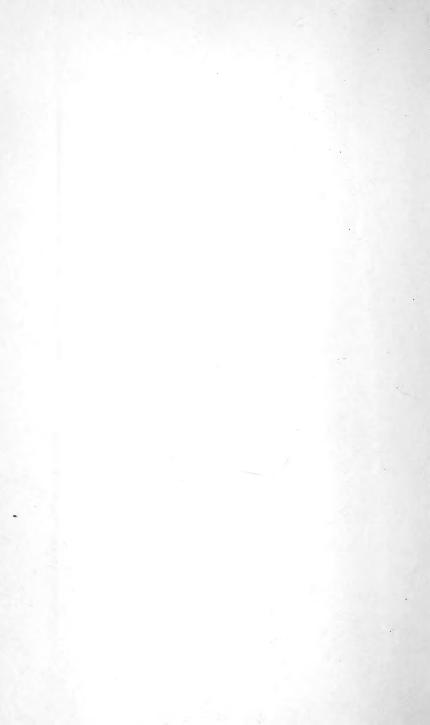
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## PROCEEDINGS

AND

# **TRANSACTIONS**

OF THE

# LIVERPOOL BIOLOGICAL SOCIETY.

VOL. XXXIII.

SESSION 1918-1919.



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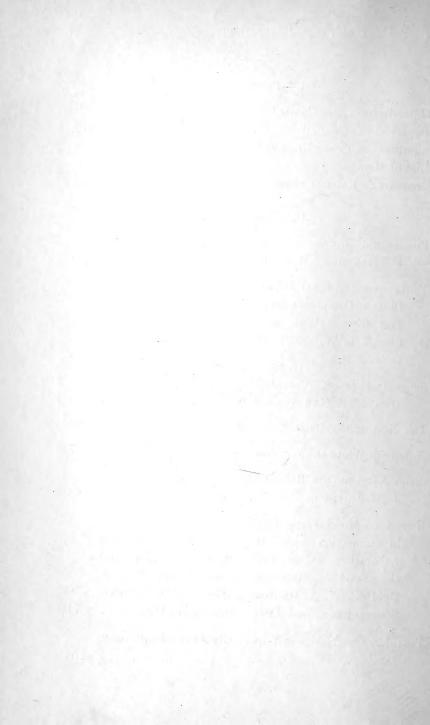
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# CONTENTS.

I.—Proceedings.	
	PAGE
Office-bearers and Council, 1918-1919	vii
Report of the Council	viii
Summary of Proceedings at the Meetings	ix
List of Members	xiii
Treasurer's Balance Sheet	xvi
II.—Transactions.	
Presidential Address—"Surface Films." By Prof. W. RAMSDEN, M.A., D.M.	3
Thirty-second Annual Report of the Liverpool Marine Biology Committee and their Biological Station at Port Erin. (With an Address upon "Periodic Change in Nature.") By Prof. W. A. HERDMAN, D.Sc., F.R.S.	25
Some Scale-Insects (Coccidæ) found in the Isle of Man. By Prof. R. Newstead, F.R.S.	63
The Nest of the Bank Vole. By George Ellison .	65
Note on a White Orkney Vole. By George Ellison	67
Some Notes on the Rats of the Port of Liverpool. By J. W. CUTMORE	68
Report on the Investigations carried on during 1918, in connection with the Lancashire Sea-Fisheries Laboratory, at the University of Liverpool, and the Sea-Fish Hatchery at Piel, near Barrow. By Prof. W. A. HERDMAN, D.Sc., F.R.S., ANDREW SCOTT, A.L.S., and JAMES JOHNSTONE, D.Sc.	71
Pearl-like Concretions in Tripe. By James Johnstone,	156



### 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. 1890—1891 T. J. MOORE, C.M.Z.S. 1891—1892 T. J. MOORE, C.M.Z.S.

1892—1893 ALFRED O. WALKER, J.P., F.L.S.

1893—1894 JOHN NEWTON, M.R.C.S. 1894—1895 Prof. F. GOTCH, M.A., F.R.S.

1894—1895 PROF. F. GOTCH, M.A., F.R.S.
1895—1896 PROF. R. J. HARVEY GIBSON, M.A.
1896—1897 HENRY O. FORBES, LL.D., F.Z.S.
1897—1898 ISAAC C. THOMPSON, F.L.S., F.R.M.S,
1898—1899 PROF. C. S. SHERRINGTON, M.D., F.R.S.
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.

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.

1914—1915 Prof. J. W. W. STEPHENS, M.D., D.P.H. 1915—1916 Prof. ERNEST GLYNN, M.A., M.D. 1916—1917 Prof. J. S. MACDONALD, L.R.C.P., F.R.S. 1917—1918 JOSEPH A. CLUBB, D.Sc.

### SESSION XXXIII, 1918-1919.

### President :

PROF. W. RAMSDEN, M.A., D.M.

### Vice-Presidents :

JOSEPH A. CLUBB, D.Sc. PROF. W. A. HERDMAN, D.Sc., F.R.S.

Mon. Treasurer: W. J. HALLS.

Mon. Librarian: MAY ALLEN, B.A.

Mon. Secretary:

W. RIMMER TEARE, A.C.P.

### Council:

R. C. BAMBER, M.Sc. (Miss). J. W. CUTMORE. G. ELLISON. W. T. HAYDON, F.L.S. J. JOHNSTONE, D.Sc. W. S. LAVEROCK, M.A., B.Sc. PROF. J. S. MACDONALD, B.A., F.R.S. PROF. R. NEWSTEAD, M.Sc., F.R.S. PROF. J. W. W. STEPHENS, M.D. EDWIN THOMPSON. EDWARD WHITLEY Mrs. GORDON WILSON

Representative of Students' Section: J. G. HAMILTON, B.Sc.

### REPORT of the COUNCIL.

During the Session 1918-19 there have been seven ordinary evening meetings. The field meeting, arranged for June 28th, had to be abandoned as a visit to Hilbre Island, in the weather then prevailing, was quite impossible. On the invitation of Dr. Clubb a visit to the Museum was substituted.

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.

The meeting on Feb. 14th was held in conjunction with the Students' Branch and Prof. Herdman gave a lecture on "The Iguanodons of Bernissart."

Owing to the difficulties created by the war it was impossible to secure a visiting lecturer, but the President filled the gap with a most interesting paper on "Vitamines in our Food."

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

The Treasurer's statement and balance sheet are appended.

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The members at pre	esent on	the roll	are as	follow	vs :—	
Ordinary members	•••	•••			•••	4
Associate members	•••	• • •		•••	•••	]
Student members, inc	luding	Students	s' Sect	tion, a	bout	6
						-
		Total	•••	• • •	•••	8

### SUMMARY of PROCEEDINGS at the MEETINGS.

The first meeting of the thirty-third session was held at the University, on Friday, October 18th, 1918.

In the unavoidable absence of Prof. Ramsden, the Retiring President (Joseph A. Clubb, D.Sc.) took the chair in the Zoology Theatre, during the earlier part of the proceedings.

- The Report of the Council on the Session 1917-1918 (see "Proceedings," Vol. XXXII, p. viii) was submitted and adopted.
- 2. The Treasurer's Balance Sheet for the Session 1917-1918 (see "Proceedings," Vol. XXXII, p. xvi) was submitted and approved.
- 3. The following Office-bearers and Council for the ensuing Session were elected:—Vice-Presidents, Joseph A. Clubb, D.Sc., Prof. Herdman, D.Sc., F.R.S.; Hon. Treasurer, W. J. Halls; Hon. Librarian, May Allen, B.A.; Hon. Secretary, W. Rimmer Teare, A.C.P.; Council, R. C. Bamber, M.Sc. (Miss), J. W. Cutmore, G. Ellison, W. T. Haydon, F.L.S., J. Johnstone, D.Sc., W. S. Laverock, M.A., B.Sc., Prof. J. S. Macdonald, F.R.S., Prof. R. Newstead, M.Sc., F.R.S., Prof. J. W. W. Stephens, M.D., Edwin Thompson, C.C., Edward Whitley and Mrs. Gordon Wilson.
- Prof. Ramsden delivered the Presidential Address on "Surface Films" (see Transactions, p. 3). A vote of thanks proposed by Dr. Caton, seconded by Prof. Macdonald, was passed.

The second meeting of the thirty-third session was held at the University, on Friday, November 8th, 1918. The President in the chair.

1. In the regrettable absence, through illness, of Prof. Herdman, Miss Bamber read the Annual Report which he had prepared on the work of the Liverpool Marine Biology Committee, and also the paper "On Periodic Change in Nature" (see Transactions, p. 39).

The third meeting of the thirty-third session was held at the University, on Friday, December 13th, 1918. The President in the chair.

- 1. Prof. Herdman exhibited and made remarks upon a Campodea found in the Strawberry.
- Prof. Newstead gave an account of Scale Insects generally with special details of specimens found in the Isle of Man (see Transactions, p. 63).

Miss Alwyn Evans contributed supplementary remarks to both papers.

The fourth meeting of the thirty-third session was held at the University, on Friday, January 10th, 1919. The President in the chair.

- Mr. G. Ellison exhibited, with remarks, some nests which
  he considered were those of the Bank Vole (see Trans.,
  p. 65).
- 2. Mr. J. W. Cutmore exhibited and discussed specimens of the various types of rat found in the Port of Liverpool (see Trans., p. 68).

3. Dr. Johnson exhibited and described a curious formation of the nature of a pearl found in the stomach wall of a cow (see Trans., p. 156).

The fifth meeting of the thirty-third session was held at the University, on Friday, February 14th, 1919, in conjunction with, and by invitation of, the Students' Section, the President of which, Miss C. M. Jarvis, occupied the chair.

- 1. Prof. Herdman gave an illustrated lecture on "The Iguanodons of Bernissart in the Brussels Museum," and informed his audience that no harm had befallen the specimens during the German occupation.
- A hearty vote of thanks to the lecturer was passed on the motion of Dr. Caton, seconded by Mr. Hewitt of the Liverpool Geological Society.

The sixth meeting of the thirty-third session was held at the University, on Friday, March 14th, 1919. Prof. Herdman occupying the chair in the absence of the President.

 Dr. Johnstone submitted the Annual Report of the Investigations carried on during 1918 in connection with the Lancashire Sea Fisheries Committee (see "Transactions," p. 71) and added an account of the history and development of British Fisheries.

The seventh meeting of the thirty-third session was held at the University, on Friday, May 19th, 1919. The President in the chair.

- Miss Allen exhibited a copy of Conrad Gesner's "Historia Naturalis Animalium," 1551-1587, and gave details of the life of Gesner.
- 2. The President gave an interesting and important illustrated address on "Vitamines in our Food."

The eighth meeting of the thirty-third session was held on Saturday, June 28th. A visit to Hilbre Island had been arranged, but owing to the inclement weather, this was abandoned, and, on Dr. Clubb's invitation, the members proceeded to the Museum. Here they were shown the preparations for new developments of an educational and popular character, and inspected the rat-breeding cages used by Mr. Cutmore in his experiments.

A short business meeting was held, and, on the motion of the President, Mr. Hugh Rathbone was unanimously elected President for the ensuing session.

### LIST of MEMBERS of the LIVERPOOL BIOLOGICAL SOCIETY.

### SESSION 1918-1919.

### A. ORDINARY MEMBERS.

(Life Members are marked with an asterisk.)

### ELECTED.

- 1908 Abram, Prof. J. Hill, 74, Rodney Street, Liverpool.
- 1919 Dr. J. G. Adami, F.R.S., Vice-Chancellor, The University, Liverpool.
- 1909 \*Allen, May, B.A., Hon. Librarian, University, Liverpool.
- 1918 Baldwin, Mrs., M.Sc., Zoology Dept., University, Liverpool.
- 1913 Beattie, Prof. J. M., M.A., M.D., The University, Liverpool.
- 1903 Booth, jun., Chas., 30, James Street, Liverpool.
- 1919 Boswell, Prof., P. G. H., The University, Liverpool.
- 1912 Burfield, S. T., B.A., Zoology Department, University, Liverpool.
- 1886 Caton, R., M.D., F.R.C.P., Holly Lea, Livingston Drive, Liverpool, S.
- 1886 Clubb, J. A., D.Sc., VICE-PRESIDENT, Free Public Museums, Liverpool.
- 1916 Dale, Sir Alfred, The University, Liverpool.
- 1917 Duvall, Miss H. M., B.Sc., Zoology Department, University, Liverpool.
- 1910 Ellison, George, 52, Serpentine Road, Wallasey.
- 1902 Glynn, Prof. Ernest, 67, Rodney Street.
- 1886 Halls, W. J., HON. TREASURER, 35, Lord Street.
- 1896 Haydon, W. T., F.L.S., 55, Grey Road, Walton.
- 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, Dr. A., Dowsefield, Allerton.
- 1903 Holt, Richard D., India Buildings, Liverpool.
- 1898 Johnstone, James, D.Sc., University, Liverpool.
- 1918 Jones, Philip, "Brantwood," St. Domingo Grove, Liverpool.
- 1896 Laverock, W. S., M.A., B.Sc., Free Public Museums, Liverpool.
- 1912 Macalister, C. J., M.D., F.R.C.P., 35, Rodney Street, Liverpool.
- 1915 Macdonald, Prof. J. S., B.A., F.R.S., The University, Liverpool.
- 1917 Milton, J. H., F.G.S., Merchant Taylors' School, Great Crosby.
- 1904 Newstead, Prof. R., M.Sc., F.R.S., University, Liverpool.
- 1913 Pallis, Mark, Tätoi, Aigburth Drive, Liverpool.
- 1903 Petrie, Sir Charles, 7, Devonshire Road, Liverpool.
- 1915 Prof. W. Ramsden, President, University, Liverpool.
- 1903 Rathbone, H. R., Oakwood, Elmswood Road, Aigburth.
- 1890 \*Rathbone, Miss May, Backwood, Neston.
- 1894 Scott, Andrew, A.L.S., Piel, Barrow-in-Furness.
- 1908 Share-Jones, J., D.Sc., F.R.C.V.S., University, Liverpool.
- 1886 Smith, Andrew T., "Solna," Croxteth Drive, Liverpool.
- 1903 Stapledon, W. C., "Annery," Caldy, West Kirby.
- 1913 Stephens, Prof. J. W. W., M.D., University, Liverpool.
- 1915 Teare, W. Rimmer, A.C.P., Hon. Secretary, 12, Bentley Road, Birkenhead.
- 1903 Thomas, Dr. Thelwall, 84, Rodney Street, Liverpool.
- 1905 Thompson, Edwin, "Woodlands," 13, Fulwood Park, Liverpool.
- 1889 Thornely, Miss L. R., Hawkshead, Ambleside.
- 1888 Toll, J. M., 49, Newsham Drive, Liverpool.
- 1918 Whitley, Edward, Bio-Chemical Laboratory, University.

### B. Associate Members.

- 1916 Atkin, Miss D., High School for Girls, Aigburth Vale, Liverpool.
- 1915 Bamber, Miss, M.Sc., Zoology Department, The University, Liverpool.
- 1905 Carstairs, Miss, 39, Lilley Road, Fairfield.
- 1914 Cutmore, J. W., Free Public Museum, Liverpool.
- 1918 Evans, Miss Alwyn M., M.Sc., School of Tropical Medicine, University, Liverpool.
- 1916 Gleave, Miss E. L., M.Sc., Oulton Secondary School, Clarence Street, Liverpool.
- 1905 Harrison, Oulton, 3, Montpellier Crescent, New Brighton.
- 1916 Horsman, Miss Elsie, B.Sc., 17, Hereford Road, Wavertree.
- 1919 Mayne, Miss C., B.Sc., 17, Laburnum Road, Fairfield.
- 1919 Sleggs, G. F., B.Sc., Zoology Dept., University, Liverpool.
- 1915 Stafford, Miss C. M. P., B.Sc., 312, Hawthorne Road, Bootle.
- 1917 Swift, Miss F., B.Sc., Queen Mary High School, Anfield.
- 1912 Wilson, Mrs. Gordon, High Schools for Girls, Aigburth Vale, Liverpool.

### C. University Students' Section.

President: J. G. Hamilton, B.Sc. Secretary: Miss M. Bowen (Contains about 30 members.)

### D. Honorary Members.

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

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

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

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

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

# THE LIVERPOOL BIOLOGICAL SOCIETY.

IN ACCOUNT WITH W. J. HALLS, HON. TREASURER.

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1918, Oct. 1st to Sept. 30th, 1919.	£ s. d.	1918, Oct. 1st to Sept. 30th. 1919.	o o
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LIVERPOOL, October 9th, 1919.

£125 4 % Debenture Stock (Commercial Cable Co.) at £73....£91 JOSEPH A. CLUBB.

### TRANSACTIONS

OF THE

# LIVERPOOL BIOLOGICAL SOCIETY.

CAMBON WED

### PRESIDENTIAL ADDRESS

ON

### SURFACE-FILMS

### By W. RAMSDEN, M.A., D.M.,

Professor of Bio-Chemistry at the University of Liverpool.

[Read to the Society, 18th October, 1918]

Some years ago I observed a very remarkable phenomenon in diluted white of egg, and it has seemed to me that it would be useful to the members of this Society to hear an account of it and of the observations to which it led, since this would serve as an introduction to the consideration of various surface-phenomena of much importance for the biological and medical sciences, and would also give me an opportunity to bring forward certain views which, although not found in the text-books, appear to me indispensable for the interpretation of the facts. I have not thought it necessary for the purpose of this address to indicate to what extent these views are novel.

### MECHANICAL SURFACE-AGGREGATES.

On pouring a dilute filtered solution of white of egg repeatedly to and fro from one test-tube into another a very extraordinary thing happens—numerous loose fibrin-like solid flakes develop in the liquid. They consist of coagulated protein, and are permanently insoluble in the mother-liquid. They are formed also when the solution is vigorously shaken up in a closed vessel, although they are then easily overlooked because they are mostly entangled in the froth. It is even possible by prolonged shaking of the solution to convert the whole of its coagulable protein into insoluble solid.

When the experiment is repeated with diluted blood-serum no such solid is visibly separated, but if the solution is first mixed with an equal volume of saturated ammonium sulphate solution and filtered, the filtrate gives threads and membranes like those obtained from white of egg, with the important difference, however, that they rapidly dissolve up again in the mother-liquid—protein molecules have in this case been simply aggregated into visible masses of solid, but no coagulation has taken place. They are presumably formed even in the absence of ammonium sulphate, but then dissolve up again so rapidly that they cannot be seen at all.

Microscopic observation of small air-bubbles in dilute egg albumin imprisoned between slide and cover-slip, and there repeatedly flattened out by pressure on the cover-slip, showed that similar solid structures were formed by this procedure, and strongly suggested that they were formed in some special connection with the air-surface of the bubbles.

This idea was confirmed by the next experiment. When a dilute solution of egg albumin is allowed to flow at an appropriate pace down the inside wall of a burette as a thin film of liquid lining its whole interior, a wrinkled membrane develops on its surface in the lower part of the burette, and this is slowly washed down and heaped up as a cake of solid protein on the surface of the liquid collecting at the bottom.

The next step was the discovery that the air surface of the protein solution, even when at rest and under conditions which entirely preclude evaporation of water, rapidly becomes rigid, and must therefore spontaneously coat itself with a thin membrane of solid protein. In this vessel containing egg-white diluted 100 times with water and filtered, are suspended two magnetised bits of watch-

spring, one on the surface, the other deeply submerged—observe that, when a magnet is brought near, the latter is freely mobile but the former is rigidly fixed. A simpler way of observing this surface rigidity is to compare drops of albumin solution and of water on a glass plate when both are lightly dusted with sulphur—the sulphur slides down the slope of the water drop and leaves the vertex clear, but on the albumin solution it is instantly fixed just where it falls.

The mystery was by these experiments to some extent solved. In this surface-rigidity of egg-albumin I had re-discovered a little-known fact described long previously by Plateau, and it became clear that the solid separated out when its solutions were shaken consisted simply of rolled-up and contorted membranes which had originally formed spontaneously at the water-air surfaces, in some cases coagulated, in others apparently unchanged.

Applying the burette method to a wide range of other solutions, mostly colloid (including amongst them solution of the glucoside saponin which Plateau had described as having rigid air surfaces), solid "mechanical surface aggregates," as they may be termed, were obtained from every protein solution tested, and also from numerous soaps, aniline dyes, quinine (the free alkaloid), saponin, methyl orange in neutral solution, phenol-phthalein in neutral or acid solution, and from suspensions of gamboge, mastic, resin, and sulphur. From bile-salt and sodium oleate only viscid gums were obtained. Colloidal silica, quinine salts, and phenol-phthalein in alkaline solution gave no aggregates.

Testing the mobility of the air surface of each of the above solutions it was found that all except bile-salts and sodium oleate developed rapidly, or only on standing, either a true rigidity or a marked viscosity in the Plateau

sense (i.e., a floating magnet brought into the East-West position took a very much longer time to return to the North-South position than in pure water), although the viscosity of the solutions remote from the surface differed very little from that of water. Bile-salts remained permanently mobile. Sodium oleate gave varying results, possibly due to impurity or chemical change, and requires further investigation.

When the surfaces are rigid it is clear that solid particles previously in solution are present there in sufficiently close contact to exhibit the properties peculiar to matter in the solid state. When the surface exhibits "Plateau viscosity," this condition would be adequately explained by the assumption that particles are present on the surface which lower its tension in some rough proportion to their number on a given area, and that the rotating magnet sweeps them up in greater density immediately in front of it and leaves the area behind it for a short time comparatively empty. If no true increase of surface viscosity accompanies the Plateau viscosity (and experimental difficulties make it almost impossible to decide this point), it must be concluded that the particles are either liquid, or are solids everywhere surrounded and isolated by intervening liquid. The complete absence of rigidity or "Plateau viscosity" on solution of bile-salts I must leave unexplained. The possibility of obtaining mechanical surface aggregates by so simple a sweeping-up process as that employed in the burette method is only intelligible on the assumption that the air surface of solutions which yield solid mechanical aggregates, no matter whether showing rigidity or Plateau viscosity or an apparently normal limpidity, becomes more or less thickly studded with relatively large solid particles which have passed spontaneously out of solution in so far that they have come into contact with both air and water.

That the remarkable property which some solutions possess of yielding more or less durable bubbles and froths when shaken was in some way connected with the presence of such particles was an obvious possibility. On testing the various solutions which had yielded mechanical surface aggregates, I found that by far the greater number of them were conspicuously capable of yielding durable thin films in air. Conversely, on testing every solution I could come across which had exceptional frothing power, it was found that all yielded solid mechanical surface aggregates, with the exception of bile-salts which yielded only viscous gummy matter, and sodium oleate which gave inconstant results. These facts strongly suggest that the ability to form good bubbles is essentially bound up with the presence of large particles at the free surfaces. When air is sucked out of saponin bubbles the surface solid actually jams together sideways (or folds up) into solid rods almost visible to the naked eye-simultaneously, as you see, the film loses nearly all its contractility and takes several seconds to re-dissolve the surface excess of saponin and recover its original spherical shape.

Taking then the studding of the air surface of most of these solutions with solid particles as an established fact, the question arises why do certain substances freely soluble in water pass spontaneously out of solution at a water-air surface? What explanation can be offered of a phenomenon so remarkable? The beginnings of an explanation were found ready to hand in the work of Willard Gibbs in the United States, and of J. J. Thomson in England. These workers had shown by reasoning from general principles of energetics that any change in the disposition of the molecules close to a surface which would result in a lowering of the surface-tension was one which would tend to take place. This deduction is commonly known as the principle

of minimal surface-energy. Among the conclusions to which it leads are the following:

- 1. Dissolved substances which lower the tension at a given surface will be more concentrated near it than in regions remote from it, and those which raise the surface-tension will tend to migrate from that surface so that there will be less there.
- 2. Chemical or physico-chemical changes which result in products lowering the tension at a given surface will tend to proceed further and take place at a faster rate there than in regions remote from that surface, and those which raise the tension will be diminished and slower. Among physico-chemical changes may be specially mentioned hydrolysis and ionisation.

On testing the various substances which in watery solution had yielded mechanical surface aggregates, it was found that all of them did lower the tension of the air surface of water, and therefore fulfilled the condition necessary to explain their surface concentration on the Gibbs-Thomson principle. That principle does not by itself suffice to explain the minute details of the observed phenomena; it does not explain why soluble substances pass actually out of solution nor, as Professor Lewis has shewn, account quantitatively for their concentration, but at least it takes us a long way in the right direction. And it offers many possibilities for explaining the fact that in some cases the mechanical surface aggregates are chemically different from the original substance in solution, as for example with egg albumin.

It was further clear that on the Gibbs-Thomson principle increased concentrations of appropriate initially dissolved substances were also to be expected at the interfaces between two immiscible liquids. It was in many cases very easy to demonstrate that abnormal rigidity or Plateau

viscosity developed at such interfaces on adding a solution of a suitable solid to one or other of the pure liquids, and also to show that there was a lessening of the surface-tension of the interfaces. It was obvious that such interface-concentrations and solid membranes must profoundly influence the durability of emulsions made of the two liquids, and it was found that they were in many cases capable of rendering the emulsions permanent, and also were invariably present when emulsions with large droplets were permanent.

In bubble-films it was clear that each of the two free surfaces would acquire its excess of "surface coating," and therefore that such films must have something like a sandwich structure.

### CONTRACTILE SURFACE-FILMS.

The explanation of the surface phenomena you have seen being thus bound up with "surface tension," let us consider briefly what is meant by the term. Everyone knows that a small mass of liquid in air or other gas tends to assume a spherical shape. This tendency might conceivably be explained in more than one way, but there is good evidence that actually it is due to the very thin stratum of liquid nearest the free (i.e., gas or air) surface, and only this stratum, being in a stretched condition, always trying to contract, i.e., always in a state of tension. A striking way of observing this is to see what happens when the end of a glass rod moistened with soap solution, or other watery solution with a much lower surface tension than water, is brought into contact with the surface of clean water previously lightly dusted with sulphur; instantly the surface stratum of the water contracts away from the glass rod, incidentally carrying the sulphur away with it to the side of the vessel,—there is a tug-of-war in which the soapcontaminated surface near the rod, with its lower tension, is hopelessly outclassed by the clean water surface with its higher tension, and is therefore rapidly pulled over the whole surface. Observe that no such rapid pulling outwards of a coloured soap solution occurs when it is introduced down the inside of a glass tube directly into the depths of the water—it occurs only in the surface layer.

Every liquid has its free surface layer down to a very small depth from the surface in a similar contractile condition, and the force which it exerts, i.e., the surface-tension, can be measured in many ingenious ways. Its magnitude has been found to differ for different liquids, and it is always lower in the warm liquid than in the cold. The addition of a soluble substance in some cases increases it, e.g., sodium chloride in water; in other cases, as you have seen, diminishes it, e.g., soap or proteins in water.

If we ask how the free surface-stratum of liquid comes to possess this contractility, we must concern ourselves at once with molecules and intermolecular forces of which our present knowledge is very inadequate. Arguing purely a priori on the basis of that knowledge it would be difficult to deduce the conclusion that the free surface stratum must necessarily be in a contractile state. Even when fortified by the knowledge that it invariably is in such a state it is at present impossible to arrive at any clear explanation of the molecular arrangements which give rise to it. It will nevertheless be profitable for other reasons to proceed as if we had such an explanation in view.

Of molecules it is safe to assume that they must often have other than a spherical shape. Of intermolecular forces it is certain that (1) they consist of attractions and repulsions, (2) they are of enormous magnitude when two molecules are very close together, but become negligible when the molecules are more than a certain minute distance apart, a distance reckoned from the centre of the molecules and known as the range of intermolecular forces (frequently represented by the symbol z), (3) they can rarely be equal along all radii between the centres of two molecules, in which case rotation of the molecules round their centres must bring about alterations in the intensity of the intermolecular forces operative between them. It is safe also to assume that (4) they increase greatly in intensity as two molecules are brought nearer together than the distance at which they first begin to be operative, and that the repulsions increase at a much greater rate than do the attractions, average equilibrium being attained when the average intermolecular distance is such that both are equal, (5) the range of the attractions extends over several molecular diameters.

Applying these assumptions to the case of pure liquid in contact with gas, and dealing with the system for convenience as if its molecules were in static equilibrium, it is clear that every molecule remote from the surface has its entire field of force occupied by similar molecules, but that every molecule in the surface stratum down to a depth z has some gas molecules within range in place of liquid, and as each of these 'occupies' a very much larger average volume than does a liquid molecule, those within range must be comparatively few; hence the intermolecular forces operative on the surface-stratum must come predominantly from the liquid, i.e., from the "cis" side of the surface. It will be clear also that one result of this specific direction of the predominant intermolecular forces must be that equilibrium could only be attained by correspondingly specific molecular spacing and strains different from those obtaining remote from the surface, and that special intramolecular strains between the atoms composing the molecules are also likely to be produced. What this spacing is and how it gives rise to a contractile state along the surface it is impossible to

say: it must suffice to note that at a free surface as a matter of observed fact it invariably is accompanied by contractility, and it is difficult to resist the conclusion that the contractile state is essentially bound up with the fact that the molecular forces operating on the surface-stratum from the "cis" side are larger than those operating on it from the "trans" side of the surface.

It will further be clear that if the surface molecules exert different attractions and repulsions along different radii they must necessarily, when exposed to the predominantly one-sided intermolecular forces, suffer rotation until they become appropriately orientated. Since such orientation must make a considerable difference to the strains in the surface-layer, and therefore to the surface-tension, the same conclusion can be drawn also from the Gibbs-Thomson principle of minimal surface energy. It should be noted also that as it can rarely happen that the intermolecular forces operative between the molecules of a solvent and of a dissolved substance are exactly equal to those between the molecules of the solvent itself, any dissolved substance present in the surface-stratum must also be specifically spaced, and if its molecules have different fields of force along different radii must also be specifically orientated.

The first suggestion of molecular orientation at a surface is due to Hardy of Cambridge, and the idea has been considerably developed by Irving Langmuir in U.S.A. with fascinating and important results. Let me point out two conclusions which follow from such orientation:

1. The rate at which a chemical reaction takes place in a surface stratum would be modified, since the mathematical chances of two reacting molecules encountering each other, so to speak "business end on" would be different at the surface and remote from it,—modified therefore in a way not taken into account by the Gibbs-Thomson principle.

2. Specific direction must be given to some chemical interactions, i.e., a preference to one out of many possible interactions, when one or both of the reacting substances are orientated, as also when specific intramolecular strains are set up by the specially directed intermolecular forces.

It follows from the above, and the reasoning is obviously equally applicable to liquid-solid and liquid-liquid interfaces, that all surfaces must tend to act catalysts. It is presumably in this direction that we must look for the explanation of the action of certain enzymes studied by Bayliss, which work even when in coarse suspension (e.g., urease, lipase). In connection with this question it is perhaps worth pointing out that since chemical affinities are essentially intermolecular or interatomic attractions they must play an important part in determining the molecular strains at a surface, and may well do so in the highly specific way manifest in many enzyme actions, as also in adsorptions. And since spatial arrangements are of fundamental importance for the strains in a surface film, the same must sometimes be true of stereo-chemical structure.

### EXPANSILE SURFACE-FILMS.

Thus far we have considered only the simple case of a free surface-stratum, a case in which the "trans" surface intermolecular forces are relatively slight. Let us turn to a liquid surface-stratum in contact with an insoluble solid, a case in which the "trans" forces may be considerable and may conceivably exceed those on the "cis" side of the surface. What does observation teach us concerning the conditions in the liquid under these circumstances? It is impossible to measure the tension in the liquid surface or to ascertain directly whether it is even contractile at all. But

by reason of the fact that the solid is rigid, and therefore itself incapable of internal movement, it is possible to draw certain highly probable conclusions concerning the liquid surface from the movements seen when completely submerged masses are brought near together.

Observation of fine solid particles suspended in liquids, and therefore in an environment where they are subject to numerous accidental approximations caused by currents and Brownian movements, shows two different types of behaviour. In some cases, e.g., kaolin in water containing lime salts, the particles run together; in other cases they remain obstinately separate, or at all events show no obvious tendency to run together, e.g., kaolin in pure water. I shall argue that in the former case the surface-stratum of the water must be actively contractile, but that in the latter it is probably, or at least possibly, actively expansile (neutrality or complete absence of surface-tension being excluded as impossible at a surface between two immissible substances).

Assuming for the sake of argument that expansile strata really exist,\* it is clear that whatever the strains set up in the two surface-strata of the liquid film separating two solid masses, whether they evoke a contractile or an expansile condition, those strains must begin to diminish as soon as the liquid is thinned below 2z so that some parts of it come within range of the solid on both sides and the molecular stresses become to that extent more equalised; consequently, either a contractility or an expansility would diminish. Diminution of the former would result in withdrawal of the liquid, since it would be no longer capable of withstanding the superior pull of the surrounding thicker films, and spontaneous approximation of the solid masses would follow; diminution in an expansility would result in

<sup>\*</sup> Compare Clerk Maxwell, Encycl. Bri., 9th edn., Art. Capillarity, and Wo. Ostwald's Handbook of Colloid Chemistry. English Translation. 1915. J. and A. Churchill.

inability of the thinned film to resist the superior sidethrust of the surrounding thicker films, so that liquid would be pushed in between the solids and cause their spontaneous separation. I must not conceal the probability, however, that the above reasoning does not convey a complete picture of the tension changes in a thinning liquid film. Such a picture is impossible without greater knowledge of the relative ranges and changes of inter-molecular attractions and repulsions with distance than is available. The one I have sketched is submitted as a working hypothesis which contains much relative truth and demands the recognition of expansile surface strata or 'negative tensions.'

Phenomena showing contractile liquid films are of daily occurrence in laboratory work. Spontaneous approximation of suspended solids is seen, for example, in the flocculation of many suspensions, in certain colloid precipitations, in the formation of rouleaux by red blood corpuscles, and in the agglutination or "clumping" of bacteria first described by Gruber and Durham and utilised for the diagnosis of typhoid fever in the well-known Widal test. Spontaneous dispersal of clumped particles of insoluble solids (presumably by negative surface tensions) may be seen on adding appropriate electrolytes to the water in which they are suspended, e.g., on adding NaOHto kaolin clumped by CaCl2. The forcible imbibition of water by many insoluble colloids may also be instanced as a phenomenon demanding a belief in the real existence of negative surface tensions. Probably the process of dissolving solids which yield colloid solutions is, in its earlier stages at least, essentially of the same nature also. As regards the electric charges shown by Hardy to be acquired or lost in the dispersals and agglutinations of suspended solids, it is assumed that, although they often play an essential part, they are only indirectly responsible for the movements and that for the most part they operate as they appear to do in the capillary electrometer, by the changes which they occasion in the surface-tensions concerned.

The existence of negative surface tensions is shown much more clearly however by some of the phenomena of capillarity. It is a familiar fact that when a capillary tube of insoluble solid is dipped vertically into a liquid that liquid either creeps up to a higher level inside the tube, e.g., water in a glass capillary, or ceases to rise before it reaches the level of the outside liquid, e.g., water in a greasy tube, or mercury in a clean glass capillary. In the former case the liquid is said to "wet" the solid and the liquid-air interface meets the solid-liquid interface at an acute angle; in the latter case the "angle of contact" is obtuse. Similarly with a plane solid plate the two surfaces meet at a definite "angle of contact," the magnitude of which is characteristic for the particular solid and liquid and is either acute or obtuse.

How is this difference in the equilibrium position of the same liquid with different solids or different liquids with the same solid to be accounted for? It is clear that ultimately it must depend upon the different magnitudes (and possibly different ranges) of the inter-molecular forces operative between the liquid stratum of thickness z and the solid within range across the surface and, since intermolecular repulsions are only secondarily brought into operation as a consequence of precedent inter-molecular attractions, that it must depend primarily upon the different magnitudes of the attractions. It is clear also on general principles of energetics that in any material system the stronger inter-molecular attractions will tend to satisfy themselves and therefore that the stronger the attractions between the solid and the liquid, the stronger would be the tendency for the solid and the liquid to acquire the maximum possible area of contact. We should therefore on a principle of minimal potential energy of inter-molecular

attraction expect a priori that a solid which is moistened by the liquid so that an acute angle of contact is formed would be one which attracts the liquid more powerfully than does a solid which is not moistened by the liquid; and further, since increase of solid-liquid contact must necessarily be accompanied by decrease of liquid-liquid contact, that moistening of the solid could only take place when the inter-molecular attraction  $K_{\rm SL}$  exceeds  $K_{\rm LL}$ .

Consider the phenomena now from another point of view, that of equilibrium of the forces\* in the region where the liquid-air surface-film intersects the solid. These forces are (1) the attraction of the solid for the liquid along a line at right angles to the solid surface,† (2) the contractile pull of the free liquid surface, (3) the force acting along the liquid at the solid-liquid interface. If the last mentioned is a contractility, equilibrium is only possible if it pulls downwards and the free liquid surface pulls upwards, i.e., if the liquid dips towards the solid and forms an obtuse angle of contact. If, on the other hand, it is an expansile thrust upwards, equilibrium is only possible if the free liquid surface pulls downwards, i.e., if the liquid rises towards the solid and forms an acute angle of contact.

An acute angle of contact must therefore, I hold, be regarded as positive proof that the liquid stratum in contact with the solid is exerting an expansile sideways thrust, i.e., has a negative tension. Carefully considered, it is also impossible to see how, without such sideways thrust, the liquid could ever creep up the walls of the solid at all, quite apart from maintaining itself there when it had crept up.

We have then an association between-

- (1) a negative tension or expansility;
- (2) an acute angle of contact;
- (3) molecular attractions across the surface greater than those of the liquid for itself.

<sup>\*</sup> Tensions of gas condensed at free surfaces are assumed to be negligible. † This opposes the component of the tension of the air-liquid surface normal to the solid.

and I would venture boldly to put forward the view that the association between (1) and (3), and between all three when an angle of contact is formed at all, is an absolute one, and therefore that whenever  $K_{\rm SL}$  exceeds  $K_{\rm LL}$  (K signifying the inter-attractions of two large slabs of matter within range across a surface) the strains and molecular spacing and orientations in the liquid surface-stratum are such as to give rise to an expansility; and conversely whenever there is a negative tension that  $K_{\rm SL}$  exceeds  $K_{\rm LL}$ .

It should perhaps be pointed out that if this view is correct, it would follow that the negative tension of liquid under the influence of solid must often be considerable since it must equal  $-\mathsf{T}_{\text{liquid-air}}$  cos  $\theta$  ( $\theta$  being the angle of contact). Thus taking Quincke's value 25.5° for  $\theta$  at a glass-water-air meeting-point\* and assuming that the air film condensed on the glass exerts neither upward pull nor downward thrust, if T<sub>water (air)</sub> equals + 81 dynes, T<sub>water (glass)</sub> would equal - 73 dynes, the bracket round the word (glass) indicating that the tension concerned is not the whole Twater-glass but only that of the water stratum in contact with glass, a "part-tension" as it may perhaps conveniently be termed. The angle of contact is assumed to be measured between the water-air surface, just beyond range of the solid, and the water-glass interface—within range alike of solid, water, and air both the tensions and the 'angle of contact' would presumably vary in complex fashion from point to point, which variations however would in no way affect the validity of the conclusion as stated.

In cases where  $K_{\text{SL}}$  sufficiently exceeds  $K_{\text{LL}}$ , we should expect that  $T_{\text{L(S)}}$  would be actually more strongly negative than  $T_{\text{liquid-air}}$  is positive, and consequently that there would then be no possibility of equilibrium at a point on the surface of the solid, and therefore no angle of

<sup>\*</sup> Modifications of  $\theta$  by gravity are eliminated by measuring it only when the slope of the solid is such that the water-air surface is horizontal.

contact—the liquid would spread over the whole solid. Whether such cases actually exist, except with soluble solids, is uncertain.

It is clear that a continuation of the subject would necessarily involve a discussion of many aspects of the phenomena of solution and gel-formation, and this I must exclude, not only for lack of time, but also because, as soon as the masses of matter become very small, surface tension considerations no longer suffice, and it becomes necessary to deal directly with the inter-molecular forces as well and to have much greater knowledge concerning these than is available.

The above conclusions concerning the part-tensions at solid-liquid interfaces must obviously be equally valid for all-liquid interfaces but, owing to the mobility of both the apposed surface-strata, clear observational proof is more difficult to obtain. It would lead me too far to attempt any adequate consideration of emulsions and of the different phenomena observable when a little liquid is placed on the free surface of another liquid in which it is insoluble.

The liquid-liquid interfaces have however one advantage over solid-liquid ones in that the total surface-tension can be measured. Taking this to be the algebraic sum of the two part-tensions, the hypothesis that the greater the 'trans' surface attractions on a surface stratum as compared with the 'cis' surface attractions the smaller would be the part tension in that stratum, and that with sufficient preponderance of the former this tension would become actually negative, requires that the sum of the two part-tensions, i.e., the total interface-tension, should always be less, and sometimes very considerably less, than the sum of the free surface tensions of the two liquids concerned. The available measurements show that this is the case. A few of Quincke's values obtained at 20°C may be quoted in illustration.

#### ADSORPTION.

Both at all-liquid and solid-liquid interfaces concentrations of previously-dissolved substances are well-known phenomena, and are then termed 'adsorptions,' when it can be proved, or inferred on general grounds (such for example as their diminution by rise of temperature) that they are attended by diminuation of the surface-energy of the system.

As it is by no means always easy to exclude the possibility that chemical combination of the adsorptum with the adsorbent, or solution in it, has occurred, it should be realised that the theoretically simplest cases of adsorption are those which occur at free surfaces—'adsorption without adsorbent' as they might be described—and also that for the practical study of some aspects of adsorption these often offer very special advantages.

It may be well to point out that the hypothesis that flocculation of solid suspensions is proof that the parttension of the water in contact with the suspended solid has increased from an initial negative, or at most feebly positive tension, into a stronger positive tension, is in no way inconsistent with the view that the flocculation results from a true adsorption, and has therefore been attended by a decrease of the total surface energy of the system. Thus in the flocculation of kaolin in water which follows the addition of a small quantity of calcium chloride, this is presumably preceded by an adsorption of the lime salt (or one of its ions or hydrolytes) and attended by a fall in the total surface energy of the system. Since none of this fall could be attributed to diminution of contact between a soluble substance like calcium chloride and water, it must be due entirely to fall in the total tension of the kaolinwater interface. To reconcile the conclusion that the rise

in the part-tension  $T_{\text{water (kaolin)}}$  accompanies a fall in the total tension, it is only necessary to assume that  $T_{\text{kaolin (water)}}$  has decreased more than sufficiently to compensate for the rise in  $T_{\text{water (kaolin)}}$ .

It is desirable also to call attention to the probability that in some flocculations and precipitations, although they doubtless result from increased tension in the water stratum near the solid, this increase is not due to active adsorption at all, but to the precipitant (or one of its ions or hydrolytes) being 'crowded' into the liquid surface-stratum in spite of its raising the total tension of the interface, as well as the part-tension of the liquid. This is probably the most considerable factor in the 'salting out' of many colloids—since forcible intrusion of a tension-raising substance into the liquid near the solid would usually occur only when large quantities of the precipitant had been added.

Discussion of the effects of adsorptions at the interfaces between the suspended liquid droplets of an emulsion and the surrounding liquid medium would take up too much time and must be kept for some other occasion.

In connection with adsorption in general, there is, however, one point of much practical importance which can be put very briefly, namely, the effects of the presence of two adsorbable substances in the solution. It is clear on the principle of minimal surface energy that if these are physically and chemically indifferent to each other, the one which is capable of producing the greater lowering of surface-tension will tend to be adsorbed preferentially and to exclude the other. This is readily demonstrable in the case of water containing both saponin and soap, the latter lowering the tension of a free water surface much more than does saponin—the solution gives typical soap-bubbles very different from saponin bubbles, it has the mobile air surface of soap solution instead of the rigid one of saponin,

and it yields mechanical surface aggregates such as are obtained from pure soap solution instead of solid saponin. The practical importance of such preferential adsorption lies in the lessons it teaches as to the necessity of—

- (1) strictest cleanliness in studying adsorption phenomena;
- (2) bearing in mind its possibilities when dealing with complex biological liquids such as blood-serum. Imagine, for example, a serum with much of a less adsorbable substance and very little of a more adsorbable one—the first rapid adsorption would be of the less adsorbable though more plentiful substance, but given time enough for the other to get to the surface this might eventually entirely displace the first-arrived—provided the two rivals did not enter into some combination or suffer a chemical change under surface catalytic influence.

### FLOATING ANIMALS.

A brief explanation may here be given of the way in which the contractile pull of a water-air surface is exploited by various small aquatic animals, and serves to support them near the surface, even though they are heavier than water. Thus the small black hairy insect Podura frisks about on the surface of a pond like a fly on quicksilver; the larvæ of the gnat hang head downwards suspended from the water surface by means of their breathing tubes, much as a ham hangs from a hook in the ceiling; numerous Entomostraca which normally live in the water, when by any accident they get on to the surface are, according to Scourfield, apparently quite incapable of getting back and float helplessly on their flat sides until they starve to death or are blown ashore. For numerous other instances the fascinating writings of Professor Miall should be consulted.

The condition necessary for the support of solids in this way is that, since the water-air surface must necessarily meet the solid at the definite angle of contact characteristic for that solid and liquid, the slope of the solid at the point of insertion must be such that at the same time the waterair surface pulls more or less upwards, i.e., it must dip towards the solid. If the solid is a horizontal cylinder or a sphere, its surface has every degree of slope, and consequently there must always be some area on its surface where a water-air surface can be inserted at its proper angle of contact and at the same time dip towards the solid. the angle be acute, this area will be entirely above the equator, and the dip of the water will be slight, but if it be obtuse, this area will extend both above and below the equator, and the dip of the water may be considerable or even vertical. As therefore it is only when the angle of contact is obtuse and the solid is not 'moistened' by water that the weight-carrying power can be notable, it may be taken as certain that this is the case with the animals which are supported by the tension of a water surface. Whether this is secured by 'greasing' the solids concerned has not been ascertained.

While talking of biological subjects, let me mention incidentally that I have examined 'cuckoo-spit,' the protective froth with which the 'frothing hopper,' Aphrophoria spumaria, surrounds itself, and find that the substance which confers 'frothability' on the water is a mucin-like protein.

Time does not permit even a brief discussion of the more complex subjects of prime biological importance concerned with surfaces, chief of which may be reckoned that of the 'plasma-haut,' the pellicle or membrane with which every living cell surrounds itself and by means of which it is defended from many undesirable losses and intrusions. It

would indeed be more profitable to deal at this stage with such subjects as emulsions and the permeabilities of porous membranes and to consider how far they support, or are reconcilable with, the unorthodox views advocated concerning the existence of expansile surface strata.

Until the simpler surface phenomena are reasonably intelligible little real progress will be made with the more complex, and I shall hope that this address will enable the student to acquire something approaching to a real grip of the fundamentals of the subject.

THE

# MARINE BIOLOGICAL STATION AT PORT ERIN

BEING THE

# THIRTY-SECOND ANNUAL REPORT

OF THE

### LIVERPOOL MARINE BIOLOGY COMMITTEE.

By Professor W. A. Herdman, F.R.S.

This is our last war-time Report. It seems probable that hostilities will have ceased and peace be assured before this Report is printed, and that we may reasonably hope for a return to more normal conditions of work at Biological Stations and other research institutions during the coming year. It will no doubt take some time before we can work up again to the same high level of prosperity and usefulness that we enjoyed in the few years previous to 1914; but the growing demand for scientific education and training in original investigation ought soon to lead to a marked increase in the number of both undergraduate students and more advanced post-graduate workers at a place offering the facilities and attractions found at Port Erin.

In 1914 we recorded ninety researchers and students occupying work-places in the laboratory. The "Station Record" and other parts of the Curator's Report that follows, shows that in 1918 we had sixteen workers, including a class of eleven senior students during the Easter vacation.

The number of visitors to the Aquarium has greatly increased during the past year, and the total given by the Curator is a good deal more than double that recorded for 1917. The work of the staff at the Biological Station has been carried on as usual, and large collections of the plankton in the bay have been made throughout the year.

As on previous occasions the statistics as to the use made

of the institution throughout the year will be given in the form of a "Curator's Report" (see below); and after that I have added, as an Appendix, some portions of an unpublished address given a few years ago in the Isle of Man, in the hope that it may be of interest to our students of Natural History at Liverpool and Port Erin.

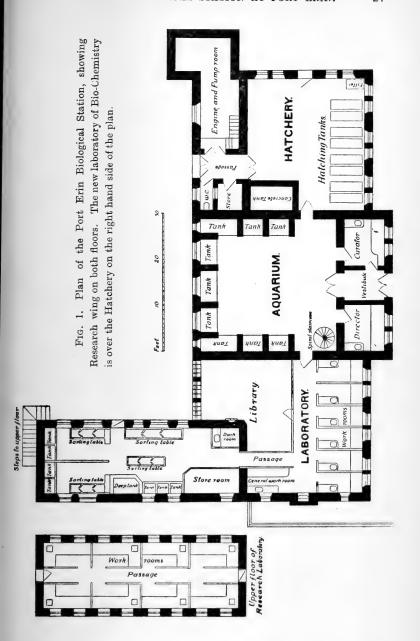
It may be useful to those proposing to work at the Biological Station that the ground plan of the buildings, showing the laboratory and other accommodation, should be inserted in this Report as on previous occasions (see fig. 1, p. 27).

#### CURATOR'S REPORT.

Mr. Chadwick reports to me as follows on the various departments of the work at the Station during 1918:—

# Station Record.

"Sixteen workers—a smaller number than in 1917—occupied our laboratories during the past year. Ten of these were undergraduates of the University of Liverpool, who undertook the Easter vacation course of instruction given by Professor Herdman and Miss R. C. Bamber, and one was a student from Newnham, Cambridge; Miss C. Mayne, Edward Forbes Exhibitioner for the year 1918, devoted herself during the Easter vacation to a study of the animal ecology of Port Erin Bay; Professor Newstead opened up a practically unworked field in the Isle of Man by collecting and identifying Scale-insects (Coccidæ), and on and within a few yards of the Station premises found several noteworthy species; while Professor Moore carried on a research on the accumulations of nitrites and nitrates in the sea in spring and autumn.



### List of Workers.

March 23rd to April 15th. Professor Herdman. 23rdMiss E. C. Herdman. Miss R. C. Bamber. 23rd 5, 6th to 19th. A prilMiss M. Bowen. 8th to 19th. Miss M. Howells. Miss M. Quayle. Miss E. M. Stephenson. Miss C. M. Jarvis. Miss P. E. Harris. Miss E. A. Dodd. Miss B. Gilman. April 9th to 20th. ,, 9th to 20th. Miss M. Hobbins. 9th to 20th. Miss L. Thorpe. 13th to 26th. Miss C. Mayne. August 7th to September 27th. Professor Herdman. ,, 7th ,, 7th ,, September 16th to 23rd. Miss E. C. Herdman. Professor Newstead. 27th.,, 20th to 27th. Professor B. Moore.

### The Library.

"The value of the Library as an adjunct of the laboratories slowly but steadily increases. In view of the growing economic importance of the study of Insects, a number of modern standard works dealing with the Class and with some of its more important Orders have been added. The governing bodies of a number of Biological Stations, British and Foreign, have again contributed their Annual Reports and Results, for which our thanks are accorded. The catalogue is now in a sufficiently forward state to be of considerable value to researchers and students.

# The Fish Hatchery.

"The stock of plaice for this year's hatching operations consisted of 51 survivors of the previous season's stock, and 23 new fish brought from Niarbyl in November, 1917. Of the latter, 12 only were of the recognised spawning size of 13 inches.

"The total number of eggs collected from the ponds during the season is 4,280,850. The first were placed in the hatching boxes on February 25th. On one occasion only, March 9th did the number skimmed from the pond exceed

quarter of a million, and the daily average was about 118,900. The number of larvæ hatched was 3,612,900, and the great majority of these were set free from Professor Herdman's motor boat, Redwing.

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

Eggs collected.			Date.	]	Larvae set free.		Date.	
140,700		Feb.	25 and $26$		113,400		March	19
268,800		,,	27 to March	2	250,950		,,	21
304,500		March	4 and $7$		284,550		,,	26
367,500		,,	9		$345,\!450$		,,	28
226,800		,,	11		178,500	• • •	April	1
245,700		,,	13 and 16		225,750	• • •	,,	3
409,500		,,	18 and 19		373,800	• • •	• •	5
245,700		,,	20 and 22		$215,\!250$		,,	8
226,800		,,	23 and $25$		$193,\!200$		,,	10
109,200		22	26		80,850		,,	12
147,000		,,	27		135,500		,,	13
212,100		,,	29  and  30		$172,\!100$		,,	16
365,400		April	2 to 5		$306,\!600$		,,	19
430,500		,,	6 to 12		338,100		,,	27
182,700		,,	13 and 16		151,200	• • •	$\mathbf{May}$	1
358,050		,,	18 to 25		212,000		,,,	6
39,900	• • •	,,	29 and May	6	35,700	•••		
4.280.850	Tota	al eggs.			3,612,900	Tota	l larva	e.

<sup>&</sup>quot;We now have in the spawning ponds 20 plaice hatched in 1914, 15 hatched in 1915, and 24 hatched in 1916. Eleven of each of the 1914 and 1915 lots were recently measured by the Assistant Curator. The largest of those hatched in 1914 was 36 cm. in length and the smallest 31 cm., the mean length of the lot being 33.9 cm. The length of the largest of those hatched in 1915 was 30 cm. and that of the smallest 28 cm., the mean length being 29 cm. The largest of the 24 hatched in 1916 was 30 cm. in length and the smallest 21 cm., the mean length of the 24 being 24.6 cm. After measurement the fish were labelled and recorded and returned to the pond.

The measurements show that, although hatched and reared under artificial conditions, the rate of growth of these fish has been equal to, if, indeed, it has not slightly exceeded, that of those hatched in the sea. Their food has consisted for the most part of mussels, with herring and mackerel, cut into small pieces, in the summer, and a very occasional meal of lug-worms.

"Several successful trips in the motor boat, *Redwing*, during September and October to the fishing grounds off Niarbyl, have resulted in the addition of 113 good fish to our stock. So that the number of spawners in the pond is now close on 200.

#### Lobster Culture.

"The supply of berried female lobsters, with nearly ripe eggs was better during the past season than for some years past, 27 having been purchased from local fishermen between June 6th and September 14th. These yielded a total of 33,022 larvæ, which gives an average number of 1,223 larvæ per lobster. The successful rearing of lobster larvæ involves the devotion of much time and labour; and though the tota number hatched was not very large, it was not found possible to deal with more than a comparatively small number of these. It was, therefore, decided to liberate 31,500 in the first stage. Of the balance of 1,522 placed in the rearing jars 273—or about 1 in 5—were reared to the lobsterling stage and set free. This number is rather smaller than last year's total, but it does not indicate any relaxation of effort. impossibility, owing to the war, of obtaining a larger supply of rearing jars, and the difficulty of collecting an adequate supply of plankton for the daily food of the larvæ were together responsible for the limitation in numbers.

#### General.

"The ephyra stage of a Pelagid medusa which occurred in large numbers in our spawning ponds during the hatching season of 1910 reappeared this year, and its progressive stages were observed every time the ponds were skimmed for plaice-eggs from the beginning of the hatching season until towards the end of April. As in 1910, efforts to rear these ephyræ in order to identify the species were unsuccessful. The large and handsome Scyphomedusa, *Pilema octopus*, usually common in this neighbourhood in March, was exceptionally abundant this year in May and June.

"Repeated efforts have been made during the past few years to induce the local fishermen to bring in from the fishing grounds the sea-anemones brought up on their long lines. In this way several specimens of two or three of the rarer British species, such as Stomphia churchiæ, Aureliana augusta and Edwardsia carnea, have been obtained and recorded in previous Annual Reports. The commonest of these deeper water species is one the identity of which has long been in doubt, and we are indebted to Mr. T. A. Stephenson of Aberystwyth, to whom specimens were referred, for the following notes upon it:- 'I do not think there can be much, if any, doubt that it is what Gosse called Bolocera eques, though a different colour variety. The question is more—what is B. eques? (1) It is certainly not a Bolocera. (2) As far as external characters go it should be a Tealia, but not the same species as the ordinary shore Tealia. In that case it should be called Urticina eques, Gosse, as Tealia is really only a commonly used synonym of the earlier Urticina. (3) It may, however, turn out to be identical with a species described by Carlgren as Rhodactinia crassicornis; if that is so, the name crassicornis, I think, has priority to eques: and the generic name would be Urticina, since there is no justification for generically separating Urticina and Rhodactinia. So, at present, I think it would be safest to call it  $Urticina\ eques$ , Gosse (? =  $Rhodactinia\ crassicornis$ , O. F. Muller).

"During the Easter vacation, the little lump-sucker, Liparis montagui, was added to our list of Fishes by Miss Bamber.

# The Aquarium.

"The Aquarium shared the large measure of prosperity which returned to the Isle of Man during the past season, 7,637 visitors—more than twice last year's number—having paid for admission. The claims of lobster culture upon the time of the Assistant Curator militated considerably against the varied display, especially of fishes, to which the visitors have been accustomed. Still, the occupants of the tanks proved interesting to many, and induced repeated visits. The seanemones, of which a large display, together with tube-building and other worms, sea-squirts, &c., has been maintained throughout the year, never fail to interest the more intelligent. It was not until September that a small octopus (*Eledone cirrosa*) was secured, the first since August, 1916.

"Several parties of pupils from Insular schools visited the Aquarium during the year.

(Signed) H. C. CHADWICK."

## REPORT OF THE EDWARD FORBES EXHIBITIONER.

An "Edward Forbes Exhibition" was founded\* in 1915, at the University of Liverpool, in commemoration of the pioneer marine biological work done in this district by the celebrated Manx Naturalist, who was born about a hundred years ago. The object of the Exhibition is to enable some post-graduate student of the University to proceed to the Port Erin Biological Station for the purpose of carrying

<sup>\*</sup> The Regulations in regard to the Exhibition will be found at p. 57.

on some piece of biological research, more or less in continuation of some line of work opened up by Forbes, or an investigation which has grown out of such work.

The Edward Forbes Exhibitioner for the year 1918 is CATHERINE MAYNE, B.Sc., who spent a couple of weeks at Port Erin in the Spring, working at some points in connection with the density of distribution of the commonest plants and animals on the sea-shore.

Miss Mayne reports as follows on her work at Port Erin :-

"As Edward Forbes Exhibitioner for this year I spent two weeks from April 13th to 29th collecting statistics relating to the density of the flora and fauna on the shores round Port Erin. My attention was devoted mainly to the animals Balanus and Patella, and the plants Fucus and Laminaria, but I noted also Spirorbis and Actinia.

"My object was to gain some idea of the *possible*, not the average, density of the population, and I therefore selected the most thickly covered parts of the shore for my purpose. The apparatus consisted of a wooden frame such as fishermen use for their lines, one-foot square, a measure, graded from  $\frac{1}{16}$  of an inch upwards, and a blue-lead pencil.

"The barnacles (Balanus balanoides) are most abundant on the rocks between quarter-tide and full-tide. They grow very thickly on the big boulders and flat-topped rocks at Fleshwick, Spaldrick, and the Miners' Bay (Bradda), and below the swimming-baths and the old Biological Station in Port Erin Bay—in all cases on the metamorphic 'Manx Slates.' On the limestone shore at Port St. Mary patches of small barnacles are to be found, but these are sparsely scattered between comparatively large areas of Enteromorpha, and the two never seem to inhabit the same piece of rock.

"The method of observation consisted in laying the frame upon as flat a piece of rock as I could find, and counting all within by hundreds, ticking them off with the blue pencil to avoid any omission or duplication. At Fleshwick I counted 2,940 in one square foot, varying in size from  $\frac{1}{16}$  in. to  $\frac{3}{8}$  in. across the base. The only part of the area not covered by barnacles was where two small limpets had established themselves. The rocks below the old Biological Station support another variety, for though they entirely covered the rock they appeared more distinct from one another, being higher in proportion from base to crown than those at Fleshwick. Here I found only 1,138 to the square foot, the average size (long axis) being  $\frac{3}{8}$  in.

"Lower down the shore, about half-tide, Fucus begins to crowd out the Barnacles. It grows in great masses chiefly on the tops of rocks, to a length of about 18 ins., and on its fronds shelter large numbers of Littorina, Purpura, and Spirorbis; the last-named I did not attempt to count, and it was difficult to obtain reliable statistics concerning the first two, as they fell off and were lost at the slightest disturbance. On the North side of Spaldrick Bay and in Trai Veg epiphytic Polysiphonia is abundant on the Fucus. My method with Fucus was to shave the rock and count the stalks of weed. When all the weed is cut away the rock is found to be by no means bare, for in one typical observation I noted, beside 35 stalks of Fucus serratus, 324 Balanus, 13 Actinia, 1 Patella and 206 Spirorbis in the square foot. Fucus vesiculosus grows more abundantly still, 352 stalks arising from 12 'roots' in one square foot.

"Although limpets are found quite commonly amongst the barnacles and the *Fucus*, they thrive best where they are entirely free of neighbours, on vertical faces of rock, and near low water mark. These three conditions are best fulfilled on the shore side of Trai Veg, where I found as many as 37 in one square foot, five of which were over 1 in. across, and in other observations, though there was a smaller number, a much larger proportion were between 1 in. and 2 in. In this

respect, again, Fleshwick proved good ground, though not as good as Trai Veg.

"Sea-anemones seem to flourish best in small rock-pools, or in cracks or under sheltering ledges of rock. Fleshwick abounds in them. I counted 20 Actinia, and 5 Sagartia to the square foot in a small pool at Fleshwick, the anemones ranging from 1 in. to  $\frac{1}{4}$  in. across.

"Laminaria is only uncovered at the lowest Spring tides, and, unfortunately, I did not have the full advantage of these. The great brown masses could be plainly seen just out of reach at the West side of Fleshwick Bay, the North side of Spaldrick Bay, and in Trai Veg. At the upper edge of the Laminaria zone I found that 6 to 8 stalks mingled with an equal number of Fucus was quite common. Two feet lower down I found the Fucus replaced entirely by Laminaria, the highest number in Fleshwick being 36 stalks to the square foot, and in Spaldrick 28 (young ones). Considering the size of the palm-like 'leaf' this is a large number, although the stalks only measure  $\frac{1}{2}$  in. to  $\frac{3}{4}$  in. across, yet it is probable that this record would be broken had the tide been even one foot lower. Port St. Mary, too, appeared to be specially rich in Laminaria.

(Signed) CATHERINE MAYNE."

This investigation is far from being complete, but seems promising; and it is to be hoped that Miss Mayne will carry it to a further stage on some future occasion at Port Erin.

SCALE-INSECTS (COCCIDÆ) OF PORT ERIN.

Professor R. Newstead, F.R.S., during his stay at Port Erin in September, devoted his attention to the Coccids of the neighbourhood, and succeeded in finding the following noteworthy forms. A more detailed paper on these insects by Professor Newstead will be laid before the Biological Society at a subsequent Meeting:—

Newsteadia floccosa (De Geer), on heather near Calf Sound.
Ortheziola vejdovskyi, Sulc., on heather near Calf Sound.
Eriococcus greeni, Newst., under stones near Biol. Station.
Pseudococcus radicum (Newst.), on grass near Biol. Station.
Ripersia halophila (Hardy), abundant on grasses and weeds.
Eriopeltis festucæ (Fonsc.), abundant on Festuca round
Port Erin.

### L.M.B.C. MEMOIRS.

Since our last report was published, no further Memoirs have been issued to the public. Himanthalia, by Miss L. G. Nash, M.Sc., is ready to print; Miss E. L. Gleave, M.Sc., has nearly completed her Memoir on Doris, the Sea-lemon; Mr. Burfield, who was writing the Memoir on Sagitta, has joined the Army; Miss Bamber has made further progress with Tubularia, and still other Memoirs 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. Tubifex, G. C. Dixon, 100 pp., 7 Pls.

HIMANTHALIA, L. G. Nash.

DORIS, E. L. Gleave.

TUBULARIA, R. C. Bamber.

APLYSIA, N. B. Eales.

TEREBELLA, C. P. M. Stafford.

BALANUS, H. M. Duvall.

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.

Pycnogonum, J. E. Hamilton.

BOTRYLLOIDES, W. A. Herdman.

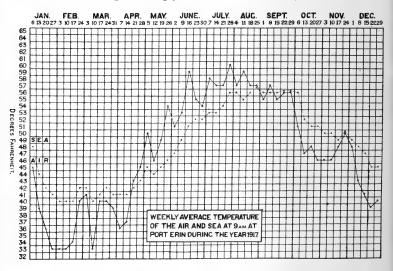
In addition to these, it is hoped that other Memoirs will be arranged for, on suitable types, such as *Pontobdella*, a Cestode and a Nematode.

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.

The diagram of sea and air temperatures for 1918, compiled by Mr. Chadwick from his daily records, is not yet completed; but that for the preceding year, 1917, is inserted here as usual.



Appended to this Report are:-

- (A) An Address on "Periodic Changes in Nature," delivered to the Biological Society, by Professor Herdman, on November 8th, 1918;
- (B) The usual Statement as to the constitution of the L.M.B.C., and the Laboratory Regulations—with Memoranda for the use of students, and the Regulations in regard to the "Edward Forbes Exhibition" at the University of Liverpool;
- (C) The Hon. Treasurer's Report, List of Subscribers, and Balance Sheet for the year.

#### APPENDIX A.

#### AN ADDRESS ON

#### SOME PERIODIC CHANGES IN NATURE.\*

GIVEN BEFORE THE LIVERPOOL BIOLOGICAL SOCIETY

# By W. A. HERDMAN, F.R.S.

There is a Manx proverb meaning "Change of work is rest," and most of us have had personal experience of the benefit of occasional change of occupation and of thought. I desire to direct your thoughts for a few minutes to some of the periodic changes in Nature around us. Along with a fundamental continuity in the series of phenomena forming the scientific history of the Universe, there is a certain amount of constant change, and to some extent periodic change, which is of enormous importance in connection with human affairs. I propose to illustrate this thesis by directing your attention to certain changes in Nature as they may be observed around Port Erin, or the Isle of Man in general.

If we take up our position on the Mull Hill to the south of Port Erin, and look N.E. towards Ramsey, we see laid out before us, like a diagram, a great deal of the past history of this island. We see the rugged series of peaks from Bradda Head, along the Carnanes, the Cronk, and South Barrule to the great central valley, and then onwards by Greeba and Injebreck to Snaefell and North Barrule—forming the backbone or central axis, the oldest part of the island—in fact, what was the original island in early Palæozoic times. Then, low down on the eastern slopes of this central "massif," and possibly on the steeper west also, we may find in places traces

<sup>\*</sup> Being portions, with additions, of an unpublished Presidential Address delivered before the Isle of Man Natural History and Antiquarian Society, on April 16th, 1914.

of the overlying much later sedimentary rocks of carboniferous date, and over these limestones and sandstones we see the boulder clays and sands of the great Ice age, bringing us within measurable distance of the Historic period.

Let us, in imagination, while gazing at this panorama, dissect our Island, remove it layer by layer, and watch for the evidence of colossal changes in nature as we pass back through the ages. Remove, in the first place, the houses and fences, and other traces of man's occupancy and industry, and then wash away from the rocks all the superficial soil forming the fields and the moorlands, and you have then reduced the surface to the condition it was in at the end of the Glacial period, and just before the first men made their appearance in this part of Britain—and conditions were certainly very different then from what they are now. The climate must have been much colder. It had recently been arctic, the whole land had been buried in glaciers—perhaps, the present condition of Greenland or Alaska gives one the best picture of what the scene then was.

Then again, sometime during the Glacial period, the land was much higher than now, and these islands were continuous with the Continent of Europe. The Irish Sea, which is now of such importance to us, so essential for our welfare and prosperity that one can scarcely imagine this land without it, did not then exist. It was temporarily filled up to the north, south, and east of the Isle of Man, and to the west the deep channel which now separates us from Ireland probably existed only in the form of a long narrow fresh-water lake or series of lakes on the course of a mighty river—perhaps the largest river in the British Isles, and far greater than any now existing. It started with the Clyde in the north, and running southwards along what is now the Firth of Clyde, between Scotland and Ireland, received as tributaries the Boyne, and the Liffey, and the Mersey, the Dee, and the Ribble, and the innumerable smaller streams from Scotland, Ireland, England and Wales,

joined the Severn lower down, and finally opened into the North Atlantic, somewhere between Finisterre and the South of Ireland.

The change from that picture to the period immediately succeeding the Ice age, when the land sank and the Atlantic flowed in and filled up the Irish Sea, and the climate became more genial, and forests covered the land, and the fauna and flora spread and multiplied, and finally neolithic man made his appearance in this island, pursuing the great Irish Elk, is, you will agree, a considerable one—a change of a striking and wholesale nature, and yet it is slight compared with some of the changes which periodically affected different parts of our country in still older geological times. We find warmer and colder climates, as indicated by the fossil remains in the rocks succeeding one another, we find land submerged and then elevated to form mountain ranges; coral reefs and deposits of coal may be found in succession in the same locality, and finally, vast volcanic outbursts, far transcending in extent anything that we know of at the present day, poured forth sheets of lava periodically, and have given rise to some of the most striking scenes of our western coasts, such as the columnar rocks of Antrim around the Giant's Causeway, and the farfamed cliffs and caves of Staffa. In one cliff on the western coast of Mull may be found, interbedded between two great horizontal layers of columnar basalt, a deposit of most beautifully preserved fossil leaves, one of them belonging to the Ginkgo tree, living at the present day in Japan. Think of the extreme changes of local conditions that that sequence of rocks indicates.

So far, I have spoken only of the superficial aspects of these great changes—matters with which I have no doubt you were all familiar; but I must now remind you that these superficial aspects represent profound changes in the fundamental nature or chemical composition of the deposits that

are being produced. Take, for example, two of the most familiar rocks to all of us-Coal and Chalk. To the east of us lie the great coalfields of Lancashire, to the N.W. the Chalk of Antrim, so both are our near neighbours, and our own limestone at Castletown and Port St. Mary is only a harder and less pure form of chalk. We associate each of these common rocks, coal and chalk, with one particular chemical element, coal with carbon, and chalk with calcium; and when the useful substances coal and chalk are used up, or disappear as such, the elements carbon and calcium are set free to reappear again later on somewhere else in nature in their beneficent scheme of world-wide circulation. Notice the periodic change. When we burn our coal we release the carbon from the condition in which it has been held or imprisoned since the Carboniferous period, perhaps twenty million years ago. The carbon is given off in the form of noxious gases which are poisonous to man, and yet-paradoxical as it may seem-without which man and all other animals on this globe could not exist—without which life, as we now know it, would not be possible—as these gases supply the small amount, only three parts in 10,000 of carbon dioxide in the atmosphere, which is required as food by the green plants upon which all animals depend for their very existence. Our coal beds represent the forests and swamps of the Carboniferous age, and the heat and light given out by the coal as we burn it to-day may be referred to as the bottledup sunshine of that far-back period by virtue of which the green plants then existing were able to appropriate and fix the carbon from the gases of their atmosphere. Nature, as it were, invested her capital in the plants of the Carboniferous period, sank it in fixed carbon, and man is now reaping the benefit and enjoying the unearned increment in the form of steam ships and railways, and all the modern manufactures that depend on coal.

Carbon, then, in the history of the world, has been some-

times free in the air and water in the form of the gas carbon dioxide, sometimes in the bodies of living things, all of which are largely formed of carbon, and sometimes stored up in solid form as coal and other materials from which it can again be set free and pass into circulation.

The same phenomenon of periodic change is seen in the case of the calcium in beds of chalk or limestone. When dissolved from that rock it is set free, and may next appear in solution in fresh or salt water, and then in the form of a tropical coral reef, the shell of an oyster, or the bone of an animal's skeleton. These corals and shells may again be consolidated on the sea-bottom to form beds of chalk and limestone. Like carbon, the calcium is a necessary part of our food—and we take it in, for example, sometimes in too large quantities, in our milk—and then those of us who are no longer young begin to store it up in solid limy form in parts of our bodies that would be better without it.

I need not give further examples in detail. I would only add that the same principle of periodic change or circulation of necessary substances in Nature is seen in many other cases. Nitrogen, a necessary part of all living things, is also found in the nitrates and other fertilisers of our soil, in the sewage poured in ever increasing quantity from our great cities into the sea, and again in the enormous harvest brought back annually from the sea to the land. When herrings and other fish are brought ashore for the markets, when the large brown sea-weeds cast ashore by storms are carted up to spread as manure on the fields, we are reclaiming from the sea some of the valuable nitrogen the land gave up in the form of sewage and other matters carried down by streams and storms.

One more case, put briefly:—The silica of our island may be in the form of a white quartz vein running through the metamorphic Manx slates of Foxdale or Bradda Head, or united with alumina in the clays and shales, or may

be a flint arrow-head dug up from a neolithic tumulus and now preserved in the Museum at Castle Rushen, or it may be in the form of the delicately-sculptured shells of microscopic diatoms swarming in the seas round our Island at certain times of the year. Silica, quartz, or flint is a most insoluble substance, but all the innumerable millions of diatoms which make their appearance in the sea every spring, must obtain the necessary material for their shells from dissolved silica in the sea-water, and the sea must obtain its supply from the land. Here again, we see the circulation or periodic change that I am demonstrating. All these important materials are kept in circulation. Gold is not the only element upon the circulation of which the prosperity of man depends. We could get on very well without the gold, but we could not exist without the constant circulation of the carbon, and calcium, and nitrogen of which I have been speaking-or, to mention another case, without the iron in our blood, and again in living plants, or even without the silica upon which it is scarcely too much to say that all life in the sea depends.

I have no doubt that these and other examples of periodic change are known in more or less detail to many. Of course, the general notion that the material making up the bodies of living organisms may at another time be lifeless, inorganic stuff, is centuries old. In Elizabethan times the genius who wrote Shakespeare's plays makes Hamlet say:—

"Imperial Cæsar, dead, and turned to clay, Might stop a hole to keep the wind away."

And Lucretius, if I am not mistaken, sixteen centuries before that expressed his belief in a general circulation of matter.

But there is the greatest difference between a somewhat vague belief and the scientific demonstration that definite chemical elements, such as I have been speaking of, do circulate periodically through earth, and air, and water around us.

Let me take as my last example those periodic changes in

our sea, of enormous amount and far-reaching importance, which have been recently discovered, and in fact, are now being investigated, and may, I think, be new to most of you. I fancy we are in the habit of thinking of the sea as constant in character and composition throughout the year. I am, of course, excluding such changes as are caused by tides and storms, and perhaps I ought to exclude also changes in temperature, for everyone knows that our sea is colder in winter and spring than in summer and autumn (the lowest temperature is usually in March, and the highest in August), but it will probably be new to you to learn that the sea differs in specific gravity and salinity from time to time, that it differs very remarkably in its microscopic contents at the different seasons, and, further, the sea changes notably in its chemical characteristic of alkalinity.

Let me tell you a little more about this alkalinity of the sea, as another example of periodic change. Several scientific men have during the last few years independently worked at these chemical variations in the sea, and one of these is Professor Benjamin Moore, who first of all discovered while working at the Port Erin Biological Station that the sea around the Isle of Man was a good deal more alkaline in spring (say April) than it is in summer (say July); and then, on examining monthly samples taken throughout the year, he was able to show that the alkalinity, which gets low in summer, increases somewhat in autumn, and then decreases rapidly during the winter, and then after several months of a minimum, begins to increase again in March, and rapidly rises to its maximum in April. That is the periodic change of alkalinity, and it will be seen to correspond roughly with certain very important changes in the living microscopic contents of the sea, and the connection between the two may be made out by inquiry into the nature and meaning of the changes in alkalinity. The alkalinity of the sea is due to the relative absence of carbon dioxide. So

here we are again brought into the presence of that all-important element carbon in one of the stages of its beneficent cycle through all the so-called elements of the ancients, earth, air, fire, and water.

When, then, the useful carbon dioxide, required as food by the developing plants of the sea, becomes stored up in increased amount, that causes a marked reduction in the alkalinity of the sea-water. The amount of difference between any two samples of water may seem small, but the volume dealt with in the sea is so vast that the annual turn-over in the form of carbon which appears and then disappears, or is used up, is colossal in amount and difficult to realise—even in the narrow seas around our little island.

The figures that follow are not final, they are still subject to correction, but even if not quite exact, will serve as an example of the order of quantities involved, and give some indication of the vast scale of the phenomenon and of the large amount of potential ultimate food-matter available in the sea. We find that at Port Erin in March the water not only on the shore, but also out in the open sea, is acid to phenolphthalein, while a month later it is distinctly alkaline to the same indicator, and this change signifies an enormous conversion of carbon in the inorganic into carbon in the organic form—a turn-over of such extent that it probably amounts to 20,000 or 30,000 tons of carbon per cubic mile of sea-water. Or if we think of the carbon as being present in the bodies of living organisms, then the weight of these organisms will amount to about ten times the above amount, viz., about 300,000 tons per cubic mile-or if we imagine these same organisms distributed along the deepest part of the Irish Channel, then they would occupy a strip of water about ten miles long by one mile wide, and 88 fathoms deep. Or we may imagine this same quantity of carbon as forming the bodies of all the organisms found in the sea all around the shores of

the Isle of Man, in which case the 300,000 tons would be distributed through the zone of water extending to about one mile out from the shore, and down to an average depth at that distance of, say, ten fathoms. Now, all of these organisms have obtained their carbon from the carbon dioxide present in the sea-water in spring, and it is absolutely certain that in the absence of this abundant supply of available carbon-food, the millions and millions of organisms in question could never have existed. This explains the connection which I mentioned above between the periodic chemical changes in alkalinity, due to variations in the carbon dioxide present, and the periodic changes in the living contents of sea-water.

In early spring there is a great awakening in the oceans comparable with the growing of the grass and the budding of the trees on land. This increase and growth of living things starts in our sea here about March, when the temperature of the water (which lags behind that of the air) is about at its lowest. So it is evident that temperature has nothing to do with this germination, although it is quite possible that the marked increase of sunlight has. These early spring organisms, the diatoms, constituting the phytoplankton, as it is called because of its vegetable nature, increases with astonishing rapidity during March and April, until it reaches what we know as the vernal maximum, when these diatoms are so abundant all through the water of the Irish Sea, that a short haul of a fine silk net towed along the surface will catch anything from one up to a couple of hundred millions of individuals. Moore's figures for the turn-over of carbon in spring indicate that there is probably a production each season of about two tons of dry organic matter per acre, corresponding to at least ten tons of moist vegetationwhich shows that we may still be very far from getting from our seas anything like the amount of possible food-matters that are produced.

These diatoms began in March, when the alkalinity was low, that is to say, when the carbon dioxide in the water is great in amount, and their vernal maximum coincides with the top of the alkalinity curve, that is to say when the carbon dioxide is relatively small in amount. The carbon that was present as that gas is not lost, but it is now present in another form, as parts of the living bodies of the diatoms, and had not the inorganic carbon been available as food these abundant swarms of living things could not have been produced.

After the vernal maximum the diatoms begin to die off, and disappear for a time, their place being taken as the dominant organisms of the sea-water, for a period in early summer, by the minute shrimp-like animals known as Copepoda.

I do not propose to follow these changes in the plankton any further throughout the year, but if the so-called "practical man" were to ask me whether all these changes in the sea, chemical and vital, have any relation to man and his industries, I would answer, "Only this—that the whole life and development of fishes in the sea seems to depend upon them "-as I shall show you presently. But, first, let me say that, personally, I do not attach much importance to demonstrating the practical utility of a scientific discovery. I am content to leave it to demonstrate itself, knowing well, that if it is true it will sooner or later be justified in the mind of even the most The history of science bristles with utilitarian of men. examples of discoveries in pure science which at first seemed to have no obvious relation to man or his material wants, and which have since formed the foundation of, it may be, a chemical industry or an electrical enterprise, or some development of radio-activity. No better instance could be taken than the change in attitude of the man-in-the-street towards the study of mosquitos and flies.

A few years ago, you will agree with me, the Entomologist was looked upon with a benevolent smile as an amiable old

gentleman, perhaps harmless, but certainly for practical purposes the most useless of cranks. And yet it is the results of these men's work—of their patient investigation of the life-histories and habits of the disease-bearing flies, as applied by the United States Sanitary Service, that have rendered possible that great engineering feat, the completion of the Panama Canal. It was not any engineering difficulties that prevented the French, quarter of a century ago, under De Lesseps, from cutting the isthmus, but merely that sanitary science, based upon the work of field-naturalists and microscopists, was not then far enough advanced to permit men to put up a successful fight against malaria and yellow fever.

However, I have no objection to point out a utilitarian connection where one can be seen to exist. Practically all our food-fishes in the sea, except the herring, produce their eggs in winter or early spring. They are hatching out in vast quantities during the time that the alkalinity is rapidly increasing and the phytoplankton of diatoms is daily growing in amount.

Now, soon after the little larval fishes have been hatched from the eggs, diatoms are exactly what they require as food. So we may put the connection this way—the increasing alkalinity in spring is a measure of the rate at which the diatoms are converting the inorganic carbon of carbon dioxide into the organic carbon of their own living bodies suitable to be the food necessary for the young cod, haddock and whiting, and plaice, soles and flounders then being hatched out.

A little later on, in early summer, when the vernal maximum of phytoplankton is over, and the diatoms have been replaced by copepoda, we find the same little fishes, now grown larger and stronger, feeding not on the vegetable food of their early babyhood, but upon the animal food in the form of copepoda, by which they are surrounded.

I excepted herring just now from the fishes mentioned,

but I only did so because of the season of the year I was dealing with—the herring and the mackerel form no exception to the rule that the fishes upon which man's food from the sea depends, are nourished in their young and even in some cases in their older stages also by the abundant swarms of copepoda of our summer seas.

It is not too much, then, to say that the fishes that form our food depend for their existence in our seas upon the copepoda, and these copepoda themselves feed upon diatoms, which also nourished the little fishes at a younger stage, and these diatoms derive their food from the carbon dioxide in the sea-water, the varying amount of which, at different seasons, according to whether or not it is then being used in the production of microscopic plants, causes those changes in the alkalinity with which I started this cycle of events. It is phenomena such as these, some of them, as you can imagine, of far-reaching importance, that are now being investigated by modern biologists—partly at our University laboratories, but partly also at such marine laboratories as our Port Erin Institution, in the foundation and progress of which, I may remind you, this Society has always taken a lively and helpful interest.

#### APPENDIX B.

# THE LIVERPOOL MARINE BIOLOGY COMMITTEE (1917).

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, J.P., M.A., 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., London.

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.

#### 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.

#### 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 steamboat 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.

## 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.

### REGULATIONS OF THE EDWARD FORBES EXHIBITION.

[Extracted from the Calendar of the University of Liverpool for the Session 1915-16, p. 438.]

#### "EDWARD FORBES EXHIBITION.

"Founded in the year 1915 by Professor W. A. Herdman, D.Sc., F.R.S., to commemorate the late Edward Forbes, the eminent Manx Naturalist (1815-1854), Professor of Natural History in the University of Edinburgh, and a pioneer in Oceanographical research.

The Regulations are as follows:-

- (1) The interest of the capital, £100, shall be applied to establish an Exhibition which shall be awarded annually.
- (2) The Exhibitioner shall be a post-graduate student of the University of Liverpool, or, in default of such, a post-graduate student of another University, qualified and willing to carry on researches in the Manx seas at the Liverpool Marine Biological Station at Port Erin, in continuation of the Marine Biological work in which Edward Forbes was a pioneer.
- (3) Candidates must apply in writing to the Registrar, on or before 1st February.
- (4) Nomination to the Exhibition shall be made by the Faculty of Science on the recommendation of the Professor of Zoology.
- (5) The plan of work proposed by the Exhibitioner shall be subject to the approval of the Professor of Zoology.

- (6) Should no award be made in any year, the income shall be either added to the capital of the fund, or shall be applied in such a way as the Council, on the recommendation of the Faculty of Science, may determine.
- (7) The Council shall have power to amend the foregoing Regulations, with the consent of the donor, during his lifetime, and afterwards absolutely; provided, however, that the name of Edward Forbes shall always be associated with the Exhibition, and that the capital and interest of the fund shall always be used to promote the study of Marine Biology."

#### EDWARD FORBES EXHIBITIONERS.

1915 Ruth C. Bamber, M.Sc.

1916 E. L. Gleave, M.Sc.

1917 C. M. P. Stafford, B.Sc.

1918 Catherine Mayne, B.Sc.

#### APPENDIX C.

#### HON. TREASURER'S STATEMENT.

The Balance Sheet and List of Subscribers are shown on the following pages.

During the year we have fortunately just been able to pay our way, as we received a little more from admissions to the Aquarium owing to the better conditions in the Isle of Man.

It is expected that next year our expenditure will be considerably heavier, as the work at the Biological Station during the War has necessarily been curtailed, so there will be more need than ever for increased financial support.

Edwin Thompson,

Hon. Treasurer.

25, Sefton Drive, Liverpool.

December 14th; 1918.

#### SUBSCRIBERS.

	£	s.	d.
Browne, Edward T., M.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., 43, Harrington Gardens, London, S.W	2	2	0
Brunner, Roscoe, Belmont Hall, Northwich	1	1	0
Clubb, Dr. J. A., Public Museums, Liverpool	0	10	6
Cole, Prof., University College, Reading	1	1	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
Hickson, Prof., F.R.S., University, Manchester	1	1	0
Holt, Dr. Alfred, Dowsefield, Allerton	1	0	0
Holt, Mrs., Sudley, Mossley Hill, Liverpool	2	2	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
Meade-King, R. R., Tower Buildings, Liverpool	0	10	0
Mond, R., Sevenoaks, Kent	5	0	0
Monks, F. W., Warrington	2	2	0
Muspratt, Dr. E. K., Seaforth Hall, Liverpool	5	0	0
O'Connell, Dr. J. H., Dunloe, Heathfield-road,			
Liverpool	1	1	0
Formand	049	15	_

	£	s.	d.
Forward	42	15	0
Petrie, Sir Charles, Oakfield, Aigburth, Liverpool	1	1	0
Rathbone, Miss May, Backwood, Neston	1	1	0
Roberts, Mrs. Isaac, Thomery, S. et M., France	1	1	0
Robinson, Miss M. E., Holmfield, Aigburth, Lipool	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, The Holt, Linton-road, Oxford	2	2	0
Yates, Harry, 75, Shudehill, Manchester	1	1	0
	£64	4	0
Add old Subscriptions received less Subscriptions still unpaid	9	19	6
	£74	3	6
			_
SUBSCRIPTIONS FOR THE HIRE OF "WORK-TA	BLES.	, ,	
Victoria University, Manchester	£10	0	0
University, Liverpool	10	0	0
University, Birmingham	10	0	0
	£30	0	0
Donations.			-
Hutton, J. A., Woodlands, Alderley Edge	5	0	0
Willey, Prof. Arthur, McGill University, Montreal,	3		Ü
Canada	3	0	0
Watson, A. T., Tapton-crescent Road, Sheffield	1	1	0
	£9	1	0
	-		

# THE LIVERPOOL MARINE BIOLOGY COMMITTEE.

IN ACCOUNT WITH EDWIN THOMPSON, HON. TREASURER.

e E

£217 18 11 æ 83 " Work Tables" By Subscriptions and Donations received ....... ", Amount received from Universities for hire of Interest on British Association (1896) Fund ... Interest on Investment ...... Sale of Guides and Post Cards...... Specimens, &c. ", Bank Interest Admissions to Aquarium-Share of £217 18 11 " Balance in hand, December, 1918 ..... Printing and Stationery ..... Boat Hire " Books, Apparatus and Supplies at Port Erin Biological Station ..... Postages, Carriage, &c. ..... Salary—Share of Curator's ...... Assistant's ..... To Balance due Treasurer, December, 1917 ", Sundries

British Workman's Public House Co. 99 Shares £1 each fully paid. Endowed Invested Fund:

# EDWIN THOMPSON,

COOK & LEATHER. Audited and found correct, HON. TREASURER.

LIVERPOOL, December 14th, 1918.

Balance December, 1917 Frisheries Research Work Expenditure..... Board of Agriculture and Fisheries Account-

Chartered Accountants.

£156 12

£37 4

Extension Fund: -Balance, as at December, 1917

£212

185

Memoir Fund—Balance, as at December, 1917 .....

Add Sales of Memoirs .....

# SOME SCALE-INSECTS (COCCIDÆ) FOUND IN THE ISLE OF MAN.

#### By Professor R. NEWSTEAD, F.R.S.

Thanks to Professor Herdman's kind hospitality I was able to devote some attention to the Coccid fauna of the Isle of Man, during a short stay at Port Erin, in September, 1918. As no attention had hitherto been given to the subject, I naturally expected that interesting records awaited discovery. In this I was not disappointed; but further search would inevitably bring additional records, more especially so of those species which live upon deciduous trees and shrubs, such as the Diaspinæ and the genus *Lecanium*. The publication of the following notes of my captures may be of value to those who take an interest in the insular fauna:—

#### Newsteadia floccosa (De Geer).

Several examples of this species were found in moss and decayed vegetable matter surrounding the stems of heather, in a sheltered spot near the Calf Sound. In some of the examples the so-called "marsupium" was fully developed; in others the waxen lamellæ forming this structure were quite rudimentary.

#### Ortheziola vejdovskyi, Sulc.

A single female of this rare and interesting coccid was found in association with the preceding species. O. vejdovskyi forms the type of the genus, and was discovered by Dr. Sulc at Prague, Bohemia, in 1894. Mr. E. E. Green has recorded it from a single locality in Great Britain; and I also found it in moss on the cliffs at the Isle of Wight in August, 1905.

#### Eriococcus greeni, Newst.

Two females. The ovisacs of both examples were attached to fragments of rock, lying partly buried in coarse grass, at

the foot of the rocky cliffs, near the Marine Biological Station. This apparently rare species has hitherto been recorded only from Budleigh, Salterton and Camberley.

Pseudococcus (Ripersia) radicum (Newst.).

Three females. One example had produced her full complement of eggs, the others were young adults. On grass near the Marine Biological Station.

Ripersia (Ripersiella) halophila (Hardy).

This root-feeding species was extremely abundant both at Port Erin and near the Calf Sound. It occurred most freely on roots of various grasses in rock fissures; but was common also on *Statice armeria*, *Plantago* spp. and *Silene* sp.

Eriopeltis festucæ (Fonsc.).

Abundant in the immediate neighbourhood of Port Erin. Small isolated plants of *Festuca* sp. were sometimes quite thickly studded with the white ovisacs; the male puparia were also not uncommon.

The examples found in exposed situations immediately above the cliffs were remarkable for their small size and the closely felted ovisacs. On the other hand those which were found in the sheltered places, between or beneath loose fragments of rock, at the foot of the cliffs near the Marine Biological Station, were exceptionally large and the ovisacs rough and decidedly woolly in appearance. The following measurements of the females and their ovisacs are noteworthy:—

Length of females from exposed situations, 1.9 to 2.8 mm.; length of ovisacs, 3.5 to 6 mm.

Length of females from sheltered situations, 4·3 to 4·6 mm.; length of ovisacs, exclusive of the outstanding filaments, 5·6 to 9 mm.; inclusive of the outstanding filaments the larger specimens measured as much as 11·5 mm.

Ovipositing was still in progress, and very few females had deserted their ovisacs.

#### THE NEST OF THE BANK VOLE.

#### BY GEORGE ELLISON.

Early in May, 1918, I was staying at a farm called Ffynontudor (Tudor's Well), Llanelidan, near Ruthin, situated on the mountain top, 700 feet above sea-level, at the head of the Vale of Clwyd. During one of my rambles I saw in a bank, at the edge of a wood, what I thought was a tuft of withered grass and leaves, about a foot from the ground. When I touched it, out jumped a vole, which I had time to see was a Bank Vole. The nest from which it had sprung, upon my attempting to handle it, fell to pieces. I searched in the neighbourhood and found, on May 12th, another nest-built, this time, amongst the stems and leaves of a bramble bush—in the nest of a whitethroat, and about eighteen inches from the ground. This nest was about four inches in diameter, and composed of dry grass and leaves. I set a trap below the bush and caught a male Bank Vole. Half a mile from here, on the same day, I found another round nest, also in a bramble bush; this nest, with the addition of some bracken to the grass and leaves, was, like the previous one, about eighteen inches from the ground and four inches in diameter.

These last two nests, to the casual observer, might easily be mistaken for small wrens' nests, which they very much resemble, and passed as such. On some waste land, near the spot where I had made my first find, I discovered a nest, oval in shape and three inches long. This was in a gorse bush, and about a foot from the ground. It was composed entirely of dried grass.

No apparent entrance exists in these nests. The voles evidently pass in and out where the wall is thinnest, and "pull the door to" after them.

In the Autumn of the same year I was again staying at the farm, and resumed my searches, but in another direction. On Oct. 1st, I was about a mile from the farm, in a lane bordered on either hand by a growth of shrubs, brambles and gorse. From one of the gorse bushes there sprang a Bank Vole, which disappeared amongst the undergrowth. Examination revealed, hidden in a dense bush, and about three feet from the ground, the nest shown in Fig. I. It is oval in shape, three inches long, and composed of withered grass and leaves. Here there is a clearly defined entrance at the far end, with a short way to it through the gorse. In a similar lane, near at hand, I found, on Oct. 6th, yet another nest, also in a gorse bush, but not so well concealed. This nest was also three feet from the ground, and three inches in length, but was composed of grass only. The entrance can be seen easily (Fig. 2).

One outstanding feature is that the nests in the bramble bushes are round, whilst those built in the gorse bushes are oval in shape. Barrat-Hamilton in his book, The Mammalia of Great Britain, states that the "nurseries" (i.e., breeding nests) of the vole may be usually found above ground, in hedgerows or hayfields, and also speaks of elevated nests. Owing to his death, it is impossible to know what he meant by this. The only figured "nursery" in his book is shown below ground. I am inclined to think that they are sleeping places, for they seem too small to hold a family. I believe they have not hitherto been recorded, and will bear further investigation.

I have presented a round and an oval nest to the Liverpool Museum; an oval one to the Manchester and to the Edinburgh Museums; and the nest built in the whitethroat's nest to the museum at Warrington.

N.B.—Further investigation since the above note was read throws some doubt upon the nests being made by the vole. It is possible that they may have been made by the dormouse.





Fig. 1 Fig. 2



Fig. 3

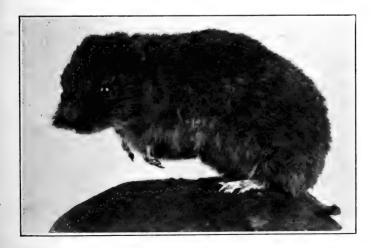


Fig. 4



# NOTE ON A WHITE ORKNEY VOLE MICROTUS ORCADENSIS, V. ALBA.

#### By GEORGE ELLISON.

The Orkney Vole (*Microtus orcadensis*), known locally as the "Cuttick," occurs in well-worn runs, tunnelling for long distances under the long grass. Mr. J. G. Millais (son of the famous artist) kept this form under observation from 1886 to 1904. He published his results in the *Zoologist*, for July, 1904, and the animal was accepted as a new species of British mammal. The vole is found on the mainland (Pomona), and several of the islands, and a sub-species differing in colour in the Island of Sanday.

The accompanying photograph (Fig. 4) shows the typical Orkney Vole, and Fig. 3, a white variety, sent to me in Oct., 1917. It is the only white specimen recorded from Orkney, and is an immature male caught near Stromness. It measures 3\frac{3}{8} inches from tip of nose to tip of tail. Instead of the pink eyes, usual in an Albino, this one had black eyes, and is possibly one out of a litter of several.

It may be seen in the the Stromness Museum, together with some black specimens (melanic), which so far have been found locally, only in the Hills of Harray on the Mainland of Orkney.

# SOME NOTES ON THE RATS OF THE PORT OF LIVERPOOL.

#### By J. W. CUTMORE

Assistant in Vertebrate Zoology, Free Public Museum, Liverpool

Much has been said and written about the large number of Rats found in Liverpool, and of their depredations; but, I think, no investigations have been made concerning the number of species and varieties, and the habits peculiar to each species. Through the help of the Staff of the Port Sanitary Authority we have been able to make certain investigations, and collect some facts. We have also been able to obtain living specimens for experimental purposes. There are three species of Rats of the *Mus rattus* type found on ships visiting the Port of Liverpool, also another and larger species *Mus norvegicus*, found chiefly in warehouses and sewers.

The Mus rattus group (or Epimys rattus, the name now generally adopted) is divided into three species:—

The Black Rat, Mus rattus rattus.

The Alexandrine Rat, M. r. alexandrinus.

The Tree or Roof Rat, M. r. frugivorus.

These are small rats with very long tails. The average dimensions are head and body,  $204\,\mathrm{mm}$ .; tail,  $235\,\mathrm{mm}$ .; ear,  $22\,\mathrm{mm}$ .; hind foot,  $32\,\mathrm{mm}$ .; the weight is 7 oz.

The Sewer Rat, Mus (Epimys) norvegicus, is a stouter animal: head and body, 240 mm.; tail, 204 mm.; ear, 20 mm.; hind foot, 36 or 40 mm.; weight, over 16 oz. It is seldom found on ships.

The following are the results of a series of catches on miscellaneous ships arriving in the Port. Out of 1,500 rats there were 576 Black, M. r. rattus; 700 Tree Rats, M. r. frugivorus; 204 Alexandrine Rats, M. r. alexandrinus; and

20 Sewer Rats, M. norvegicus. Other names for this last species are "Wharf Rat," "Barn Rat," "House Rat," "Water Rat," "Hanoverian Rat." It is abundant everywhere in the British Isles. A melanic variety is found in Ireland, known as the Irish Rat. It is undoubtedly the original of the Domestic tame white Rat. I have at present a Mus norvegicus paired with a tame white Rat, and hope to confirm this statement. Mus norvegicus is readily distinguished from the Mus rattus group by its stouter build, shorter and thicker ears, and its tail which is shorter than the body. The skull is larger, and differs in form in the parietal and inter-parietal regions. It is not so good at climbing, and prefers the addition of flesh to its diet.

The general colour of *Mus r. rattus* is slaty—darker on the back, paler below; some specimens are of a rich black. These were described by Millais as *Mus r. ater* (*Zoologist*, 1905).

Mus. r. alexandrinus only differs in coloration—the upper parts brown and ventral slaty or light brown.

Mus r. frugivorus has the upper parts grey or brown, sometimes russet; under parts pure white or pale yellow, the line of demarcation sharply defined along the flanks.

In Britain, Mus r. rattus was formerly wide spread both on the mainland and islands. It has been carried by shipping from Europe to all parts of the world.

Mus r. alexandrinus is introduced from ships.

Mus r. frugivorus is the common rat of the Mediterranean region and N. Africa. It has a world-wide range. We found it on the Island of Socotra, where it bred true to type, there being no other rats in the Island.

The appearance of *Mus norvegicus* in a district is generally followed by the disappearance of mice and the smaller species of rats.

I find all the varieties clean animals to keep. My results in cross-breeding have not yet reached their final stage. So far

I have failed to procure any results from crossing M. norvegicus with M. rattus. I think no result has ever yet been obtained.

My results from crossing M.r. rattus (Black Rat) with both M.r. alexandrinus and M.r. frugivorus have been successful, producing about an equal number of each colour, with this exception. The pair, Black male M.r. rattus and Brown dark bellied female M.r. alexandrinus, produced one white-bellied brown M.r. frugivorus in each litter.

Dr. Bonhote found on crossing M. r. frugivorus with M. r. alexandrinus that the former was apparently a simple mendelian dominant to the latter.

The experiment of de L'Isle seem to indicate that M. r. rattus behaves in turn similarly as a dominant to M. r. frugivorus.

Morgan's experiments (American Naturalist, 1909) showed that which ever way the cross was made the progeny of M.r. frugivorus  $\times M.r.$  rattus were black.

Hagedoorn (American Naturalist, 1917) found that the Black  $M.\ r.\ rattus$  behaves as a simple dominant.

My experiment is now in the Mendelian stage F. 2. I hope to publish the results later.

THE INVESTIGATIONS REPORT ON CARRIED IN CONNECTION WITH ON DURING 1918 THE SEA-FISHERIES LABORATORY LANCASHIRE ATTHE 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.

	CONTENTS.	PAGE
1.	Introduction. By W. A. Herdman	71
2.	On the Monthly Occurrence of Pelagic Fish Eggs. By A. Scott	85
3.	Intensive Study of Irish Sea Plankton. By W. A. Herdman, A. Scott, and H. Mabel Lewis	95
4.	The Dietetic Value of Sprats and other Clupeoid Fishes. By J. Johnstone	106
5.	The Probable Errors of Bacteriological Analyses By J. Johnstone	134

#### INTRODUCTION.

Although hostilities have now happily ceased and assured peace is within sight, the year to which this Report refers is still a year of war, and the work has been carried on under war conditions. The steamer is still in Government service and our scientific fisheries work at sea is, therefore, still impossible; but the reduced scientific staff has carried on such work as could be done in inshore waters, along the coast and in the laboratory, and that is what is reported on in the following pages.

Mr. Scott has been engaged on his usual useful work at the Piel laboratory, and that requires no further comment. He contributes to this Report an article on the monthly occurrence of pelagic fish eggs in the Port Erin plankton.

Dr. Johnstone, as in the previous two years, has been lent for a portion of his time to the Board of Agriculture and

Fisheries, in order to carry on for that Government department investigations of national importance on the best methods of utilising the immense shoals of sprats that frequent our coasts at certain times of the year. Through the courtesy of the Assistant Secretary at the head of the Fisheries Division of the Board we are permitted to publish in this report so much of Dr. Johnstone's work as is completed. Dr. Johnstone also contributes an article on some bacteriological methods.

With the assistance of Mr. Scott and Miss Lewis I have been able to draw up a further instalment of observations on the distribution of the plankton of the Irish Sea throughout the year, with some conclusions as to which are the dominant organisms of most importance in connection with the food of young fishes.

I sincerely hope that before the next of these Annual Reports is due to appear, normal scientific fisheries work will have been resumed throughout the Lancashire and Western Sea-Fisheries district. It is with the view of pointing out the importance of losing no time in the necessary reconstruction of our staff and full programme of work that I have written the following paragraphs.

#### RECONSTRUCTION.

In the period of reconstruction and necessary development of all material resources that lies before us, Sea-Fisheries Research and Education must play an important part. The industries, such as those of Agriculture and Aquiculture, which have to do with the production of food-matters for the people must ever be of prime importance, and anything that affects their prosperity ought to receive full consideration and public support.

It is a notable sign of the times that in a "Memorandum" for the consideration of the Minister of Reconstruction, the Scottish Steam Drifters Association representing the Herring

Fishing Industry of Scotland, lays special emphasis on the need for Fishery *Research* and Fishery *Education* in order to ensure the prosperity of the industry.

All Fishery statistics show that Great Britain in the years before the war occupied a very dominant position in the fisheries of North West Europe. For example, in the year 1912 this country's share of the produce of the North Sea represented over 68 per cent. of the total catch. In Herrings alone, Great Britain's share was 70 per cent. of the whole. Now, these practical men, the Scottish Herring Fishers, in this Memorandum drawn up with a view to "the reconstruction of the fishing industry after the war," give as their considered opinion that "If Great Britain is to hold her present position in the Fisheries of Northern Europe it can only be done by providing higher education and scientific knowledge for those intending to enter the Fishing Industry, and closer co-operation between the expert and those carrying on the Industry."

There are other notable passages expressing sound and enlightened views in the Memorandum, such as "Pure science is the fountain from which applied science draws its life and force," and again "The Fishing Industry . . . has now reached such dimensions and is so complex in its activities as to demand special educational facilities on the same lines as is provided for agriculture." "The managers of the future," they point out, "engaged in the various departments of fishery enterprise have not time to serve an apprenticeship in all the branches of business which they require to know . . . To meet the wants of these managers, of all who mean to hold high place in the Fishing Industry, of fishery officers, and of teachers for coast schools, it is now a clamant necessity that suitable provision should be made."

These and other considerations lead up to the demand "That a College of Fisheries should be established in Scotland on the same lines as the College of Agriculture," and it is

recommended that the College should be established in Aberdeen in definite and organic connection with the University.

The avowed object is naturally to promote the Scottish Fishery industries, but the wider application of their views is recognised both in relation to research and education. recommend that "Each [Government] Department should have power to enter into arrangements with any Educational Institution whereby facilities should be afforded to such Institution for Higher Scientific Education in the laboratories and by the staff of the Department"; and that "Research and Education should proceed in matters affecting the preservation and development of the fisheries, the distribution, growth, and habits of fish, the catching, curing, and preserving of fish (including refrigeration and cold storage), the utilisation of fish, including by-products, and the marketing and transport of fish, including refrigeration." Moreover, they emphasise the point that "The combining of Fishery Research with the practical conduct of the Industry is of such vital importance that centralisation of Fishery Research in London could not be accepted"—a statement that may possibly require some expansion and definition before it can be universally accepted.

There are many other matters dealt with in the Memorandum in relation to fishery administration and control, both in Scotland and in the United Kingdom generally, which do not concern us at the moment; but the final statement that "The Association are convinced that if the schemes for Education and Research as outlined above are carried out, the Scottish Fishing Industry will continue to hold its high place among the nations of Europe" must not be allowed to apply to Scotland only. The quotations I have given in regard to the value of research and education to the further development of the fishing industries are of general application and should receive careful consideration from every responsible and enlightened Sea-Fisheries Authority.

Whatever may be the future of Sea-Fisheries administration and control in this country, and there are various more or less conflicting schemes in print and in the air at present, there can be no doubt as to the value and urgent need of more, and still more, research and education. If that great need be admitted, it is surely foolish, if not criminal, to delay until more comprehensive or ideally perfect schemes can be evolved. Neither research nor education need be confined to one or a few national centres. Wherever and whenever research is possible all right-minded men will say let it be carried on, and surely education in fishery matters is desirable at every great fishing port.

The Lancashire and Western District has in the past made at least a good beginning in both fishery research and fishery education. For many years the scientific staff has organised and conducted practical classes in marine biology and navigation for fishermen at the Piel laboratory and hatchery near Barrow, and up to the summer of 1914 the scheme of scientific work at sea and in the University Department at Liverpool was constantly enlarging in scope and improving in methods. War conditions and restrictions necessarily put an end for the time being to both fishermen's classes and research work at sea. The reports on the scientific work for the past four years show that with reduced grants, depleted staff and restrictions at sea it has only been possible to carry on a portion of the investigations—that portion, in fact, which can be done in the laboratory or on the sea-shore. A great gap of over four years at least thus exists in our series of accumulated observations and statistics at sea-a gap which ought not to be allowed to become any greater. At the earliest possible moment after the return of the Lancashire Sea-Fisheries steamer from her special Admiralty duties to her normal civil life the full programme of scientific work at sea ought to be resumed.

The necessary preliminary steps to be taken are :-

- 1. The renewal of the grants for scientific work which were obtained previous to 1914 with such increase as may be required under present conditions;
- 2. The re-appointment of the necessary assistants, so as to fill up the depleted scientific staff to at least the degree of efficiency supported by the Development Commission and approved by the Board of Agriculture and Fisheries in the programme of work printed in the Introduction to the Report for 1912;
- 3. Such revisal of the programme of scientific work—including observations and statistics at sea and laboratory work at Piel, Port Erin and Liverpool—as may seem desirable in view of the break in our records, the work that has been done since both here and elsewhere, the new developments that have taken place in connection with sea-fisheries and the possibility of future consolidation and expansion of scientific work in the Irish Sea under a University department of Oceanography.

In regard to this last point (3), the scientific staff are prepared to do their part whenever required, and it is hoped that the Committee will, as soon as may be practicable, take the necessary steps to secure the renewal of the grants and the re-appointment of the Assistants. If the Lancashire and Western Sea-Fisheries Committee is to regain and maintain the position it held in relation to the scientific exploitation of the district and the Fishing Industries under its charge, no time should be lost in resuming active work at sea and in promoting a sound progressive policy in both Fishery Research and Education.

In response to a recent request for a Memorandum designed to give an outline of the scope and nature of sea-fisheries research, and its relation to administration, as the result of our experience in this district, the following paragraphs have been drawn up in consultation with Dr. Johnstone, to whom I am indebted for many of the examples. It may be useful to place these views on record at this time when an increase in fisheries research is keenly desired and strongly advocated, and when schemes for the promotion of such work are being freely discussed in many quarters. After reading and carefully considering several such schemes I am led to the conclusion that there are good points in all, that they need not be mutually exclusive, but might, in part at least, be usefully combined. After all, men are more important than measures. Especially in such a growing ever-changing business as sea-fisheries research and oceanography it is more important to have the right men at work directing the progress and co-ordination of the investigations than to insist on having from the first an ideally perfect scheme.

Another point that seems clear is that all possible use should be made of existing institutions before running any risk of duplicating laboratories, marine stations, equipment, men and their efforts.

#### MEMORANDUM ON FISHERIES RESEARCH.

Sea-Fisheries Research is of a most varied nature embracing, as it does, the subject matter of different sciences (Biology, Chemistry and Physics), and requiring in some cases the co-operation of several kinds of scientific men. This indicates at once the conclusion that much of it can best be carried out at institutions of University rank, where laboratories and libraries of the various sciences are provided and where many scientific experts are available for consultation.

It is possible, however, for practical purposes, to divide these varied researches into two main categories:—

- (1). Those investigations which are directed towards supplying information required immediately for administrative purposes and for advancing the fishing industries; and
- (2). The more academic researches which may not be obviously of direct practical importance, but which aim at extending knowledge by way of filling up gaps, affording explanations of ascertained facts and obtaining scientific evidence as to conditions of life in the sea.

It is difficult, however, to draw a strict line of demarcation between these two categories. The academic research may at any moment lead to some conclusion of direct practical importance, and the investigation of some industrial or administrative point may branch off into an inquiry of a purely scientific nature.

Consequently, it is no easy matter to decide how far research should be combined with administration, and which particular researches should be relegated to University laboratories and Biological Stations.

Moreover, as much sea-fisheries investigation must be of a local character and be carried out locally, it is difficult to determine to what extent researches around the whole coast should be controlled by a central Committee or may be left in the hands of competent local organisations.

To take a concrete example, the scientific fisheries work carried out at the University of Liverpool and in the Irish Sea in connection with the Lancashire and Western Sea-Fisheries Committee during the last twenty years, has included both categories of research mentioned above, and has been in close relation with both the Administrative Authority and the University laboratories.

- (1). Under the first category may be grouped the following, amongst other, investigations:—
  - (a) Trawling observations and experiments with various nets on different grounds, measuring and marking experiments with fish set free and subsequently re-captured;
  - (b) Researches into the life-histories of fishes, their rates of growth, spawning periods, seasonal migrations, feeding grounds, etc.
  - (a) and (b) involve work at sea. These investigations were devised for the purpose of ascertaining the need of regulations or to test existing bye-laws. Without such experimental results the statistical data collected officially are of little use as a guide to administration.
  - (c) Plankton work so far as it includes a study of the distribution and occurrence (both quantitative and qualitative) of fish eggs and larvae.
  - (c) also involving work at sea, gives valuable information as to the spawning grounds of fish, their seasons and variations in relation to physical conditions and the fate of the young fish.
  - (d) The study of sewage pollution with respect to the shell-fish beds, including both topographical surveys and sampling on the shore and the subsequent bacteriological research in the laboratory.
  - (d) is necessary for sound administration which ought to involve not merely restriction, but also measures of cultivation and purification. The Local Government Board and the Local Health Authorities may take steps to prevent the distribution of polluted shell-fish, but this is not enough. Such administrative

action merely prohibits and punishes and is not constructive and productive. It is only the Sea-Fisheries Authorities (Central or Local), so far, that have attempted constructive work in the interests of the industry, and all such work must be based upon detailed scientific local investigation.

These are fair samples of research having a direct bearing upon administration: no doubt other examples from other parts of the coast could be given.

- (2). On the other hand there are many researches of less direct and immediate interest to the administrator but possibly of great value in the end, such as:
  - (a) Purely oceanographic investigations such as the study of drifts and currents, the distribution and variation of salinities and temperatures and other seasonal and annual changes in the physical and chemical characters of the water. All such work has to be done partly at sea on research vessels (possibly the same vessels carrying on the work under 1, a, b, c, above), and partly in the University laboratory or at a well-equipped Biological Station.
  - (b) Life-histories in the technical, zoological, sense—that is the study of embryonic and larval stages of marine edible or economically important animals, and also morphological studies of the adults.
  - (c) Faunistic research, such as determining the associations of edible and inedible animals and plants on various grounds, and investigating their relations to one another as food, as enemies and as competitors in the struggle for existence. Also the determination of the amount of fish food at different localities and seasons. This is an example of a natural-history research which might easily lead

when a little more advanced to results of direct value to the administrator

- (d) Plankton research in general, including cycles of production of ultimate food matters in the sea, and the bearing of plankton production upon the nutrition and the migrations of plankton-eating fishes. This again, starting as a purely academic research, might at any time lead to results of importance in administration.
- (e) Parasitology and Diseases of Fishes and other marine edible animals. In this the laboratory is sometimes brought into relation with the markets when fish condemned by the Authorities are sent for investigation and report.

These, again, are only some examples of the researches undertaken. Other subjects might, no doubt, be stated.

It may be noted that practically the same methods that are necessary to give the results required for 1, a, b, c, will supply the material for the investigations under 2, b, c, d. After the material has been collected by the research vessels and used for the purposes of 1, a, b, c, the grant-aided laboratories could take up the work and study the collections with their special objects in view. This tends to show that there is no real distinction in nature, but only one of convenience, between our two categories of research.

All the investigations mentioned in Category (1) involve work at sea in research vessels or on the shore in association with fishermen or fishery officers with expert local knowledge. All the work done is in the closest possible relation with the actual fishing industries of the district, and with the administrative staffs, both police and statistical. It is difficult to see how some, or even most, of it can be separated from this close

contact with local administration without causing it to lose much of its immediate practical value. It ought to be remembered also that the severance of all research from administration might have an unfortunate effect upon the spirit in which statutory duties are carried out by depriving it of a progressive outlook and an attitude of enquiry. In the case of the local Fishery Authorities, the combination of a certain amount of research (such as Category 1) with administration has resulted in an increase of local interest and efficiency which might be lost, if all research were centralised under the control of a Government Department or Central Council.

On the other hand such research as is included in Category (2) need not have any direct relation to administration, and much of it might well be regulated by a central department or Council of Scientific Research, so long as enlightened views in regard to the nature and results of research prevail and the laboratory workers are not tethered by red-tape and coerced as to methods and duration of work. Some of the more academic researches would, no doubt, be suggested by those working at the administrative problems under Category (1). Some (such as oceanographic and faunistic researches) would require to be done at sea from the research steamers alongside the administrative work. But that need present no difficulties. The determination of salinities and temperatures, for example, can easily be combined on the same expedition with experimental trawling and plankton work. Some of the researches under (2) would naturally be carried on at marine biological stations, such as the investigation of life-histories and work on the living plankton. The rest of the work would be done best in University laboratories. There also the results of the Oceanographic researches under both (1) and (2) would be collated and put into final form for publication.

For the direction and co-relation of the work as a whole a University with its various laboratories, libraries, museums

and experts in the different sciences involved is obviously the best centre, although much of the practical work must necessarily be done at sea or on the shore.

A Government Central Research Department or Council would perform a useful function in referring problems from the Administrative Authorities to University laboratories, and by assigning grants to the Universities and Biological Stations undertaking such research work. The Central Council might also undertake the publication of all the results of the fisheries researches in a uniform manner issuing the separate papers as soon as they are ready (as is done by the United States Bureau of Fisheries), and then subsequently in an Annual Volume.

Whether all Fisheries research, Category (1) as well as (2), should be placed under the control of a Central Research Council is a very difficult question. The removal from Administrators of all opportunities for making investigations bearing on their own regulations might be prejudicial to good administration; and, as is stated above, certain researches gain much by the close co-operation of the scientific worker with the administrative staff and the fishermen.

Purely industrial research seems to be adequately provided for by the Privy Council Committee's Scheme for "Research Associations" under the Department of Scientific and Industrial Research.

This is the last Report that I shall have the honour of submitting to the Committee as Professor of Zoology; but that does not mean that any change—save happily for the better—need be made in the direction or the conduct of the scientific work. The recent establishment of a Professorship and Department of Oceanography in the University of Liverpool is intended to consolidate and perpetuate the local marine biological and other oceanographic investigations and

their applications to sea-fisheries research. The Council of the University has invited me to occupy the Chair of Oceanography for the first year, from October, 1919, after which period of reconstruction and arrangement I shall gladly resign the Professorship into the competent hands of my colleague, Dr. Johnstone, who has for so many years been associated with the scientific work of the Lancashire Sea-Fisheries. It is the earnest hope of the founders of the Chair that the work of the Port Erin Biological Station and the scientific investigations of the Lancashire and Western Sea-Fisheries Committee may for the future be united under the Oceanographic Department of the University, and that this union may conduce to the advancement of our knowledge of the sea and its products, and the promotion of the local fishing industries.

W. A. HERDMAN.

Fisheries Laboratory,
University of Liverpool,
March 17th, 1919.

# ON THE MONTHLY OCCURRENCE OF PELAGIC FISH EGGS IN PORT ERIN BAY IN 1918.

# By Andrew Scott, A.L.S.

In the previous reports on the pelagic fish eggs from the area of the Irish Sea adjacent to Port Erin, a definite classification for the adult fishes was adopted. The general distribution throughout the year was then given under each heading. This system, no doubt, was the correct one. If, however, we wish to know what eggs are likely to occur in any particular month, it is necessary to go through the complete lists and tabulate the records. This takes time, and single records may be overlooked.

For the sake of simplicity I have made use of a rather different arrangement in this report. The number of hauls of the coarse, fine and vertical nets in each month is first given, and then the numerical distribution is set out. One sees at a glance when the eggs are likely to be present and also their relative abundance. It also shows the different catching powers of the various nets. One naturally expects that the coarser nets will catch a greater number of the larger organisms than the fine nets, especially when diatoms happen to be plentiful. Fine nets soon choke up and their fishing power much restricted. Diatoms and other microorganisms pass through the meshes of the coarse nets and the efficiency continues much longer. The vertical nets very rarely capture eggs. It occasionally happens, however, that pelagic fish eggs are captured by this method that are not represented in the horizontal surface hauls. This may be due to the eggs having acquired a slightly higher specific gravity than the surface water so as to sink to deeper layers.

Bi-weekly collections were taken in the bay by the staff of the Biological Station as far as weather and unforeseen circumstances permitted throughout the year. These were greatly augmented by the special collections taken by Professor Herdman from his motor launch working in the Bay itself, and, occasionally, as far as two and three miles outside it, during the University vacations at Easter and Midsummer. The outside collections were not very numerous, but they help to indicate how the eggs were distributed.

There is no easy and rapid method for working through a sample of plankton containing fish eggs. The work can be facilitated to some extent by washing the collection through a series of three sieves. The first one should be of brass wire, 12 meshes to the inch, which allows everything, except sagitta, crab megalopa and other larger forms, to pass through. intermediate sieve is of silk, of 36 meshes to the inch. retains all the fish eggs and plankton organisms down to, but generally excluding, anything about the size of newly hatched nauplii of barnacles. The last sieve is also of silk, 200 meshes to the inch. It retains the diatoms, dinoflagellata and all the smaller organisms caught. The residue in the three sieves is turned out into flat glass dishes, similar to those used for bacteriological cultures, mixed with a little water and placed against a black background. That from the brass sieve is taken first, and the number of sagitta, etc., noted. It may contain a few fish eggs, entangled amongst the larger things. The eggs of the long rough dab and the plaice sometimes just fail to pass through the wire gauze. The residue from the intermediate sieve is next examined, and the fish eggs are picked out by means of a pipette. Collections taken in March and April frequently contain over 100 fish eggs. The residue from the fine sieve is also examined to make quite sure that no very small eggs have escaped into it. The residue from the two silk sieves is afterwards transferred to graduated tubes, and the estimation of the organisms proceeded with in the manner described in the Report for 1907 (p. 97). The eggs require to be

examined individually. If necessary they are first separated into two groups, distinguished by the presence or absence of oil globules. Each group is next arranged in a series of rows of approximately similar size on a glass slide with the aid of a pocket-lens, and then measured under the microscope with the micrometer. The size of the oil globule has also to be measured, and its colour noted. The oil globule in the eggs of some species of fish, such as the weever and sole, is broken up into a number of small globules scattered through the yolk or arranged into groups. These details all aid in identifying the species of fish. None of the pelagic fish eggs, with the exception of perhaps the long rough dab and the plaice, can be identified without careful examination. The eggs of the long rough dab can be determined almost at a glance by the large size and peculiar appearance of an egg within an egg, due to the large perivitelline space. The plaice egg is also large. It has a thick corrugated capsule. After some practice it is possible to separate the eggs of the dragonet, even with the pocket-lens. In all cases it is advisable to examine the eggs with the microscope, and measure them with the micrometer.

### JANUARY

Material: Coarse net, 6; Fine net, 6; Vertical net, 6 = 18 hauls.

Rockling eggs Green cod eggs		•••	•••	Coarse. 4 1	Fine. 0 0	Vertical.  0 0	Totals. 4
Totals	•••	•••		5	0	0	5

These eggs were only present once in the January series. The rockling eggs occurred on January 21st, and the green cod on the 30th. In some years no pelagic fish eggs have been obtained in this month. In others, we have found rockling eggs as early as January 2nd.

FEBRUARY.

Material: Coarse net, 7; Fine net, 7; Vertical net, 7 = 21 hauls.

				Coarse.	Fine.	Vertical.	Totals.
Rockling eggs	•••	• • • •		38	-11	0	. 49
Green cod eggs			•••	6	1	0	7
Plaice eggs	•••	. • • • .	•••	0	. 0	1	1
Totals		•••	•••	44	12	1.	57

A slight increase in the number of pelagic eggs usually occurs early in February, and becomes more marked towards the end of the month. The species of fish represented do not appear to materially increase. We find this to be the case year after year, and only about three or four species are represented by their eggs in the plankton. The coarse nets caught nearly 6·3 eggs per haul, and the fine nets nearly 1·7 eggs per haul. The average catch of the coarse and fine horizontal surface hauls amounted to 4 eggs for each combined sample. The rockling eggs were fairly uniformly distributed throughout the month. The single specimen of the plaice egg was taken on February 19th.

#### MARCH.

Coarse net, 9; r	me ne		rucar net				
,, 8;	,,	8;	,,	8; Bay			47 hauls
" 2;	,,	2:	>>	0; 2 an	d 3 miles	out	
	Bay.				Outsi	DE.	
		Coarse.	Fine.	Vertical.	Coarse.	Fine.	Totals.
Rockling eggs		1,378	506	2	14	22	1,922
Whiting eggs		243	172	2	88	46	551
Bib eggs		108	89	0	12	12	221
Cod eggs		45	40	0	12	1	98 .
Green cod eggs $\dots$		11	1	0.	0	0	12
Dragonet eggs $\ \dots$		23	7	0	2	0	32
Plaice eggs		0	0	0	2	2 .	4
Sail fluke eggs	•••	1	0	0	1,	1	3
Totals		1,809	815	4	131	84	2,843

Professor Herdman began collecting his special hauls on March 25th, and added 16 to the official set. Four hauls, two and three miles outside the bay, were also taken by him. pelagic eggs show a great increase compared with February, and average 74.7 per haul of the whole of the horizontal coarse and fine surface collections. The bay collections averaged 77 eggs each, and the outside hauls 53.7 eggs. The coarse nets captured 106.4 eggs each in the bay, and the fine nets 48 eggs each. Outside the bay the coarse and fine nets averaged 65.5 and 42 eggs respectively. Rockling eggs were more abundant in the bay than outside. The other pelagic eggs, with the exception of the green cod and the dragonet, appeared to have been better represented outside the bay, especially the whiting. No green cod eggs were identified in the outside collections. Plaice eggs occurred outside the bay only.

## APRIL.

Coarse net, 9; Fine net, 9; Vertical net, 9; Bay official

,,	17;	22	17;	,,	0; Bay			: 75 hauls.
,,	7;	,,	7;	,,	0; 2an	d 3 miles	out	
				BAY	ζ.	Ours	SIDE.	
			Coarse.	Fine.	Vertical.	Coarse.	Fine.	Totals.
Rockling eg	gs		1,329	550	4	618	141	2,642
Whiting egg	gs		314	118	0	693	123	1,248
Bib eggs			113	24	0	133	40	310
Cod eggs	•••	***	59	15	0	63	13	150
Dragonet eg	ggs		51	5	0	31	24	111
Plaice eggs	•••		2	1	0	2	-2	7
Sail fluke eg	ggs		. 3	1	0	9	3	16
Topknot eg	gs		21	7	0	22	0	50
To	tals		1,892	721	4	1,571	346	4,534

Professor Herdman completed his special collections on April 13th, and added 34 samples to the official set. He also took 14 collections at two and three miles outside the bay. The total number of eggs captured was considerably higher

than in March, but when the number of hauls are taken into account we find that there is a slight decrease in the average. The average for the whole of the horizontal coarse and fine surface nets is 68.6 eggs. Pelagic eggs were evidently more abundant outside the bay than inside. The bay collections averaged 50.2 eggs each, and the outside ones 137 eggs. coarse surface nets worked in the bay captured 72.7 eggs each, and the fine nets 27.7 eggs. Outside the bay the coarse and fine nets averaged 224.4 and 49.4 eggs respectively. pelagic eggs of the green cod (Gadus virens) were not observed in any of the April collections. The eggs of one of the topknots made their appearance in this month. The vertical hauls did not capture any eggs not represented in the surface collections either in March or April. The very few that were taken belonged to the most abundant of the series, and may well have entered the net just as it reached the surface.

MAY. Material: Coarse net, 9; Fine net, 9; Vertical net, 9 = 27 hauls.

				Coarse.	Fine.	Vertical.	Totals.
Rockling eggs	•••	•••	•••	64	. 13	1	78
Whiting eggs	•••	•••	•••	17	3	1 .	21
Cod eggs	•••	•••		1	/ 0	0	1
Haddock eggs	•••	•••	•••	. J-	0	0	1
Dragonet eggs	•••	•••	•••	17	4	0	21
Sprat eggs	•••	•••	•••	20	0	. 5	25
Topknot eggs	•••	•••	•••	21	4.	. 2	27
Totals		•••	•••	141	24	9	174

The usual official collections were taken in May. The figures show that a very considerable drop in the total number of pelagic eggs captured has taken place, and the averages are greatly reduced. The eggs of three species of fish have ceased to be represented, and two others take their place. The haddock is the most interesting of the two additions. The

movements of the adult fish in the Irish Sea are very irregular. In some years great shoals of haddock are present throughout the area, and prove very valuable to the fisherman. In other years no haddocks are captured. It will be rather interesting if the occurrence of haddock eggs in what may be regarded as the centre of the Irish Sea in the early summer of 1918 indicates an important invasion of the adults at an early date. The coarse and fine horizontal surface nets averaged 9·1 eggs for each combined haul. The coarse nets caught 15·6 eggs each, and the fine nets 2·6 eggs. The vertical hauls taken during May averaged one egg each. That is the highest number captured in any month throughout the year by this method of collecting plankton.

 ${\bf JUNE.}$  Material: Coarse net, 8; Fine net, 8; Vertical net, 8 = 24 hauls.

				Coarse.	Fine.	Vertical.	Totals.
Rockling eggs	•••		٠	66	5	1	72
Haddock eggs	•••		• • •	1	0	0	1
Dragonet eggs				10	0	0	10
Sprat eggs				7	0	3	10
Gurnard eggs				1	0	0	1
Topknot eggs	***	•••		15	1	. 0	16
Totals				100	6	4	110

These figures show a further reduction in the number of eggs present. The whiting and the cod have disappeared. One of the gurnards is now represented for the first time by a single egg captured in one of the coarse net hauls. One haddock egg was also taken in a coarse net collection. The coarse and fine horizontal surface nets averaged 6.6 for each combined haul. The coarse nets captured 12.5 eggs each, and the fine nets 0.75 eggs per haul.

Totals

### JULY.

Coarse net, 9;	Fin	e net, 9;	Vertical net,	9; Bay o		= 30 hauls.
			BAY.		SPECIAL.	
		Coarse.	Fine.	Vertical.	Coarse.	Totals.
Rockling eggs	• • •	289	32	1	82	404
Sprat eggs	• • •	2	2	0	1	5
Topknot eggs	•••	116	8	0	23	147

42

106

556

1

407

Three special hauls in addition to the official collections in the bay were taken in July. The number of species of fish represented in the plankton by their eggs has been reduced by half compared with June. The number of eggs captured in the 21 horizontal surface hauls show a very marked increase over the record of the previous month. This may be due to one or two causes,—(1) either to a prevailing westerly drift into the bay, or (2) the finer and warmer weather had induced the spawning adults to come nearer the shore, and the eggs have had less chance of being widely scattered in the area. The whole of the surface collections averaged 26.4 eggs each. The bay coarse nets captured 45.2 eggs, and the fine nets 4.6 eggs. The three special collections averaged 35.3 eggs per haul. One coarse net collection contained 132 rockling eggs and 12 topknot eggs.

#### AUGUST.

Coarse	net, 9 26 4	;			0; Bay	special	out	85 hauls.
				BAY.		Outs	SIDE.	•
			Coarse	Fine.	Vertical.	Coarse.	Fine.	Totals.
Rockling	eggs		 109	59	2	. 0	. 1 .	171
Mackerel	eggs	•••	 9	. 0	. 0	0	0	9
	Totals		 118	59	2	0	. 1.	180

Professor Herdman took a second series of special hauls in August and September. He commenced work on August 9th, and added 52 to the official set. He also took 6 hauls two and three miles outside the bay. The area was therefore well investigated. The pelagic eggs were apparently confined to the bay as only a single rockling egg was captured by the 6 hauls taken outside. The combined coarse and fine horizontal surface hauls in the bay averaged 2.5 eggs each. The coarse nets captured 3.3 eggs each, and the fine nets 1.6 eggs.

## SEPTEMBER.

Coarse net, 8;	Fine net, 8;	Vertical net, 7;	Bay official	= 73 hauls.
,. 25;	,, 25;	,, 0;	Bay special	= 15 nauis.

			Coarse.	Fine.	Vertical.	Totals.
Rockling eggs		•••	 7	4	0	11
Mackerel eggs	.***	• • •	 1	0	0	1
Totals	•••	•••	 8	4	0	12

Professor Herdman's series of special hauls came to an end on September 26th. He was able to add 50 collections to the official set taken in the bay. No hauls were taken outside the area. The pelagic eggs have become very scarce, and were only represented in six coarse net collections and two fine net collections. One coarse net collection contained three rockling eggs. Two fine net collections contained two eggs each. The average for the whole horizontal surface collections is 0·18 eggs. The coarse nets averaged 0·24 eggs, and the fine nets 0·12 eggs respectively.

#### OCTOBER.

Material: Coarse net, 6; Fine net, 6; Vertical net, 4 = 16 hauls.

Results: No fish eggs.

### NOVEMBER.

Material: Coarse net, 8; Fine net, 8; Vertical net, 8 = 24 hauls.

Results: No fish eggs.

## DECEMBER.

Material: Coarse net, 7; Fine net, 7; Vertical net, 7 = 21 hauls.

Results: No fish eggs.

From the above records it will be seen that pelagic eggs were present in the plankton of Port Erin Bay for nine months The maximum occurred in March. out of the twelve. The increase at the beginning was very rapid, rising from 0.83 per haul of the coarse net in January to 106:4 in March. reduction set in during April, which was continued to June. A well defined increase took place in July, which was largely due to the presence of the eggs of two species of fish. After that the pelagic eggs ceased to be conspicuous amongst the plankton organisms, and finally disappeared in September. The fine nets although catching many fewer eggs show the same rise and fall that is revealed by the coarse nets. The hauls taken outside the bay indicate that the maximum was rather later there than nearer the shore. The coarse net collections two and three miles outside in April captured 224.4 eggs each against 72.7, and the fine nets 49.4 against 27.7 in the bav.

AN INTENSIVE STUDY OF THE MARINE PLANKTON AROUND THE SOUTH END OF THE ISLE OF MAN.—PART XI.

By W. A. Herdman, F.R.S., Andrew Scott, A.L.S., and H. Mabel Lewis, B.A.

This research is extending over a longer period than was at first contemplated, but new developments of the work have opened up and the value of cumulative evidence has been impressed upon us, and, moreover, the work has been greatly interrupted and hampered during the last four years of war. Up to 1914 eight Annual Reports upon our observations were published in full. The Reports (Nos. IX. and X.) for 1915 and 1916 were issued in brief abstract only. In the Report for 1917, we had no separate article on the Plankton work, but gave merely a couple of paragraphs in the "Introduction" on the Sea-Fisheries investigations in general. Consequently, there is now a considerable accumulation of material in the form of statistics of hauls, notes, curves, &c.; but the time has not yet come for a full and final discussion of these data, so we propose in the present Part to deal mainly with the results obtained in 1918, and the research will in all probability be wound up in next year's Report.

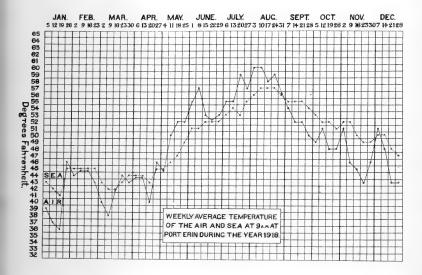
The work during 1918 has been carried on in the same manner as in previous years, so as to be comparable, and 459 samples have been collected, preserved and examined. These bring the total number of samples for the 12 years' work up to 5,962. In most months of 1918 well over 20 samples were taken, and in the more critical months (March, April, May, July, August and September) there were many extra hauls. The work at sea was carried out from the 27-foot motor-boat "Redwing," fitted with a 7 horse-power Kelvin motor, which

is found to be a very suitable and handy boat for all such work within a few miles of the shore in ordinary weather. The coarse and fine standard horizontal nets used (of No. 9 and No. 20 grades of millers' silk, respectively) were towed, simultaneously, one on each side of the stern, and worked within a few feet of one another. They were frequently renewed, so as to be constantly in good condition.

The full results of the work have been written out in 459 lists, analysed and synthesised into eight tables of statistics, and represented graphically by fifteen sheets of curves, in some of which six or eight organisms (such as the prevalent Diatoms or Copepoda) are shown in the one diagram. It is hoped that all these data may be printed eventually, but in the meantime, for reasons of economy, we must be content to publish the following summary of the leading characteristics of the year's work, with some conclusions bearing on the connection between plankton and the sea-fisheries which naturally follow.

In 1918 the spring maximum of the total plankton was again in May, when the month's average haul was 85.8 c.c., and the largest individual catch 158 c.c., on May 9th. Diatoms show a regular rise in the monthly average from 65,000 in January to  $26\frac{1}{2}$  millions in May, then a rapid decline to the minimum of over 13,000 in August, a second rise to over 319,000 in October, and finally, a fall to the winter condition at the end of the year-which, however, is not so low as the summer minimum. The curve of these monthly averages is beautifully regular and typical, with its greater vernal and lesser autumnal maxima; and it is noteworthy that Diatoms are present in greatest abundance when the sea is nearly at its lowest temperature, and are at their minimum in the warmest month of the year (see accompanying temperature chart for 1918). The greatest individual number of Diatoms was over 73 millions in one haul on May 21st.

The monthly average curve for the total Dinoflagellata is not so regular, and reaches its maximum of nearly 225,000 in July, the largest individual haul being 543,000 on July 1st. These are relatively high numbers for the Dinoflagellata, and are chiefly made up of Ceratium tripos which is at least four times as abundant as the species of Peridinium taken together. The monthly average curve for Peridinium rises gradually from units in January through tens, hundreds, thousands and tens of thousands to 50,000 in June, and then declines to a minimum of 100 in September, with a slight secondary rise in



October, falling to the end of the year in a most regular manner. *Ceratium*, with a monthly maximum of 180,000 in July, is not so regular, but shows the same form of curve.

The highest monthly average for the Copepoda (both in the adult form and as Nauplii) was in August (when the Diatoms were at a minimum), although the September averages were nearly as high, and the greatest individual Copepod haul was 191,288 on September 6th. Looking into the statistics for the individual more important genera of Diatoms, we find that the numbers of *Biddulphia* were very much higher this spring than we have ever seen them before in the 12 years of this intensive work, and reached a maximum of 1,389,000 in one haul on April 18th. The interesting form *Biddulphia sinensis* was very variable in character this year, and showed again figures such as we gave in the Report for 1912.

Chaetoceras was as usual very abundant, with a high monthly average of nearly 22 millions in May. As an example of the way in which a single form may on occasions become dominant and almost monopolise the plankton, we find that in the haul of May 21st, out of about 58 millions of Chaetoceras over 51 millions belonged to the one species C. debile. Rhizosolenia usually attains its maximum in June, but this year it was in May, fully a month earlier than it has ever been before in our experience. The great crest of the Chaetoceras curve, extending from the middle of April to late in May, covers the maxima of five out of the remaining six selected genera. Guinardia has the only maximum that is outside the Chaetoceras curve.

In a survey of the monthly averages of the seven more important genera of Diatoms we find that *Biddulphia* and *Coscinodiscus* reach their spring maxima in April, while the other five, *Chaetoceras*, *Rhizosolenia*, *Thalassiosira*, *Lauderia*, *Guinardia*, have the maxima in May.

Amongst those Copepoda which are of greatest importance in connection with the food of fishes, we find that *Calanus finmarchicus*, which seems to be associated with the local mackerel fishery, attained its greatest numbers in July this year, but remained in fair abundance up to the middle of September. *Temora longicornis*, which we found in 1917 to be definitely related to the summer herring fishery off the Isle of Man, showed in 1918 a perfectly regular curve of monthly

averages rising from zero in January to a maximum in June, and then falling to the minimum at the end of the year. The greatest number in one haul was over 8,000 on June 27th.

"Temora longicornis is, on occasions, one of the most abundant of our Copepoda in the Irish Sea, and must be of considerable importance as a food for fish, and especially for the herring in summer. In the latter part of July and the first half of August, 1917, the shoals of herring to the west of the Isle of Man came unusually close to land, and even penetrated into bays and creeks; and during this time they were feeding mainly, if not wholly, on Temora. Late in July this Copepod

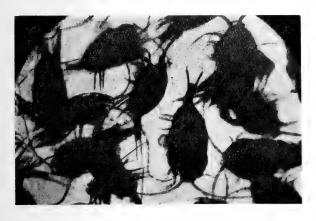


Fig. 1. Temora longicornis, from the "red patches." From a photo-micrograph by A. Scott.

was so abundant that its presence caused large patches of a red colour on the surface of the sea, off Port Erin and around the Calf Island. These red patches were noticed by the fishermen, and were spoken of amongst them as being 'fish-food' or 'spawn.' A large jarful from such a red patch, obtained by one of the fishermen, was brought to the laboratory and found to be swarming with small Copepoda, which on examination proved to be almost wholly *Temora longicornis* (fig. 1).

About one-fourth part of the contents of the jar was preserved, and on being counted later on was found by Mr. Andrew Scott to amount to 50 c.c. of Copepoda, consisting of 33,340 *Temora* and 2 *Calanus*. Mr. Scott estimated the oil present in 9 c.c. of the dried *Temora* at 2.47 per cent. of the weight, which was 0.925 gramme.

"During this same time the men were catching herring in quantity unusually close inshore in the neighbourhood of the red patches, and on examining, in the laboratory at the Biological Station, the stomach-contents of a number of these herrings, I found in every case that the stomach contained

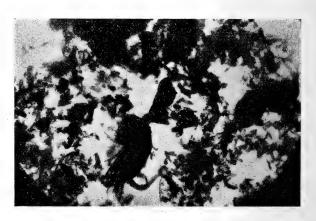


Fig. 2. Temora remains from the stomachs of the Herring, From a photo-micrograph by A. Scott.

a mass of red material which was obviously, under the microscope, the broken-down remains of Copepoda. A few Crab zoea were recognisable, but the bulk of the material consisted undoubtedly of the Copepoda. Mr. Scott examined 5 c.c. of the stomach-contents for me, and found that it contained 975 easily recognisable specimens of *Temora*. A photograph (fig. 2), which Mr. Scott has made from one of the microscopic preparations, shows appendages that undoubtedly belong to this Copepod, while here and there in the stomachs complete

specimens of *Temora* are to be seen. It is not possible to doubt that during these weeks, at the height of the summer herring fishery in the Irish Sea, the fish were feeding mainly upon this species of Copepod.

"We recorded a similar occurrence off the Lancashire coast a few years ago, when in July, 1913, at the time of an abundant mackerel fishery off Walney Island, the stomachs of some of the fish were found to be full either of *Temora* alone or of *Temora* mixed with *Isias* and a few other Copepoda. A few herrings from the Port Erin fishery of July, 1916 were found by Mr. Scott to be feeding mainly on *Calanus*." \*

Amongst note-worthy large catches of the other more important Copepoda, we find :—

Pseudocalanus elongatus—34,800 on March 28th and 22,200 on September 6th.

Oithona similis—60,100 on July 15th; 53,600 on August 5th; 88,800 on August 8th and 51,410 on December 21st.

Paracalanus parvus—71,300 on September 6th.

Acartia clausi—62,200 on September 6th; 37,600 on August 5th and 39,000 on August 22nd.

In December there was a decided invasion of our district by the minute northern and deep-water Copepod *Microcalanus* pusillus, the distribution of which was discussed in one of our former Reports.

Turning now to other less important groups of the plankton, we find that Oikopleura was, as usual, present throughout the year, and was especially abundant in June and August. The greatest number recorded for one haul was 26,000 on June 6th. In August the cast-off "houses," or cuticular investments, were so prevalent in the plankton of Port Erin as to clog the nets and increase greatly the difficulty of picking out and counting the other organisms.

In regard to the various larval forms:—Echinoderm
\* Herdman, Spolia Runiana III, Journ. Linn. Soc. Bot., XLIV, p. 199 (1918).

larvae reached a maximum of 8,700 on March 25th and were also abundant in September. Polychaet larvae were especially abundant in the first half of the year; amongst the largest hauls were 147,600 on March 1st and 190,000 on April 9th. Balanus nauplii were most abundant from middle of March to late in April (299,230 on April 22nd), while the Cypris stage ranged from middle of April to the end of May. Crab zoeas reached their maximum at the end of March (810 on March 28th). Molluscan larvae were present in all months, but most abundantly in spring, and again in the latter part of the year. The largest haul is 130,200 Lamellibranch larvae in April.

Fish eggs ranged from February to August inclusive, with the maximum at the end of March; but the fish eggs of the plankton throughout the year have been discussed in a separate article by Andrew Scott, at p. 85.

One conclusion that is becoming clear from our accumulated observations of the last ten years is the surprisingly small number of different kinds of organisms-both plants and animals—that make up the bulk of the plankton that is of real importance in relation to fish. Our food from the sea seems to depend, in great measure, ultimately upon comparatively few species of Diatoms and Copepoda respectively. A very large proportion of the Diatoms in the spring plankton and of Copepoda in that of late summer belong in each case to a very few different kinds, so that one can select about half-a-dozen species of Copepoda which constitute by far the greater part of the summer zooplankton and about the same number of Diatoms which similarly make up the bulk of the spring phytoplankton year after year. These few species, belonging to these two very widely separated groups, thus come to be the most significant organisms in relation to the annual metabolic cycle of our seas and the food supply from our coastal fisheries. It is this fact that gives great economic importance to the attempt to determine with as much precision as possible

the times and conditions of occurrence of these dominant factors of the plankton in an average year.

Our confidence that these samples upon which we are reporting in this series of papers are adequate and representative, receives support from the fact that the same organisms are recorded in much the same quantities year after year, and that practically no new forms that really matter turn up. Rare species are no doubt of scientific interest, but it cannot be too emphatically stated that it is the common species that are of most importance, those species which by their abundance in nature play their part in providing fish-food for man or in affecting the public health either by keeping the sea clean or by causing plagues.

For the purpose then of arriving at conclusions as to the distribution throughout the year of these really significant organisms, we may pick out from our records the following six species of Copepoda as being undoubtedly the most abundant and economically the most important representatives of that section of the plankton:—Oithona similis, Pseudocalanus elongatus, Acartia clausi, Temora longicornis, Paracalanus parvus and Calanus finmarchicus. In the case of the Diatoms it is better to deal with genera, and we may choose the following as being the most important representatives of the Diatoms in our plankton:—Biddulphia, Coscinodiscus, Chaetoceras, Rhizosolenia, Thalassiosira, Guinardia and Lauderia. In some of these genera, such as Chaetoceras and Rhizosolenia, there are usually several allied species occurring together in any large gathering.

Now, these are the selected Copepoda and Diatoms to which we have been paying special attention for the last decade, and in regard to which we have given many details in these successive Annual Reports.\* We shall, therefore, merely add

<sup>\*</sup> For further details and a summary of the whole matter see Herdman, "Spolia Runiana, III," in Journ. Linn. Soc. (1918); and for similar information in regard to the English Channel, see Lebour, Journ. Mar. Biol. Assoc. (1917).

here a few examples illustrating the abundance of some of these common forms and some characteristics of their occurrence.

The Diatoms often attain their greatest profusion successively, not simultaneously, so that single genera, or it may even be single species of a genus, make up on occasions the bulk of the phytoplankton. For example, on May 29th, 1916, the haul consisted mainly of Chaetoceras, of which genus there were 19,461,600 individuals, and of these 19,396,000 belonged to the species C. sociale leaving only 228,900 as the number of all the other Diatoms present taken together. On such occasions, and this is only one example of many, a single species makes up nearly the whole of the plankton. The maximum on the Diatom curve ranges in different years from March to May. In 1907 it was in March, in 1908 in May, and in 1909 in April. In some years the Diatom maximum may be divided into two parts, an earlier due mainly to Chaetoceras and Thalassiosira, and a later in June due to Rhizosolenia and Guinardia. A common order of succession for the species which contribute most largely to the Diatom maxima is-Biddulphia mobiliensis and Coscinodiscus (C. concinnus and C. radiatus) in early April, Chaetoceras debile in late April, Chaetoceras sociale in May, Chaetoceras teres and Rhizosolenia shrubsolei in early June, and Rhizosolenia (several species) and Guinardia in later June. The autumn Diatom maximum is constituted mainly in the Irish Sea by Chaetoceras subtile and Rhizosolenia semispina.

A comparison of the monthly averages for the selected Diatoms throughout the years shows that *Chaetoceras* and *Rhizosolenia* are by far the most abundant forms, and that sometimes the one and sometimes the other outnumbers its fellow in the proportion of nearly ten to one. But they do not occur in quantity together. *Chaetoceras* in March, April and May is succeeded by *Rhizosolenia* in May and June. A comparison of the largest hauls of Diatoms and of Copepoda in each year brings out how greatly the Diatoms outnumber

the Copepoda—in some cases in the proportion of about a thousand to one. It must be remembered, however, that the Copepoda are individually much larger and so may be of greater importance in the plankton as a food material.

In looking at the total numbers recorded of the selected Copepoda we find that *Oithona* is the most abundant and *Pseudocalanus* comes next. *Calanus* is the least numerous in individuals, but that is probably made up for by its very large size. It certainly seems to be of great importance as a food for migratory pelagic fishes. We have given an example above of the connection between *Temora* and the herring.

The Copepoda, as a whole, are a summer and autumn group, all the crests of their annual curves being found between May and October—the time of year when the post-larval fish of that season are growing up and when shoals of herring and mackerel appear in the coastal waters. The association of shoals of fish with abundance of zooplankton is the result of the fact that, in order to get an adequate quantity of planktonic food, the fish must seek out and capture the Copepoda. In other words, the plankton-feeding fish must go where the plankton is sufficiently abundant and must in their migrations follow the movements of the swarms of Copepoda.

Consequently, it is of importance to show, as we now can, that in our coastal waters at least, where the fisheries we are interested in take place, the plankton is not uniformly distributed. Many kinds of Copepoda occur very definitely in local swarms, and various localities and depths are characterised at the different seasons by particular assemblages of plankton. It is, therefore, reasonable to believe, in view of the facts given above as to the association of fish and plankton, that these variations in the distribution must have a marked effect upon the presence and abundance of, at least, such migratory fish as herring and mackerel, and also of the shoals of post-larval young of many of our other food fishes.

# THE DIETETIC VALUE OF SPRATS AND OTHER CLUPEOID FISHES.

By Jas. Johnstone, D.Sc.

## Introduction.

The following observations are published by permission of the Board of Agriculture and Fisheries. They relate to some investigations made for the Board which were begun early in 1918 as part of some war emergency proposals. Various lines of enquiry into methods of fish preservation and into the rationale of the commercial processes were marked out, but nothing need be said here about these. In the course of these enquiries, however, a number of analyses of fresh and preserved Clupeoid fishes were made and these results have general interest and may usefully be put on record. It became apparent, while these analyses were being made, (1) that there is a great lack of knowledge regarding the seasonal variation in composition of the flesh of many kinds of fish; (2) that next to nothing is known with regard to the chemical changes undergone by the flesh of fish cured, salted, smoked, hermetically preserved or generally conserved in the various ways at present practised; and (3) that even the routine methods of analyses of the tissues of fishes are imperfectly developed. The observations made here have reference mainly to these latter points.

# Material Examined.

# 1. Sprats.

Attention was first directed to the sprat which is one of the most abundant and least valuable (from the commercial point of view) of British fishes. Obviously, it was of particular importance, as events appeared to be shaping themselves in January of 1918, to develop this source of food to the highest degree possible and the investigations instigated by the Fish Food Committee had this for their object.

Sprats are caught principally off the coasts of Devon, Kent, Essex and Suffolk in the south, and in Morecambe Bay in the north-west. There are Scottish fisheries, mainly in the Tay, and there is little doubt that the fish occurs elsewhere. The supply is indefinitely large, being limited only by the catching power employed, which depends again on the demand. The latter is very limited. Before the war sprats landed at British ports were (a) sold for manure; (b) salted and exported to the continent of Europe; (c) packed by one or two canning factories; (d) consumed fresh; and (e) salted and smoked and sold in this state, or made into fish pastes.

The fish were but little appreciated in the fresh condition, and were probably rather a nuisance than otherwise to the wholesale and retail fishmongers, who probably preferred, as a rule, to "handle" herrings and other larger, more profitable, and more easily kept fish. The trade with the Continent (mainly Norway) had developed to a considerable extent before 1914, the sprats being salted, somewhat after the manner of pickled-herrings, packed in barrels and exported. There appears to be some doubt as to their further use and it is said that they were "de-salted" by some process involving osmosis and then packed after the manner of sardines. They also appear to have been repickled in the style of anchovies or used otherwise as "delikatessen," and it is probable that the bulk of the English and Scottish sprats exported were used in this way.

Smoked and dried sprats, sold in bundles and boxes, were a familiar article of food in London in the years before the "Fish (Prices) Order" of 1918. The cure varied greatly, some sprats being lightly salted and smoked for immediate consumption, and others being "dipped" in annatto and

<sup>\*</sup>See Fish Trades' Gazette for March 1st, 1919, where recipes for the pickles are published.

highly-salted and smoked, in which condition they "kept" for many months (like red herrings or dried salted cod). They could be eaten (head, bones, skin and all) in this condition, or lightly de-salted in warm water for a few minutes and then eaten. Doubtless, there were also many ways in which they could have been prepared. Given adequate mechanical drying and smoking apparatus—an easy enough thing to elaborate, it would appear—there seems to have been no reason why enormous quantities of sprats could not have gone into trade in this form.

In the highly-salted and highly-smoked cure of the red herring the fish would generally have lost about one-third of its weight in drying. Thus:—

Brightlingsea Fresh Sprats.

Water, 69; Fat, 12; Proteid, 17; Ash, 2 = 100 %

Brightlingsea Highly-smoked Sprats.

Water, 35; Fat, 24; Proteid, 30; Ash, 10 = 99 % \*

Thus about one half of the water in the fish is lost in the drying and smoking process, so that roughly, about  $1\frac{1}{2}$  lbs. of fresh sprats were required to produce 1 lb. of smoked sprats. In the smoking and drying process there was also a considerable amount of labour, and this was relatively greater the smaller the fish handled. Obviously, more labour was involved in drying and smoking 100 sprats than the same weight of herrings (say 100 sprats weigh 2 lbs., roughly, and 8 herrings weigh 2 lbs.).

Now, "the Fish (Prices) Order" of 16th January, 1918, fixed 6d. as the maximum price per lb. of fresh sprats, and 8d. as the maximum price per lb. of smoked sprats. The price of 1 lb. of fresh herrings was fixed at 8d., and the price of 1 lb. of kippered herrings at 1s. Say there are about 5 to 7 kippered herrings in 1 lb. The labour-cost of producing 1 lb. of kippered

herrings from, say,  $1\frac{1}{2}$  lb. of fresh herrings is evidently relatively less than that of producing 1 lb. smoked sprats from  $1\frac{1}{2}$  lb., say 75, of fresh sprats. The result that might have been expected was that which actually occurred—the Fish (Prices) Order stimulated the production of kippered herrings, but, the factor of profit in private production being what it is, it drove smoked sprats out of the market. On the other hand, the price of fish pastes was uncontrolled, and the production of these was greatly stimulated. No doubt the substitution of fish paste for butter, margarine and jam in the early months of 1918 had something to do with this, but the Fish (Prices) Order was probably also a factor from the point of view of profitable enterprise.

Smoking and drying of sprats being, therefore, out of the question in 1918, there remained the canning factories and the manure works as the outlets for the "gluts" of fish which could not be sold fresh or exported. Only one factory in England canned sprats in the sardine fashion in 1918, and the quantity of fish turned into paste in the smaller establishments on the Essex coast and elsewhere was probably exceedingly small in comparison with the total quantities landed—for these small establishments "carried on" under great difficulties because of the Military Service Acts. The output of the single English factory canning sprats increased greatly in 1918, but, obviously, it could take only a very small fraction of the fish placed at its disposal.

How the Fish (Prices) Order affected the quantities of sprats sold fresh in the shops it is impossible to say with the information now available. But it seems to have been the case that the fisheries on the Kent and Essex coasts, in Morecambe Bay and in the Firth of Tay were (relative) failures in the winter of 1918-19. Sprats were much less in evidence in the smaller fish shops and probably much more in evidence in the manure factories.

Obviously, the problems involved in the attempt fully to utilise the enormous quantities of sprats available in British seas are (1) overcoming the difficulties of an intensive, short seasonal fishery everywhere—the disposal of "gluts" of fish; (2) industrial enterprise directed to conserving large quantities of fish by smoking, canning, and the manufacture of paste; and (3) some means of preserving the *fresh* fish either for consumption as such or for further treatment in the factories. The problem (3) has been attacked by study of the brine-freezing methods and the solution may be regarded as unlikely of attainment when the cost of the treatment is fully considered. The problem (2) is touched upon later in this report.

## 2. Whitebait.

A few samples of whitebait were examined in connection with questions of conservation. It would be premature to consider these results here, and the analyses are given later in the report.

# 3. Hermetically-Sealed Herrings, Brisling and Pilchards.

Herrings, Brisling, French (Clupanodon pilchardus) pilchards and Californian (Sardinia caeruleus) pilchards, packed in tins in various media and under various conditions, were examined. The object was to find in what ways the chemical composition of the flesh was changed by the various processes and cooking media employed. Details of the analytical results are given later on and are discussed—to a certain extent. It soon became evident that more subtle methods than those employed in the determination of "proximate food stuffs" were necessary, and until such are elaborated much discussion is needless.

The processes employed in the production of "hermetics" are, in very general terms, as follows:—The fish are trimmed (gutted, beheaded, or otherwise prepared), salted, pickled and (more or less) dried or smoked. The pickle varies in com-

position and (like many other details of the processes) may be a trade secret. They may be cooked or fried in oil prior to being packed in the tins (the French method) or cooked in the process of sterilisation after the tin is sealed (the Norwegian method). They are packed in the tin (with various essential precautions) and the oil or sauce is added. The composition of the sauce varies and may be a trade secret. The tin is then sealed, a "vent-hole" being left, or not: when there is a vent-hole the latter is closed by solder after the sterilisation. The sterilisation is effected in autoclaves, the time and pressure of steam necessary being suggested by the experience of those responsible. The product is then allowed to "ripen" or mature.

Doubtless the processes employed in Great Britain in the preparation of hermetically sealed fish products are capable of improvement, but these improvements must, in the main, be the result of purely industrial research: improvement of the tin-making machinery, introduction of labour-saving devices, automatic plant for "handling" the fish in the operations of trimmimg, automatic smoking and drying machinery and ovens, economy of material and in general process research which aims at increasing production in relation to establishment and labour costs and the cost of raw material by speeding up machinery and eliminating unnecessary manual operations—with all this, though it is the most important part of the whole matter, the scientific man, as such, has little to do.

The improvement of the product itself, apart from economy in producing it, is another matter and most British "hermetics" are certainly capable of improvement. In some cases the raw materials—summer-caught herrings and mackerel and Cornish pilchards—are probably the best that can be obtained: they are certainly equal to the French or Californian pilchards. In many other cases the raw materials—winter-caught herrings and sprats—are inferior, being not so suitable for canning as

the French pilchards or Norwegian autumn-caught brisling. There is certainly a field here for industrial research directed towards the discovery of processes (such as variations on the formulae of the pickles, spices and sauces employed, or variations in the methods of smoking and cooking) which can be utilised so as to compensate for the natural "wateriness" and lack of fat in the inferior winter-caught fish: English factory processes are certainly apt to become stereotyped. But we must always remember the factor of profit—one which plays an all-important part in the factory, but does not matter at all in the laboratory. It is here that the experimental factory ought to intervene between the laboratory and the commercially-run factory.

Scientific research must at first, at all events, be restricted to a study of the actual results of established processes and, later on, to the investigation of the results of varying those processes in the ways suggested above. It should endeavour to answer the question why different results follow upon variations in factory processes. If such answers can be given it would generally follow that the research, after the adoption of a change in process, would react on the factory methods and suggest other changes—at least, that is what one hopes will be the case.

# Remarks on the Analyses made.

The methods adopted in the preparation of this report are, in the main, similar to those followed in the Report of 1917.\* A routine of methods was adopted in order to save time and be ready to deal with more numerous samples than were actually obtained. The samples were always composite ones. From a sample of fresh sprats or whitebait about 12 to 20 fishes, representative of those sent, were selected, and sections

<sup>\*</sup> Report for 1917 of the Lancashire Sea-Fisheries Laboratory, pp. 13-59, 1918.

through the "shoulders," trunk and tail parts of each individual were made. These were placed in a weighed and numbered weighing bottle containing a small Soxhlet extraction thimble having a number written in pencil on the outside, and a small cotton-wool plug. In the cases of tinned fish a paste was made from parts of each fish, the latter being taken from the tin and placed on filter paper so as to absorb the oil or sauce adhering to their surfaces. The weight of the wet flesh was found by difference. The water was estimated by drying for at least 48 hours in a steam oven (temperature 98° C.). Absorption of the oil and water by the thick walls of the thimble materially aided the process of drying.

Some general remarks on the water estimations may be made:

(1) The percentage in fish taken from the same shoals and at the same time varies with the length (and thus the age) of the sprats. Thus:—

Devonshire Sprats.

9-10 cms. long,  $68^{\circ}0~\%$  ; ~13-14 cms., long  $60^{\circ}1~\%$  of water.

Sprats from Menai Straits.

8-10 cms. long, 76.5 %; 10-12 cms. long, 74.6 % of water.

Morecambe Sprats.

6-8 cms. long,  $75\cdot0$  %: 8-10 cms. long,  $72\cdot8$  %; 10-12 cms. long,  $71\cdot2\%$  of water.

Whitebait, which are either herrings or sprats, and vary between 45 and 80 mm. in length, had from 78.4 to 80.4 % of water, although the fish were collected during the summer and autumn months, when the percentage of water should have been minimal.

(2) The percentage of water *plus* the percentage of fat is approximately constant, thus:—

Devonshire Samples.

Water and fat = 79.5 to 82.4, though water varies between 60.1 and 67.1, and fat between 15.1 and 22.3 %.

Menai Straits Samples.

Water and fat = 78.8 to 80.7. Water varies between 69.7 and 75.5, fat between 5.0 and 11.0 %.

Morecambe Samples.

Water and fat = 80.7 to 81.6. Water varies between 68.0 and 71.9, and fat between 9.6 and 14.6 %.

Whitebait.

Water and fat = 80.3 to 81.1. Water varies between 78.4 and 80.4, fat between 0.7 and 1.9 %.

Estimates of the water contents alone would, therefore, give measures of the fat contents accurate to about 1%, as a rule; thus percentage of fat = 80.0 – percentage of water.

The paper thimbles containing the dried substance, were next taken from the weighing bottles. The cotton-wool plugs were inserted into the openings of the thimbles so as to prevent the detachment of any small fragments of tissue during the extraction. Carbon tetrachloride was used for the latter operation, and three or four extractions were usually made simultaneously. The solvent was distilled off from the extracted oil, and the latter was dried at 98° C. until the weight just appeared to rise—usually a matter of two to four hours. The thimble with the extracted material was then replaced in its weighing bottle, dried for at least 12 hours, and weighed. The water (by difference), the dried oil, and the dried residue, expressed as percentages of the original wet substance, usually added up to about 100·05 %.

When the extraction was made on recently dried material the CCl<sub>4</sub> solution was always clear, and more or less brown in colour. Occasionally, the dried material was stored in stoppered or corked weighing bottles for two to six months, re-dried and re-extracted, and in such cases the extract was always more or less turbid, something coming through the cotton-wool plug in the mouth of the extraction thimble. Some of this insoluble, detached substance was separated: it consisted of fine, needle-shaped crystals, insoluble in CCl<sub>4</sub>

and ether. Its nature is being investigated and apparently it is some substance formed by oxidation of part of the dried unextracted tissue.

The oil extracted from the sprats and whitebait was apparently similar to that obtained from herrings, but no chemical or physical examinations were made.

As a rule, from 7 to 9 gms. of wet tissue were taken for the analysis. From 0.5 to about 1.5 gms. of oil were usually obtained—enough for a nitrogen estimation. About 1.5 to 2 gms. of dried residue remained in the thimble. This was detached, ground up into a fine powder and replaced in its own weighing bottle. About 0.5 to 1 gm. was taken and ignited in a porcelain capsule for the estimation of non-volatile, mineral matter, and enough remained for nitrogen analyses: 0.500 gm. being taken for the latter estimation. When the dried residue had been stored for some days it was always re-dried before the estimation.

The digestion of the fat-free dry residue was carried out in long-necked Kjeldahl flasks, three operations, as a rule, proceeding simultaneously. Several grams of dried sodium sulphate were used to raise the temperature, and 20 c.c. of sulphuric acid were always used for the digestion. Copper sulphate was used to accelerate the process, which took from three-quarters to one hour as a general rule.

The clear solutions in the Kjeldahl flasks were allowed to cool and were diluted. As a rule, the flasks were corked and the distillations were made on the day following that of the digestion. The distillation was made in the usual manner, deci-normal sulphuric acid and deci-normal sodium hydrate being used. Cochineal was employed as an indicator and gave better results than litmus or methyl-orange. Even then the discrimination of the best colour end-point was a source of some uncertainty and error. The ammonia distilled off was received into 75 c.c.  $N/10~\rm H_2SO_4$  and about 17 to 25 c.c.  $N/10~\rm H_2SO_4$  and about 17 to 25 c.c.  $N/10~\rm H_2SO_4$  and about 17 to 25 c.c.  $N/10~\rm H_2SO_4$ 

sodium hydrate were required for neutralisation. The process, as a routine one, might be improved by using a bulb-burette, the stem being graduated in 20ths of a c.c., and 5th-normal acid and alkali. The precise end-point to be taken ought to be investigated and this and the indicator standardised in some way. As a very general rule, however, the error of analysis did not exceed  $\pm~0.05\%$  of N. (which corresponds, however, to  $\pm~0.3\%$  of "proteid").

Remarks on the Nitrogen Analyses.

The estimation of N in these products may be a matter of much importance, and some considerable attention was directed to methods.

It is to be noted, first of all, that the order in which the operations described above are carried out is of significance. It will be seen that the sum of the percentages of water,  $CCl_4$ -extract, "proteid" and ash seldom add up to approximately 100%, and (especially in the cases of some of the hermetically-sealed products) the deviations from 100% are much greater than can be reasonably attributed to experimental error. Now, the tissue may be dried and extracted and the residue then examined for N; this was the routine usually followed. But the unextracted tissue might be examined directly for N (in which case the digestion is more difficult, and the clear solution must be diluted and filtered off from the small quantity of fatty acids usually left, so as to avoid the formation of soaps in the ammonia distillation). Rather different results are obtained in such contrasted results. Thus:—

į	$\%$ of $N \times 6.25$ in the unextracted substance	% of $N \times 6$ : in the extracte substance
Herrings, in tomato sauce, matured since 1914	20.3	19.0
Herrings, in mustard sauce, matured since 1908 (Percentages calculated on the w	24.0	22•6
Salted Sprats	59·2 52·7	56·1 45·3
Devonshire brine-frozen Sprats French (Rodel) Pilchards	59.8	61.0
French (Rodel) Pilchards (dried material kept 6 months) (Percentages calculated on the d	58.7 ry weights of substa	59·2 ance).

These results are rather puzzling: most of them are to be accounted for by assuming that N in some form is extracted by the carbon tetrachloride when estimating the oil present. Thus, apparently, more N can be obtained from the unextracted than the extracted material, except in the case of the Rodel sardines, where the quantities extracted may be regarded as the same.

The  $CCl_4$  extract was then examined. About 1 grm. of the oil was weighed out into the broken-off end of a test-tube, and this was digested in the usual way—the operation was rather troublesome on account of the frothing in the initial stages—and the quantity of N present was estimated, particular attention to the results of blank analyses being given. The results apply to "hermetics," and are:—

## Percentage of N in CCl<sub>4</sub> extracts from fat-estimations.

Matured Morecambe Sprats, the	extrac	t contair	ıs		 0.31 % N.
Unmatured Devonshire Sprats	"	,,	***		 0.27 % N.
Matured (Rodel) Sardines	99	,,		• • •	 1·1 % N.
Unmatured Essex Sprats	27	,,			 0·3 % N.
Matured Herrings, tomato sauce	,,	29			 0·16 % N.
Matured Herrings, mustard sauce	29	,,			 0.26 % N.

Thus, quite a significant quantity of nitrogen is contained in these extracts: it corresponds to from 1.0 to 6.9% of "proteid." Now, it was rather surprising to find that the oil in the various tins contained no significant quantity of N. Thus:—

## Percentage of N in the oils from various tins of "hermetics."

Matured Morecambe Sprats, the	 	0.001 % N.		
Matured (Rodel) Sardines	22	22	 	0.09 % N.
Matured Herrings, tomato sauce	,,	,,	 	0.08 % N.
Unmatured Portuguese Sardines				0:04 9/ N

These results are not far from the limits of experimental error, and we cannot say that the oil in the tins contains an appreciable amount of N, though the oil in the fish certainly does.

The oil used in the preparations—olive, pea-nut, cotton-

seed, whatever it may be—either contains no proteid matter, or contains such as an insignificant trace resulting from the methods of preparation, but the oil in the fish tissue does contain nitrogen in some form, and this nitrogenous oil does not easily diffuse out from the tissue into the medium in the tin. In hermetics that are long matured there may be some diffusion out from the fish—thus, the highest results in the cases of the four samples examined were those of the French pilchards and the herrings in tomato sauce, both matured for about 4 to 5 years. In the latter case the medium in the tin was a mixture of watery sauce and oil that had been squeezed out from the flesh of the fish.

When pilchards, herrings and sprats are packed in oil in tins the oil is always watery. As a rule, the mixture is apparent, and the oil and water segregate at once, but in some cases (notably that of the Rodel sardines) the mixture was such as to pass through a dry Swedish filter paper as a pellucid liquid. After some weeks this separated into a thin layer of turbid watery solution and a layer of clear oil. The water thus incorporated in the oil may amount to 4 to 7% of the weight of the mixture.

The percentage of nitrogen in the dry, fatfree, ash-free residues.

Obviously, the use of the term "proteid" in these analyses is to be regarded as rather a convention than as indicative of some definite chemical substance: it is, however, employed as useful from the dietetic point of view, and may be regarded as some substance, or mixture of substances, containing from  $14\cdot3$  to  $17\cdot4\%$  of nitrogen. In considering the composition of fresh and preserved fish from the point of view of methods of conservation it is better to speak of the percentage of nitrogen present rather than that of "proteid," assuming this to be nitrogen  $\times$  6·25 where the factor, 6·25, is an arbitrary one,

which may, but usually does not represent the composition of the proteid constituents of the "flesh." Now, the quantity of mineral matter present in the "flesh" of fresh and preserved fish is rather variable: it is difficult to sample the musclesubstance of a Clupeoid without including some small bones and the hermetically-sealed sprats, herrings, &c., had to be sampled as they were likely to be eaten, for many people are not fastidious enough to remove the skin, backbone, &c., of the sardines that they consume: these products attained to a more important place in dietaries during the last five years than hors d'oeuvre did formerly. The fish from tins were, therefore, made into a paste, bones, skin and all, and this paste was sampled. Again, a very variable percentage of salt is added in the course of preservation in different ways, thus kippers may contain about 3.5%, bloaters up to about 10%, pickled herrings from 6 to 12%, and red herrings as much as 12%. Hermetically sealed sprats, herrings and pilchards contain from about 3 to 5% (though sometimes the proportion was found surprisingly low). Brine-frozen fish, which were supposed to be "fresh," may have had from 3 to 9% of salt in the tissues. Quite fresh, untreated herring and sprat "flesh" contained, as a rule, about 1.5% of non-volatile mineral matter, some of this being certainly derived from the bones.

The fat being extracted, and the mineral matter being estimated by ignition, the dry, fat-free, ash-free substance present in 0.500 grm. of the dry residues was then calculated and the nitrogen found by the Kjeldahl process was expressed as a percentage of the dry, fat-free, ash-free substance. This is the "N" of the detailed tables of analyses and its values are summarised in the following statement, where some results taken from last year's Report are included.

Manx Sum	mer-ca	ught	Herring	s ( $Fresh$ ).		
			Water.	Fat.	Water + Fat.	N:4mayaan
July			water. 40.2	38·6	+ Fat. 78.8	Nitrogen. 14·8
August	•••	•••	43.5	36·6	80.1	14.7
Septem	hon	•••	49.2	27·6	76.8	14.3
June		• • •	55·3	22.1	70.8	14.3
May	•••	•••	70.8	3.2	74.0	14.1
Means	•••	•••	51.8	25·6	77.4	14.5
In eans	•••	•••	31.9	20.0	11.4	14.9
Pickled (pro	bably	all)	Summer-	caught Herri	ngs.	
Means			47.7	20.9	67.6	14.2
Hermetically	j-seale	d, m	natured H	Terrings in vo	rious med	ia.
Means	•••		62.9	12.8	75.7	14.8
Hermetically	ı-seale	d, $u$	nmatured	Herrings.		
Means		•••	63.6	12.2	75.8	15.2
Smoked Her	rrings	and	Sprats.			
Means	-		45.1	17.7	62.8	15.1
Hermetically	ı-seale	d, u	nmatured	Sprats, in	oil.	
	•••	•••	58.3	15.7	74.0	15.1
Hermetically	y-seale	d, n	natured I	Pilchards in	various me	edia.
Means	•••		58.2	10.8	69.0	15.6
Fresh Wint	er-cau	aht .	Herrinas.			
~ -	•••		62.4	21.7	84.1	15.6
Fresh Wint	er-cau	aht i	Sprats.			
> 20 %			60.1	22:3	82.4	15.9
15-20 %	, ,,	• • •	63.3	17.8	81.1	16.2
10-15 %			68.8	12.4	81.2	. 15.9
5-10 %			73.3	7.0	80.3	16.2
0-5 %	) ),		79.6	3.1	82.7	16.6
Means	•••	• • •	69.0	12.7	81.5	16.2

These statements seem to point the way to lines of further investigation as to the rationale of methods of fish conservation. The results are set out roughly in order of magnitude of the percentage of nitrogen in the residue which we may take to be "proteid" in nature and this percentage is so stated as to be unaffected by the variable quantities of salt that are added

to the fish in the course of the conservation process. This salting, together with other factors in the conserving process alters the percentage of nitrogen present in the proteid fraction of the fish, and is no doubt responsible, to some extent, for the quality of the product. But the variable percentage of nitrogen also depends on the initial condition of the fresh fish: mainly, upon the quantity of oil contained in it, and upon the sexual condition with respect to the season. Thus, a wintercaught, sexually ripe fish containing say 20% of oil is not the same thing as a summer-caught, sexually ripe fish containing approximately the same proportion of oil (though, as a general rule, the ripe winter fish would contain less oil than the ripe summer fish).

The sum "water and oil" depends on the degree of drying of the fish in the process of conservation. When this sum is relatively low the percentage of "proteid" will be relatively high.

# Details of the Analyses.

#### 1. Fresh Sprats.

Collected from Penrhyn Weir, Menai Straits, North Wales. The fish are mostly immature and are about 8 to 10 cms. in length. Throughout the season they are mixed with small herrings, and "whitebait" are abundant during the summer and autumn months. Towards the spring the sprats become relatively few and are replaced by herrings about 8 to 12 cms. in length.

_						
Date.	Water.	Fat.	Proteid.	Ash.	Total.	Nitrogen.
20.9.18	71.3	8.6	19.5	1.2	100.6	17.1
4.10.18	75.4	5.1	17.7	1.6	99.8	15.8
18.10.18	73.6	5.2	22.0	$2 \cdot 2$	103.0	16.4
21.11.18	71.6	8.7	18:3	1.7	100.3	16.2
6.12.18	69.7	11.0	18.5	1.5	100.7	16.4
19.12.18	72.8	7.9	18.0	1.7	100.4	16.3
22.1.19	74.6	5.9	17.7	1.6	99.8	15.8
6.2.19	75.5	5.0	17:3	1.9	99.7	15.8

Evidently, these sprats are not very suitable for preserving in tins, the percentage of oil being rather low, and the fish being small and rather unequal in size. The mixture of whitebait and small herrings with the sprats is also a disadvantage. It should be noted, however, that the method of catching was a very special one—fixed weirs traversed by tidal streams—and it may be the case that appropriate fishing experiments with stowbut nets and drift nets of restricted mesh would be successful in obtaining sprats of suitable size. As regards fat-contents the fish are, however, of poor quality.

# 2. Fresh Sprats.

Collected from "fishing-baulks" and stowbut nets at Morecambe. The sprats are rather unequal in size (though this disqualification, from the packing standpoint, might be avoided by the use of drift-nets instead of the stow nets). Those sampled were mostly fine fish of about 9 to 12 cms. in length. Towards the end of the season they become sexually mature, and ripening ovaries and testes may be seen, though spawning does not appear actually to occur in the shallow inshore area of Morecambe Bay. The season begins about October and may last till March, but it came to an end prematurely in 1919—much earlier than in 1918. It is doubtful whether the cause of this was natural or economic.

Date.	Water	Fat.	Proteid.	Ash.	Total.	Nitrogen.
22.2.18	71.9	10.2	17.5	0.5	100.1	16.0
4.10.18	67.0	14.6	17.1	1.3	100.0	16.2
26.11.18	68.0	13.1	17.5	1.5	100.1	16.0
17.12.18	68.3	12.4	16.9	1.6	99.2	15.2
23.1.19	71.9	9.6	16.8	1.7	100.0	16.0

Apart from the variability in size these sprats are highly suitable for conservation processes. The percentage of fat in the tissues is moderately high, for winter-caught fish. The fish themselves are plump and well-shaped, and they are usually handled by the fishermen with great care. The

quantities of sprats, of this kind, obtainable from the sea off-shore from Lancashire and Cumberland is probably indefinitely great, being limited only by the catching power employed.

### 3. Fresh Sprats.

Collected at Torquay and Sidmouth, coast of Devonshire. The samples of 4.10.18 and 19.10.18 consisted of "flesh" only, as it was desired to make comparisons between these fish and the Manx Summer Herrings reported upon previously. The other samples consisted of the "whole fish." The sample of 4.10.18a consisted of fish of about 9 to 10 cms. in length; all the others were about 14 cms. long. The samples of 18.11.18a had been brine-frozen and cold-stored for about a month.

Date.	Water.	Fat.	Proteid.	Ash.	Total.	Nitrogen.
4.10.18	60.1	22.3	16.7	0.3	99.4	15.9
4.10.18a	67.1	13.8	18.1	1.3	100.3	16.3
19.10.18	62.6	18.9	17.7	1.2	100.4	16.1
18.11.18	61.2	18.3	17.3	2.5	99.3	15.5
18.11.18a	66.0	15.1	17.1	1.8	100.0	16.0

These sprats are eminently suitable for canning. The fish are large and plump and evidently they can be obtained in nearly uniform sizes. The percentage of fat is high. Sample 18.11.18a is interesting as it shows that sprats can be brine-frozen without excessive salting, though whether the cost of this process and that of the subsequent cold-storage would not be prohibitive is a question with which we have nothing to do here.

# 4. Fresh Sprats.

Caught at Brightlingsea. The season was a very short one, and had almost came to an end before arrangements for sampling could be made.

Date.	Water.	Fat.	Proteid.	Ash.	Total.	Nitrogen.
16.1.19	67.8	13.5	16.6	1.8	99.7	15.6
10.2.19	70.7	10.3	17.0	1.6	99.6	15.8

These sprats are also suitable fish for canning, though not so fine as the Devon ones.

# 5. "Fresh" Yawling.

These are immature herrings about 10 to 14 cms. long. They were caught off the Essex coast about 28.3.18, brine-frozen and cold-stored till 3.5.18. Sample 1 consisted of the whole fish and Sample 2 of the "flesh" only. Each was, as usual, a composite sample.

	Water.	Fat.	Proteid.	Ash.	Total.	Nitrogen.
1	59.1	0.4	30.9	9.2	99.6	17.9
2	82.3	0.7	12.9	3.4	99.3	15.5

These fish have been canned in oil and tomato sauce, but they are certainly the least suitable of those examined so far. They are very poor in fat, the flesh is thin and watery and the fish are, as a rule, carelessly handled by the fishermen and stand transport very badly. The brine-freezing, in this case, added to the quantity of salt in the tissues, but even in quite fresh fish the mineral matter would be relatively high because of the influence of the bones in thin fish such as these.

# 6. Whitebait, fresh and brine-frozen.

- 1. Sprats from Menai Straits, 45 to 60 mm. long, 23.7.18.
- 2. Herrings from Menai Straits, 65 to 80 mm., 23.7.18.
- 3. Sprats and herrings from Menai Straits, 50 to 60 mm., 4.10.18.
- 4. Sprats from Morecambe, caught August 18th, brine-frozen and cold-stored till 7.2.19. 60 to 80 mm.

	Water.	Fat.	Proteid.	Ash.	Total.	Nitrogen.
1	79.9	0.8	17.9	1.4	100.0	16.6
2	80.4	0.7	17:3	2.5	100.9	16.7
3	78.4	1.9	18.4	2.0	100.7	16.6
4	69.6	0.8	20.2	8.2	98.8	15.1

These are fish which are of little value for canning purposes (though "Sardine Whitebait" has been packed in oil in tins). The flesh is watery and almost wanting in fat. Whitebait are difficult to "handle," and brine-freezing with subsequent

cold-storage might be attempted with success, because of the relatively high value of the fish in the fresh condition. Some experiments were made with inconclusive results, but it is evident that there are no difficulties that could not be easily overcome.

- 7. Hermetically Sealed Sprats.
  - 1. Morecambe sprats in oil. Unmatured. Packed 22.2.18.
  - 2. Morecambe sprats in oil. Unmatured. Brine-frozen March, 1918. Cold-stored till 15.5.18.
  - 3. Morecambe sprats in oil. Matured. Packed November, 1917. Examined 21.6.18.
  - 4. Morecambe sprats in oil. Unmatured. Brine-frozen March, 1918. Cold-stored till 15.5.18. Examined at various dates.
- 5, 6, 7. Morecambe sprats in oil. Unmatured. Brine-frozen March, 1918. Cold-stored till 15.5.18. Examined at various dates.
  - 8. Morecambe sprats in oil. Matured. Packed November, 1916. Examined 20.12.18.
  - 9. Devonshire sprats in oil. Unmatured. Brine-frozen 16.10.18. Cold-stored till 16.11.18. Examined 3.12.18.
  - 10. Norwegian brisling in oil. Matured.
  - 11. Brightlingsea sprats in oil. Unmatured. Packed January, 1919. Examined 7.2.19.

	Water.	Fat.	Proteid.	Ash.	Total.	Nitrogen.
1	56.3	16.5	22.9	3.3	99.0	15.1
2	49.8	7.8	30.5	10.6	98.7	16.1
3	58.8	13.0	23.1	4.5	99.4	15.7
4	52.9	14.2	24.2	8.8	100.1	16.0
5	69.3	7.4	15.7	6.7	99.1	14.6
6	53.3	11.4	25.2	9.3	99.2	15.0
7	54.3	11.7	23.6	8.5	98.1	14.3
8	59.3	14.4	21.7	5.9	101.3	17.0
9	54.0	18.9	21.6	5.0	99.5	15.5
10	60.3	12.4	25.3	3.6	101.6	17.1
11	60.4	14.9	20.7	3.3	99.3	15.2

- 8. Hermetically Sealed Herrings.
  - Brightlingsea yawling, in tomato sauce. Brine-frozen March, 1918. Cold-stored till 15.5.18.
  - 2, 3. East Coast herrings, in tomato sauce. Summer-caught. Matured fish.
- 4, 5, 6. East Coast herrings, in tomato sauce. First of season's catch. Unmatured.
  - 7. Manx Summer herrings, in oil. Packed by French process. Unmatured.
  - 8. East Coast Summer herrings, in tomato sauce.
    Packed in 1914. Examined 14.2.19.
  - 9. East Coast Autumn herrings, in mustard sauce. Packed in 1908. Examined 14.2.19.

	Water.	Fat.	Proteid.	Ash.	Total.	Nitrogen.
1	72.8	1.0	22.8	4.6	101.2	16.8
2	64.4	11.6	20.2	2.7	98.9	15.0
3	$62 \cdot 4$	13.7	18.3	2.0	96.4	13.3
4	71.8	8.1	17.5	0.8	98.2	15.1
5	66.3	7.8	25.9	2.1	102.1	17:4
6	$65 \cdot 4$	8.4	22.5	0.3	96.6	15.7
7	55.4	16.3	23.4	2.7	97.8	15.1
8	62.6	15.1	19.0	1.5	98.2	15.0
9	62.3	10.7	22.6	4.2	99.8	15.8

- 9. Hermetically Sealed Pilchards.
  - 1. Californian pilchards (Sardinia caeruleus), in mustard sauce. Matured.
  - 2. Portuguese pilchards (*Clupanodon pilchardus*), in oil. Summer-caught fish. Matured.
  - 3. Portuguese pilchards (Clupanodon pilchardus), in oil. Winter-caught fish. Matured.
  - 4. French pilchards (C. pilchardus), in oil. "Rodel fils." Packed in 1913. Examined 30.11.18.
  - 5. Portuguese pilchards (C. pilchardus), in oil. Unmatured. Winter fish.

	Water.	Fat.	Proteid.	Ash.	Total.	Nitrogen.
1	58.8	11.3	24.0	3.7	97.8	14.8
2	55.3	13.6	25.6	5.2	99.7	15.4
3	58.2	9.8	25.9	5.9	99.8	16.0
4	59.9	11.7	24.5	3.2	99.3	16.0
5	58.6	7.8	27.4	5.6	99.4	15.7

### 10. Highly Smoked Sprats.

Brightlingsea fish. (1) the whole fish; (2) the "flesh" only.

	Water.	Fat.	Proteid.	Ash.	Total.	Nitrogen.
1	35.1	23.7	29.6	10.0	98.4	15.1
2	39.1	16.1	32.2	9.8	97.2	14.7

In all the above analyses "Proteid" means "nitrogen  $\times$  6.25," and "Fat" means "Carbon-tetrachloride-extract." "Nitrogen" is the N found by Kjeldahl analysis and calculated on the quantity of ash-free, fat-free, dried residue taken.

If the "totals," that is, % water +% fat +% proteid +% ash, be examined, it will be found that they may vary between about 97 and 103 as extremes, a difference of +3% from that which should have been obtained on the assumptions that everything present in the tissues taken can be estimated as water, fat, ash and  $N\times 6.25$ . Where the total is much less than 100 % the "nitrogen" is markedly less than 16 %, and vice versa. It was assumed that fish proteid contains 16 % of N though it was evident that it did not—in general.

In fresh summer-caught herrings such as those examined here, the N%, as defined above, was about 14.5. In fresh winter-caught herrings it was about 15.5. In fresh winter-caught sprats it was about 16.2.

In salted, smoked and hermetically-sealed sprats and herrings the N % depends, to some extent, on the season of capture of the fish canned, and on the methods of conservation.

The apparent errors of analysis as indicated by the above tables are, therefore, to be traced to the use of the factor 6.25,

that is, to the assumption that fish proteid always contains 16~% of nitrogen. But, since the fat solvent always seem to extract something which is not a true saturated or unsaturated oil or fat, but which contains nitrogen, it must be overestimated and the "proteid" must always be underestimated for the same reason. The manipulative error of estimation of "fat" is very small; that of estimation of N may be taken as about + 0.05~%, and that of estimation of "Proteid" as about  $+ 6.25 \times 0.05~\%$ .

From the point of view of "dietetics," "calories" and the like, in connection with public food problems these errors do not matter since they must be negligible when compared with other sources of incertitude that arise.

#### The Nature of the "Maturation" Process.

It is well known that pilchards, herrings, sprats, mackerel and other Clupeoid fish, which are processed and packed "à la Sardine" must be matured or ripened. When newly packed the bones are not softened, and the taste and smell are not those expected. The fish are "raw"—even if they have been cooked in the ordinary sense of the term, and the taste is only that of the specific cooked fish. Maturation, or ripening is the result of simply allowing the tins to stand unopened for a considerable time, 6 months to 4 years, during which the fish continually improve, and the flavour becomes increasingly "richer." This process of maturation may continue for about 10 years.

The bones soften until they can hardly be distinguished (in the mouth) from the flesh. The scales (in the case of pilchards) do not seem to undergo any change. The flesh becomes very soft and pasty, so that fish in a watery medium, like tomato sauce, or the media used in "marinaded" products become so tender that they can hardly be handled without

breaking. This extreme softness and pastiness does not proceed so far in the case of fish packed in oil. Oil oozes out from the fish into the watery media, and water comes out into the oil from fish that have been packed in an oil medium. In the latter case (with olive oil, at all events) the watery fluid that oozes out from the fish may form an apparently homogeneous mixture with the oil in the tin, so that the weight of the latter can be reduced on drying at  $100^{\circ}$  C., even if the presence of water does not become apparent by the mechanical separation of the latter.

The richness of the flavour may pass into something that (to the trained palate of the trade expert) suggests decomposition, just as game that has been "hung" becomes "gamey," or as cheese may become too rich in taste. But nothing in the appearance, smell or taste of such over-matured tinned fish suggests putrefaction in the usual sense, and one has, of course, no reason to expect that this condition would be set up by well-packed hermetically-sealed fish, in oil, or other suitable media.

The internal surface of the tin undergoes change. In marinaded goods, which may contain acid, or produce acid (at first) the tin may be etched. It is probable that no means of protecting the tin, such as by shellac varnish would be permanently effective. Both in oil and tomato sauce the tin-plate becomes slowly blackened, and this change is progressive with the length of maturation. The film of tin is apparently removed and the iron blackened, and it is easy to show that the black substance is a film of ferrous sulphide. The film of tin on the plate may be regarded as a solution of iron in tin and the iron is slowly attacked by sulphuretted hydrogen, or some sulphur compound liberated in extremely small quantity from the maturing fish.

The decomposition is, apparently, not due to bacterial action. In the processes of cooking and sterilisation the contents

appear to be rendered sterile for the tins are heated in steam under pressure to a temperature well over 100° C. The sealed tins may be incubated at 37° C. for weeks without "blowing," or giving any indications of growth of micro-organisms when they are opened. Of course, the possibility is not entirely excluded of certain spores remaining alive in the oily parts of the fish or medium even under the conditions of the sterilisation. Also some tins are septic and become blown on keeping, but this does not appear to occur when the factory management is all that it ought to be.

One may, therefore, exclude bacterial action as a cause of ripening, or if it does occur the precise nature and conditions must be very different from those usually associated with anaerobic putrefactive processes. Now, the changes that occur in the process of maturation do seem to suggest an autolytic process, though there is the difficulty of supposing intra-cellular enzymes to remain active after having been submitted to temperatures that are sufficient to produce sterility with regard to bacterial micro-organisms and ordinary moulds. Assuming, however, that there are enzymes in the flesh or media that are not destroyed by the processing, the changes that occur as the results of ripening seem, at first sight, to be such as would fit this hypothesis—supposing that these enzymes are present in very small quantities and that they do not increase, for the assumed autolytic changes go on extremely slowly in the usual conditions.

If this is so, if the ripening is a process of autolysis due to specific, intra-cellular enzymes normally present in the flesh of the fish, and not entirely destroyed by the sterilisation, then we ought to expect that the change that occurs is a proteolytic one, the proteids of the flesh being partially split up with the appearance of amino-acids. The flavour of the matured product would then be due to the traces of certain amino-acids produced as the results of proteolysis. This is what occurs in the process of ripening of salted herrings. The

strong brine penetrating the tissues of the fish prevents growth of bacteria: it does not render the fish sterile for the brine may putrefy when it is diluted with sterile water, but it inhibits growth. It permits some degree of autolysis as the result of the intra-cellular enzymes of the fish, and this autolytic change results in the formation of amino-acids, which dissolve out into the brine.

A quantity of filtered herring brine obtained by Professor Herdman from a barrel of salt herrings which he prepared at Port Erin in September of 1917, and which had been kept in a bottle from June of 1918, was examined by Sörensen's method for estimating amino-acids. The amino-acid contains a COOH group, which may take up a basic radicle, and an NH2 group which may take up an acid radicle. NH2 group is neutralised by the addition of previously neutralised formaldehyde and the amino-body, which now has an acid reaction, is titrated with deci-normal sodium hydrate, using phenol-phthalein as indicator. Several estimations of the herring brine referred to above were made, and it was found, as a mean, that 5 c.c. required, for neutralisation, 18.2 c.c. N/10 NaOH. This gives us (roughly) about 0.5 % of nitrogen in the brine, nitrogen present in amino-acids split off from the proteid of the fish flesh by autolytic action.

So also the watery liquid filtered off, through a wet filter paper, from a tin containing long-matured herrings, in tomato sauce, contained about 0.07 % of nitrogen.

It was assumed that the process of proteolysis was similar in the case of hermetically-sealed herrings to that indicated above. Exactly 4 grms of fresh herring flesh were weighed out and ground up with clean sand in a mortar. The fine emulsion was washed out from the same into a measuring flask and made up to 250 c.c., and successive portions of 50 c.c. each were titrated as described above. The mean quantity of N/10 NaOH required for 50 c.c. was 1·1 c.c.

Brine-frozen sprats packed in oil, matured herrings in

tomato sauce, long-matured herrings in tomato sauce, and herrings matured 10 years in mustard sauce were all examined by the same method. In all cases 4 grms. of the flesh were ground up into a very fine emulsion, and made up to 250 c.c. with water, and then several portions of 50 c.c. each were titrated. In all cases the quantity of N/10 NaOH required for neutralisation was from 1·3 to 1·4 c.c.

Similar estimations were made with the same quantity of flesh made into an emulsion with 50 % alcohol, but just the same results were obtained.

Amino-acids were, therefore, present in exceedingly small traces—so small that one may neglect them, and the quantity of these substances present was not affected by the length of the maturation period, not even by a prolonged period of maturation. Whatever is the nature of the latter process it is, therefore, not the same as the process of ripening of herrings pickled in brine.

Experience shows that the excellence of a canned pilchard, herring or sprat depends primarily on the nature of the raw material—the "processing" may make the quality of the product better or worse, how much better or worse depends on the extent to which the profit-factor is allowed to influence the details of factory management. Thus, herrings deep-cooked in oil (the French process) at a higher temperature than that of the autoclaves employed for sterilisation and cooking combined (the Norwegian process) make a better (though a different) product. But which of the two processes may be employed is a matter of policy in which the profit-factor is, perhaps, the main consideration. Many brands of herrings in (the almost unique) tomato puree, which the British public have apparently been trained to regard as the only possible sauce, at present on the market are very inferior stuff. In

the absence of factory experiments (and of competition from more carefully thought-out Continental and American factories), these products are hardly likely to improve. This, however, is a matter with which we have little to do here, except in so far as scientific research, which is intended to be industrial in its applications, ought to consider factors apparently unrelated to it as pure scientific research.

Details of processing do, however, improve a naturally poor raw material—fat-poor herrings or sprats for instances—and *some* English tinned herrings are certainly very superior products. Then the maturation certainly leads to a better product. Why? Why should a naturally fat-rich fish make a good article and *vice versa*? Why cannot the missing fat be supplied in the added oil? What is the nature of the interaction between the proteid and fat of fish flesh, an interaction suggested by the observations recorded here? These questions are very interesting and answers to them *might* react usefully on the factory processes at present employed.

# THE PROBABLE ERROR OF A BACTERIOLOGICAL ANALYSIS.

By Jas. Johnstone, D.Sc.

There has been a tendency of late years to depreciate the value of bacteriological analyses in relation to questions of shell-fish pollution, and to attach greater significance to conclusions based upon topographical and epidemiological investigation. This tendency is due partly to the lack of any really convincing and generally accepted method of analysis; partly to the reaction against an exaggerated estimate of the utility of bacteriological tests, and partly to the fact that analysts have not usually attempted to make estimates of the magnitude of the error to which their conclusions are always subject. This error may be a rather large one, and neglect of it has led to apparently anomalous results which have done much to create the tendency to which I allude.

But one must recognise that topographical data are very difficult to apply, for it is most difficult to foresee exceptional conditions and to reckon on the probability with which these may be expected to occur. Epidemiological evidence is still more difficult to interpret even when it has been collected with scrupulous care—and the local public health services cannot be said to be organised yet with such thoroughness as to justify us in expecting this scrupulous care. There are several well-known examples—Bulstrode's investigation of the Winchester and Emsworth outbreaks, and Hamer's work on the Bethnal Green epidemic-where the enquiry was a really fine piece of research. But the majority of cases of illness attributable to shell-fish pollutions are sporadic, and it must be exceedingly difficult to trace the cause to shell-fish alone, eliminating all other possible explanations. Naturally, a local authority will tend to make such an hypothesis as will remove the cause of disease outside its own area, and so inland medical officers of health will incline to fix on shell-fish as the source of infection. Much of the evidence accepted in such cases is not convincing. If the cause of illness were made a criminal issue it is very doubtful whether a Judge would regard the statements made by local sanitary inspectors and medical officers of health as legal evidence—at any rate that has still to be tested.

It is quite proper that evidence of marked bacteriological pollution should be made the basis of ameliorative work—the better disposal of sewage, the erection of purification plant, the adoption of systems of cleansing the mussels, voluntary certification of "pure" mussels, sterilisation of the shell-fish, etc. But one may reasonably object to analytical results being made the basis of penal action, that is, leading to restrictions and prohibitions, infraction of which may be punished with fine or imprisonment, unless these results are criticised far more severely than has usually been the case in the past.

A bacteriological analysis is generally a very definite statement—there are so many Bacillus coli per mussel, for instance. Scientific men know quite well that any quantitative result is always an approximation, and chemists say as much when they give the "limits of experimental error." If the data on which a bacteriological result is based are stated, one may often find some measure of the degree of error. But these data are not always given. There is a single numerical estimate of the degree of pollution, and the administrative officials for whose use the analysis has been made are left to make what allowance they like for error.

Some methods of estimating the "margin of error" are considered here. These methods are applications of the "laws of probability" in relation to samples "taken at random." There is nothing of a very abstract nature about

them—everyone applies them every day in the ordinary affairs of life, in a practical, though unsystematic way, and there is no reason why we who apply them in private matters should not systematise, and apply them in public affairs, especially when our business is such as to lead us to interfere with the industry and liberty of other people.

One or two particular methods of analysis are taken to illustrate the simple statistical precautions considered. These statistical devices are, however, general ones, and may be modified and extended so as to fit any other methods of analysis that may be employed.

# Method of Analysis.

A shell-fish bed usually includes various, more or less marked sub-areas in each of which the liability to sewage pollution varies. Each sub-area must, therefore, be sampled and considered individually.

A sample consists of 5 mussels collected "methodically at random" from different parts of the sub-area. Whether the sample consists of 5 or 10 or 20 mussels is, of course, a matter of practical convenience—a compromise always.

The mussels are opened and the liquid in the shells is drained into a small mortar. The soft parts are cut out, minced up with scissors and put into the mortar. The whole is ground up into a thin paste, and this is poured into a 250 c.c. flask, which is then filled up to the mark with water. The flask is well shaken, and the coarser particles are allowed to settle to the bottom. A number of samples, of one c.c. each, are taken from the supernatant liquid, and each sample is placed in a separate Petri dish. Liquid neutral-red, bile-salt, lactose agar (McConkey) is then poured into the dishes (10 c.c. into each), and agar and infected liquid are well mixed. The plates are set and incubated.

One c.c. of the liquid in the flask is taken as containing 1/50th part of an average mussel.

#### The Experimental Error.

The mussels are unequal in volume, so that 5 taken now or here do not necessarily have the same quantity of liquid in the shell, or the same mass of soft parts as 5 taken afterwards or elsewhere. The suspension of ground-up tissue in water is not uniform, for the hard muscle fibres do not disintegrate easily, and these fall to the bottom, while the lighter and softer tissues make a more stable suspension. There are errors of calibration in the pipettes and measuring flasks. Much more important than the above sources of error is that due to the variable composition and reaction of the agar medium. The materials, as bought, vary from time to time, and the preparation is usually the work of laboratory assistants who cannot always be overseen.

If the number of micro-organisms inoculated is large—some grow so slowly as not to be recognised, the excessive number of organisms may diminish the nutritive capacity of the medium. Thus, a series of inoculations from "decimal dilutions" do not necessarily give multiples of 10 organisms, for the lower dilutions usually give fewer colonies than they ought to give.

On the whole we may neglect these experimental errors as likely to be insignificant in comparison with those attributable to random sampling.

#### The Original Error of Random Sampling.

The mussels sampled are variable with respect to the quantity of micro-organisms taken up and retained, just as they are variable with respect to any morphological or physiological character that can be measured. Therefore, any sample of n mussels will differ from any other sample of n mussels collected from the same small area. Even if that sub-area is quite small it will still contain a very great number of mussels.

This error might be minimised by taking a larger sample or several small samples, but, since the area (and those contiguous to it) must be sampled many times in different conditions, one small sample at a time will usually be all that can be managed.

# The Error of Random Sampling of the Emulsion.

The plates made are "read" after a standard period of incubation. The following numbers represent the counts of colonies growing on 20 plates, each of which had been inoculated with one c.c. taken from 250 c.c. of the emulsion of 5 mussels in 250 c.c. of water:—7, 24, 40, 15, 22, 20, 17, 9, 16, 29, 7, 9, 10, 26, 15, 11, 21, 17, 10, 41. Now what we are trying to find is the number of micro-organisms contained in 250 c.c. of emulsion, that is, in 5 mussels. We can only (conveniently) count the number contained in a rather small fraction (1/250th) of this volume of liquid, and we find that there are 7 or 24 or 40, and so on, organisms in this "unit volume" of one c.c. Each of these is a real count, an actual fact, a possible value for the number of micro-organisms, of a certain category, contained in 1/250th part of our sample.

The obvious procedure is now to find the mean of the 20 counts. This is about 18, and multiplying 18 by 50 we find the mean number of organisms in our average mussel, that is, 900. We might let the result go at that, but we may carry the investigation a little further by finding one of two statistical functions, the probable error of the mean, or the probable error of the distribution of the deviations from the mean. To obtain these functions we must calculate the standard deviation of the series given, and this is a very simple matter. Having obtained the standard deviation,  $\sigma$ , we find  $\pm 0.6745\sigma/\sqrt{20}$ ,

which is the probable error of the mean, or  $\pm 0.6745 \, \sigma$ , which is the probable error of the deviations from the mean.

If our desired result had been some single-valued, natural constant, say a molecular weight, we should have expressed it as mean  $\pm 0.6745\sigma/\sqrt{20}$ . We should have assumed that

there could only be one value for the physical constant, but (because of our experimental methods) there were errors in our estimation of this value, and that these errors were as often in excess of the "real" value as they were in defect of it. The most probable value of the constant would have been the mean of our multiple estimations, but this could only have been the most probable value, and there were other admissible ones. By a recognised convention we "commute" these other admissible values, restricting them to the very small range given by our expression for the probable error of the mean.

But whenever we deal with a number of organisms individually, variable in their morphology and functioning, every measurable character is a variable: it has many values. We cannot examine all the organisms in the "population," and so we take a sample consisting of only a few. Any sample will differ from any other, and so the error of random sampling arises. So, likewise, there are 250 one-c.c. samples in our measuring flask, and we can only examine (say) 20. The micro-organisms are not evenly distributed throughout the whole contents of the flask, and so any sample of 20 c.cs. will differ from any other sample of 20 c.cs.

In such a sample of 20 c.cs. the mean is only an abstraction and does not represent anything "real." Each of the c.cs., with its count, is real, and each is a possible and admissible value, but some of the counts are more probable than others. So we form the function  $\pm 0.6745~\sigma$ , the probable error of the series. The mean we take as about 18,  $\sigma$  is about 10, so that the probable error =PE is about 7. The convention is to say that our number of organisms per c.c. is  $mean~\pm PE$ , that is,  $18~\pm~7$ , or 11 to 25. It can be shown that (given certain conditions)

one half of all our counts lay between the limits 11 to 25 and one half outside those limits, that is between 7 and 11, and between 25 and 41. Usually it would be said that the number of colonies on a plate was from 11 to 25, any one of the values included in this range being (conventionally) an equally admissible value. Expanding these figures we get 550 to 1,250 as our mean number of organisms present in a mussel.

Now this estimate would be a fallacious one: it depends on the assumption that the mean is the most probable of all the values, and that the deviations in defect of the mean are generally equal to those in excess of the mean. In other words, the curve of variability of counts of colonies on a plate is assumed to be symmetrical (and in theory Gaussian).

The curve is really J-shaped, that is, extremely asymmetrical, as we see by grouping the counts, as follows:—

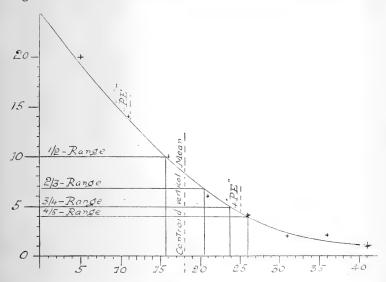
No. of Colonies = 
$$6-10$$
,  $11-15$ ,  $16-20$ ,  $21-25$ ,  $26-30$ ,  $31-35$ ,  $36-40$ ,  $41-45$   
Frequency =  $6$  3 4 3 2 0 1 1

The most probable range of counts is 6-10, and the least probable is 41-45. This means that we must find some other estimate of error due to random sampling. Curves of variability are often so nearly symmetrical as to make the method just described an admissible one, but in this case (and theory suggests that the result is to be expected) the curve is asymmetrical to the extreme degree, and the concept of a standard deviation has no meaning and is misleading.

We can deal with these figures by a graphic method which appears to be as accurate (all the circumstances being considered) as it need be. We make a new "summational" curve by adding, in succession, the frequencies from the right-hand term, thus:—

```
No. of Colonies = 6-10, 11-15, 16-20, 21-25, 26-30, 31-35, 36-40, 41-45
Frequency = 6 3 4 3 2 0 1 1
Summed frequencies = 20 14 11 7 4 2 2 1
```

Then we plot the summed frequencies, as in the following figure:—



The point 1 being plotted against 41 on the horizontal axis, 2 against 36, 2 against 31, 4 against 26, and so on. A smooth curve is then drawn as evenly as possible and as near as possible to all these + points. This is an approximation to the integral curve, that is, if f(x) be the function representing the frequencies,  $\int f(x)dx$  is the function representing the summed frequencies, and it is given approximately by the summational curve.

Now it is easy to see, from the construction of the figure, that any point on this curve gives us the sum of the frequencies from the right-hand limit up to that point, and so we can interpolate graphically for any desired range of counts of colonies. The mode, or greatest frequency, is at the extreme left of the curve. Now the range of the vertical scale (No. of colonies on a plate) is 20, and we take  $\frac{1}{2}$ ,  $\frac{2}{3}$ ,  $\frac{3}{4}$ ,  $\frac{4}{6}$ , etc., of this and measure off these distances from above downwards, thus

getting the vertical points 10, 13·3, 15 and 16, read from above downwards. From these points horizontal lines, "½-range," etc., are drawn to cut the curve, and from the latter points of intersection vertical lines are drawn to cut the horizontal axis. The points on the latter axis thus obtained, and read on the horizontal scale, give us various ranges of counts with their respective probabilities of occurrence. Thus:—

```
1 of all the counts lie between 0 and 16,
2 0, , , , 0 and 21,
3 1, , , , 0 and 24,
4 1, , , , 0 and 26,
```

#### and also :--

It is 1 to 1 that there were less than 16 colonies on a plate.

It is 2 to 1	"	"	21	,,	23
It is 3 to 1	,,	,,	24	,,	.,
It is 4 to 1	,,	. ,,	26	,,	,,

We have now clear ideas as to the degree of pollution, for any one of the admissible estimates is qualified by its degree of probability. If we wish to minimise the pollution we might say that there were less than 16 colonies on a plate, that is, less than  $16 \times 50 = 800$  organisms per average mussel, but we ought also to say that the chance that there are less than this number, and not more, is only 1 to 1—"the odds are even." If, on the other hand, we wish to make the pollution as grave as possible we might say that there were not less than 26 colonies per plate, that is, not less than  $26 \times 50 = 1,300$  organisms per average mussel, but the probability that the pollution is less than this is 4 to 1.

Suppose that we make 1,000 organisms per average mussel a "standard of permissible impurity." This corresponds to a count of 20 colonies on a plate. Reading the graph inversely we see that 20 colonies per plate corresponds to a range of  $\frac{2}{3}$  of the total range of counts, and thus the probability that the number of organisms found was less than the standard was

2 in 3 chances, while the probability that the number was greater than the standard was 1 in 3 chances.

Even if the curve of variability is not an exponential (as it is above) but the ordinary curve of organic variability, rising from zero to a maximum and falling to zero again, we can still apply the foregoing method. The summational curve will then tend to flatten out at its extremities and to have a change of curvature (a point of inflexion) somewhere. The abscissa of this point of inflexion gives the maximum, and then we can find the corresponding point on the vertical scale and measure off distances on the latter on either side of this point giving the various ranges. The rest of the work is as in the figure.

Evidently, therefore, one plate cannot give us a reliable measure of the pollution, and the meticulous care with which plates are sometimes counted is labour wasted. We must have a measure of variability, and even 20 plates, as in the above experiment, is sometimes not enough.

Another method which is now often employed is the inoculation of tubes of bile-salt broth by unit quantities of a polluted liquid diluted in various degrees. Thus the analysis referred to above might have been made in the following way:—

1 c.c. of the liquid in the flask is put into a tube of bile-salt broth. If a positive reaction occurs there must have been at least one organism in 1/50th mussel;

1 c.c. is mixed with 9 c.c. of sterile water, and then 1 c.c. of the dilution is inoculated as before. If a positive reaction occurs there must have been at least one organism in 1/500th mussel;

And so on, through a series of decimal dilutions.

# The Error of Random Sampling in Houston's Method of Decimal Dilutions with Bile-Salt Broth.

Let the liquid of the measuring flask be diluted 10, 100, 1,000, etc., times: we thus get a series of liquids, one c.c. of which contains respectively 1/50th, 1/50th, 1/5,000th, 1/50,000th, etc., part of an average mussel. One c.c. of each dilution is put into a tube of bile-salt broth, and the cultures are incubated. The tubes containing 1/50 and 1/500th mussel give (say) positive reactions, and the tubes containing 1/5,000th and 1/50,000th show no change, that is,

1/50th +, 1/500th +, 1/5,000th -, 1/50,000th -.

There must, therefore, have been at least one micro-organism in the c.c. of liquids inoculating the first two tubes, and there could not have been any in the c.c. used to inoculate the third and fourth tubes. The inference is that there are at least 500 organisms present in an average mussel, but there are less than 5,000. If only one tube is inoculated from each dilution this method gives apparently definite results. The limits of dilution used above are wide ones, but obviously they can be narrowed so that we could apparently get closer approximations to the degree of pollution.

Now each c.c. of the 1/500th dilution was assumed to contain at least one micro-organism, and in making the 1/5,000th dilution at least one organism must have been introduced. One c.c. of the 10 was taken, so that it is not absolutely certain that the higher dilution is sterile. Therefore this method only gives a probability of infection.

This becomes quite evident when a number of tubes are inoculated from each dilution.

The following examples illustrate this:—

(1) Dilution 0 ... ... ... ... ... ... +++++++++ ,, 1/10th ... ... ... ... ++++++++++ ,, 1/100th ... ... ... + a a a a a a a a a ,, 1/1000th ... ... ... ------ Where "dilution 0" represents 1 c.c. of a liquid suspension, made as indicated above, and is taken as equivalent to 1/50th part of an average mussel. The sign "+" represents formation of acid and gas in the bile-salt broth; "a" represents formation of acid only, and the negative sign indicates absence of any reaction.

0.00010111							
1 c.c. poll	uted	sea-wate	er	***			+++ $a$ $a$
1/10th	. ,,	,,			•••		
1 c.c. poll	uted	water			• • • •		++ a
1/10th	,,	,,,					a $a$
1/100th	,,	,,		•••			
1 c.c. poll	$_{ m ited}$	water			•••		+++++
1/10th	,,	99			• • •		+ + + + +
1/100th	99	,,	* *.*				+++++
1/1,000th	,,	29					+ a a a -
1/10,000th	,,	,,			, • • •		a
1 c.c. polle	ited	water		• • •			+++++
1/10th	,,	,,					+++++
1/100th	,,	**					+++
1/1,000th	,,	٠.,					
1 c.c. pollu	ited	water				+++++	+++++
1/10th						+ + a a a	a
1/100th	,,	,,					
	1/10th 1 e.e. polls 1/10th 1/100th 1 c.e. polls 1/10th 1/1,000th 1/1,000th 1 e.e. polls 1/10th 1/1,000th 1/1,000th 1/1,00th	1 c.c. polluted 1/10th ,, 1 c.c. polluted 1/10th ,, 1/100th ,, 1 c.c. polluted 1/10th ,, 1/100th ,, 1/1,000th ,, 1/10,000th ,, 1/10,000th ,, 1/1,000th ,, 1/1,000	1 c.c. polluted sea-water 1/10th ,, ,, ,, ,, ,, ,, ,, ,, ,, ,, ,, ,, ,,	1 c.c. polluted sea-water 1/10th ,, ,,  1 c.c. polluted water 1/10th ,, ,,  1/100th ,, ,,  1 c.c. polluted water 1/10th ,, ,,  1/100th ,, ,, 1/1,000th ,, ,,  1/10,000th ,, ,,  1/10th ,, ,,  1 c.c. polluted water  1/10th	1 c.c. polluted sea-water 1/10th ,, ,,  1 c.c. polluted water 1/10th ,, ,,  1/100th ,, ,, 1/100th ,, ,, 1/1,000th ,, ,, 1/10,000th ,, ,, 1/10th ,, ,, 1/10th ,, ,, 1/10,000th ,, ,, 1/10th ,, ,, 1/100th ,, ,, 1/100th ,, ,, 1/100th ,, ,,	1 c.c. polluted sea-water	1 c.c. polluted sea-water

In all these examples the tubes were incubated for 24 hours. In the following experiment the tubes were incubated for 4 days, and "read" at intervals of 24 hours:—

			1 Day.	2 Days.	3 Days.	4 Days.
(7)	1/50	mussel	5+	5+	5+	5+
	1/500	99	4+, 1-	4+,1-	4+, 1-	4+,1-
	1/5,000	,, .	2+, 3-	5+	5+	5+
	1/50,000	99	5 —	1+, 2a, 2-	1+ , $2a$ , $2-$	1+, $3a$ , $1-$
	1/500,000	0 ,,	5 —	5	1+, 4-	1+,4-
	1/5,000,0	000 ,,	5 —	5 —	5 -	5 -

Now all these tests, simple and conclusive as they may appear to be when only one culture from each dilution is made, become more difficult to interpret when they are so made that

the error of random sampling appears in the result. From (6) we conclude that organisms of the "Bacillus coli" category are present in 1 c.c. of water and absent in 1/100th c.c. Whether they are present in 1/10th c.c. is a matter of probability. If we had made 10 separate experiments, and considered each separately, we should have found the organisms in 1/10th c.c. in 2 cases, and should have failed to find them in 4 cases. In 4 cases the reaction is ambiguous. So also with the other water examples: there is always a rather wide interval between the test which gives unequivocal evidence of the presence of organisms and that which is unequivocal as to absence.

When we employ a "factor" to convert the experimental results into numerical estimates of the pollution this margin of error becomes greater. In Example (7), organisms fermenting glucose and growing in bile-salt media are present in 1/5,000th part of a mussel, and are absent in 1/5,000,000th part. They may be present or absent in 1/50,000th and in 1/500,000th part, and whether they are present or absent is a probability easily read from the results as they are stated. This conclusion applies to the cultures examined after 4 days of incubation. If the 1-day cultures had been all that were made, the conclusion would have been that polluting organisms were present in 1/50th mussel, present or absent in 1/500th and 1/5,000th mussel, and absent in 1/50,000th mussel. The chance that they were present in 1/500th was 4 in 5, and that they were present in 1/5,000th was 2 in 5.

# The Error of Expectation.

In the last example there were 5 samples of 1 c.c., each taken from a flask containing 250 c.c. Of these 5 samples 2 were positive and 3 negative. It is a rather näive assumption to expect that  $50 \times 2$  c.cs. in the flask would be positive and  $50 \times 3$  would be negative.

Let there be an experience of n cases, and let p/n of these be positive and q/n negative, p/n + q/n = 1. Now let there be a second experience of m cases: what percentage of these may we expect to be positive? If n is very great compared with m we expect to find—

$$100 \frac{p}{n} \pm 67.45 \sqrt{\frac{p/n \times q/n}{m}},$$

but if, as in this case, m is greater than n we expect—

$$100 \frac{p}{n} \pm 67.45 \sqrt{\frac{p}{n} \times \frac{q}{n} (\frac{1}{m} + \frac{1}{n})}$$

When we sample the sea-water in the neighbourhood of a mussel bed we usually examine a few c.cs., say 10 (though it is not often that the sampling is so thorough as this). Ten plates, let us say, are made, and these give n,  $n_1$ ,  $n_2$ , etc., colonies. The mean is  $m \pm PE$ , when we consider the probable error. Now there are certainly very many cubic metres of water in the neighbourhood of the spot where our 10 c.c. samples were collected. Are we to assume every c.c. of this volume contains  $n \pm PE$  organisms? The treatment of the error of expectation will follow the lines already indicated.

The statistical precautions suggested here are in no way fantastic: one really does all this sort of thing, in a quite irregular way of course, every day in the ordinary affairs of life when we "take chances." It is all done in a systematised and skilled way by actuaries and insurance companies, and in a practical, unscientific way by bookmakers.

#### The Indicators of Faecal Pollution.

What is looked for in such analyses as we have discussed are not the germs that cause enteric fever, but indications that such germs may, in certain circumstances, be present simul-

taneously. If there were no enteric fever in England, or if all enteric cases were rigorously and successfully segregated, and all "carriers" isolated in some way not yet suggested, the case against shell-fish as disseminators of the disease would have very little force, and interference with the industry would have no utility or purpose.

Glucose-fermenting organisms growing in bile-salt broth, lactose-fermenting organisms growing on McConkey, Bacillus coli, "coliform" organisms, or whatever else we call them, are indicators of risk only, not actual causes of risk. They are assumed to be "faecal organisms," which have entered the mussels via sea-water, sewers and drains, and water-closets from the human intestine. If that was where they came from, then the actual virulent germs of enteric fever may also travel along the same paths and also enter the mussels. Shell-fish beds are "closed," not because the molluscs contain Bacillus typhosus, but because, containing "faecal organisms," they may also contain B. typhosus, which is a faecal organism in its most characteristic and significant environment.

The cogency of a bacteriological case for the closing of a mussel bed, or for any other penal restriction or prohibition, depends, therefore, on the recognition of organisms infecting mussels as truly faecal organisms. They must have come originally from the human intestine, or from an animal intestine, if there are animals that suffer from enteric fever communicable to man, and from no where else. It is useless to show that they proceed from street washings, or cattle or horse manure, or the faeces of fish and sea-birds, or sewage-contaminated soil, unless it is known positively that the germs of enteric fever may persist with undiminished virulence in such materials. An organism growing on McConkey would have no significance for us in this connection, if we could show that it is capable of maintaining itself in a medium apart from any possibility of contamination with human excreta.

#### What is an Exclusively Faecal Organism?

In the Lancashire Sea-Fisheries Laboratory Report for 1914, I gave a list of some of the characters of organisms isolated from mussels. Since then this list has been extended, and the fuller details accumulated may be given here. The methods of identification of lactose-fermenting species evolved by McConkey have been utilised since, these seem to be the most useful and satisfactory. A source of incertitude experienced was the conclusive recognition of "motility," and the cases where this was doubtful are omitted.

The sign + means the production of acid and gas in the medium, and the sign - indicates either no reaction at all or an incomplete one. In the case of the indole reaction, the Voges and Proskauer reaction, "V. and P.," and the motility observation, the symbols mean respectively a definite, positive reaction and no change. The numbers "M. 71," etc., in the first column relate to McConkey's identifications.\* In all cases the reaction to inulin was tried, but this was invariably negative so it has been omitted from the tables.

<sup>\* &</sup>quot; Lactose fermenting organisms," Journal of Hygiene, Vol. IX, No. 1, 1909.

Red Colonies identifiable from McConkey's Tables.

Organism.	Glucose.	Lactose.	Cane.	Dulcite.	Adonite.	Indole.	V. and P.	Motility.	From polluted mussels.	From polluted sea-water.
М. 101	+	+	+	_	+	+	_	_	20	8
B. neapolitanus	+	+	+	+	_	+	_	-	7	19
B. vesiculosus	+	+	_	-		+		-	12	8
M. 71	+	+	+	+	_	+		+	9	3
B. acidi lactici	+	+	_	-	+	+	-	-	7	4
B. lactisaerogenes,	+	+	+	_	+	_	+	+	10	1 '
B. dysenteriae vitellorum										_
B. grunthal,									7	3
B. castellus	+	7			_	+	_	+	,	3
B. coscoroba	+	4	_			_		_	6	1
M. 106	+	+	+			+	_	+	- 5	1
B. coli communis		'				•		•		-
B. cavicida	+	+	_	+	_	+	_	+	0	6
M. 67	+	+	+	+	+	_	+		6	0
М. 66	+	+	+	+	+	+	_	_	3	2
M. 100	+	+	+		+	+		+	2	2
B. cloacae	+	+	+	_	-	_	_	+	4	0
B. schafferi	+	+		+		+			3	1
B. rhinoscleroma	+	+	+	+	+	_		-	$\frac{2}{1}$	1
M. 74	+	+	+	+	_			+		2
M. 1	+	+		_	+	+	-	+	3	0
B. capsulatus	+	+	+	_	+	-	+		1	0
M. 73	+	+	+	+			+	+	1	0
М. 6	+	+	-	-	_	+	+	-	1	0
									110	62

247 organisms; 51 categories, including unidentifiable forms. In 19 cases motility was not observed, or was so doubtful that it was not recorded.

Red Colonies unidentifiable from McConkey's Tables.

Organism.	Glucose.	Lactose.	Cane.	Dulcite.	Adonite.	Indole.	V. and P.	Motility.	From polluted mussels.	I	From polluted ea-water
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26	+++++++++++++++++++++++++++++++++++++++	+++++++++++++++++++++++++++++++++++++++	++++++++++++++++++	+++-++	++++ -+++	+++	+ + + + + + + + + + + + + + + + + + + +	-++	5 6 7 7 4 4 0 0 4 0 0 1 1 1 1 1 1 1 1 1 1 1 1 1		3 1 0 1 2 1 5 0 2 2 1 1 1 1 0 0 0 0 0 0 0 0 0 0 0 0 0
									53		22

White or Transparent, Colourless, Surface Colonies isolated from Mussels.

	Clucose. Lactose. Cane.		Adonite.	Sorbite.	Mannite.	Indole.	V. and P.	Motility.	Milk.	95.
1										
	a	U	0	0	0		+	+	(l	3
2	a	0	0	0	a	-	+	+	a	$\frac{2}{2}$
3	a	(l	0	a	0	-		+	ac	2
4	a	(t	0	et	$\epsilon t$	+		+	u	1
5	a	(t	re	0	0	_	+	+	$\alpha$	1
6	a	u	0	a	0	_	witness		0	1
7	$\alpha$	0	0	0	()	_	+	+	0	1
8	a	rt.	0	0	0		+	+	0	1
9	a	11	0	12	0	-	+	+	a	1
10	a	0	O	a	0		+	+	a	1

The first thing that would occur to a systematist on looking at these lists is the extraordinary number of "species." There are 247 organisms in the first two lists, and these fall into 51 categories. It would be safe to say that no one could collect, at random, 247 individual animals or plants belonging to (say) the same order, and find 51 separate species. Now in these identifications there are 8 reactions, each of which may be positive or negative, that is, 16 characters in all. At first the results seem to suggest that we are dealing with a series of combinations of the given characters, which are simply chance combinations, but this is not the case. We may compare the series of cases given in the tables with the cases that would arise on tossing 8 coins simultaneously. The first coin may fall in two ways, and each of these ways is to be associated with the two ways in which the second coin may fall, and so on, giving the number of possible results =  $2 \times 2 \times 2$ , etc., or  $2^8 = 256$  combinations. But our first reaction, that with glucose, which ought to occur in two ways, only occurs in one way (the coin is "loaded" so that it always falls heads), and this is also the case with the reaction to lactose. reaction to inulin was always negative (the coin always falls tails). The reaction to cane sugar occurs in the positive way far oftener than in the negative way (the coin is loaded so that it falls more often heads than tails), and just the opposite is the case with the V. and P. reaction. So likewise with the other reactions. The number of actual results is far less than the number of possible results.

Looking at the frequencies of occurrence of the various results we see that these are 28, 26, 20, 12, 11, 11, 10, 7, 6, 6, 6, 5, 4, 4, 4, 3, 3, 3, 1, etc., where the first five terms represent the relative frequencies of the organisms, McConkey's No. 101, B. neapolitanus, B. vesiculosus, B. acidi lactici and B. lactis aerogenes, or we may look at the results in another way: one-third of all the organisms identified, and nearly one-half

of all those studied are glucose +, lactose +, indole +, V. and P. -, immotile forms. The method of analysis certainly enables one to describe the majority of the bacteria isolated as belonging to fairly well-marked categories, each of which is distinguished by a set of similar characters, the number of which is, to some extent, a compromise.

The next step towards making bacteriological analyses afford definite results is to associate each of these categories with some combination of physical characters. We may suppose that "typical" B. coli communis, and other organisms, exist in large numbers in the human alimentary canal and reproduce there, preserving their characters. But before entering the shell cavities and alimentary canals of mussels they traverse drains, sewers and sea-water, so that their further multiplication occurs in various environmental conditions. Probably they are highly mutable, and since a large number of successive generations may appear in a relatively short time there is opportunity for selection, and so for the production of new strains in a comparatively short time. If the period elapsing between the voiding of typical B. coli communis, or other truly faecal organisms, and the infection of shell-fish were a very short one, we might expect to recognise these organisms in the latter habitat in relatively large numbers, but if this period is prolonged we ought to find the descendants of mutants of the original strain. We ought to be able to conclude, from identification of the prevalent "species" of bacteria found in the shell-fish, what were the environmental histories of the infecting organisms; whether the latter had had their origin in the human intestine, or in some other source, uncontaminated by faeces, and what was the probable duration of the period after leaving the human and entering the molluscan alimentary canal. This recognition of the relative significance of the various kinds of bacteria that may be found in polluted mussels or sea-water is the really important

thing at the present time. It is improbable that we have the necessary knowledge to enable us to make the recognition, but it is quite certain that well-planned "team-work," carried on by some public authority for a year or two, would afford the knowledge.

As it is, our existing methods of assessing the significance of bacterial pollution of shell-fish only give us rather crude indications of possible or probable risk to the public health. In some cases the pollution may be gross and patent, and in others it may be so slight as to be negligible, but the majority of cases that arise lie between these extremes. Crude indications, such as existing methods of analysis give, are good enough evidence to justify public authorities in constructing and using purification plant, or in making better disposal of their sewage.

But it may reasonably be urged that they are not good enough to be the bases of penal restrictions. They ought to be interpreted in the way a statute is construed when the issue is a criminal one, that is, literally. An Order closing a mussel bed creates a new offence, which is only immediately the infraction of the Order, but which is really the selling, as human food, of commodities which involve risk to the public health. It ought to be demanded of those making such a new offence what is the degree of risk which the Order attempts to obviate or minimise, and if this cannot be stated with some precision there is really no excuse for making a restriction or prohibition that involves fine or imprisonment.

Experience of such Orders as have been made, or attempted, during the last few years shows that the evidence on which the creation of the new offence is based is (sometimes at least) vague and ambiguous. It may not even be necessary for the authority making an Order to disclose its evidence, and it is left for those whom the authority proposes to restrict under pain of fine or imprisonment to "show cause" why

they should not be restricted. This procedure may well be in the spirit of much recent legislation, but it seems, none the less, arbitrary and unfair, and scientific men who make analyses for public authorities ought to take care to express their results in such a way as not to commit themselves to facilitating it.

#### PEARL-LIKE CONCRETIONS IN TRIPE.

By Jas. Johnstone, D.Sc.

Early in 1919 Dr. Hanna, Medical Officer of Health for the Port of Liverpool, sent me some pieces of tripe which formed part of large consignments reaching this country from America. The tripe had been cleaned, boiled and frozen, and came in a tightly packed condition. Each of the pieces examined contained hundreds of small glittering, perfectly spherical pearl-like bodies varying between one and two millimetres in diameter. They were very hard in texture and easily detached from the tissues in which they were embedded.

Sections of the tissue containing these pearl-like bodies were made, but the results were not satisfactory. processes of cleaning the mucous coat of the intestine had been removed and the submucous coat was very greatly altered by the freezing. The loose, spongy texture of the latter had disappeared and was replaced by a honeycomb-like structure with large spaces which became filled with air. The areolar texture had disappeared and the tissue was itself compact and hard. This is the well-known effect of relatively slow freezing of meat and fish tissues. The tissues shrink, forming hard and compact parts with large interstices between them, and their juices gather in the spaces, from which they drain away in the processes of thawing and subsequent handling, with the result of a loss of savoury constituents. It was, therefore, difficult to see what were the relations of the pearl-like bodies to the structures in which they were situated.

Later on, however, Dr. Hanna obtained some fresh tripe, prepared from a cow's stomach, containing similar intrusive bodies. These were examined and sections were made successfully, and it was seen that the conditions were the same as in

the frozen tissues. Even in the fresh tripe the mucous layer had entirely disappeared but the submucous layer and the muscular and serous coats were little changed. The "pearl" was easily cut into sections and the latter showed all the typical structure of a fine pearl, such as may be found in shell-fish except, of course, that it was composed of concentric laminæ of hard elastic tissue instead of limy substances. It was embedded in the submucous layer.

Round it the ordinary areolar tissue of the submucosa had been converted into a thin, compact, fibrous cyst containing flattened nuclei. This appears to have been the formative sac. The submucosa thinned out over the "pearl" but was everywhere continuous so that it held the latter in its place, whereas in the frozen material the submucosa had been rubbed away, leaving the pearls lying loose in little cavities from which they were easily dislodged. In the latter case the pearls were very easily seen because of their iridescence, but in the fresh tissue they were covered and were recognised only as protuberances above the general level.

There was no nucleus at the centre of any of these bodies, or at least none that could be recognised, so that it is impossible to say what was their origin. It is almost certain, however, that they are the result of an infection of the tissues of the alimentary canal by a great number of organisms which had gained admittance into the blood stream, had been carried into the small vessels of the submucosa and had been arrested there. Irritation followed, no doubt with the formation of a formative sac which then acted precisely like the similar structure that is the cause of pearl-formation in the tissues of shell-fish. This process of death and degeneration of a larva may often be seen in fish some species of which are particularly liable to infection by Cestode larvae. In the case of such an infection (and doubtless others) the events that occur follow two lines: (1) The infecting larvae or eggs find their specific

host and develop normally in its tissues, thus many skates, rays and dogfishes eat other food animals which contain Tetrarhynchus larvae and the latter undergo transformation and develop into adult tapeworms in the intestine of the fish which is their final host. (2) The same infected food animals may be eaten by a halibut, gurnard or some other fishes which cannot serve as the final hosts. In such a case the Tetrarhynchus larvae become encysted and finally die without undergoing development. That seems to be what has occurred in the tripe-infection now recorded: the organisms (Cestodes or Sporozoan parasites) that acted as the nuclei of the pearl-like bodies did not find their true, final host and so became encysted and destroyed with the results described here.





