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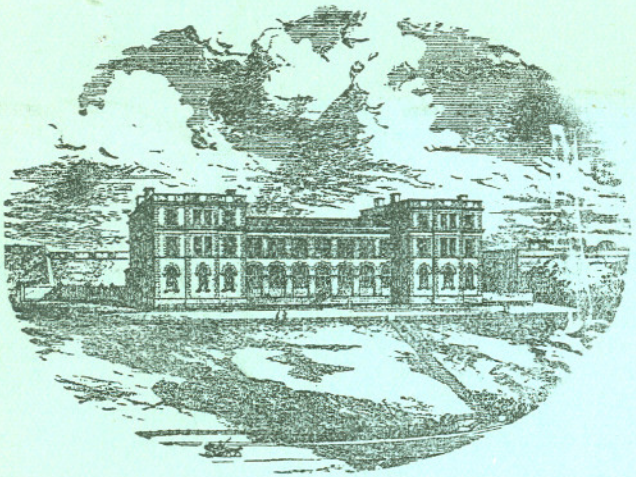

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Rays and Skates (*Raiæ*).

No. I.—Egg-Capsules and Young.

By

Robert S. Clark, M.A., B.Sc.,

Naturalist at the Plymouth Laboratory.

With Figures 1–20 in the Text.

CONTENTS

INTRODUCTION.

THE family Raiidæ belongs to the division Batoidei of the sub-order Hypotremata, and is represented by the principal genera Raia, Psammobatis, and Sympterygia (Regan, 1906). The genus Raia (an altered spelling of Raja) was instituted by Linnæus, after Artedi, for the type *Raja batis* in the Systema Naturæ, Ed. X, Vol. I, 1758. The species are numerous and mostly of northern distribution. They are not very well defined, and the synonymy of many of them is distinctly doubtful, and in some cases quite erroneous, e.g.

(1) *R. bathyphila* Holt and Byrne=*R. ingolfiana* Lütken=*R. lintea* Fries.

(2) *R. falsavela* Smitt (non. synonymy)=*R. circularis* Collett=*R. fyllæ* Lütken.

(3) *R. miraletus* Le Danois (non. syn.)=*R. nævus* Müller und Henle.

The extraordinary amount of variation in individuals of the same species has led the more recent systematists to adopt a classification according to geographical distribution, e.g. Jordan and Evermann (1896) and Garman (1913). Commercially, they are known to the Trade as Rays and Skates, but this distinction is not sharply defined, though the idea might be copied with advantage in making a subdivision of the genus.

Little is known of the life history and rate of growth of these fishes.

The present paper deals with the egg-capsules and young of nine British species, and is based on living eggs which were secured at Plymouth from known adult fish, and which were kept alive through the period of incubation under the circulation of sea water in the Laboratory tanks. The particular objects of the investigation were to obtain more precise information of the less known features of embryonic development, to determine the period of incubation of the embryo, to secure the newly hatched young fish, and to fix the character and extent of the post-embryonic changes.

The more interesting and important points with reference to the structure and function of the branchial filaments and to the absorption of the caudal fin require more comprehensive treatment than has been possible in the present work.

The photographs are reproduced from non-contact prints which were taken from the writer's negatives by Mr. C. Gill, Press Photographer, Plymouth.

The writer is greatly indebted to Mr. A. J. Smith, of Plymouth Labora-

tory, for securing a regular series of egg-capsules from Plymouth fish quay; to Professor A. Meek and Mr. B. Storrow for a consignment of *R. batis* capsules from North Shields; and to Dr. A. Bowman, Aberdeen, for the loan of an excellent series of young forms of rare species from the deeper waters of the North Sea and from the North-West of Scotland.

Explanation of Measurements of Capsules and Post-embryonic Stages.

Capsules.

Length	Length along the median longitudinal axis of the shell, excluding the horns.
Width	Greatest width of the shell, excluding attachment threads.

Post-embryonic Stages.

Total length	Tip of snout to tip of caudal.
Length of disc	Tip of snout along median longitudinal axis of the fish to the line joining the posterior margins of the pectorals.
Width of disc	Greatest width across the outer angles of the pectorals.
Snout	Tip of snout along the median axis to line joining anterior margins of orbits.
Interorbit	Narrowest width between the orbits.
Snout to tip of ventrals	Tip of snout along the median axis to line joining the posterior margins of the distal lobes of the ventrals.
Snout to vent	Tip of snout along median axis to anterior margin of vent.
Tail	Tip of caudal to junction of tail and ventrals.
2nd dorsal to tip of tail	Posterior margin of base of second dorsal fin to tip of caudal.
Præoral	Tip of snout along median axis to middle of closed mouth (meeting of middle series of teeth of both jaws).
Internasal	Narrowest width between inner margins of nostrils (anterior groove).
Prænasal	Tip of snout along median axis to line joining anterior margin of nostrils.
Teeth	Vertical rows in upper jaw.

To obtain a certain amount of uniformity in the measurements, each

fish was placed ventral side downwards on a flat surface and the outline traced out. The measurements were then taken from the tracing. Those measurements which were recorded in this way were total length, length of disc, and snout to tip of ventrals.

Stage 1 represents the newly hatched young.

„ 2 represents the young a few months old undergoing changes in shape of disc and in length of caudal.

Stages A and *B* of *Raia fyllæ* represent stages approximate to 1 and 2.

GENERAL SCHEME.

The following scheme was adopted on the strength of our knowledge that the egg is fertilised in the upper reaches of the oviduct, is completely enclosed with its yolk and albumen in a capsule which is formed by the shell gland, passes down the oviduct comparatively quickly as a complete fertilised product and is ejected from the cloaca.

Most of the egg-capsules which are treated here were taken from the cloaca of the adult fish as they were landed on the fish quay. After being received at the Laboratory, they were measured and labelled and transferred to tanks under the usual circulation of sea water. Small opal-glass labels were used with numbers and dates appended in pencil, and these were attached to the egg-capsule by silk thread, which had previously been immersed in liquid paraffin wax. This precaution is necessary owing to the decomposing effect of sea water and to the injurious action of bacteria. Marking with waterproof Indian ink on the flat portion of the shell between the long horns is just as efficient.

The mortality in the eggs was high, and this may be explained largely by rough handling of the adult fish and by artificial extraction of the capsule and the resultant jar on the yolk, even allowing for the possible use of the thick albuminous layer as a "buffer." A small percentage of perfectly formed, undamaged egg-capsules contained some albumen but no yolk, and these had evidently been closed before the eggs had passed down from the upper reaches of the oviduct. There were also a few very abnormal egg-capsules, obviously too small to receive fertilised eggs, and yet in two cases (*R. clavata*) contained yolk about the size of a pea. Unfortunately, the adult fish from which these eggs were taken were not secured for examination. In two species only were sufficient numbers of capsules obtained for a reliable percentage of successes. *R. clavata* gave 13% out of a total of 240 capsules, and *R. brachyura* 15% out of 127.

One must not lose sight of the fact that these eggs were reared under conditions more or less abnormal, where the temperatures and salinities were, on the average, a good deal higher than in natural conditions. The

results, however, are interesting and show that rearing under artificial conditions is quite possible. They show quite definitely that fertilisation is effected before the egg is enclosed in its shell. There are a few facts to look out for in the process. After about two months (the period varies with the species and with temperature and salinity conditions) in sea water, the albumen of the egg is absorbed, the slits at the base of the horns are then open, and it is dangerous to remove the eggs from the sea water. Bubbles of air collect inside the capsule and these prove fatal to development.

Capsules of *Raia clavata* were found to be quite easy to rear, and as the development covers only a few months, it is suggested that they would be more convenient for developmental studies than a larger form like *R. batis*, in which development is slower.

It was found to be more convenient to pull off the attachment filaments from those egg-capsules which had them, as they collected bubbles of air, which seemed to disturb the equilibrium of the shell.

SPECIES OCCURRING AT PLYMOUTH.

1. Raia clavata Linn. Thornback.
2. R. maculata Montagu. Homelyn.
3. R. brachyura Lafont. Blonde.
   syn. *R. blanda* Holt & Calderwood.
4. R. microcellata Montagu. Small-eyed Ray.
5. R. undulata Lacépède. Painted Ray (non. Couch).
   syn. *R. picta* Lacépède.
6. R. nævus Müller & Henle. Cuckoo Ray.
7. R. circularis Couch. Sandy Ray (Couch).
8. R. fullonica Linn. Shagreen Ray (Skate).
9. R. batis Linn. Blue or Grey Skate.
10. R. marginata Lacépède. White-bellied Skate. Bordered Ray (Young).
    syn. *R. alba* Lacépède. (Bottlenose Skate.)
11. R. vomer Fries. Long-nosed Skate.

Numbers 1, 2, 3, 4, 6, 8, 9 are of frequent occurrence in the neighbourhood and are taken at all stages.

Numbers 5, 7, 10, 11 are periodic in their appearance, but the young of 5 and 10 occur commonly on the Outer Grounds.

Numbers 7, 10, and 11 increase in frequency with deeper water towards the western entrance to the Channel.

The commercial distinction between Rays and Skates is not very well defined at Plymouth. All the white-bellied forms are grouped as Rays, while the blue or dark-bellied forms, even in the immature stage, are known as Skates.

They are gutted and " winged " and find a ready market. The lateral jaw muscles of the large skate are often extracted as " knobs " and sold commercially as food.

ORIENTATION OF EGG-CAPSULE IN THE UTERUS.

The long horns are directed towards the cloaca and the more convex face of the capsule towards the dorsal side of the fish. This is the normal orientation, and has been determined in situ in several hundred examples. Vaillant made the same observation in 1885. The embryo emerges at hatching between the long horns. In Dogfish, the blind end of the capsule is the first to appear at the cloaca, and the future open end is the last to be " presented."

DEPOSITION OF EGG-CAPSULES.

A large female Blonde (*Raia brachyura*) was caught by otter trawl on the inner Eddystone fishing grounds on 5th April, 1922, and was transferred alive to one of the large tanks in the Aquarium. The bottom of this tank had a thick covering of gravel and pebbles. The other occupants of the tank were a large Cod (*Gadus morrhua*), several flat fish (Turbot, Plaice, and Dab), and a few immature Rays (*R. clavata*, *maculata*, and *brachyura*). All male Rays were transferred to another tank, to obviate the risk of vitiating the experiment. Egg-capsules began to be deposited on 12th April, 1922, and continued to be extruded singly at, more or less, regular intervals. At the beginning of the experiment, capsules periodically became visible in the cloaca of the fish, but disappeared rather strangely. It was supposed that the Cod was responsible for their disappearance, but on raking up the bottom of the tank, they were discovered completely buried in the " sand." With a few exceptions, this method of depositing the eggs was adopted by the fish, which extruded twenty-five capsules up to May 31st, 1922. The fish was then still alive, and the process of egg-laying was being continued. The actual method of deposition was not observed.

The capsules, as they appeared in the cloaca of the fish, were marked with string, which was attached to one of the protruding long horns. By this means, more definite information was obtained as to the time occupied in getting rid of the capsule and the period which elapsed before another made its appearance. This point also had an important bearing on the fertilisation of the egg and the degree of development of the germinal disc. Occasionally a capsule would be hung up for a few days in the

cloaca of the fish, but generally one capsule would be followed immediately by another, when a short period of rest would intervene, about twenty-four hours, before the process would be repeated. This appearance of eggs, spawned in pairs, as it were, is in keeping with the maturation of a single ovum from each ovary.

Dean, in "Fishes Living and Fossil," remarked that the eggs of oviparous skates were said to be deposited on sand-flats near the mark of low water, and he gave an observation by Mr. V. N. Edwards, Woods Holl, who believed that they were implanted vertically in the sand. From the occurrence of beds of skate eggs, the latter supposed that the fishes were singularly local in their places of spawning.

Williamson (1913) recorded the capture of skate eggs in considerable numbers on ground sixteen miles S.S.E. of Aberdeen by the trawl towing off the shoal water on Aberdeen bank, but records of living egg-capsules, after deposition, are extremely few. They have probably escaped capture, because of the inaccessibility or the roughness of the grounds on which they have been deposited. The occurrence of spawning fish and the young stages in shallow water tends towards accepting the view that spawning, in most of the species, takes place close inshore.

A few capsules with living embryos have been dredged in Plymouth Sound in water from five to six fathoms. The capsules were those of *R. brachyura* and *R. clavata* and the attachment filaments on the more convex face of the shell were not attached to living seaweeds or rocks in situ, but were firmly fixed to a mass of debris, which included pieces of dead mollusc shells, loose algal fronds, sand, and gravel. The capsules, which were spawned in the Aquarium, quickly picked up loose foreign objects, such as broken pieces of shells and small pebbles, immediately after deposition.

FERTILISATION OF THE EGG.

The large female *Raia brachyura*, which was kept alive and which spawned in the Aquarium, afforded an excellent opportunity for testing the fertility of the eggs. As has already been stated, no males were kept in the same tank, so that all possibility of impregnation since being placed in the tank was eliminated. The fish was captured on 5th April, 1922, and transferred alive to the Aquarium on the same day. One egg was deposited on board ship. Capsules began to be extruded on April 12th and have continued to be deposited up to the time of writing, when a total of twenty-five capsules has been recorded. On 17th May, 1922, more than a month after the extrusion of the first egg in the tank, eight capsules were opened and examined. The series was so arranged that it should include stages from the appearance of the capsule in the cloaca to the full period of thirty-five days after deposition. All these eggs

were fertilised, the oldest having embryos from 5 to 6 mm. in length. Six other capsules were extracted as soon as they appeared in the cloaca of the fish, and the eggs were examined. All showed the circular white halo of the germinal disc, the early cleavage stage as figured by Balfour, Plate 6, Fig. 2. It is worthy of note that all the capsules were perfectly formed. A few capsules, which were taken from the cloaca of adult fish as they were landed on the fish quay, were also opened and showed the same halo condition of the germinal disc. In most cases, it occurred at the long horn end of the yolk and was comparatively of large diameter, ca. 5 mm. On being fixed with osmic acid and corrosive sublimate, it lost the circular shape and became more oval. Undoubtedly, the egg is fertilised just before it is enclosed in its capsule, and the further process of completion and extrusion of the egg-capsule must be fairly rapid. Fertilisation, in this case, must be effected by the sperm, which are stored somewhere, probably in the upper reaches of the oviduct, and which in some way become functional when the ovum matures and passes down to be enveloped by its shell.* Dean, in "Fishes Living and Fossil," notes the appearance of sperm in the upper reaches of the oviduct in Chimæra.

The alternative suggestion of a simultaneous fertilisation of ripe ova in the ovary must be ruled out, as there is nothing to suggest retardation of development after fertilisation.

Lo Bianco found the same thing at Naples, where the eggs of *R. asterias* and *R. undulata*, which were spawned in the tanks, were nearly all fertilised. He suggested the same two alternatives, and laid stress on the greater possibility of a receptaculum seminis. The question is one that may lead to great possibilities and is well worth following up.

EGG-CAPSULES.

STRUCTURE AND COMPOSITION.

The egg-capsules of Rays and Skates have been frequently described and figured. They differ considerably in shape and in size in the various species, but their structure and composition are identical. The chemical composition of the shell has been determined by Hussakof and Welker as resembling Keratin. It consists of several layers of closely packed fibres, which show definite longitudinal striation. The shape is more or less rectangular with the corners prolonged into hollow tubes or horns. These horns may be drawn out into fine points, but the tendril formation of the dogfish capsules is absent entirely. Each horn, as a rule, is provided with a definite slit, which is closed in the early stages by a thick

* Storage of sperm is a common phenomenon amongst invertebrates, but has not been observed very often in vertebrates. Schmidt (1920) describes an interesting case in the viviparous teleost, *Lebistes reticulatus*.

plug of albumen, or by a delicate membrane. These slits vary in position according to the type of capsule. There are always two long and two short horns. The embryo emerges normally on hatching between the long horns, and this is the end which is oriented towards the cloaca of the fish. Authors are far from agreeing as to the naming of each end of the shell, and the term inferior or posterior as used by some for the long horn end is rather confusing. This is the end which is oriented towards the cloaca of the adult, but it is also the end from which the embryo emerges. Thus it may be defined as posterior in relation to the fish or apical in reference to the embryo.

The shell, after being exposed to the action of sea water, becomes brittle. Osmosis undoubtedly takes place, and, as Peyrega's experiments show, a definite equilibrium is reached, both ways, between the outside medium and the internal fluid. As development of the embryo proceeds, the albumen disappears or is absorbed and the capsule is aerated through the open slits on the horns. A certain amount of weathering takes place, chiefly between the long horns, as the embryo gets ready for hatching. The shell at this stage is easily opened and more easily broken in handling. Its life corresponds with the period of incubation of the embryo.

AERATION OF THE EGG-CAPSULE.

Opinions are divided in regard to this interesting feature in the life of the capsule. One school represents the view that there is an inward and outward flow of water through slits on the tubular horns, and the other, with modifications, that such a current is impossible. Wyman (1867) gave definite observations on older capsules of *Raia batis*, a species which has since been determined as *Raia diaphanes* Mitchill. He stated that the outer edge of each horn was the more rounded, and near the free end had an oblong slit for the inward and outward flow of the water which passed through the egg during incubation. He admitted that there might have been an albuminous covering at an earlier period and that this had been absorbed. Owen (1866) had the same opinion when he remarked that in the oviparous sharks the branchial filaments reacted on the streams of water admitted into the egg by the apertures.

On the other hand, Beard (1890) had no hesitation in stating that such an inward and outward current was non-existent, and that there was no mechanism present in the egg which would cause such a current to flow. His explanation was that the ordinary laws of endosmosis and exosmosis were quite sufficient to account for the presence of sea water in the egg-capsule and to provide for its aeration. His opinion of the function of the slits was that they were intended to counteract the effects of pressure. Dumeril (1865) and Moreau (1881) held that the slits were closed by an

excessively thin membrane and that the yolk and embryo were separated from the shell by a thick layer of albumen. Nordgaard (1917), in describing the capsules of *Raia radiata*, was unable to find the tube fissures, and remarked that the gelatinous mass would prevent any ingress of water.

In all the capsules which the writer has examined, *Raia batis* probably excepted, the longitudinal slits are definitely marked and easily recognised in capsules before and after deposition. At first, these fissures are tightly closed by albumen, which may or may not have a delicate covering membrane. Capsules taken from the cloaca of a fish show, when cut open, a thick gelatinous mass of albumen, which is in close contact with the whole of the shell and fills the tubular horns. In the middle of the central cavity a liquid fluid surrounds the yolk. After the capsule is deposited and has been in sea water for some time, the albumen begins to disappear. This disappearance coincides with the growth of the embryo, and especially with the development of the vascular system and the specialised branchial filaments. There is reason to suppose that the branchial filaments may help in the absorption of albumen. With the growth and expansion of the pectorals and their junction with the snout, the albumen has practically vanished. The slits are now open, and on lifting the capsule, at this stage, out of the tank, the internal fluid rapidly drains off. When replaced, the capsule floats on the surface of the water, and unless care be taken to expel every particle of air, it results in the death of the embryo. Experiments with finely powdered carmine grains were made on capsules with well-developed embryos, and a definite current was found to move away generally from one of the long horns. On the capsules being opened, after being submitted to the experiment for a few minutes, carmine grains were found inside the shell and on the spiracles of the embryo. Observations on a living embryo of *Raia nævus*, whose capsule is transparent, showed the use of the elongated caudal fin in expelling the enclosed water.

General osmosis undoubtedly occurs, but it can hardly be accepted as providing for the needs of a growing embryo, whose period of incubation may extend over several months. The slits are probably a supplementary adaptation for aiding in the respiration of the embryo, and are analogous to the respiratory perforations on the shell of Chimæra, cf. Dean (1902).

ORIENTATION OF THE EMBRYO IN THE CAPSULE.

References are given in the present paper to observations on particular embryos at different periods of growth, but the material examined has not been sufficiently large to draw definite conclusions, except in regard to the orientation of the embryo just before hatching. Egg-capsules

of *R. nævus* (Cuckoo Ray) and of *R. marginata* (White-bellied Skate or Bottlenose) were periodically examined in the living condition up to the time of hatching of the embryo. The capsule of *R. nævus*, being transparent, was examined without being disturbed, but in the large skate capsule, observation windows were cut in the shell. In both cases, the embryo had its head end generally facing the short horns of the capsule for the period of development preceding the curling of the outer pectoral angles and the tail, a curling which takes place as growth in width and in length exceeds the dimensions of the central cavity. The embryo, however, was observed to undergo complete turning movements on the horizontal plane, but preserved a constant dorso-ventral position. The embryo emerged normally head first between the long horns. One example, however, of *Raia clavata* proved an exception by emerging between the short horns, which end, as a rule, is firmly closed. The dorso-ventral aspect of the embryo in relation to the flat or the convex side of the capsule, varied considerably, but there appeared a greater tendency, especially in capsules with hooked short horns, for the dorsal side of the embryo to face the more convex side of the shell, cf. Nordgaard (1917), for capsules of *R. radiata*. The orientation of the embryo seemed to bear a definite relation to the position assumed by the capsule after deposition.

PERIOD OF INCUBATION OF THE EMBRYO.

Species.	Average Period in months.	Range in days.	No. of Specimens.	Months.	Years.
Raia clavata	$4\frac{1}{2}$	121–154	22	IV–X	1921
	$5\frac{1}{2}$	167–168	2	VI–XI	1920
R. nævus	8	212	1	VI–II	1920–1
R. maculata	5	145–172	5	V–X	1921
R. brachyura	7	189–219	4	VI–II	1921–22
R. microcellata	ca. 7	240	1	VI–I	1921–22
R. marginata	$14\frac{3}{4}$	449	1	IV–VII	1921–22

The period of incubation refers to embryos which were reared under artificial conditions in the Laboratory tanks. Newly hatched young, approximately the same size, and with similar characters, were secured by trawl in shallow water off Plymouth, about the same time as these tank specimens hatched out. The following table shows the range in width of disc for each of the monthly captures. The number of fish captured is here neglected. The figures represent the range in width of disc in millimetres, while those in thick-faced type show the width of disc of the artificially hatched fish as they occurred.

Examination of the following table gives a good index in regard to the period of spawning at Plymouth of the three species: *R. clavata*,

RECORDS OF SIZES OF EARLY YOUNG FISH CAUGHT IN TRAWL (PLYMOUTH) DURING THE YEARS 1920, 1921, AND 1922, ARRANGED MONTHLY.

Months.	I.	II.	III.	IV.	V.	VI.	VII.	VIII.	IX.	X.	XI.	XII.
R. clavata	84–99	86–100	80–100	82–100	83	80–100	80+	90+	***75–83***	***71–86***	79–90	88–94
R. maculata	99	90	78–95	81–88	—	70–90	—	100	—	***71–79***	—	—
R. brachyura	***103–106***	***115***–151	117–157	130–139	—	—	100–150	110–150	120–150	124	—	***100***
R. nævus	—	***62***	63–88	—	—	—	70–74	—	70–86	—	—	—
R. microcellata	***86***	—	110	—	—	80	—	—	—	—	—	—
R. marginata	—	—	266–276	253–265	—	***190***	—	—	—	—	240	—

R. maculata, and *R. brachyura*. From the small sizes which were caught practically in all the months of the year, from the known period of incubation of the embryo and from the occurrence of the eggs, one arrives at the conclusion, which seems justifiable, that the spawning for these species is prolonged for the greater part of the year. Similar observations of a prolonged spawning period have been recorded by Beard for the common Skate (*R. batis*) and the Cuckoo Ray (*R. nævus*) in the North Sea, by Borcea for the Cuckoo Ray at Roscoff and by Lo Bianco for *R. punctata* (syn. *R. asterias* Delaroche) in the Mediterranean.

The following table gives the record of the occurrence of egg-capsules at Plymouth during the period of the present investigation. It will be noted that the maximum months for the more frequently occurring species were April, May, and June. This is probably the true state of things, but it must be understood that these months represent the period of intensive fishing for Rays and Skates at Plymouth. It coincides with the season for boulter or long-line fishing, which fills in the gap between the winter herring and the summer mackerel fisheries. The numbers tabulated are the frequency numbers and represent the capsules examined during the years 1920, 1921, and part of 1922.

OCCURRENCE OF EGG-CAPSULES AT PLYMOUTH.

Months.	I.	II.	III.	IV.	V.	VI.	VII.	VIII.	IX.	X.	XI.	XII.
Raia nævus	—	3	10	10	19	2	—	—	—	—	—	—
R. undulata	—	—	—	—	—	—	2	—	—	—	—	—
R. microcellata	—	—	—	2	4	3	3	2	—	—	—	—
R. brachyura	—	1	15	32	43	79	34	—	—	—	—	—
R. clavata	—	—	1	15	44	159	51	—	—	—	—	—
R. batis	—	—	—	1	1	—	—	—	—	—	—	—
R. circularis	—	—	—	1	4	—	—	—	—	—	—	—
R. vomer	—	—	—	—	—	—	—	—	2	—	—	—
R. marginata	—	—	—	4	—	2	—	—	—	—	—	—
R. maculata	—	—	—	6	21	5	1	—	—	—	—	—

AVERAGE SIZES OF EGG-CAPSULES SECURED AT PLYMOUTH.

Measurements in mm.

Species.	No. of capsules.	Length (without horns) along the median line.	Width (greatest).
Raia nævus	28	63·4	36·8
R. undulata	2	81·5	52·0
R. microcellata	15	90·8	57·2
R. brachyura	177	128·4	78·5
R. clavata	255	74·9	57·1
R. batis	2	143·5	80·8
R. circularis	3	89·3	50·3
R. vomer	2	133	79·5
R. marginata	6	180·3	138·6
R. maculata	24	70·7	41·8

The foregoing table gives the average sizes of the capsules of ten species, all of which were secured from the Plymouth area. It is interesting to compare these sizes with those recorded by Lo Bianco for Mediterranean forms. The method of measurement is the same in both cases.

SPECIES RECORDED BY LO BIANCO AT NAPLES.

	Length (without horns).	Width (greatest).
R. asterias Rond.	105	60
R. clavata Rond. (*R. petrosa*)	60	45
R. maculata Mont.	65	35
R. oxyrhynchus L. (*R. monaca*)	140	115
R. punctata Risso (*R. d'arena*)	45	30
R. undulata Lacep.	90	45

Considerable variation is apparent in these Mediterranean capsules from the Plymouth specimens. There is the strong suggestion that the mature fish of the same species are of smaller size in the Mediterranean, as Borcea (1905–6) has also remarked, but there is an alternative suggestion that we are dealing with different species. With *R. asterias* Rondelet, the writer is not familiar and cannot give its true synonym until specimens have been examined. *R. oxyrhynchus* L. seems to be distinct from *R. vomer* Fries, to which it is closely allied. *R. punctata* Risso is identical with *R. asterias* Delaroche, which is a definite species, and probably confined to the Mediterranean.

The size of the egg-capsules from the same fish shows considerable variation. Twenty-nine capsules from a female *R. brachyura* which spawned in Plymouth Aquarium supplied the following data :—

Arithmetic means	Length (without horns)	130·5 mm.
	Width (greatest)	77 mm.
Variations from the means	Length	+12·5
		− 6·5
	Width	+11·0
		− 3·0

Nordgaard (1917) remarks that the eggs from the same fish need not be of the same size.

BRANCHIAL FILAMENTS.

The term, branchial filaments, here applies to those temporary external embryonic structures, the elongations of the gill-lamellæ, cf. Goodrich (1909). The name, however, is still open to question as their function has not been definitely determined. As Southwell and Prashad (1919) remark, the name is less open to confusion than external gills or gill-filaments, which would denote homology with true external gills, or trophonematous filaments, a name which Alcock and Wood Mason (1891) applied to the uterine villiform papillæ of certain viviparous Elasmobranchs. In the Rays, they arise from five-gill arches, being absent from the spiracle. They are extremely delicate and are highly vascular. It seems feasible in oviparous Elasmobranchs to believe that they may assist in the absorption of albumen, but it is more difficult to accept the view that they absorb nutriment from the yolk sac, which pours its secretion into the spiral valve through the medium of an internal yolk sac. They are present also in viviparous Elasmobranchs, and are there supposed to be of use for the absorption of nutriment.

In the living eggs which the writer examined, they were observed to decrease in length with the growth of the broad flat pectorals, the consequent ventral position of the gill-clefts and the overgrowth of the anterior part of the gill-arch. They do not quite disappear even at the end of the embryonic period, though they are more or less invisible on the surface of the cleft, but are pushed outwards to the outer margin of the gill-arch with the development of the permanent gills. At the end of the embryonic period, one or more may be seen to extrude, generally from the last gill-cleft. Further investigation appears to be necessary to determine their true function.

TEMPERATURE OF LABORATORY TANKS.

The temperatures of the sea water in the experimental tanks in the Laboratory have been recorded daily since the beginning of 1920. Readings are taken generally about 10 a.m. and 5 p.m., and the thermometers are weighted so as to register the temperatures at the bottom of the tanks. The work was begun by Dr. J. H. Orton, and has been continued by Mr. A. J. Smith, who has been responsible for the daily readings. It will be seen from the times of observation and from the absence of self-recording instruments that the temperatures are only approximate, that they do not record the actual minimum, but are probably nearer the true maximum. The figures are tabulated in the above table and represent the monthly averages for five of the tanks during the periods of incubation of the egg-capsules. The incubation period for eggs of the same species was successfully obtained for *Raia clavata* in separate years. Two

TEMPERATURE OF LABORATORY TANKS.

Average Monthly Temperatures of five Laboratory Tanks in degrees Centigrade.

Months. Year.	I.	II.	III.	IV.	V.	VI.	VII.	VIII.	IX.	X.	XI.	XII.
1920.	—	—	—	—	—	14·8	14·9	14·8	14·8	14·4	12·2	10·3
1921.	11·5	10·9	11·1	12·1	13·4	15·6	18·0	16·6	16·6	15·4	12·7	11·8
1922.	10·6	9·9	10·1	10·0	13·6	—	—	—	—	—	—	—
Average Surface Temperatures at Stations L. 1 and L. 2.												
1921.	—	—	—	—	12·9	—	—	15·7	—	15·9	12·3	11·0
1922.	10·0	8·5	8·4	—	13·7	—	—	—	—	—	—	—
Average Surface Temperatures at Stations L. 3, L. 4, L. 5.												
1921.	—	—	—	—	12·0	—	—	15·4	—	15·9	14·2	12·2
1922.	10·5	9·2	8·9	—	12·7	—	—	—	—	—	—	—
Average Surface Temperatures at L. 6 and E. 1.												
1921.	—	—	—	10·1	13·4	—	14·7	15·6	15·8	15·6	14·4	12·8
1922.	11·2	9·9	9·4	—	12·7	—	—	—	—	—	—	—
Average Bottom Temperatures at Station E. 1.												
1921.	—	—	—	9·8	10·7	—	12·5	13·2	13·8	15·3	14·9	13·1
1922.	11·3	10·5	9·6	—	10·0	—	—	—	—	—	—	—

embryos were incubated from June to November, 1920, and twenty-two embryos in 1921 from April to September. The temperatures for 1920 were lower than those for 1921, though the average for the period showed only a difference of 1 degree Centigrade. The rate of development in the 1920 embryos was correspondingly slower, being one to two months longer than in 1921. Experiments, however, on a larger scale are required before making any definite deductions.

The average temperatures of the sea in the immediate neighbourhood of Plymouth at fixed Hydrographical Stations have been added for comparison. Stations L. 1 and L. 2 are taken together, L. 1 being below the Laboratory and L. 2 at the western end of Plymouth Breakwater. Stations L. 3, L. 4, and L. 5 occur at equal distances from L. 2 to the Eddystone rocks, and thus represent the area involved as the Inner Eddystone fishing-grounds. Stations L. 6 and E. 1 are outside the Eddystone rocks, and have been taken to represent the Outer Eddystone fishing-grounds. Only surface temperatures at Stations L. 1 to L. 5 were obtained, but there is probably not a great deal of difference from temperatures on the bottom, as the water is more or less homogeneous. A general comparison between the tank temperatures and those of the sea shows that the former are higher from March to September (inclusive) and lower from October to March, except in the case of L. 1 and L. 2, which are lower throughout the year than the Laboratory tanks. This is probably what one might expect to happen.

The correlation of these sets of temperatures distinctly shows that the tank temperatures differ very little from those of the Sound and of the Inner Eddystone grounds, which represent the conditions for the normal habitat of the eggs of the fish which are here recorded.

Salinity observations, unfortunately, have not been obtained. These might have brought out some interesting facts, for the water circulation in the Laboratory, owing to the periodicity in fresh supplies, develops an abnormally high degree of saltness.

Raia clavata Linnæus.

Common Names.—Thornback, Roker.

Egg-capsule (Fig. 1).

The egg-capsule of the Thornback, Fig. 1, is reproduced approximately ¾ actual size. The tips of the four horns end as delicate fibrillæ. One side is decidedly more convex than the other, which has a tendency to become almost flat. The more convex side appears ventrally in relation to the enclosed embryo in the figure. This is the side of the capsule which is oriented towards the dorsal side of the adult fish. It is covered with a tight-fitting felty mass of fibres, which become looser at each end of

the shell. The side margin of the shell is projected horizontally for most of its length as a flattened keel, from which springs a mass of loose fibres, which serve undoubtedly as attachment processes. This felty mass was present in all the specimens examined, but became much reduced with

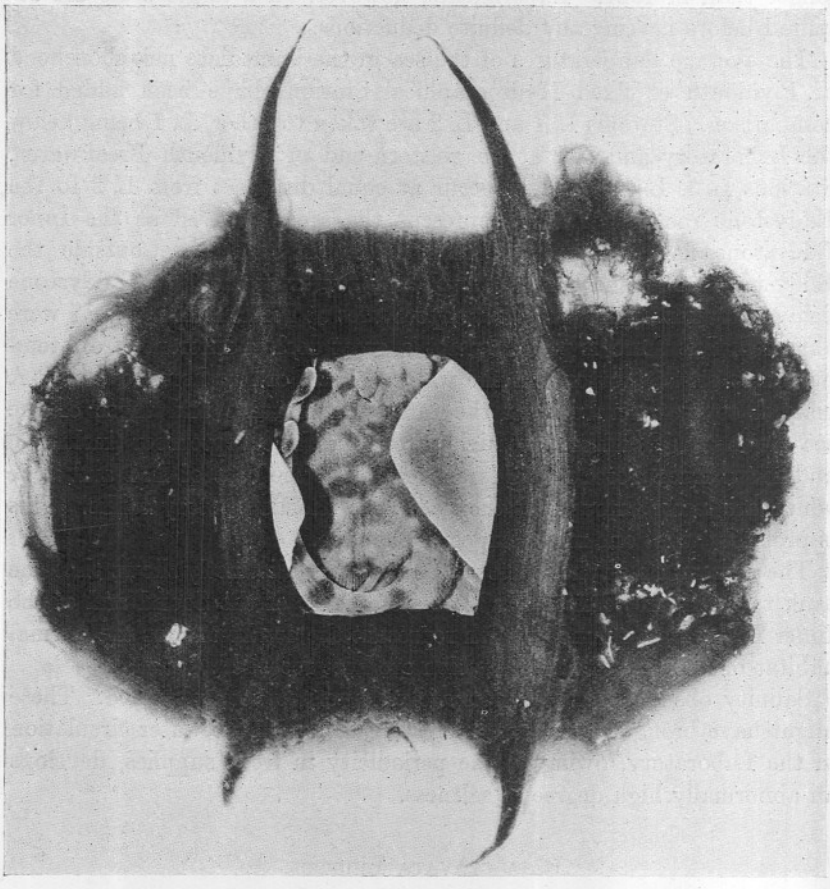

Photo. R.S.C.

FIG. 1.—EGG-CAPSULE OF RAIA CLAVATA L.

Length (without horns) 80 mm. Width (greatest), without attachment threads, 62·5 mm. Typical orientation of embryo just before hatching.

long exposure in sea water. Each horn has a slit near its tip on the outer side, not on the inner base of the horn, as has so often been assumed. These slits help in the aeration of the egg, but are closed with a thick plug of albumen during the early stages.

The number of capsules examined amounted to 300. The average

sizes were 74·9 mm. in length along the median line of the capsule, and 57·1 mm. in width (greatest). The length ranged from 63 to 90 mm., and the greatest width from 49 to 68·5 mm.

A few abnormal capsules were obtained, which were badly twisted, and had the central cavity so much reduced that the reception of a normal egg was impossible. Unfortunately, the adult fish could not be secured for examination. Their measurements were as follows :—

	Length (without horns).	Width (greatest).
1.	48 mm.	55
2.	45	59
3.	51	55
4.	46	41
5.	47	43

Fig. 1 shows the embryo in situ just before hatching. The orientation is normal, with the head of the embryo pointing diagonally towards the long horns, the end from which it would finally have emerged. The writer, however, has one example of an embryo of this species which hatched from the short horn end, which, as a rule, is tightly closed.

The dorsal side of the embryo faces the flat side of the capsule, but several examples show the reverse orientation. The embryo is capable of considerable movement within the capsule, as the writer has observed repeatedly, but the position assumed by the capsule may have a definite bearing on the final orientation of the embryo, cf. Beard and Nordgaard.

As the embryo increases in size, it becomes too big for the cavity of the capsule and it adopts the overlapping of the angles of the pectoral fins, while the tail curls round towards the head. The writer has not observed the embryo actually in the process of emerging from the capsule, but he has a strong suspicion that the curl of the tail is used as a fulcrum against the short horn end of the cavity in spasmodic efforts to force the head end out between the long horns.

Post-embryonic Stages.

Stage 1. (Fig. 2).

The period of incubation of the embryos ranged under artificial conditions in the Laboratory tanks from 4 to 5½ months. Temperature seemed to be an important factor, but the periodic high salinities of the Laboratory circulation might have had a deleterious effect. Twenty-three embryos were hatched, of which 15 were females and 8 males. The mean sizes of these at hatching, or soon after hatching, were 125·9 mm. in length and 79·5 mm. in width of disc, with a range round the mean from −7·9 mm. to +10·6 mm. in length, and −8·5 mm. to +6·5 mm. in width.

The means of 8 males were 126·9 mm. in length and 79·1 mm. in width, and of 15 females 125·4 mm. in length and 79·7 mm. in width.

Fig. 2 shows a male a few days old. The claspers are well defined.

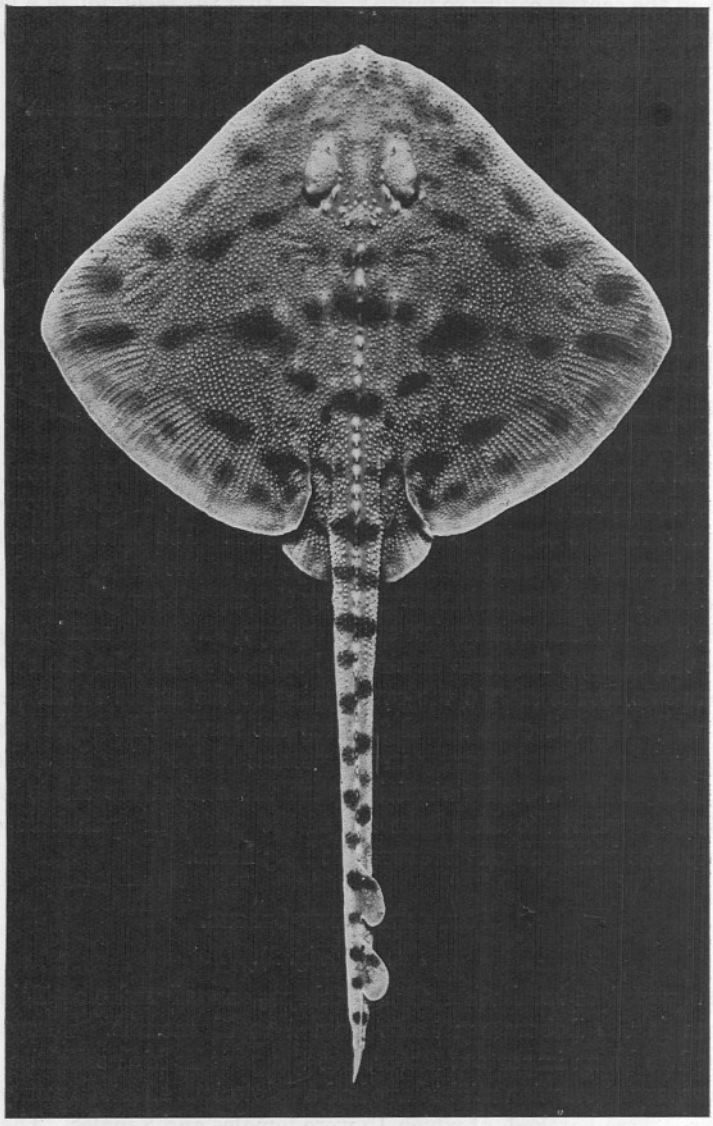

Photo. R.S.C.

FIG. 2.—RAIA CLAVATA L.

Newly hatched. Sex ♂. Period of incubation 155 days. Tota Length 133·5 mm. Width of Disc 82 mm.

The external yolk is fully absorbed, and only the flattened remains of the sac, ca. 1 mm. in diameter, can be seen. The internal yolk sac is still very large and occupies more than half the space of the body cavity. The spiral valve has been pushed to the left side of the fish and overlies the stomach. The date of hatching was 30th September, 1921, and the egg-capsule was taken from the adult fish and placed under circulation on 29th April, 1921. The period of incubation was thus 155 days.

STAGE 1 (Fig. 2).

Measurements are in mm.—on preserved specimen.

Total length.	133·5
Length of disc	62·5
Width of disc	82·0
Snout	14·0
Interorbit	6·0
Snout to tip of ventrals	69·0
Tail	70·0
Præoral	17·0
Internasal	10·5
Prænasal	12·0
Width of mouth	10·0
2nd dorsal to tip of tail	12·5

Teeth in upper jaw very irregular, both in shape and in position. There are less than forty rows, but these are difficult to define in vertical series. They are quite flat and embryonic, but a few show a short point posteriorly.

Upper surface entirely spinulose, except for a narrow bare longitudinal space on each side of the median ridge of spines. Thirty-five median spines on the body and tail, of which two only are in front of the shoulder and one between the dorsals. There are two præ-orbital and three post-orbital spines and two smaller inner orbital spines on each side. The two endolymphatic canals are open and appear as short tubes behind the head, in front of the first large median spine. There is one spine on each shoulder. The side margins of the tail are spinulose. Colour of upper surface approximates to Klinksieck and Valette, code No. 134, with small irregularly shaped dark patches scattered over the disc and tail, and a few cream spots more or less circular in shape near the central area of the body. The lower surface is entirely smooth and white except for a margin of grey round the angle of the pectorals and along the posterior border of the disc and pelvics. The tip of the tail is darker brown.

There is a considerable amount of variation in spinulation. In sixty-one specimens the variation in the total number of median spines ranged from twenty-six to thirty-eight, with a mean of thirty-one.

Pigmentation is very unstable. Some are more or less uniform, others have dark and light spots, others have a black bar on each wing, cf. Figs. 3 and 4.

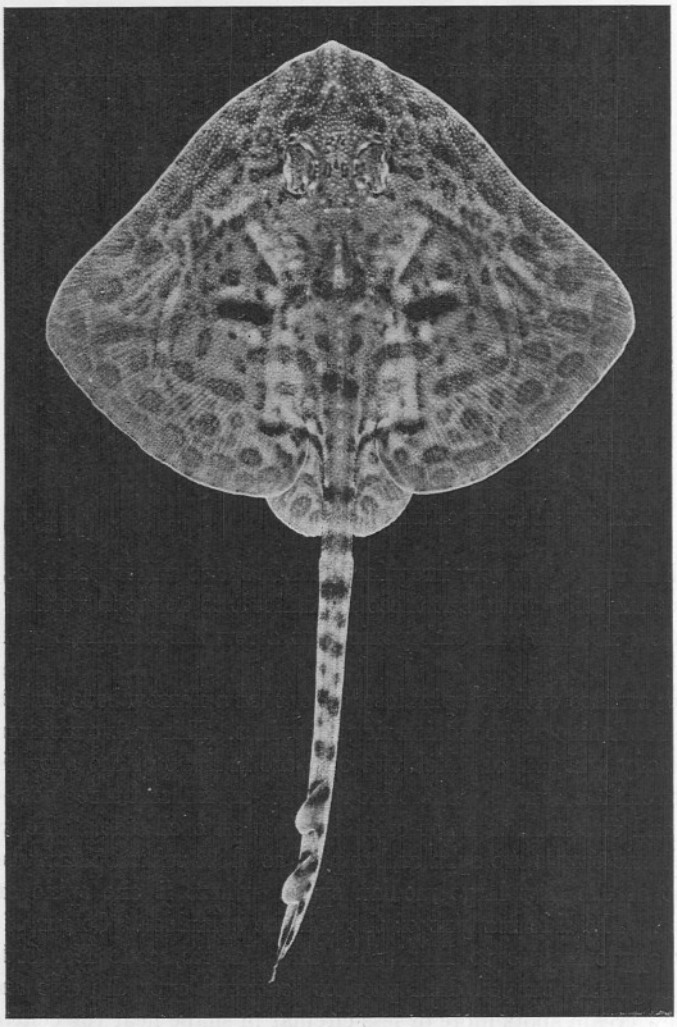

Photo. R.C.S.

FIG. 3.—RAIA CLAVATA L.

Age, after hatching, 4 months. Sex ♂. Reared in Laboratory Tank. Total Length 121 mm. Width of Disc 75 mm.

STAGE 2 (Fig. 3).

Fig. 3 represents a male Thornback, age four months. It was hatched in October, 1921, and lived till 12th February, 1922.

Measurements in mm.

Total length	121
Length of disc	57·5
Width of disc	75
Snout	13·5
Interorbit	5·5
Snout to tip of ventrals	62·5
Snout to vent	48
Tail	63
2nd dorsal to tip of tail	11
Præoral	15·5
Internasal	10
Prænasal	12
Width of mouth	10
Teeth in upper jaw	ca. 40 rows.

Upper surface entirely spinulose. A narrow bare patch along each side of the median row of spines. Total number of median spines thirty-two, of which two are in front of scapula and two between the dorsal fins. Two præ-orbital and three post-orbital spines, with two smaller inner orbitals. Endolymphatic tubes prominent and open. A narrow projecting margin of skin along the greater part of the tail. Colour of upper surface mottled, as the figure shows, with two pairs of dark spots transversely elongate, one larger pair near the middle of the body, the other pair smaller and narrower near the base of the disc. A narrow white border surrounds the disc, snout and ventrals. The lower surface is smooth and white, with a darker margin round the angle and posterior border of the disc and ventrals. The teeth are still irregular in shape and position, but the middle rows show a tendency towards a sharp posterior point.

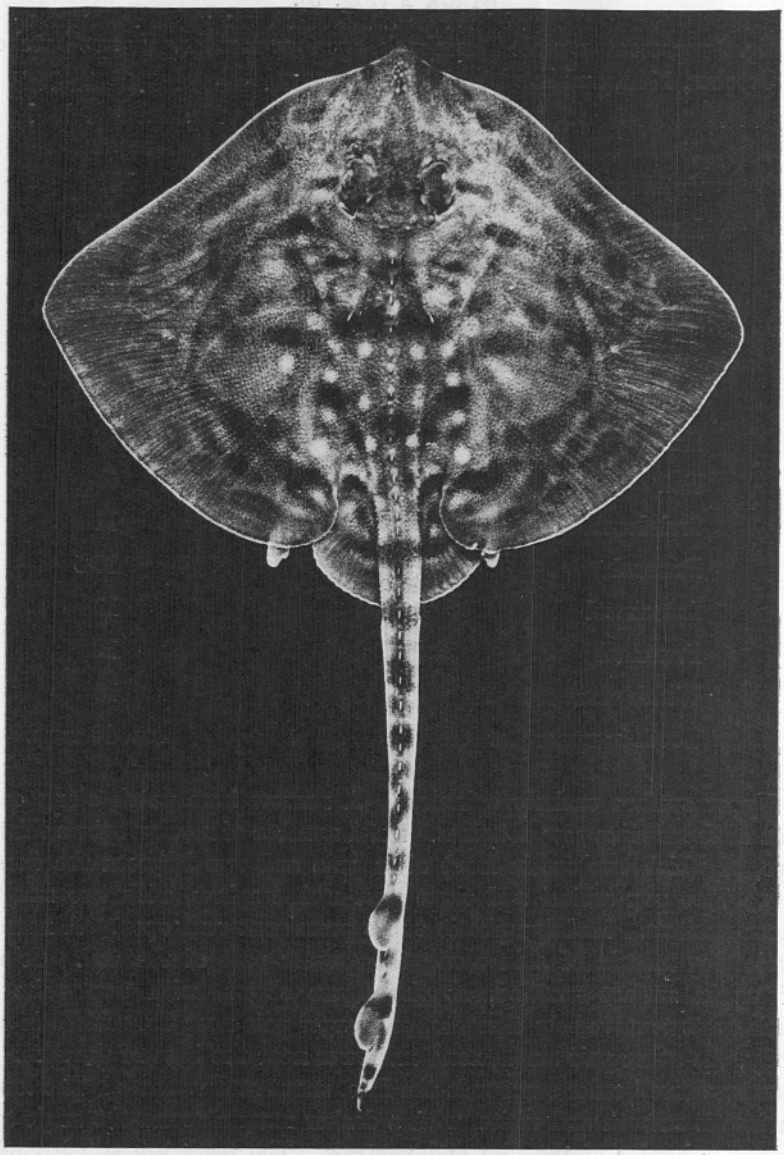

Photo. R.S.C.

FIG. 4.—RAIA CLAVATA L.

Age, after hatching, ca. 4 months. Sex ♀. Reared in Laboratory Tank. Total Length 146 mm. Width of Disc 95 mm.

STAGE 2 (Fig. 4).

Fig. 4 represents a female Thornback, also about four months old. It was hatched in October, 1921, and killed on 6th February, 1922.

Measurements in mm.

Total length	146
Length of disc	69
Width of disc	95
Snout	16·5
Interorbit	6
Snout to tip of ventrals	77
Snout to vent	59
Tail	75
2nd dorsal to tip of tail	10·5
Præoral	19
Internasal	12
Prænasal	13
Width of mouth	12
Teeth in rows in upper jaw	ca. 44

This fish was observed to feed freely on Amphipods which were placed in the tank. It shows the typical shape and characters of the young Thornback. The disc is broad, tail long, dorsals widely separated, upper surface entirely spinulose. There are thirty-six median spines on the body and tail, of which three are præ-scapular.

The anterior margin of the disc is slightly undulated. The pigmentation scheme is less pronounced than in Fig. 3, and the dark bars are absent. The circular cream white spots are more pronounced near the middle of the body. The tip of the caudal is much reduced. Otherwise, the characters are the same as in the preceding.

RAIA MACULATA Montagu.

Common Names.—Homelyn, Spotted Ray.

EGG-CAPSULE (Fig. 5).

The egg-capsule, Fig. 5, is narrower in proportion to its length than that of the Thornback, which is shown in Fig. 1. The general characters are identical, but the texture of the shell is much more delicate and the mass of attachment threads much smaller. The capsule more nearly

resembles that of *Raia undulata*, Fig. 18, but is of much smaller size. Both sides of the shell are convex, the side which is dorsal in relation to the adult fish being covered with a close-fitting network of fibres, and the ventral side being perfectly smooth. Each horn has a definite longitudinal slit on its outer side and near its distal end, in the region of the bend of the tube. There is no lateral horizontal prolongation of the

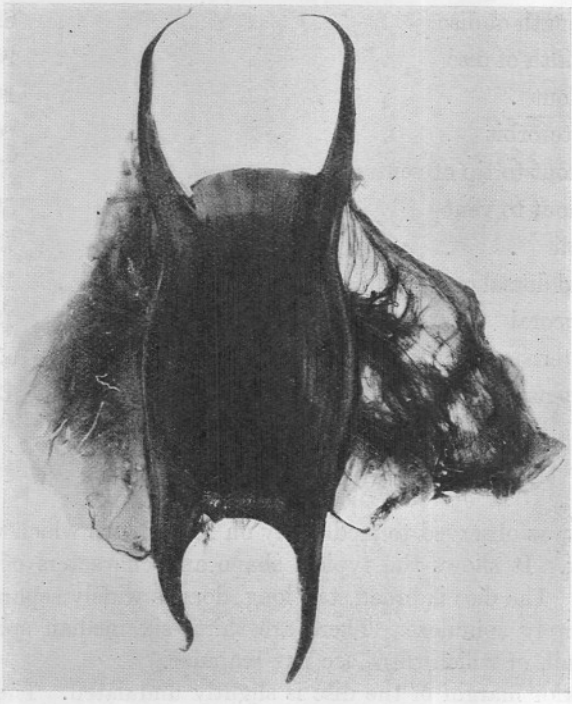

Photo. R.S.C.

FIG. 5.—EGG-CAPSULE OF RAIA MACULATA Montagu.
Length (without horns) 69 mm. Width (without attachment threads) 42 mm.

capsule into a flattened keel as in the Thornback. The figure reproduced by Holt and Calderwood belongs to this species, but appears rather broad in relation to its length. Beard could find no difference between the eggs of the Thornback and the Homelyn, but this is not convincing. The average sizes for twenty-four capsules were 70·7 mm. in length without horns, and 41·8 mm. in greatest width. Mediterranean examples, according to Lo Bianco, are much smaller, the length being given as 65 mm. and the width 35 mm.

Post-embryonic Stages.

Stage 1 (Fig. 6).

The following are the measurements in mm. of the young Homelyn, reproduced in Fig. 6, which has just emerged from its capsule :—

Total length	116
Length of disc	54·5
Width of disc	67·5
Snout	11·75
Interorbit	6
Snout to tip of ventrals	61
Snout to vent	47·5
Length of vent	3
Præoral	14·5
Internasal	8·5
Prænasal	10
Width of mouth	9
Tail	60
2nd dorsal to tip of tail	11
Teeth in upper jaw	ca. 40

The period of incubation in five examples ranged from 145 to 172 days, from May to October, 1921. Of the five examples, two were males and three females. The average total length of these fish was 127·2 mm. and width of disc 75 mm. The upper surface is smooth, except for the characteristic median row of spines and a narrow border of spinulæ on the anterior margin of the disc. There are a few small spines on the rostrum and on the interorbit, two præ-orbital and two post-orbital spines and one smaller inner orbital. Twenty-eight median spines extended from the neck region to the dorsal fins, and of these two only were in front of the shoulder and one between the dorsals. A single spine is present on each side of the shoulder. The tail is also provided on each side with a single row of marginal spines, which are less pronounced than the median series. The colour is light fawn with black spots, which do not extend on to the edge of the disc. A narrow line, white in colour, surrounds the outer margins of the disc and ventrals, while the tail has a horizontal prolongation of the skin as a flattened keel, which extends longitudinally or most of its length.

The mucous canals are well shown in the figure. The endolymphatics occur as short open tubes behind the head. The characteristic feature of this newly hatched fish is the long caudal tip behind the second dorsal

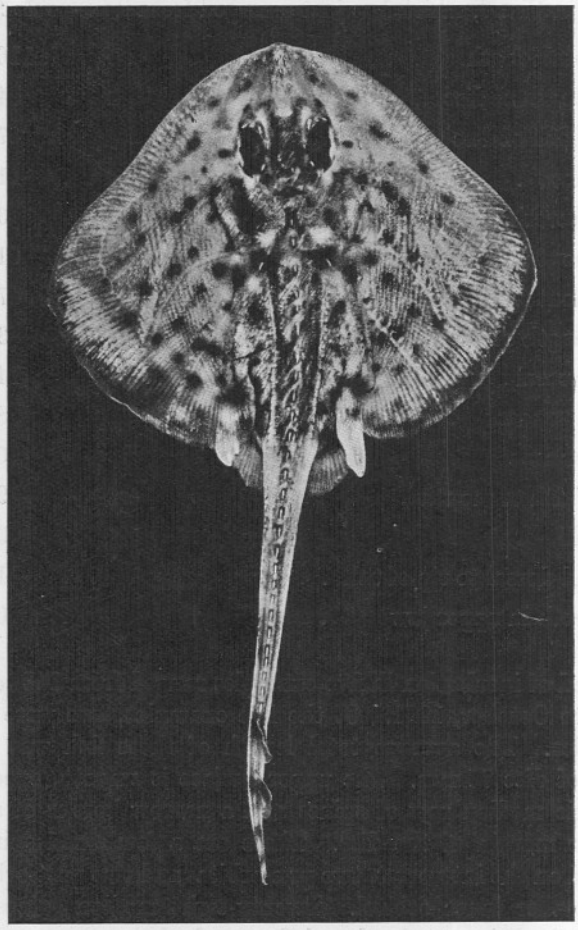

Photo. R.S.C.

FIG. 6.—RAIA MACULATA Montagu.
Newly hatched. Sex ♂. Period of incubation 172 days. Total Length 116 mm. Width of Disc 67·5 mm.

fin. The extreme tip has begun to show signs of shrinkage. The lower surface is entirely smooth and white, except for the usual border of pigment round the angle and the posterior margins of the disc and ventrals. The internal yolk sac is still la ge.

STAGE 2 (Fig. 7).

Measurements in mm. of fish reproduced in Fig. 7.

Total length	126
Length of disc	65
Width of disc	86·5
Snout	16
Interorbit	6
Snout to tip of ventrals	72
Snout to vent	51·5
Tail	63·5
2nd dorsal to tip of tail	6·5
Præoral	18·5
Internasal	10
Prænasal	13·5
Width of mouth	9·75
Teeth in upper jaw	ca. 40

This specimen was hatched on 17th October, 1921, and lived till 16th March, 1922. Its length on hatching was 132 mm. and width of disc 79 mm., and the distance from the end of the 2nd dorsal to the tip of caudal fin was 6·5 mm. The reduction in length of the caudal is a pronounced feature of all these post-embryonic Rays. It begins to shorten before the end of embryonic life, and the process of absorption is carried on for some time after hatching. It shows a wrinkled appearance, while the extreme tip curls and shrivels up, cf. Figs. 6 and 7. The fish, Fig. 7, has undergone considerable changes. The snout has been pushed out and now projects slightly beyond the margin of the disc. The pigmentation is intensified, and an ocellus on each pectoral is beginning to show more prominently, as a circular cream-coloured spot, surrounded by a few irregularly shaped black spots. The general shape of the fish approximates more to the adult condition, but the characters are still more or less embryonic.

The upper surface shows the same spinulation as in the younger stage. There is a median row of thirty spines, of which two are præ-scapular. No spine is as yet developed between the dorsals, which are separate. The inner orbital spine is absent, but spinulæ are more frequent on the interorbit. The upper surface is covered with irregularly shaped black spots, which are large in comparison with the size of the fish. The cream-

coloured spots are circular and very prominent near the middle of the body. The tail has a definite lateral keel.

The lower surface is entirely smooth and white except for a greyish border round the angle and posterior margin of the disc and ventrals.

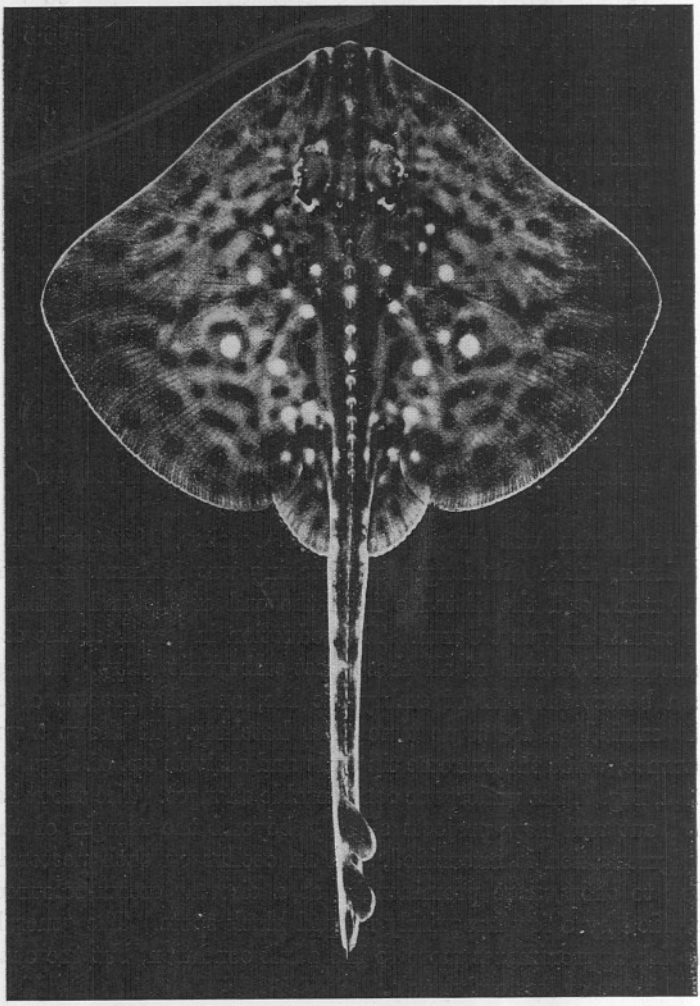

Photo. R.S.C.

FIG. 7.—RAIA MACULATA Montagu.
Age, after hatching, 5 months. Sex ♀. Reared in Laboratory Tank.
Total Length 126 mm. Width of Disc 86·5 mm.

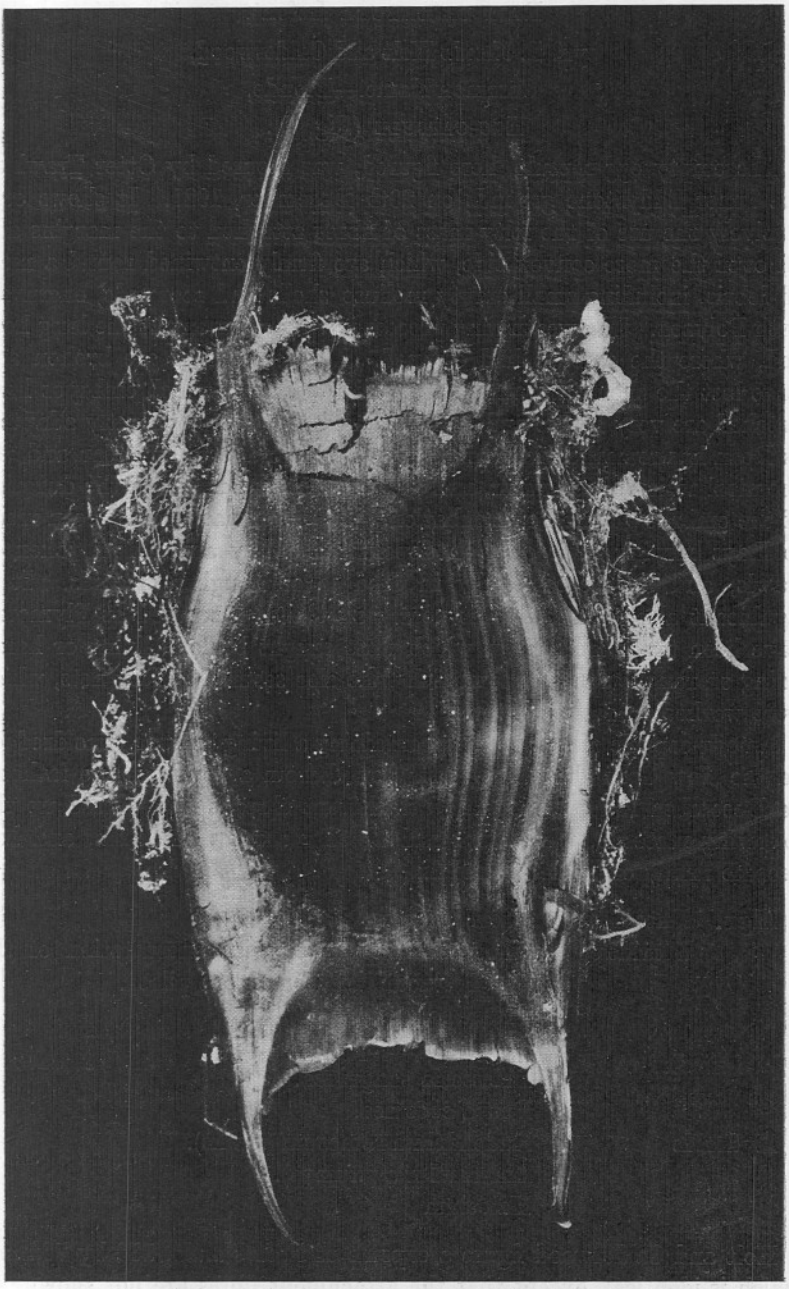

Photo. R.S.C.

FIG. 8.—EGG-CAPSULE OF RAIA BRACHYURA Lafont.
Length (without horns) 121 mm. Width (without attachments) 79 mm.

RAIA BRACHYURA Lafont.

syn. *R. blanda* Holt and Calderwood.

Common Name.—Blonde.

EGG-CAPSULE (Fig. 8).

The empty egg-capsule, here figured, was secured by Otter Trawl on the Inner Eddystone grounds on 20th December, 1921. It shows conclusively the use of the felty mass of fibres attached to the margins and all over the more convex side, which are firmly entwined round a mass of debris, including small empty mollusc shells and seaweeds, encrusted with Polyzoa. The weathering process is very apparent at the long horn end, where the embryo had escaped. The long horns are broken and have lost the short filamentous tip. Capsules from which embryos have hatched are very brittle and easily broken. They are evidently adapted, as Dean has remarked for Chimæra, to withstand wear and tear and the chemical action of sea water, and their life corresponds with the duration of the embryo's period of incubation. Reference has already been made to Rays burying their eggs on deposition. They are seldom taken n trawls, and only occasionally in dredges on rougher grounds. It is significant also to record that nearly all the eggs secured at Plymouth have been taken from Rays which were caught by long lining or hand-lining on rough ground. Some fish were also secured in Ray nets, which were worked on rocky bottom.

An egg-capsule of this species, containing a living embryo, was dredged on the New Grounds in Plymouth Sound from a depth of five fathoms on December 12th, 1911. The attachment threads had picked up the following material, which was undoubtedly a mass of debris, and had become firmly entangled round it.

Algæ.	Fucoideæ	Fucus serratus.
	Florideæ	Rhodymenia palmata with encrustation of the Polyzoan, Membranipora pilosa.
		Gigartina stellata.
		Nitophyllum sp.
		Delesseria sanguinea.
		Dasya coccinea.
Hydroids		Eudendrium ramosum (stalks).
		Antennularia (stumps).
		Halecium ,,

Holt and Calderwood figure a capsule of *Raia blanda*, 136 by 76 mm., which is identical, except that the attachment fibres are not shown.

The average of 177 capsules gave a length of 128·4 mm. without horns, and a greatest width of 78·5 mm.

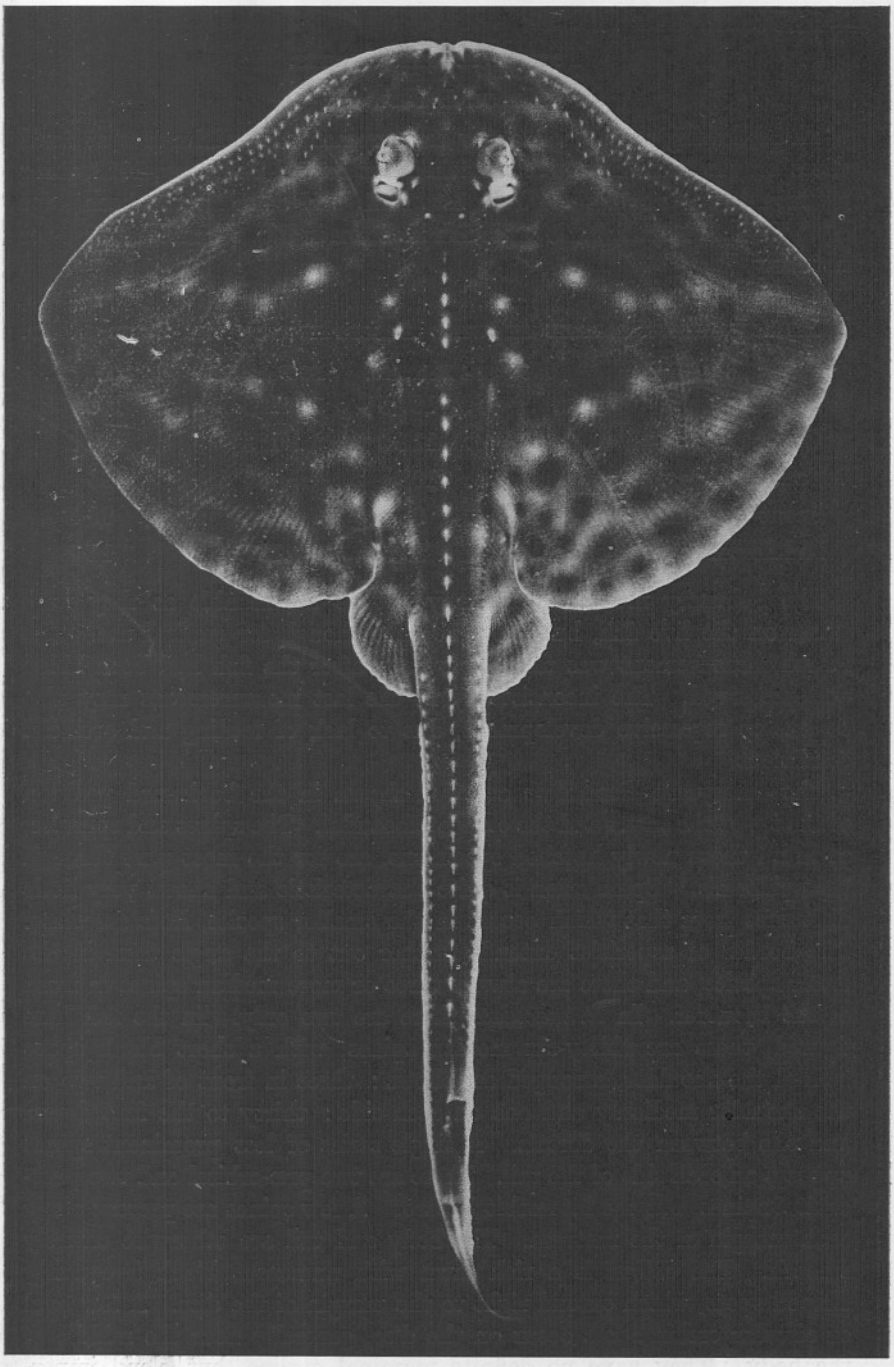

Photo. R.S.C.

FIG. 9.—RAIA BRACHYURA Lafont

Newly hatched. Sex ♂. Period of incubation 218 days. Total Length 187·5 mm. Width of disc 115·5 mm.

Post-embryonic Stages.

Stage 1 (Fig. 9).

The following table gives a few measurements in millimetres at fixed dates on the same fish which hatched out on 3rd February, 1922, after an incubation period since July 1st, 1921, and after having been kept alive till 20th March, 1922. The fish is shown in Fig. 9 immediately after hatching.

Table of Measurements showing Absorption of the Caudal Tip.

	4 Feb., '22.	26 Feb., '22.	22 Mar., '22.
Total length	187·5	188·5	186
Width of disc . . .	115·5	123	122·5
Tail	103	94	93
Snout to tip of ventrals . .	95	102	90·5
2nd dorsal to tip of caudal .	22	13·5	14

The February measurements may not be too reliable as they were taken on the live fish, but they are near enough for general use. The March measurement was taken two days after the fish had been killed and preserved. The inconsistency in the last measurements may also be due to regressive development, due to lack of feeding and artificial conditions.

The embryonic characters are well shown in the backward position of the tip of the snout and the elongated caudal. The spines and spinules, as in all those newly hatched specimens, are for the most part through the epidermis. Spinulation is quite characteristic and shows very little difference from that which occurs in the smaller species, *Raia maculata*. The border of asperities extends further out to the angle than in the latter species, and there are always three or more median spines in front of the shoulder. In *R. maculata* there are two only. In the fish here figured, there are thirty-six median spines, with a short blank space behind the shoulder, arranged as follows : three in front of and two on the shoulder medianly and one on each side : thirty to the first dorsal and one between the dorsals. A single series of less prominent spines appears on each side of the tail. There are two præ-orbital and two post-orbital spines, but the interorbit is bare. A few spines are present along each margin of the rostrum. The body and tail are otherwise entirely smooth. The colour is a rich fawn with numerous black irregularly shaped spots, which extend right to the margin of the disc. A few oval or circular cream spots occur on the disc, generally near the middle of the body. They have no definite marginal outline, but blend imper-

ceptibly with the ground colour. There is a narrow white border line round the disc, and a soft white keeled expansion on each side of the tail, extending longitudinally from near the tip of the ventrals to a point just behind the second dorsal fin. The tip of the tail in these early stages is generally reabsorbed to this point.

The arrangement of the mucous canals is well shown in the figure, where the ends appear as white dots. A pair of endolymphatics occurs as short open tubes in front of the first median spine. They are shown as white dots. The lower surface is entirely smooth and is white except for the usual marginal band round the angle and posterior disc and pelvics.

Stage 2 (Fig. 10).

A few measurements are recorded for this fish, which was kept alive from 21st December, 1921, the date of hatching, to 21st February, 1922. The period of incubation was ca. 7 months. The external yolk sac after the embryo hatched out had a diameter of 6 mm., but very little yolk was left. The internal yolk sac could be seen through the skin and occupied most of the body cavity. In the majority of cases, the external yolk sac is reduced to the size of a pin's head at the time of hatching, but occasionally it was found a few millimetres in diameter. In the above fish the spasmodic movement of the long caudal end was observed at intervals, after the fish had left the egg-capsule.

Measurements in mm. of the same fish at regular intervals.

Date of measurements	24.12.21.	22.1.22.	5.2.22.	21.2.22
Total length	175	181	184	187
Width of disc	100	116	122	122
Length of disc	—	88	—	—
Tail	—	92	—	95
2nd dorsal to tip of tail	22·5	14	13	12·5
Snout to tip of ventrals	—	98	101	102

Growth appeared to be about normal. The reduction in the distance from the base of the second dorsal to tip of caudal is very pronounced. This is shown clearly in the figure. The table proves, if another were needed, that the tip of the caudal is absorbed. It confirms Jensen's opinion that the embryonic tail end behind the horizontal skin fold at each side of the tail is the part that is reduced. This is one of the most interesting features in the present investigation, and has occurred definitely in all the species which have so far been examined. Another post-embryonic change occurs in the snout region. The newly hatched fish, vide Fig. 9, shows the tip of the snout a little behind the anterior margin

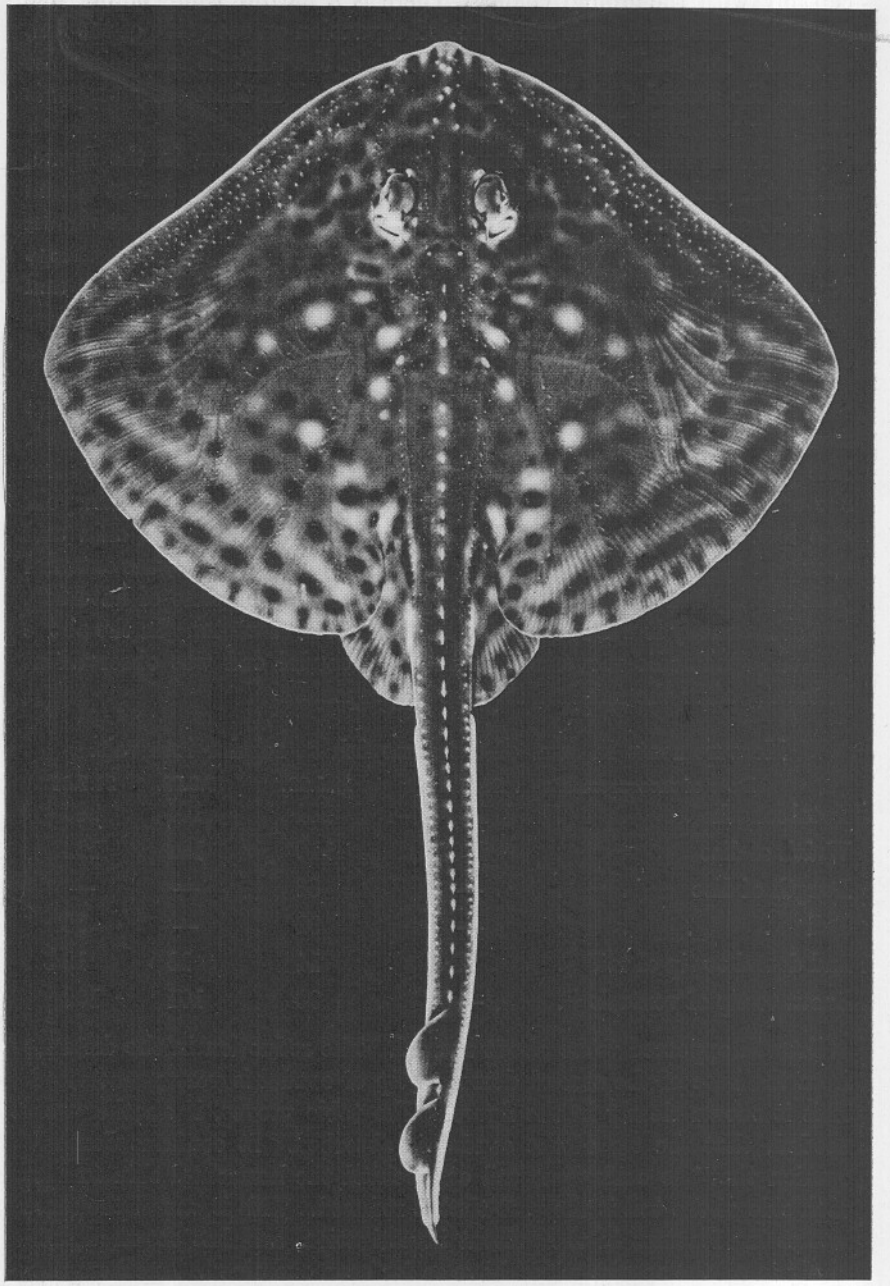

Photo. R.S.C.

FIG. 10.—RAIA BRACHYURA Lafont.
Age, after hatching, 2 months. Reared in Laboratory Tank. Sex ♀. Total Length 187 mm. Width of Disc 122 mm.

of the disc in a small gap, but, with growth, the snout soon projects beyond the disc and assumes the adult shape. The teeth are the last to assume the adult condition, the median series being the first to show change. Internally, the embryonic structures show a rapid change. At first the internal yolk sac is very large, but is quickly absorbed with the functioning of the stomach. Young specimens have been examined with the internal yolk sac still persisting, though much reduced, and with the stomach full of small Crustacea—Amphipods and Crangonids. With the reduction of the internal yolk sac, which communicates directly with the top of the spiral valve, the latter gradually assumes the normal position on the right-hand side of the fish. The life of those Rays which were hatched and reared in the Laboratory tanks averaged from two to seven months, the older fish surviving with additional food. The difficulty has been not so much to get the required food, but to get the fish to feed under artificial conditions.

Measurements in mm. of the fish reproduced in Fig. 10.

Total length	187
Length of disc	93
Width of disc	122
Snout	21·5
Interorbit	8
Snout to tip of ventrals	102
Snout to vent	74·5
Length of vent	4·5
Præoral	25
Internasal	12
Prænasal	17·5
Width of mouth	14·5
Teeth in upper jaw	ca. 60
2nd dorsal to tip of tail	12·5
Tail	95

Teeth irregularly placed, flat, and embryonic.

Fig. 10 requires little by way of description. The characters are similar to those of the newly hatched fish, with the exceptions already noted. The general scheme of spinulation is the same, variation occurring in the number of median spines which are here 33. In the newly hatched stage of the same fish the spine between the dorsal fins was absent. In 22 fish, early stages with width of disc ranging from 117 to 179 mm.,

the total number of median spines ranged from 30 to 36. There were never less than 3 in front of the shoulder, while between the dorsals the range was 0 to 3. In slightly larger fish the spines immediately behind the shoulder are the first to disappear.

The ground colour of the upper surface approximates to Klinksieck

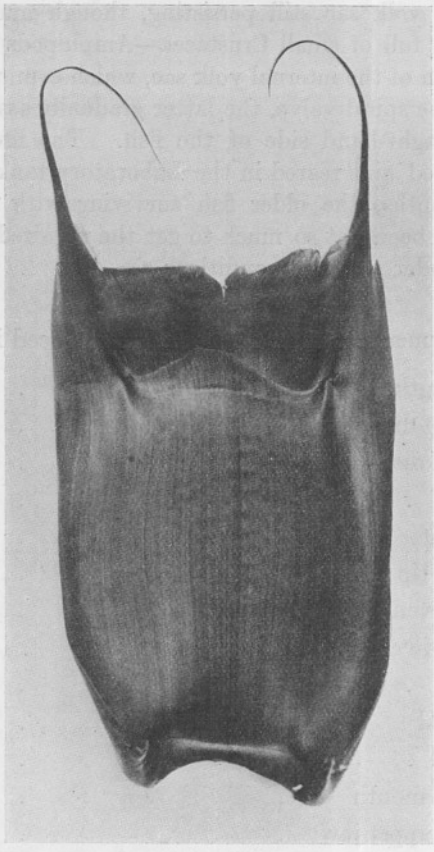

Photo. R.S.C.

FIG. 11.—EGG-CAPSULE OF RAIA MICROCELLATA Montagu. Length (without horns) 93 mm. Width (greatest) 56 mm.

and Valette, Code Nos. 134 and 135. The disc is covered with black spots, while there are a few lighter spots near the middle of the body. The upper surface is quite smooth, except for the spinules on rostrum and anterior margin of disc, the median spines on the body and tail, the marginal row on the tail, two præ-orbital and two post-orbital spines, and one small inner orbital. The horizontal skin fold on the margin of the tail is very pronounced, and is white in colour.

Raia microcellata Montagu.

Common Name.—Small-eyed Ray.

Egg-capsule (Fig. 11).

The capsules of this species are smooth and without attachment filaments. One side is considerably more convex than the other, the more convex side being the dorsal in relation to the adult fish. Both sets of horns tend to curve away from the dorsal side of the capsule, and give the egg the appearance of curving longitudinally. The long horns are elongated into thin tubes, while the others are very short and strongly hooked. The widest part of the capsule is across the base of the long horns. The hooks in the short horns may probably serve as anchors. The lateral margin of the shell is produced horizontally into a keeled flange. The average sizes of fifteen capsules were 90·8 mm. in length along the median line and 57·2 mm. in greatest width. Slits are present on the outer edge of the long horns, near the base of the exposed part where the lateral keel shows as a spur. They open alternately, one towards the dorsal side, the other towards the ventral aspect of the capsule. The slits in the short horns are more median in position and open longitudinally for almost their whole length, one on each face of the shell. The ventral opening on the long horn coincides with the ventral opening on the short horn.

Post-embryonic Stages.

Stage 2 (Fig. 12).

The stage figured (Fig. 12) corresponds to Stage 2 of the general scheme adopted in the present report. The patchy effect of the figure is due to the post-mortem sloughing of mucus, which carried the pigment with it when an attempt was made to clear the fish. Unfortunately, it was the only available example of this species. It was first observed on 31st January, 1922, in a tank, where there were also several newly hatched young of *Raia clavata*, but it had obviously been hatched for some time. As proof of this, one may instance the shortening of the caudal tip, the advanced development of the spinulation and the disappearance of the internal yolk sac. The period from the date of placing the egg-capsule under circulation to the date of observation amounted to eight months, so that one may assume the period of incubation for this embryo to be about seven months.

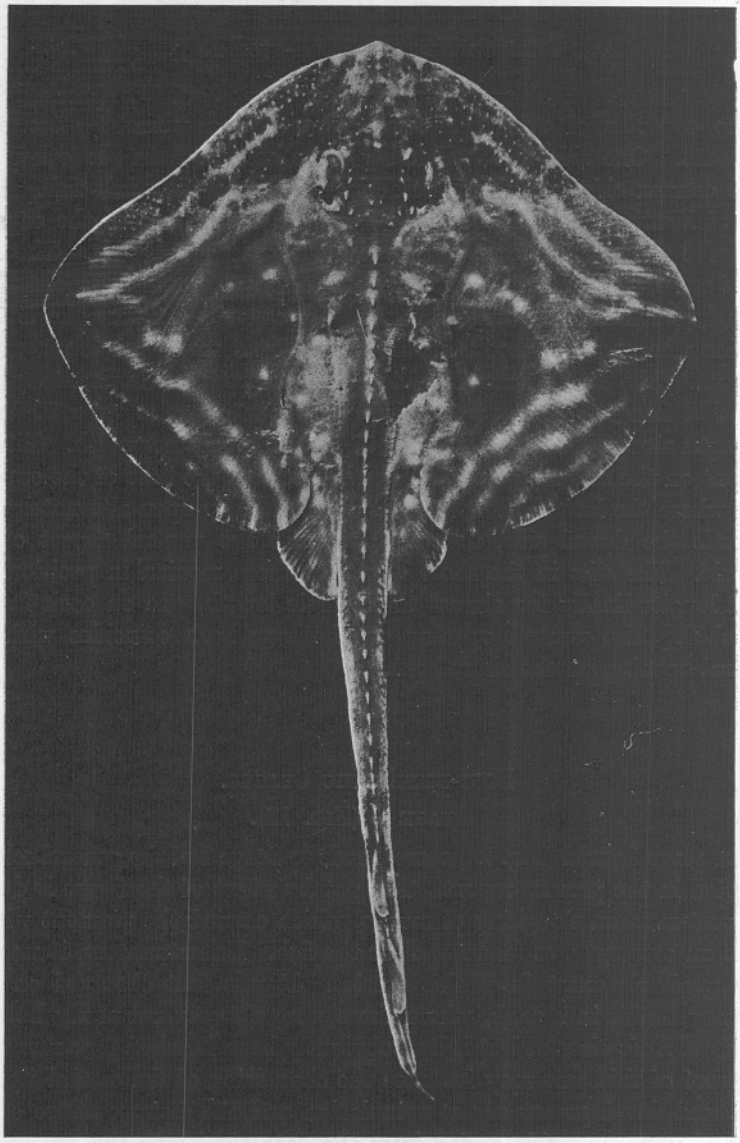

Photo. R.S.C.

FIG. 12.—RAIA MICROCELLATA Montagu.
A few weeks after hatching. Reared in Laboratory Tank. Sex ♀. Total Length 144 mm. Width of Disc 86 mm.

Fig. 12. Stage 2. Measurements in mm.

Total length	144
Length of disc	66
Width of disc	86
Snout	16
Interorbit	7
Snout to tip of ventrals	75
Snout to vent	57
Tail	75
2nd dorsal to tip of tail	13·5
Præoral	18·5
Internasal	10·5
Prænasal	14
Width of mouth	12
Teeth in rows in upper jaw	ca. 42

The spinulation of the upper surface is much the same as occurs in *Raia brachyura*, except that there is a definite and prominent triangular patch of densely packed spinulæ across the snout and anterior region of the pectorals, including the interorbit. The base of the triangle may be taken as a line drawn across the eyes and extending outwards to the margins of the disc at right angles to the main longitudinal axis of the fish. There are thirty-three median spines on the body and tail, of which three are in front of the shoulder and two between the dorsals. There are two præ-, three post-, and two inner orbital spines, and one on each side of the shoulder. The tail is furnished with a lateral row of smaller spines, which is irregularly double on the proximal half of its length.

The colour of the upper surface approximates to Klinksieck and Valette, Code Nos. 130, 135, with a few white spots and long narrow white bands which follow the outlines of the disc, both anteriorly and posteriorly. The tail has a narrow, white, longitudinal keel.

The lower surface is entirely smooth and white except for a margin of grey round the angle and the posterior border of the disc and ventrals.

RAIA NÆVUS Müller und Henle.

Common Name.—Cuckoo Ray.

EGG-CAPSULE (FIG. 13).

The egg-capsule of this species has been figured by Holt and Calderwood as *Raia circularis* (Günther). It is definitely the egg of the Cuckoo Ray. The capsule is relatively very small, and so far as can be gathered from

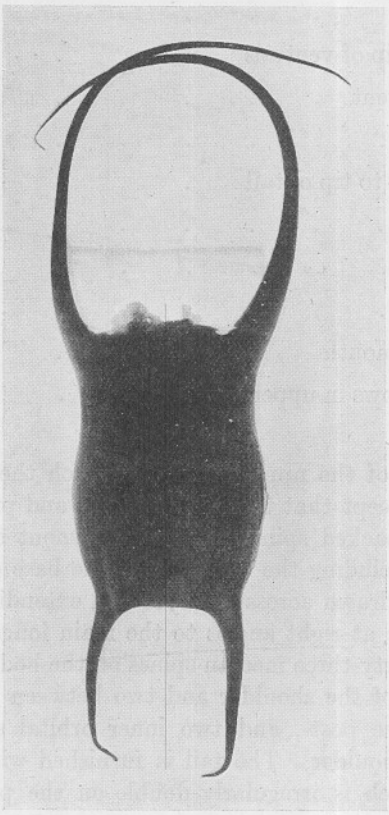

Photo. R.S.C.

FIG. 13.—EGG-CAPSULE OF RAIA NÆVUS M. und H.
Length (without horns) 60·5 mm. Width (greatest) 36 mm.

the number examined, is devoid of any loose threads. The shell is more or less transparent and a convenient one in which to watch the development of the embryo, though the whole structure is delicate and requires careful handling. Both sides of the capsule are convex, the short horns projecting outwards from one side, the ventral in relation to the adult

fish, and the long horns tending towards the opposite direction. The short horns have their tips curled almost in hook fashion, while the long horns nearly always cross one another. The slits occur on the outer margins near the tip of each horn, generally where the tube bends. The slits on the short horns are on the ventral side of the capsule. The end between the short horns is tightly closed and almost concave, but the long horn end is convex and easily opened. This is the end from which the embryo escapes on hatching.

The average sizes of twenty-eight egg-capsules were 63·4 mm. in length and 36·8 in breadth.

Post-embryonic Stages.

Stage 1 (Fig. 14).

The period of incubation for a single record was 243 days. The following observations were made on the developing embryo. The egg was taken from the cloaca of an adult fish on 5th June, 1920, and placed in a tank under circulation in the Laboratory. It was suspended from a glass rod at the crossing of the long horns. The outer surface of the capsule retained its fresh glossy appearance throughout development, showing only a darkening in colour from a yellowish brown to a dark reddish brown. The yolk lay at the short horn end of the inner cavity, while the embryo, as it developed, became suspended above the yolk sac near the long horn end. The external branchial filaments developed from five gill arches, being absent from the spiracular cleft. They increased in length enormously and seemed to hang in close contact with the yolk sac. As development proceeded, the embryo lay suspended by its yolk stalk, head downwards diagonally across the cavity towards the yolk sac, while the tail end moved freely upwards towards the base of one of the long horns. The movement was lateral and rhythmic along the whole length of the embryo, but more apparent in the tail region, whose length was considerable. As the embryo increased in length, the elongated caudal region entered the base of the long horn and continued its rhythmic movement. At this period the slits were found to be open and not obstructed by albumen, as on lifting the capsule out of the water the internal fluid was seen to drain off from the slits at the tips of the horns. There seems no doubt that the phenomenon observed was an adaptation to secure aeration of the egg. As this happened to be the only successful attempt to rear the embryo of this species, the capsule was not disturbed, as it was thought equally important to hatch out the fish to secure the definite characters of this species for systematic purposes. A capsule, however, of *Raia clavata*, with a well-developed embryo, was placed in a dish of water and treated experimentally with powdered carmine grains.

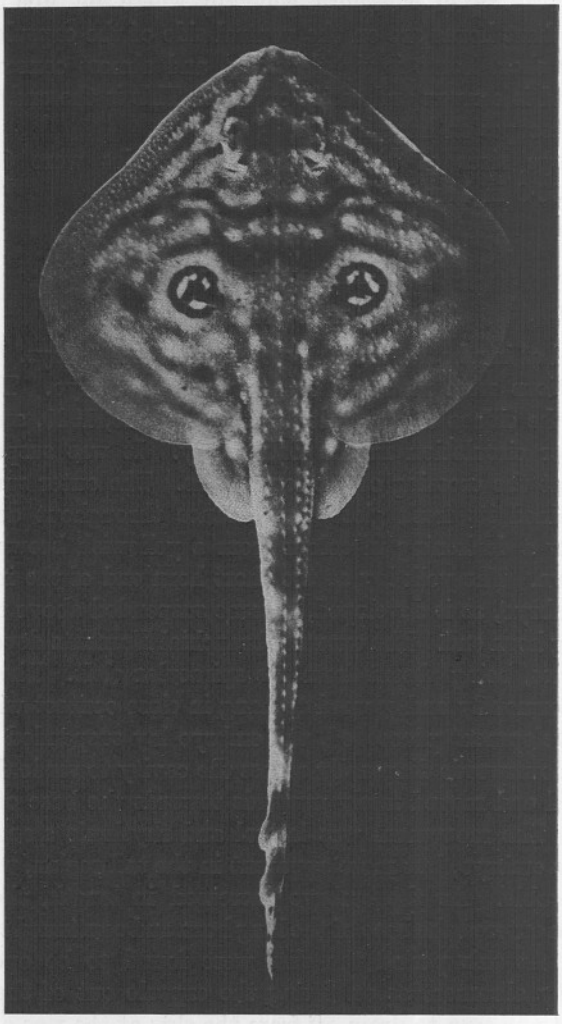

Photo. R.S.C.

FIG. 14.—RAIA NÆVUS M. und H.
Newly hatched. Sex ♂. Period of incubation 243 days.
Total Length 119 mm. Width of Disc 62 mm.

A definite current was found to move away from one of the long horns, but no other movement was visible. After a few minutes, the capsule was opened and carmine grains were found on the spiracle, on the inner lining of the shell cavity, and were also forcibly ejected from the gill clefts. Observations were interrupted in September, October, and part of November, but were continued at the end of the last month. The embryo had now assumed the definite form of a Ray, with broad flat pectorals. As these increased in size and became too large for the narrow cavity, the outer angles of the pectorals curled over on the dorsal side of the embryo, while the tail was stowed away round the left side of the embryo and with its tip towards the long horn end. The embryo thus became definitely oriented with the snout pointing diagonally towards the long horn end. The external yolk sac was greatly reduced. Pigment was gradually acquired on the dorsal side, and the ocelli could be seen as rings of black dots near the middle of the disc. The egg case showed signs of weathering between the long horns and to a small extent along the sides. Pigmentation was well developed before the embryo hatched out on 3rd February, 1921. Immediately after hatching, the external yolk sac was reduced to the size of a pin head, but the abdominal region was greatly distended by the internal yolk. The points of the larger median spines had penetrated the skin, but the tips of the spinulæ were just beginning to show through. This explains to some extent the poor definition of the photograph.

Fig. 14 shows the characteristic scheme of pigmentation. The general ground colour is light fawn with bands of sepia. There are also a few lighter coloured patches and some smaller oval to circular spots. A large ocellus, in the form almost of a simple spiral, occurs near the middle of each pectoral. The outer ring and the core are dark brown to black, while the intervening space is cream coloured.

The upper surface is entirely spinulose, with the median series of spines on the body and tail prominent. There are thirty-three median spines, of which three are in front of the scapula. A lateral row of less prominent spines extends from the shoulder to the dorsal fins, and is supplemented on the tail proximally for part of its length by another series. There is a triangular patch of spines in front of the shoulder with the apex of the triangle pointing anteriorly. The dorsal fins are almost confluent.

Stage 2 (Fig. 15).

The newly hatched embryo, Fig. 14, was kept alive for three months, and the following measurements were taken, at intervals, the last on 8th May, 1921, after the fish had been a short time in preservative :—

Measurements in millimetres.

	4 Feb., '21.	7 Mar., '21.	10 April, '21.	8 May, '21.
Total length	119	125	125	122·5
Length of disc	50	58	—	58·5
Width of disc	62	65	67	66
Snout	8	—	—	12
Interorbit	5	—	—	5
Snout to tip of ventrals	59	66	—	66·5
Snout to vent	—	—	—	50
Tail	64·5	63	—	62
2nd dorsal to tip of tail	13	9	7	4·5
Præoral	—	—	—	19
Internasal	—	—	—	9
Prænasal	—	—	—	12
Width of mouth	—	—	—	9
Teeth in rows in upper jaw	—	—	—	ca. 44

Growth was maintained probably entirely at the expense of the internal yolk sac for about two months, as the fish was not observed to feed. The shrinkage of the caudal area is well seen in the above measurements.

The upper surface is entirely spinulose, except for a narrow margin on the posterior edge of the disc and pelvics. There are thirty-four median spines on the body and tail, but none between the dorsals, which are practically confluent. A single small spine is present on each side opposite the meeting of the dorsals. A triangular patch of large spines occurs on the shoulder, which includes three medium spines in front of and one on the shoulder, and two on each side. The first spine behind the shoulder is minute. The median series of spines on the body and tail is very prominent, but later, as the fish reaches maturity, these spines entirely disappear and the lateral series becomes more pronounced. A row of spines extends along each side of the median series on the body and tail. On the proximal half of the tail, there is an additional outer row. A ring of spines is present on the inner orbit. All the large spines are hooked.

The colour of the upper surface approximates to Klinksieck and Valette, Code No. 574, with narrow transverse bands of a dark brown colour on the snout, disc, and tail. There are also a few whitish spots on the disc, but these have no definite outline. The colour of the fish

was lost in preservative, and is not fully represented in the figure. A prominent marbled ocellus, dark brown to black and creamy white, occurs on each wing.

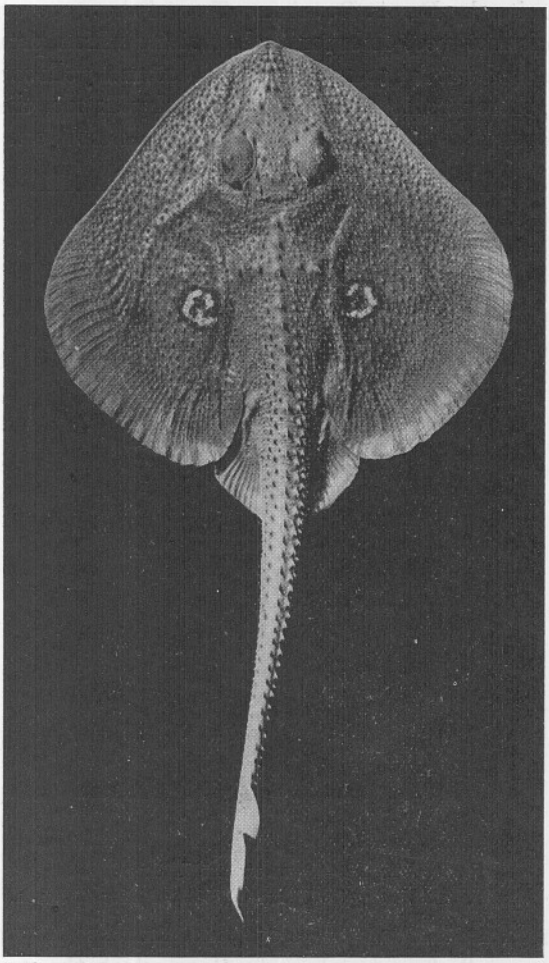

Photo. R.S.C.

FIG. 15.—RAIA NÆVUS M. und H.
Age, after hatching, ca. 3 months. Reared in Laboratory Tank.
Total Length 122·5 mm. Width of Disc 66 mm.

The lower surface is entirely smooth and white. A narrow greyish margin is present on the outer angle and along the posterior border of the disc and ventrals. There are a few grey patches on the tail.

Raia fyllæ Lütken.

Post-embryonic Stages.

Stage A (Fig. 16).

Three small specimens of this rare deep-water species were kindly lent to the writer by Dr. A. Bowman. Two of these are very small, but they have distinct post-embryonic characters, which prove that they had been hatched out for some time. The external yolk sac had entirely disappeared, there was no trace of an internal yolk sac, the spiral valve was in its normal position on the right side of the fish, and the stomach

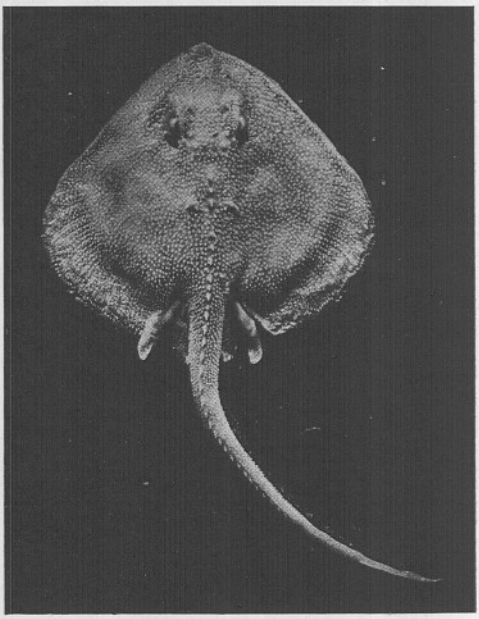

Photo. R.S.C.

Fig. 16.—Raia fyllæ Lütken.
Sex ♀. Total Length 80 mm. Width of Disc 40·5 mm.

contained small crustacean food—Copepods, Amphipods, and Schizopods. They are the smallest post-embryonic Rays which the writer has seen, smaller even than the diminutive Starry Ray, *Raia radiata*. The egg case, or its cavity, at least, must be of very small size.

There is a suggestive resemblance in the shape of these young fish to the young of *Raia nævus*. From Lütken's descriptions of *Raia fyllæ* adult No. 1 and young No. 2, the writer has provisionally ascribed the present series to the same species.*

* Pigmentation and spinulation strongly suggest a specific difference between Stages A and B, but more material is necessary for verification.

The two smallest fish are very thin and delicate, especially round the margin of the disc. On the anterior margin of the disc near the snout there is a tendency, in these preserved specimens, for the skin to curl ventrally, and thus to give the figure a slightly sharper appearance than it really has. There is distinct sinuation, as Lütken has remarked, though no definite notch. Lütken, it is presumed, had in his mind the deep sharp notch of the adult, of which he gives, in the same report, an excellent figure of a mature male. Maturity in this species is reached at a very small size. The measurements of the two smaller fish are given here in millimetres. One was a female, the other a male. Both were captured with a small trawl on 23rd August, 1910, at a depth of 1448 metres in Latitude 58° 43′ N. and Longitude 9° 6′ W.

	Sex ♀	Sex ♂
Total length	80	72·5
Length of disc	34·5	32·5
Width of disc	40·5	39·5
Snout	8	7
Interorbit	4·5	4
Eye	4	4
Eye+spiracle	5	5
Snout to tip of ventrals	38·5	34
Snout to vent	30·5	27·5
Tail	44·5	40·5
2nd dorsal to tip of tail	ca. 3·5	ca. 4
Præoral	9·75	8·5
Internasal	5·75	5·5
Prænasal	7	6·5
Width of mouth	6	6
No. of median spines	35	37

Fig. 16 represents the larger of the two specimens, which is a female. The upper surface is entirely spinulose, the spinulæ having a very sharp delicate point. Larger spines stand out prominently on the orbit, scapula, and median line of the body and tail to the first dorsal. The dorsal fins are practically confluent and are small and delicate structures. There are two præ- and three post-orbital spines, one to three spines on each side of the shoulder, two in front and one on the shoulder. The anterior lobe of the pelvics is long.

Colour of upper surface is uniform (in formalin), and approximates to Klinksieck and Valette, Code No. 128 C. Lower surface a dirty white.

The lower surface is smooth except the margins of the tail, which are spinulose for most of the length.

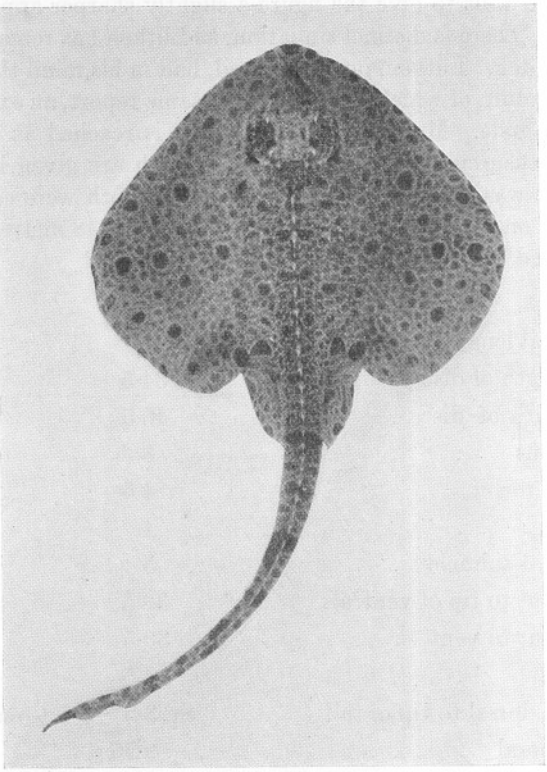

Photo. R.S.C.

FIG. 17.—RAIA FYLLÆ Lütken.
Sex ♀. Total Length 104·5 mm. Width of Disc 53 mm.

STAGE B (FIG. 17).

The third specimen, a female, is represented in Fig. 17. It was captured by small trawl at Scottish Station 13 A, on 9th July, 1913, at a depth of 630 metres. It is obviously much older. The following are the measurements in millimetres :—

Total length	104·5
Length of disc	44·5
Width of disc	53
Snout	9·5
Interorbit	4·5

Eye	5
Eye+spiracle	6
Snout to tip of ventrals	52
Tail	57
2nd dorsal to tip of tail	4
Præoral	12
Internasal	7
Prænasal	9
Width of mouth	7·25
Teeth in rows in upper jaw	ca. 26
No. of median spines	34

The general ground colour, in formalin, corresponds to Klinksieck and Valette, Code Nos. 133 and 138, while the round or oval spots are darker brown and correspond to Code No. 143. The larger oval spots have a distinct halo of lighter colour round them. The whole of the upper surface except a narrow margin on the posterior edges of the disc and ventrals is covered with spinulæ, which have a very sharp point. There are two præ- and three post-orbital spines and one inner orbital. Of the thirty-four median body and tail spines, two are in front of and one on the shoulder. A group of three spines is present on each side of the shoulder. There are no spines between the dorsals, which are closely set. The rows of spinulæ on the sides of the tail are beginning to enlarge, the outer row being the largest. All the spines are hooked, and the median series most prominent.

The lower surface is of uniform dirty white colour and is smooth, except for a few spinulæ on the margins of the tail near its base.

Raia undulata Lacépède.

Syn. *R. picta* Lacépède.

Common Name.—Painted Ray (non Couch).

Egg-capsule (Fig. 18).

The capsule of this species is very similar to that of *Raia maculata*, but is much larger and more robust. The filamentous attachments are well shown in Fig. 18. They arise from a horizontal flattened keel at the margin of the shell. The capsule is convex on both sides and there is little difference in the degree of convexity. However, one side is definitely smoother than the other, which is covered with a close felty mass of loose fibres. At both ends of the median line of the capsule these fibres are set more loosely and more thickly. Each of the horns

bears a longitudinal slit. It is present nearer the tip of each horn, occurring on the outer side near the bend of the long horns and near the middle of the short horns on the same face as the smooth surface of the capsule. Only two capsules have come under observation at Plymouth.

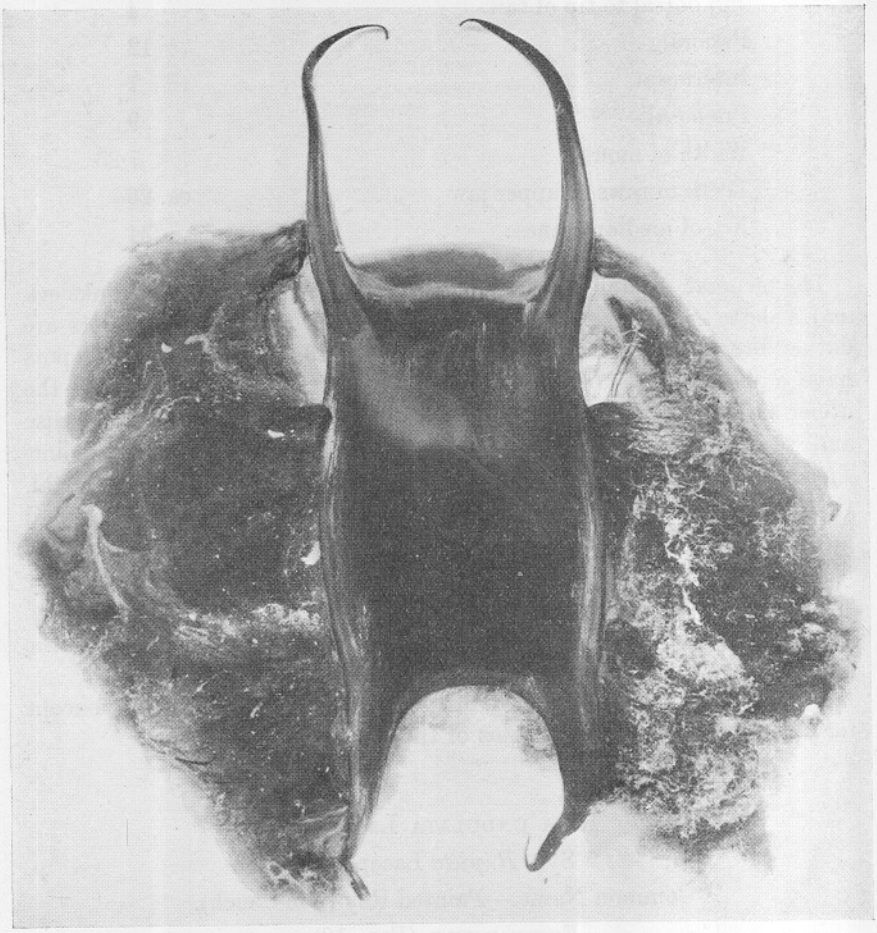

Photo. R.S.C.

FIG. 18.—EGG-CAPSULE OF RAIA UNDULATA Lacép.
Length (without horns) 81 mm. Width (without attachment filaments) 52 mm.

The species is not uncommon in the outer fishing-grounds of the Channel, and these capsules were secured on 15th July, 1920. The average sizes of these two capsules were 81·5 mm. in length (without horns), and 52 mm. in breadth (greatest). The size of this capsule agrees closely with that for Mediterranean specimens, for which Lo Bianco gives 90 mm. by 45 mm.

Raia batis Linnæus.

Common Names.—Skate, Blue Skate, Grey Skate.

Egg-capsule (Fig. 19).

Fig. 19 represents a capsule which was sent, with five others, to the writer by Mr. B. Storrow, of the Cullercoats Marine Laboratory. These were secured from the adult fish on North Shields fish quay on 18th February, 1922, and were kept in the Laboratory tanks for more than a week, when they were despatched to Plymouth and arrived on 4th March, 1922. Five of these eggs did not develop, but the sixth, represented in the above figure, was opened on 25th May, 1922, and showed a well-developed embryo with a length of 39·5 mm. The branchial filaments were well developed, being longer than the head, and showed the red blood circulation streaming round the loop of the filaments. Albumen still closed up the capsule, but a watery liquid surrounded the yolk and embryo in the central cavity. No trace of slits could be found on the horns, but the hollow tube could be traced out to the tip, with a delicate thin membrane on the upper and lower face of the horn, through which the albumen penetrated with a little pressure. It seems suggestive that this membrane slits at a later period in the age of the capsule. Beard described the slits as occurring at the extremity of the horns on the inner side. Along each side of the capsule the outermost layer of the shell projects horizontally, showing a central longitudinal furrow. Near the base of the long horns a patch of very long attachment filaments arises. These become twisted in rope fashion for most of their length, and end in an expanded mass of very fine filaments. The weight of this wet knotted end when lifted out of water is considerable, and it has the property of fastening on to any solid object.

The average sizes of these six capsules were length 159 mm. along the median line, without the horns, and greatest width 80·5 mm. The averages of two Plymouth specimens were 143·5 mm. in length and 81 mm. in width. The length of the attachment filaments in these last two specimens was about 200 mm. The tips of all the horns end as very thin delicate filaments, and are easily broken off. The capsule is bi-convex and thicker towards the short horn end. The whole of the shell is covered with a felty mass of tightly packed fibres. Strengthening is given to the margins by a solid inner rod, triangular in cross section, running longitudinally along each margin of the shell. The walls of the capsule are formed of several layers, most of which are membranous, except the outermost, where the fibres are more loose. All the layers have definite longitudinal striation.

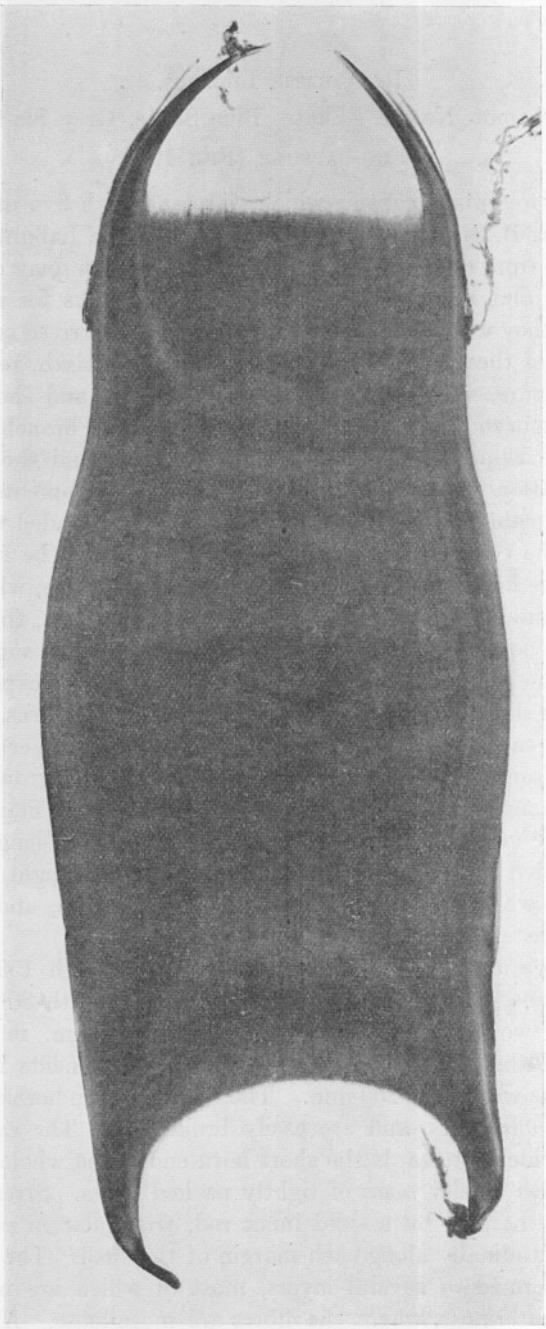

Photo. R.S.C.

FIG. 19.—EGG-CAPSULE OF RAIA BATIS L.
Length (without horns) 163 mm. Width (greatest) 82 mm.

Young Stages.

No early post-embryonic stages have been hatched at Plymouth, but the following description is based upon sixteen young stages, ranging in width of disc from 219 to 394 mm. These were caught by trawl in the immediate neighbourhood of Plymouth.

Disc broader than long. The ratio, width to length of disc averaged 1·31, with a range from 1·26 to 1·39. Anterior margins of disc emarginate, with very slight undulation. Outer angles rounded. Snout blunt and obtusely rounded, its length 4·06 in width of disc (range 3·85 to 4·42). Interorbital width 3·8 in length of snout (range 3·42 to 4·29). Internasal width 2·28 in præoral length. Teeth close set, flat, broader than long, with short obtusely pointed posterior cusp. Forty-five to fifty-four rows in upper jaw.

Upper surface smooth. One præorbital spine, sometimes 2, 0–2 postorbital spines. No other spines on the body. Tail, beginning at the junction of the pelvics, with thirteen to twenty median spines, of which there may be 0–3 between the dorsals. Only one specimen had marginal tail spines, and these were limited to three on right and three on left side near the dorsals. Colour of upper surface varies considerably in different fish. The general ground colour approximates to a mixture of Code Nos. 65, 120, 154, and 155 (Klinksieck and Valette), with or without oval or circular spots, Code Nos. 153 and 162. A large oval ocellus may or may not be present, but it is very faint in colour and with no prominent margin. It appears generally in fresh specimens as a yellow halo round a light brown centre, but it is by no means a constant feature. The pigment both of upper and lower surfaces has a tendency to rub off very easily. This is even a more pronounced feature in the young of *Raia vomer* Fries, where the dark grey pigment sloughs off with mucus as a greyish black ink.

Black ends of mucous pores show a definite arrangement on the upper surface. They occur as a single line—duplicated at points—along the anterior margin of the snout and disc and for some distance round the angle : scattered dots along the outer edges of the rostrum : a densely packed aggregation just outside the præ-orbit and extending along the junction line of the disc and snout to the anterior edge : a small semicircular area on each side of the shoulder and a few scattered dots along the sides of the median line on to the base of the tail.

Lower surface entirely smooth in the youngest fish ; but later, with a narrow band of spinulæ round the anterior snout and margin of the disc, extending about half-way out to the angle. Colour, K. and V., Code Nos. 69, 120, 474, and 475, with scattered black dots—the ends of the

mucous pores. A broad band round the angle and posterior margins of the disc and ventrals free from black dots. Short black streaks show through the skin. These are the pigmented sections of the ends of the mucous canals.

Raia marginata Lacépède.

Syn. *R. alba* Lacépède.

Common Names.—White Skate, Bottlenose Skate, Bordered Ray (young).

Egg-capsule (Fig. 20).

Holt has given a description of the capsule of the Bottlenose Ray, but without any illustration. Emphasis was laid on the beaded structure of the capsule as a ready method of identification. This beaded character, however, is a misnomer, except in the newly spawned condition. The surface of the capsule becomes later a honeycombed mass, with the longitudinal ridges and transverse crests as the walls of the pits. This honeycombing disappears on the lateral strengthened keels, which are striated longitudinally. The capsule has one side distinctly more convex, the side which faces the dorsal aspect of the adult fish before extrusion, and with the curling of the horns away from this side, gives the other side a flattened effect at least near the base of the long horns. The slits are present in the middle of the short horns on the more convex side and in the middle of the long horns on the opposite side. They extend along the horns for most of their length. There are no accessory attachment fibres on the capsule, and anchorage, if one may suggest such a thing, may be effected by the strongly hooked short horns. The long horns are ribbon-shaped.

Le Danois gives an outline figure of the same capsule under the name of *Raia batis*. The average sizes for six capsules were length 180·3 mm. along the median line (exclusive of horns), and greatest width 138·6 mm. Four of these eggs were secured on Plymouth fish quay in April and two in June, 1921. It is the largest Skate capsule which has come under observation.

Out of six eggs, one has been reared successfully almost to the hatching stage. It was placed in a Laboratory tank under circulation on 14th April, 1921, the date on which it was taken from the cloaca of the fish on Plymouth fish quay. A small window was cut open on the capsule on 27th February, 1922, to watch the further development of the embryo. At that time the embryo was well developed and had the pectorals and snout united. The head end pointed towards the short horns, and the expanse of the pectorals did not occupy the full width of the cavity. It was then capable of turning round inside the capsule cavity, for at a subsequent date the snout became oriented towards the long horn end.

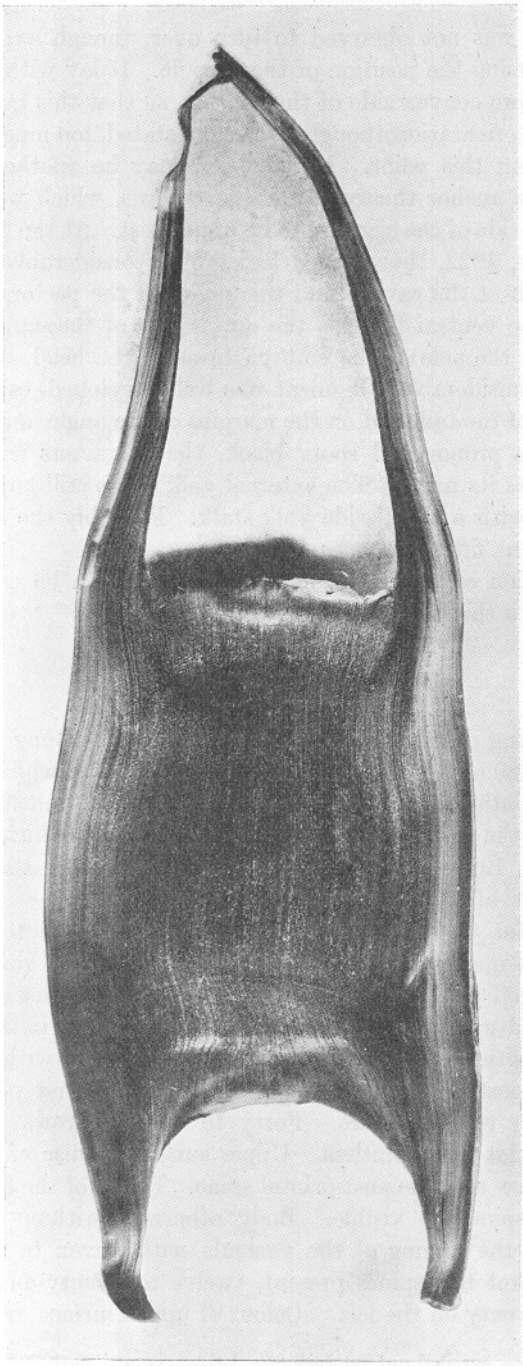

Photo. R.S.C.

FIG. 20.—EGG-CAPSULE OF RAIA MARGINATA Lacép.
Length (with horns) ca. 390 mm.; (without horns) 176 mm.
Width (greatest) 131 mm.

The embryo was not observed to turn over, though experiments were tried in reversing the position of the capsule. It lay with its dorsal side facing the more convex side of the capsule, so that this may be assumed as its normal orientation, though, as already stated, too much stress should not be laid on this point. It, however, may be another argument in favour of the anchor theory of the short horns, which would cause the more convex side of the egg case to lie uppermost. At the time of writing, on 25th May, 1922, the embryo had grown considerably. It occupied the full width of the cavity, and the angles of the pectorals were curled over from the ventral towards the dorsal side of the embryo. The tail curled round the side of the embryo towards the head, and the caudal length was considerable. Pigment was well developed, especially on the ventral side of the tail, and on the margins of the angle and posterior disc which were a pronounced sooty black, the characters from which this species derives its name. The external yolk sac is still large, ca. an inch in diameter with a short wide yolk stalk. Probably the full incubation period is about fifteen months.

A description of this fish will be given if it can be carried through successfully to the hatching stage.*

Young Stages.

The following description is based on a series of young stages ranging from 238 to 389 mm. in width of disc, the youngest of which are probably not many months old. These were taken in March and April, 1921 and 1922, except the smallest, 238 mm. in width of disc, which was captured in November, 1921. All were taken by Otter Trawl in the Outer Eddystone grounds off Plymouth.

Disc broader than long. Its length contained 1·3 to 1·4 times in breadth. Its margin undulated, outer angles slightly rounded. Snout acutely pointed and projecting considerably beyond the disc, its length 4 to 4·3 in width of disc. Interorbital width 3·2 to 3·5 in length of snout. Internasal width 2·1 to 2·2 in præoral length. Teeth with base rounded and with a posterior conical cusp, which is long and pointed. Rows closely set in vertical series. Forty to forty-six rows in upper jaw. Outer series flat and pointless. Upper surface of disc entirely smooth, except one præ- and one post-orbital spine. Traces of the base of another post-orbital spine are visible. Body otherwise without spines. Tail, beginning at the joining of the ventrals, with eleven to fifteen median spines. Lateral tail spines present, twelve to twenty-one on the right and ten to twenty on the left. Colour of upper surface approximates to

* This embryo hatched out on July 6th, 1922. Total length 292 mm., and width of disc 190 mm.

Klinksieck and Valette, Code Nos. 129, 130, 154, with or without oval or round spots of a light colour, Code No. 153 D. No ocellus.

Lower surface with the præoral area strongly spinulose. A narrower margin of spinules along the anterior edge of the disc half-way out to the angle. Body and tail smooth. Colour of lower surface white, with a broad margin of sooty black round the angle, along the posterior edge of the disc, narrower on the ventrals and covering most of the tail.

FOOD OF THE EARLY YOUNG OF FOUR SPECIES OF RAIA.

Number of fish examined	100		50		27		32	
Range in width of disc in mm.	79–119		78–360		117–336		63–254	
Species.	Clavata.		Maculata.		Brachyura.		Nævus.	
Stomach contents.	No. in which present.	%	No. in which present.	%	No. in which present.	%	No. in which present.	%
Polychæta	7	7	11	22	—	—	8	25
Crustacea—								
Amphipod	69	*69*	42	*84*	16	*59*	16	*50*
Isopod	—	—	3	6	1	3	—	—
Cumacea	5	5	3	6	—	—	—	—
Schizopod	8	8	3	6	10	37	7	21
Crangonid	47	*47*	32	*64*	17	*62*	12	*37*
Nika edulis	3	3	3	6	1	3	1	3
Galathea sp.	4	4	3	6	—	—	—	—
Upogebia sp.	4	4	1	2	—	—	—	—
Hippolyte sp.	1	1	1	2	1	3	—	—
Pandalus sp.	1	1	3	6	—	—	1	3
Portunus sp.	—	—	4	8	—	—	—	—
Mollusca	1	1	7	14	—	—	—	—
Cephalopoda—								
Sepiola sp.	2	2	—	—	—	—	—	—
Pisces	2	2	1	2	8	29	9	28
Empty	10	10	—	—	2	7	5	15

The above table shows the predominant types of food of the early young of four species of Rays. It will be seen that Amphipods and Crangonids form a very high percentage in all the species. Examination was also made of a few available stomachs of the following six species: *R. radiata*, *microcellata*, *fyllæ*, *fullonica*, *batis*, and *marginata*, and the results were more or less similar, except that in the Skate species, *R. fullonica*, *batis*, and *marginata*, small fish and the larger crustacean types, Galathea and Portunus, occurred more frequently than in the smaller Ray species. In the youngest stages, where the internal yolk sac still persisted, or had just been absorbed, the food consisted, almost without exception, of small Amphipods and Crangonids, with the occasional occurrence of Isopods, Cumaceans, or Schizopods. As the young fish

increased in size, the food consisted more frequently of the larger Crustacean types and of small fish.

The specific identification of the stomach contents was a matter of considerable difficulty, owing to the immaturity of the specimens and their consequent decomposition. The commonest Amphipods were *Nototropis vedlomensis* (Sp. Bate) (syn. *Ampelisca spinipes* Bœck), which were identified for the writer by Mrs. E. W. Sexton. A large percentage of the specimens was ovigerous. The Crangonids caused considerable trouble, as they were nearly all immature forms. The most frequently occurring species were *Pontophilus spinosus* (Leach), *Crangon vulgaris* Linn., *Philocheras trispinosus* (Hailstone), and *Philocheras sculptus* (Bell). The nomenclature is that of Kemp, in Decapoda Natantia of the coasts of Ireland. The fish belonged to three types, of which the following were easily determined :—

Gobius jeffreysii and *G. minutus.*

Arnoglossus laterna—post-larval and early bottom forms.

Callionymus lyra—young specimens.

RECORDS OF EGG-CAPSULES IN THE STOMACHS OF OTHER FISH.

Jensen, in his report on the Selachians of Greenland, p. 19, states, under *R. radiata*, that the egg-capsules were frequently taken in the stomachs of other fishes (especially Halibut). Again, on p. 25, a footnote says that some egg-capsules of *R. hyperborea* have been taken from the stomachs of other fishes (Greenland shark) which had swallowed them. These are the only records which the writer has, so far, been able to obtain.

SUMMARY.

Eleven species of the genus Raia are landed by fishermen at Plymouth. Seven of these occur frequently, and are known to spawn in the immediate neighbourhood.

The species are all oviparous, and the eggs are enclosed in rectangular horny capsules. The capsules vary in size in different species.

Capsules spawned by the same fish show also considerable variation in size.

Egg-capsules have a definite orientation in relation to the adult fish.

Spawning extends over a considerable part of the year, but the maximum probably occurs during the early summer months.

Eggs appear to be deposited in " beds " and the fishes may be local in their places of spawning.

There is a strong suggestion that the eggs are buried in sand on deposition, or that the fish selects sheltered spots between rocks.

The eggs are fertilised in the upper reaches of the oviduct, and the egg-capsule passes rapidly down the oviduct to be extruded from the cloaca.

A single experiment has shown that sperm may be stored for the fertilisation of ripe ova as they pass down to be enclosed in the shell.

The egg-capsule is aerated by general osmosis and by the special adaptation of slits on the horns. The aeration is greatly assisted by a rhythmical lateral movement of the whole body, and especially of the tip of the elongated tail.

Temporary external branchial filaments are developed by the embryo. They are highly vascular, but may also be used for the absorption of nutriment.

The period of incubation of the embryo has been determined for six species under artificial conditions from four to ca. fifteen months.

The newly hatched fish differs considerably in character and in shape from the adult, and undergoes definite post-embryonic changes.

The tip of the tail behind the second dorsal fin is gradually reabsorbed about the end of the period of incubation, and more quickly after the embryo has hatched out.

The embryo absorbs the yolk from the external yolk sac through the medium of an internal yolk sac which opens directly into the anterior end of the spiral valve.

The food of the early post-embryonic stages consists almost entirely of small Crustaceans, Amphipods and Crangonids, which are supplemented, soon after the fish hatches, by a reserve store of yolk from the internal yolk sac.

SUMMARY OF SPECIFIC CHARACTERS OF EGG-CAPSULES AND POST-EMBRYONIC STAGES.

Egg-capsules.

R. clavata. Capsule medium, average 74×57 mm. range 63 to 90×49 to 68. Shell stout and rough, being more or less covered with loose fibres. One side more convex, the other almost flat. Lateral horizontal keel pronounced.

R. maculata. Capsule small. Narrow in proportion to length. Average 70×41 mm. No horizontal keel. Loose fibres not voluminous. The less convex side smooth. Similar to *R. undulata*, but smaller.

R. brachyura. Capsule large. Average 128×78 mm. Thick mass of loose fibres on convex side and on lateral margins. Other side smooth and nearly flat.

R. microcellata. Capsule medium 90×51 mm., curved longitudinally and much narrower at base of short horns, which are stumpy and hooked. Long horns filamentous. Shell almost transparent.

R. nævus. Capsule small, average 63×36 mm. No loose fibres. Shell entirely smooth and transparent. Long horns curved and greatly prolonged. Bi-convex.

R. undulata. Capsule medium. Similar to *R. maculata*, but much larger, 81×52 mm. Lateral fibres voluminous. Outside smooth and slightly rounded, the other more convex with felty mass of fibres.

R. batis. Capsule large. Velvety texture. Loose fibres confined to one spot at the base of the long horns, extremely elongated, being longer than the capsule. Golden yellow colour when spawned. Average 143×80 mm.

R. marginata. Very large capsule. Average 180×138 mm. Beaded structure when spawned. Honeycombed and pitted on long exposure in sea water. Short horns stumpy and hooked. Long horns flat and ribbon shape.

POST-EMBRYOS.

R. clavata. Width of disc in newly hatched young, 71–86 mm. Upper surface entirely spinulose. Median spines 26–38, of which two are in front of the shoulder. Colour of upper surface, K. and V., Code No. 134, with scattered dark patches or a few circular cream-coloured spots. Dark bars present or absent. Teeth ca. 40 in vertical series in upper jaw. Tail long, dorsals far apart.

R. maculata. Width of disc in newly hatched young 71–79 mm. Upper surface smooth, except for a narrow border of asperities on anterior margin of disc, præ-orbital and post-orbital spines, spinules on the interorbit, a median series of body and tail spines, twenty-eight to thirty in number, one spine on each shoulder and a single series of smaller spines on each tail margin. Two spines only on the median line in front of shoulder. Upper surface fawn colour with scattered black and some circular cream-coloured spots. Older specimens show a definite ocellus, with light-coloured centre, and surrounded by a few irregularly shaped black spots. Teeth ca. 40 in vertical series in upper jaw.

R. brachyura. Width of disc in newly hatched young 100–115 mm. Spinulation of upper surface with the same pattern as in *R. maculata.* Median series of 30–36 spines. Never less than three spines in front of the shoulder. Interorbit smooth. Colour of upper surface light fawn, with black spots extending close to the margins of the disc, and with lighter coloured spots showing no definite margin but an imperceptible blending with the general ground colour. No ocellus. Snout very blunt in newly hatched stage and more obtuse in later stages than in *R. maculata.* A single row of less prominent spines on each side of the tail. Teeth ca. 60 in vertical series in upper jaw.

R. microcellata. Width of disc ca. 80 mm. in newly hatched young. A large triangular patch of spines on the snout area, including the interorbit, and extending right across the disc to a line passing through the middle of the orbits. Body otherwise smooth. A median series of about thirty-three spines, of which three are in front of the shoulder. Tail with a double row of spines near the base on each side of the proximal half. Colour of upper surface, K. and V., Code Nos. 130, 135, with white lines parallel to the outer margins of each side of the disc and with a few indefinite white spots. Teeth ca. 42 rows in vertical series in upper jaw.

R. nævus. Width of disc in newly hatched young ca. 60 mm. Upper surface entirely spinulose. A triangular patch of spines on the shoulder. Tail with one or more marginal rows on each side of the proximal half. Colour of upper surface light fawn with wavy bands of sepia colour. Also lighter patches and a few smaller oval or circular light spots. A large marbled ocellus on each wing. Teeth ca. 44 rows in vertical series in upper jaw. Dorsal fins practically confluent.

R. fyllæ. An extremely small fish. Width of disc 39 to 40 mm. Upper surface entirely spinulose. Spinulæ with needle-shaped points. A group of one to three large spines on each shoulder, and a median series of thirty-four to thirty-seven retrorse spines. Dorsals small and practically confluent. Colour of upper surface marbled in later young stages, with oval or circular brown spots surrounded by a light margin. Teeth ca. 26 rows in vertical series in upper jaw.

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The Food of Plankton Organisms.

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With 3 Figures in the Text.

INTRODUCTION.

Following the researches on the food of young fishes, it was thought advisable to investigate the food of the planktonic invertebrates. Whenever possible, therefore, the food of the larger animals brought in by the tow-nets was noted, and also that of many of the smaller creatures down to some of the unicellular organisms, such as the tintinnids and those members of the Peridiniales which are holozoic.

With the larger animals it was hoped to find which of them actually ate the young fishes, and by investigating their food in general ascertain how much they were actually competitors with the fishes.

The present work is offered as a preliminary, and it is hoped to continue it, following it up especially with more experimental work on the living animals.

The tow-nettings were examined fresh and the food noted. Sometimes a few hours elapsed between the taking of the sample and the examination, so that some of the catch was moribund in the jar. There is always the objection that the food might have been taken in the jar whilst the plankton was being brought in, and it is a matter of general observation that many medusæ and various pelagic animals will devour young fishes and almost anything living when crowded up with them in the hauls. In many of the organisms examined, however, the nature of the food was so consistent in various hauls and from various localities that it seems almost impossible to believe that it is merely accidental, and it is probable that what food one usually finds inside any planktonic animal is natural to it.

The Cœlenterates are the most important of the larger plankton organisms, and it is especially in these that one finds young fishes, often almost

wholly digested. The smaller medusæ (*Phialidium*, *Obelia*, etc.), which are very miscellaneous feeders, and also Ctenophores, frequently contained fishes, all of which were very small, either newly hatched or in the young post-larval stages. Clupeoids were commonly taken, also whiting, wrasse, *Cottus* and many others.

The question arose as to whether these cœlenterates catch the fishes alive and eat them in the sea under natural conditions, or only take the moribund fishes when caught in the hauls. In the interesting experiments of Delap (1903–1905), where she reared various medusæ, although a variety of food was taken, no fishes are mentioned. Browne (1898), who also reared many medusæ, gave them no fishes.

In order to ascertain whether medusæ could catch and eat live fishes a large plunger jar was put up in the Laboratory similar to the original plunger apparatus of Browne (1898). The preliminary results of this experiment are shown in the first part of the present paper under the heading of *Experimental Work*. Although this has only been going on for about two to three months the results show that certain medusæ can, and do, catch and eat live fishes on which they thrive, and there seems no doubt that this is a perfectly natural diet.

Sagitta perhaps comes next in importance amongst the larger animals, its food being sometimes its own species, more usually copepods, but young fishes (herring) have also been found in these.

It was interesting to ascertain what such extremely delicate and transparent worms as *Tomopteris* and larval *Pœcilochætus* feed on. These very seldom have any food at all inside, but a few records show that this food is of the most minute kind. Larval annelids of different species vary as to their diet, most of them being principally diatom feeders, but an interesting case occurred where the larva of *Magelona* was found to feed exclusively on larval bivalves.

Amongst the copepods a good deal of work has already been done by other investigators, but in the present records it is shown that some feed differently from others. The majority are diatom feeders, but a few feed on Crustacea, and it is the same with the decapod larvæ. When kept alive in the plunger jar both the diatom-eating copepods and decapod larvæ would eat the débris (chiefly organic) collected at the bottom of the jar.

Such larval forms as *Actinotrocha*, on the one hand, and *Cyphonautes*, *Tornaria*, Echinoderm larvæ and larval mollusks, on the other, feed differently; *Actinotrocha* living almost entirely on Peridinians, the others on diatoms. The tintinnids are also almost entirely Peridinian feeders.

In many cases there have not been sufficient specimens examined to show that all the above-mentioned facts hold good, but the records are

given as a preliminary, and this year it is hoped to continue them more fully. At present the records for 1922 agree entirely with those given here.

The records have been divided into those from the *Inner Grounds*, including the Sound to Rame Head, and the *Outer Grounds*, including those from Rame Head outwards. The food does not apparently change much in inner and outer grounds.

Certain seasonal changes in the quality of the plankton do affect the general food. For instance, in the spring and throughout the early summer the alga *Phæocystis* is very abundant in the area investigated, forming large gelatinous masses, which clog all the nets and interfere very much with fishing. *Phæocystis* serves as a food for many of the plankton organisms, including *Calanus*, *Temora* and *Evadne*. The very young flounder *Pleuronectes flesus* has been shown to feed on *Phæocystis* (Lebour, 1920), and it is also much eaten by unicellular organisms, such as the unarmoured peridinians. At the time when very large quantities of *Rhizosolenia Shrubsolei* and *R. alata* were present in the plankton they were eaten by *Calanus* to the exclusion of the other diatoms usually taken, such as *Coscinodiscus* and *Thalassiosira*. In the autumn, when *Sagitta* was abundant, it was much eaten by the medusæ *Phialidium* and *Obelia*.

Some organisms seem to be only abundant at the time when the food that they usually eat is abundant. Thus the tintinnid *Cittarocyclis serrata* is specially common in the summer, when the peridinians on which it feeds abound, and it disappears when they disappear.

Flagellates seem to form a large part of the food of the organisms that eat diatoms and peridinians, but as these are very easily destroyed it is difficult to identify them. *Phæocystis*, as is shown, is a most important food. Coccoliths are so frequently found inside the diatom-eating species that the coccospheres from which they come must be much commoner than we are led to believe from the examination of centrifuged water in the district. The coccoliths usually occurring apparently belong to a small species of *Coccosphera*, found fairly frequently in water samples and not as yet identified.

In previous papers (Lebour, 1918–19–20) it has been shown what kind of organisms the young fishes chiefly feed on. As all the food ultimately depends on the plant life, it is interesting to find out the various chains of food formed from the plants upwards until we reach the fishes. For instance, young Clupeoids in their very early stages feed chiefly on larval mollusks. So far as we have examined them all larval mollusks feed on diatoms. Those most commonly found inside them were small forms of the naviculoid type ; also other single-celled species, such as *Coscinodiscus* and *Pleurosigma* and colonial diatoms, such as *Thalassiothrix* and

Thalassiosira. These are all kinds found all the year round, and not specially dependent on the seasons. Records for 1922, not yet published, show this much more fully than those given in the present paper. In order that the young Herring may have a sufficiency of mollusk food, it is thus essential that there should be plenty of these diatoms. Later on the small post-larval Clupeoids and nearly all the other post-larval fishes eat copepods. These, as is already well known, depend largely on the diatoms for food, in fact many seem to be almost wholly diatom feeders. This seems to be the case with *Pseudocalanus*, which is the main food of so many of the little fishes, also with *Acartia Clausi*, *Paracalanus parvus* and probably many others. *Temora longicornis*, although feeding largely on diatoms, also sometimes eats copepods, and it is the same with *Calanus finmarchicus* and *Centropages typicus*. All of these also eat flagellates more or less, probably more than we can prove, on account of their being easily destroyed. With all these diatom-eating copepods, however, it is striking that disc-shaped forms, such as *Coscinodiscus* and *Thalassiosira*, are by far the most commonly eaten. *Paralia sulcata* is also common. This when broken up is also disc-shaped. It is possible that it may be an easier shape to be drawn in by the currents created by the mouth parts. All the copepods seem to crush up the diatoms that they eat. A few cases occur of many *Calanus* eating the needle-like species of *Rhizosolenia* when they are very abundant, and these also occur occasionally in other copepods. Dakin (1908) found in the copepods at Kiel that *Coscinodiscus* and *Thalassiosira* were the commonest food. He also found that *Calanus* ate *Biddulphia*. Although this genus is so common it is very seldom found in the Plymouth copepods, probably being too large except to be taken occasionally (e.g. *Calanus* in November in these records). Esterley (1916), who gives an account of the mechanism of feeding in copepods, also finds *Coscinodiscus* to be the commonest food of *Calanus*. He shows how currents created by the copepod bring food to it which is formed into a ball and swept into the mouth, the mandibles probably crushing the larger diatom shells. It is almost certain that in the absence of the disc-shaped diatoms those of other shapes would be taken instead;* but both *Coscinodiscus* and *Thalassiosira* are common most of the year, and there is a distinct preponderance of the disc-shaped diatoms as food for the diatom-eating copepods in the Plymouth district and also at Kiel and in America.

The copepods so far examined seem to group themselves into three natural groups according to the food taken, always allowing for the fact that a sufficient number has not yet been examined for this to be anything

* The *Calanus finmarchicus* reared by Crawshay in the Plymouth Laboratory were fed on a pure culture of *Nitzschia closterium*.

but a suggestion, and that flagellates are also much eaten by those that eat diatoms and a mixed diet :—

Diatom Feeders.	*Mixed Diet* (chiefly diatoms & copepods).	*Copepod Feeders.*
Pseudocalanus elongatus.	Temora longicornis.	Anomalocera Pattersoni.
Acartia Clausi.	Centropages typicus.	Labidocera Wollastoni.
Paracalanus parvus.	Calanus finmarchicus.	
Oithona similis.		
Corycæus anglicus.		

It is to be noted, however, that the food given above is the predominant food, and other organisms generally occur as well in less amount. The records for 1922 so far seem to confirm this. In the same way we seem to be able to divide the larval decapods according to their food, but in this case they are nearly all predominantly diatom feeders. The only striking exception seems to be the larval lobster, which in two specimens examined were full of crustacea remains. Most of the crab zoëæ eat diatoms, but in one case bits of copepod were found. One megalopa was found to be full of bits of decapod larvæ. Calcareous remains of what are probably larval mollusks are often found in the decapod larvæ, together with diatoms ; also bits of the spines of Echinoderm larvæ, showing Metazoa as well as unicellular organisms are eaten. We can roughly divide most of the plankton organisms into diatom feeders (including other minute unicellular organisms), Peridinian feeders (Peridinians being the predominant food, but also other unicellular organisms being taken), Mollusk feeders, Crustacea feeders and Miscellaneous feeders (the miscellaneous being chiefly Cœlenterates and *Sagitta*) :—

Diatom Feeders.

Copepods (most of the common species excluding Anomalocera and Labidocera and probably most of the Harpacticids).

Decapod larvæ (excluding the larval lobster and crab megalopæ).

Echinoderm larvæ.

Mollusk ,,

Annelid larvæ (most of the common forms excluding Magelona).

Cyphonautes.

Tornaria.

Tomopteris heligolandicus.

PERIDINIAN FEEDERS.

Actinotrocha.
Tintinnids.

MOLLUSK FEEDERS.

Magelona larva.

CRUSTACEA FEEDERS.

Anomalocera.
Labidocera.
Larval lobster.
Crab megalopa.
Sarsia tubulosa.
Many other medusæ.

MISCELLANEOUS FEEDERS.

Most medusæ.
Pleurobrachia.
Beroë.
Sagitta.

Amongst these are many that eat fishes :—

Aurelia (including the ephyra), ephyra of Chrysaora.
Phialidium.
Obelia.
Turris.
Arachnactis larva.
Rathkea.

With these so-called miscellaneous feeders there seems to be generally some food more frequently taken than the rest, the reason probably being that it is present at the moment more abundantly. Thus when *Beroë* and *Pleurobrachia* are both commonly present the latter is eaten by *Beroë*, but it is not always the commonest creatures in the tow-nets that are taken as food. *Magelona* larva manages to obtain bivalves, although gastropods may be much commoner, and the tintinnids nearly always eat Peridinians in spite of the fact that diatoms are more numerous.

Experimental Work.

A large glass aquarium (about 50 litres capacity) was put up in the Laboratory early in February, 1922. This was fitted up with a glass plunger, as described by Browne (1898), and the water has not yet been changed (at the end of April). In this it was possible to keep medusæ and other plankton organisms alive and study their food and methods of feeding. The following notes for the first three months are given as a preliminary, and it is hoped to continue experiments with other animals. So far those studied were *Aurelia* ephyra and the metamorphosed form, *Phialidium hemispherica*, *Turris pileata*, *Sarsia tubulosa* and *Arachnactis Bournei*. A variety of mixed plankton was put in with these, including many Crustacea. Whenever possible newly hatched and very young fishes were put in also alive, the latter from tow-nets and Young Fish Trawl, the former hatched in the Laboratory, and these were eaten by all the above-mentioned Cœlenterates except *Sarsia*, which always ate copepods.

The following young fishes were used :—

Ammodytes tobianus, with small amount of yolk-sac, about 6–7 mm. long, from tow-nets and Young Fish Trawl.

Cottus bubalis, newly hatched in Laboratory, ca 5 mm. long.

Agonus cataphractus, probably a few days old, from tow-nets, ca 8 mm. long.

Solea vulgaris, newly hatched from eggs in the Young Fish Trawl, ca 3–3·5 mm. long.

Gobius minutus, newly hatched in the Laboratory, ca 3 mm. long.

Blennius pholis, newly hatched in the Laboratory, ca 4 mm. long.

Nerophis lumbriciformis, newly hatched in Laboratory, ca 12 mm. long.

Aurelia aurita Lam.

Delap's (1905) experiments showed the ephyræ of *Aurelia* to eat when very young *Obelia* and *Phialidium*, small copepods and fish eggs, afterwards small Ctenophores and Pteropods and big *Calanus*. Gemmill (1921) has recently shown that the newly liberated ephyræ can feed upon ciliate Infusoria, which they catch by means of stinging cells on the lappets and carry to the stomach by means of currents set up by ciliary action. This method of feeding went on for at least two weeks.

Ephyræ of Aurelia in the plunger jar in the Laboratory at a very young stage, and probably not more than a few days old, ate young fishes. As they required a great deal of food, only one was allowed to remain in the

Aquarium. This was put in on February 9th and measured about 5 mm. It repeatedly caught and ate the young fishes, the method of catching them being apparently just to sting them with the lappet's edge, and then envelope them with the whole of the umbrella until the fish is well fixed in the manubrium, when it is digested. The live fish brushes against the edge of the lappets and is caught, but usually struggles for some time before it is eaten. Occasionally a crab zoëa was eaten, and this was caught in the same way. On March 13th the ephyra had grown into an *Aurelia*, which still ate the young fishes, but with a greatly increased appetite. These are now caught by the marginal tentacles, and presumably stung, and then helped by the long lips into the manubrium. These lips may also sting the fish, which are sometimes seen attached to them by a thread. The *Aurelia* was, on the 24th of March, transferred to another jar, in which it continued to eat fishes whenever these were given to it. One dab, just metamorphosed and measuring about 16 mm., was put into the jar and not eaten, presumably because the *Aurelia* could not catch it when it was close to the glass, as it usually was. Another dab, 15 mm., not yet fully metamorphosed and quite transparent which swam about freely, was eaten by the Aurelia. Sixteen small fishes, *Cottus* or *Blennius*, made a usual meal for the Aurelia, and would take less than half an hour to catch. When fishes were not available, amphipods, crab zoëæ and other crustacea, even small copepods were taken, and also medusæ. The *Aurelia* is still alive (April 24th), measuring about 25 mm., and the dab is also alive in the jar with it.

The following notes show the food of this *Aurelia* :—

Date.	*Time.*	*Food.*
Feb. 17.	9.30 a.m.	Young *Ammodytes tobianus*. Not wholly digested at 7 p.m.
,, 21.	9.30 a.m.	Young *Ammodytes tobianus*, 12.30, not completely digested. Disappeared by 4.30.
,, 27.	10 a.m.	Young *Cottus bubalis*, very lively, 3 p.m. From tail to anus digested. Disappeared next morning.
,, 28.	9 a.m.	Young *Cottus bubalis*.
March 1.	11.30 a.m.	Young *Cottus bubalis*, digests up to near its head, gets rid of the rest.
	6 p.m.	Another young *Cottus bubalis*.
,, 2.	9 a.m.	Young *Cottus bubalis*, eats half of it.
,, 3.	8 p.m.	Young *Cottus bubalis*.
,, 6.	9.15 a.m.	Remains of a *Cottus* inside it.
,, 10.	11 a.m.	A crab zoëa (very few fish in jar).

Date.	*Time.*	*Food.*
March 13.		Changed into young *Aurelia*. Eats 6 live gobies (*Gobius minutus*).
	2.30 p.m.	At least two more eaten, later the stomach was full of them.
,, 20/21.		Still eating gobies. Caught and ate a newly hatched *Nerophis lumbriciformis*.
,, 22.	9.30 a.m.	Two *Nerophis*, now partly digested.
	12.30 p.m.	Another *Nerophis*.
,, 23.	9.30 a.m.	One *Nerophis*.
	3 p.m.	Another *Nerophis*.
,, 24.	9.30 a.m.	One *Nerophis*, *Sarsia tubulosa*. It is now transferred to another jar.
,, 30.		Eats 20 newly hatched *Cottus* within half an hour, and goes on eating them until they are finished.

After this there is a period when fishes are scarce. The dab, 15 mm., is put in but not eaten. Amphipods, crab zoëæ (including Corystes) and other Crustacea are eaten.

April 21. Several newly hatched *Blennius pholis* eaten.

,, 22. Several *Blennius* and a dab not yet completely metamorphosed (15 mm.) eaten.

,, 23/24. Continues eating blennies until they are all done.

It is thus evident that fishes can form an important part of the food of *Aurelia* from very early ephyra stages up to a large size. The records stop here (the MSS. going to press), but the *Aurelia* is still feeding freely on fishes and growing fast. We do not know if it will continue to eat fishes in its adult stage. Further observations will be interesting.*

A young ephyra of *Chrysaora* also ate young fishes in the same way as *Aurelia*.† Unfortunately it disappeared, being probably eaten by some medusa in the jar.

* Since writing the above, Orton's observations on the feeding of *Aurelia* (*Nature*, August, 5, 1922) show that adults feed normally on plankton which is collected on the bell surface and transferred by ciliary currents to the margin of the bell and removed by the lips of manubrium to the mouth.

† Delap (1901) reared *Chrysaora isoscelcs* up to 13 inches. This caught a young fish about an inch in length, but released it without doing it any harm. Other small fish were also kept with it, which it did not attempt to catch. Its chief food was cœlenterates.

PHIALIDIUM HEMISPHERICUM (Gron.).

Many *Phialidium* were seen to catch and eat the young fishes. In the plunger jar they throve well, growing to a large size. The medusa would float about in the jar with its tentacles outstretched to the finest threads, several times longer than the diameter of the bell, and wait for some living thing to come along. Directly this touched the tentacle it reacted and presumably stung the prey. Several more tentacles then came into use and together entangled the fish, or whatever the food caught might be. The fish would struggle, and the long tentacles would play it until it was exhausted. Very rarely the fish would escape and run away with the tentacle. Usually it would be caught and killed in a few minutes.

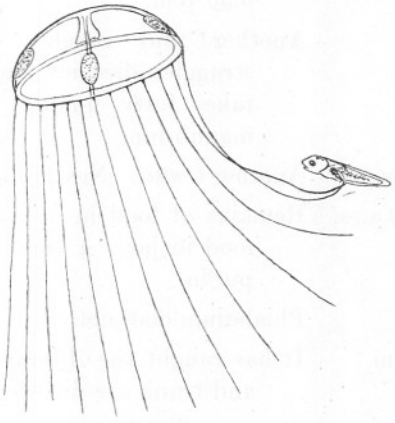

FIG. 1.—Phialidium (ca 10 mm. across) catching a *Cottus*.

The tentacles would then, helped by the umbrella folded in, proceed to deposit the fish in the manubrium. Sometimes this would only take a few minutes; at other times more difficulty would be encountered, and after twenty minutes or more the umbrella would turn upside down and the fish would be dropped into the manubrium. From a few hours to half a day or, rarely, more was taken to digest the fish, sometimes the head part being disgorged. *Phialidium* was seen to eat young *Cottus bubalis*, *Ammodytes tobianus*, *Agonus cataphractus*, *Solea vulgaris*, *Gobius minutus* and *Blennius pholis*.

The following notes were made on one put into the jar on February 17th, which measured about 6 mm. across. This grew to about 12 mm. across when it had 24 tentacles. After March 15th it disappeared, probably eaten by something else.

Date.	*Time.*	*Food.*
Feb. 22.		A solid mass, probably a fish, in manubrium.
,, 27.	9.15 a.m.	Two *Cottus bubalis*, one half digested.
	4.30	Almost completely digested.
,, 28.	9 a.m.	A partly digested *Cottus*.
March 1.	10.15 a.m.	Caught a live *Ammodytes tobianus*. Inside manubrium by 10.30, within an hour it is an opaque mass, only distinguishable as a fish by its eyes. Almost entirely digested by 3 p.m.
	6 p.m.	Caught a *Cottus*.
March 2.	9 a.m.	Another *Cottus* caught, completely digested by 6.30 p.m.
,, 3.	9.20.	Another *Cottus* caught, which after frantic struggles dies in about ten minutes. It takes forty minutes to get it into the manubrium.
,, 4.	12 noon.	Another *Cottus*. Nearly digested at 6.30 p.m.
,, 5.	11.30 a.m.	Remains of food in manubrium. Hardly any food in jar. A small *Agonus cataphractus* put in.
,, 7–9.		Phialidium does not eat.
,, 10.	11 a.m.	It has caught the *Agonus*. At 6 p.m. the tail and trunk are digested, the rest got rid of.
,, 13.		Some small *Gobius minutus* put in.
	12 noon.	It has caught one.
	12.15 p.m.	It has caught another. These both helped by the umbrella edge into manubrium.
	3 p.m.	It has caught 2 more.

It is now about 10 mm. across with 24 tentacles. Disappears on March 15th.

Other specimens of *Phialidium* ate *Solea vulgaris* and *Blennius pholis* besides the above-mentioned fishes. One which was very lively on April 2nd fed on several young fishes, and on April 10th caught and ate a live *Sagitta* in just the same way as it caught and ate the fishes. Small Crustacea were also occasionally taken, but these more often escaped, especially the copepods. On the whole when the young fishes were abundant it seemed that they were much more often taken as food than Crustacea, which were also present in numbers.

On the other hand, *Sagitta*, if put into the jar, was caught and eaten almost at once. Unfortunately only few *Sagitta* were obtained alive in these months. It is hoped to put in more later, for judging from the specimens examined from the tow-nets, *Sagitta* is an important food of *Phialidium*. Amongst *Crustacea* taken Cirripede nauplii were noticed.

We thus see from these notes that *Phialidium* can, and does, capture live fishes and eat them, and if these are present in any quantity they seem to be able to serve as its whole diet. The fishes are all very young,

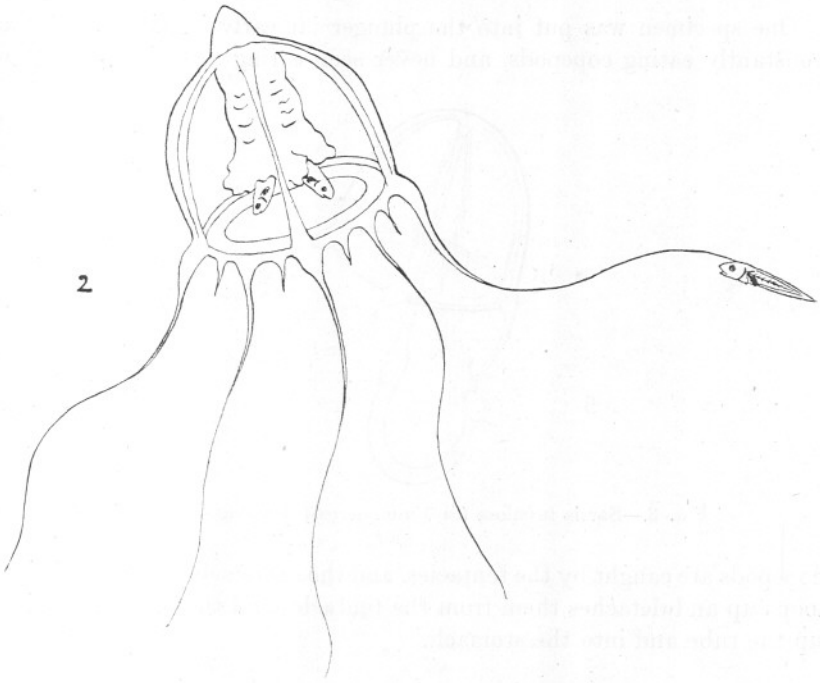

FIG. 2.—Turris (ca 10 mm. across) catching a *Cottus*.

but may measure more than half the diameter of the medusa's umbrella. From the records of the food from the tow-nets given below most of the fishes were taken in the spring and summer, *Sagitta* and other organisms more especially in the autumn.

TURRIS PILEATA (Forskål).

It was difficult to keep *Turris* alive in the plunger jar. One specimen which lived several days ate a young squid, which was longer than its own body and which completely filled its stomach and took several days to digest. Three days afterwards (March 30th) it had two young *Cottus bubalis* inside it, and with its tentacles greatly elongated it was seen to

catch another in the same way as *Phialidium*. After playing it for over ten minutes the medusa lost the fish, which ran away with the tentacle. Another *Turris* caught and ate young *Blennius pholis*, and there seems no doubt that this is its natural method of feeding, and that it can, and does, catch and eat live fishes, although it certainly does feed upon a great variety of other organisms, especially Crustacea. One of these was seen to eat crab zoëæ.

Sarsia tubulosa (Sars.).

One specimen was put into the plunger jar early in March. It was constantly eating copepods, and never seen eating anything else. The

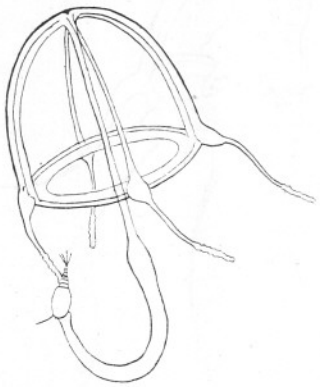

Fig. 3.—Sarsia tubulosa (ca 7 mm. across) catching a Copepod.

copepods are caught by the tentacles, and the extremely long manubrium loops up and detaches them from the tentacle, and thus they are taken up the tube and into the stomach.

The following notes were made on its food from March 13th to the 24th, on which day it was eaten by the *Aurelia*.

Date.	*Food.*
March 13.	Caught and ate 2 *Pseudocalanus elongatus*.
,, 14.	,, ,, ,, several *Acartia Clausii*.
,, 17.	,, ,, ,, ,, ,, ,,
,, 20.	,, ,, ,, a *Calanus finmarchicus*.
,, 22.	,, ,, ,, several *Temora longicornis* and *Acartia Clausii*.
,, 23.	,, ,, ,, several Acartia and Pseudocalanus.
,, 24.	,, ,, ,, eaten by Aurelia.

Mr. Smith (1898) noticed large *Sarsia tubulosa* up the River Tamar by the quay above Saltash Bridge, feeding on mysid larvæ. Forbes (1848) describes this species devouring small Crustacea, and also attacking and beginning to swallow *Lizzia* (*Rathkea*) *octopunctata*. It seems evident that small Crustacea are its natural food, and it was never seen to eat a fish, although many were in the jar with it.

Arachnactis Bournei Fowler.

This is the larva of a *Cerianthus*, probably *Cerianthus Lloydi* Gosse. Fowler (1897) shows that it is different from *Arachnactis albida* both in structure and colour. Several of these were kept in the plunger jar and grew to a large size with from 10 to 12 marginal tentacles. They also ate the young fishes, several being observed inside them. Only once was one seen actually catching a fish, a young *Solea vulgaris*. Whilst catching it two long threads were protruded from somewhere between the tentacles and the fish caught by one of these. It is probable that these were long threads of stinging cells. They were, however, so long that they had the appearance of the fine tentacles in Phialidium. *Cottus bubalis* and *Blennius pholis* were also eaten by *Arachnactis*.

The above notes on the feeding of plankton organisms in a plunger jar are only a beginning. From them we see that the following Cœlenterates catch and eat young fishes :—

Aurelia aurita (including the ephyræ).

Chrysaora isosceles (ephyra).

Phialidium hemisphericum.

Turris pileata.

Arachnactis Bournei.

And that *Sarsia tubulosa* lives almost entirely on copepods and other small Crustacea.

FOOD RECORDS.

In the following notes each group is taken and all the records of food given. Some of these are only single and most are entirely inadequate at present, but they are offered as a preliminary and it is hoped to continue the work in more detail.

A short summary for each month throughout the year 1921 follows :—

January.

Very young herrings were abundant in the plankton. These were eaten by *Phialidium*, which also ate sprat eggs. *Coscinodiscus excentricus*

was the commonest diatom, probably the species of *Coscinodiscus* chiefly eaten by *Calanus*, *Pseudocalanus* and *Acartia*.

February.

Coscinodiscus, chiefly *C. excentricus*, is again the commonest diatom in the plankton, and is eaten by terebellid, spionid and polynoid larvæ, *Pseudocalanus* and *Temora*, Cirripede nauplius, *Galathea* larva and zoëa of *Carcinus mœnas*. *Phialidium* took *Sagitta*, whilst *Sagitta* ate *Pseudocalanus*.

March.

Phæocystis appeared abundantly, and was eaten by *Temora* and also *Evadne*, which had just begun. *Aurelia* ephyra was eating *Gobius*. Amongst the *Phæocystis* were many small creatures feeding on it.

April.

Phæocystis predominates with many *Amphidinium* and other unicellular organisms feeding on it; also *Calanus* eating it largely, but also eating *Coscinodiscus* and *Thalassiosira*. The green cells in *Evadne* and in the Cirripede nauplii and also in many of the copepods are probably *Phæocystis*. *Magelona* larva begins and eats larval bivalves, other larval annelids eating *Coscinodiscus*, *Thalassiosira* and *Skeletonema*, all common in the plankton. Peridinians plentiful, *Actinotrocha* feeding on them.

May.

By the middle of the month *Phæocystis* is nearly all gone. *Actinotrocha* continues feeding on Peridinians. *Centropages typicus* takes a variety of diatoms with a few copepods. *Phialidium* abundant, eating fish, and many *Sagitta*. Obelia very common, eaten by *Phialidium*, and *Obelia* itself eating *Sagitta* and *Calanus*.

June.

Obelia and *Phialidium* still eating fish as well as Crustacea. *Turris* abundant and chiefly eating crab zoëa. *Cyphonantes* very common and eating a variety of small diatoms, but not *Rhizosolenia*, which is the commonest diatom this month. *Actinotrocha* still eating Peridinians. Many larval decapods eating larval mollusks, larval echinoderms, diatoms and coccospheres.

July.

Much *Rhizosolenia*, chiefly *R. alata* and *R. Shrubsolei*, these are eaten by *Calanus* in large quantities, but a few have eaten other diatoms, principally

Coscinodiscus, and bits of copepods. *Cittarocyclis* eating many peridinians, especially *Protoceratium* and *Dinophysis*, which are very abundant. *Porcellana* larva eating a variety of diatoms, including *Rhizosolenia*, with coccospheres, and is itself eaten by *Pleurobrachia*. *Obelia* and *Phialidium* eating fish and Crustacea. *Oikopleura*, which is common, eaten by *Sarsia*, *Stomotoca* and *Phialidium*.

August.

Many *Cittarocyclis* eating chiefly *Prorocentrum*. Many *Sagitta* chiefly, eaten by *Phialidium*, and themselves eating *Calanus*. *Calanus* eating diatoms (Skeletonema), flagellates and bits of copepods. Decapod larvæ eating diatoms (*Calocaris* larva containing *Rhizosolenia setigera*). Echinodum larvæ eating small flagellates, peridinians and diatoms (*Thalassicthrix*), *Magelona* larva still eating larval bivalves. *Pleurobrachia* eating many *Calanus*.

September.

No records for September.

October.

Very rich plankton. *Rhizosolenia Shrubsolei* and *R. alata* still very common, also *Biddulphia sinensis*. *Rhizosolenia* eaten by larval terebellids, *Calanus* and *Anomalocera*, *Biddulphia* by terebellid larvæ. Larval bivalves very common, eaten by *Magelona* larva. *Calanus* eating much *Thalassiosira* and *Coscinodiscus*. Larval decapods eating many small diatoms, chiefly *Coscinodiscus*, *Navicula* and *Paralia*, also coccospheres. Many *Sagitta* eaten by *Phialidium*, *Obelia*, *Cosmetira* and *Pleurobrachia*.

November.

Rhizosolenia Shrubsolei and *R. alata* still common, eaten by terebellid larva, *Paracalanus*, *Ebalia* zoëa and *Galathea* larva. Other Crustacea, including *Calanus*, eating chiefly *Coscinodiscus* and *Thalassiosira*. *Sagitta* common, eating copepods, and eaten by *Obelia*, *Phialidium* and *Stomotoca*. *Biddulphia sinensis* eaten by *Calanus*.

December.

Much *Sagitta* eating *Sagitta* and eaten by *Obelia* and *Phialidium*. Copepods and larval decapods eating chiefly *Coscinodiscus*, *Paralia* and *Thalassiosira*. *Actinotrocha* still present, but eating more diatoms, as there are fewer peridinians.

PROTOZOA.

Peridiniales.

Amphidinium sp.

Whenever *Phæocystis* was present in abundance, a small *Amphidinium*, apparently closely related to *A. crassum* Lohmann, to be worked out later, was feeding on it and was nearly always full of remains of the spores. Other unarmoured peridinians also fed on it (*Gymnodinium triangularis*, Lebour, 1917; *G. rhomboides* Schütt), and it seems to collect many animals round it. As has been frequently pointed out, the unarmoured peridinians ingest solid food to a large extent, and various unicellular organisms have been recognised inside many species. In *Polykrikos*, which is armed with large nematocysts, various other peridinians have been found, notably a pink peridinian, probably *Peridiniopsis asymmetrica*, and Kofoid (1921), who figures many other Peridinians with food, states that many planktonic organisms have been recognised inside it, both Metazoa and Protozoa. A diatom (*Thalassiosira*) was found once (Lebour, 1917) in *Gymnodinium rhomboides*, but usually the food recognised in these unarmoured peridinians are peridinians also, except when *Phæocystis* is eaten, and probably many of the greenish brown masses to be seen in these are remains of flagellates.

It is well known also that Noctiluca (now regarded as a peridinian) devours many micro-organisms, such as diatoms, flagellates and Peridinians, and Stein (1883) has figured some of these (Plate XXV). The following organisms were found in *Noctiluca* from Wembury Bay, June, 1898: Halosphæra viridis in many, Paralia sulcata in 3, Pleurosigma in 2, Pleurosigma, Paralia sulcata, Prorocentrum micans in 1.

The only other unicellular organisms examined for food were the tintinnids, chiefly Cittarocyclis serrata. It was interesting to find that in these the food was almost entirely peridinians.

Cittarocyclis serrata (Möbius).

Inner Grounds, 1921, *July*. Prorocentrum micans in 1, Dinophysis acuminata, Peridinium pellucidum in 2, Protoceratium reticulatum (1–7) in 8, Protoceratium, Dinophysis reticulatum, Exuviella perforata in 1, Dinophysis acuminata, Protoceratium, Prorocentrum micans in 1, Dinophysis acuminata in 1, Dinophysis acuminata, Peridinium indet. in 1, remains of Peridinians indet. in 5, Skeletonema costatum in 1, Goniaulax sp., small triangular bodies indet. in 1. *August*, Prorocentrum micans, Peridinium sp., small Peridinians indet. in 1, Dinophysis sp. in 3, Tintinnoposis beroidea, Prorocentrum micans in 1, Prorocentrum micans in 2, Prorocentrum micans, Goniaulax spinifera in 1.

Outer Grounds, 1921, *July*. Prorocentrum micans in 2, Peridinium sp., Peridinian remains in 1, Peridinian remains in 1, Dinophysis sp. in 1, Prorocentrum micans, Peridinium pallidum, Dinophysis sp., triangular bodies indet. in 1.

Thus out of 36, 35 contained Peridinians, one also a Tintinnopsis and one a diatom.

Remains of Peridinians also occurred in one specimen of Tintinnopsis beroidea and one of Tintinnopsis campanula (July, 1921).

As the tintinnids occur in large numbers only in the summer and also the Peridinians, their abundance would be easily explainable by the amount of Peridinians present.

CŒLENTERATA.

Hybocodon prolifer L. Ag.

Outer Grounds, 1921, *April*. Calanus finmarchicus in 1. In 1922 nearly all were eating copepods.

Steenstrupia rubra Forbes.

Inner Grounds, 1921, *May*. Fish eggs in 2, Temora longicornis in 1, Porcellana larva in 1, Corycæus anglicus in 1, Gebia larva in 1.

The presence of fish eggs in 2 out of 6 is interesting.

Sarsia prolifera Forbes.

Inner Grounds, 1920, *June*. Labrus sp. (bergylta type) juv. in 1. *July*, Oikopleura dioica in 1, Centropages typicus in 1.

Outer Grounds, 1920, *July*. Large annelid indet. in 1. Copepod egg in 1.

The Labrus in one of these tiny medusæ was very large for it and much more than filled the manubrium.

Sarsia gemmifera Forbes.

Outer Grounds, 1921, *July*. Calanus finmarchicus in 1.

Sarsia tubulosa (Sars).

As is shown above this medusa in the plunger jar fed entirely on copepods (page 656).

Stomotoca dinema L. Ag.

Inner Grounds, 1920, 1921, *June*. Cosmetira pilosella in 1. *July*, Oikopleura dioica in 1. *October*, Calanus finmarchicus in 1. *November*, Phialidium sp. in 1, Sagitta bipunctata in 2.

Outer Grounds, 1921, *October*. Calanus finmarchicus in 2. *December*, Sagitta bipunctata in 1.

TURRIS PILEATA (Forskål).

Inner Grounds, 1920, 1921, *June*. Carcinus mænas zoëa in 2, Carcinus mænas zoëa and Cosmetira pilosella in 1, Carcinus mænas zoëa and Porcellana larva in 1, Crab zoëa indet. in 3, Crab zoëa indet. and Centropages typicus in 1, Acartia Clausi in 2, Galathea larva in 1, Cottus bubalis in 1, Phialidium sp. in 2. *July*, Crab zoëa indet. in 3, Phialidium sp. and Hybocodon prolifer in 1, Cosmetira pilosella in 1. *August*, Pseudocalanus elongatus in 1. *September*, Crab zoëa indet. in 2, Calanus finmarchicus in 6, Pseudocalanus elongatus in 1. *October*, Temora longicornis and Calanus finmarchicus in 4, Calanus finmarchicus in 1.

Outer Grounds, 1920, 1921, *June*. Gebia larva in 1. *July*, Calanus finmarchicus in 1. *August*, Calanus finmarchicus in 3.

Those examined, which came chiefly from the Inner Grounds, 39 in all, had principally fed on Crustacea, in one case a young fish was present and 5 contained medusæ. Crab zoëa were the commonest food.

As is shown above (page 655), *Turris* in the plunger jar caught and ate a cephalopod and young fishes.

RATHKEA OCTOPUNCTATA Hæckel.

Inner Grounds, 1921, *February*. Oikopleura in 1. *April*, Crab zoëa indet. in 2, Pseudocalanus elongatus in 2, Poecilochætus larva in 1.

Outer Grounds, 1921, *April*. Young Pilchard in 2.

LAODICEA CRUCIATA L. Ag.

Inner Grounds, 1920, *August*. Calanus finmarchicus in many.

A large number were in the sample and all had eaten Calanus.

OBELIA sp. (including *geniculata* Allman and *nigra* Browne).

Inner Grounds, 1920, 1921, *March*. Oikopleura dioica in 1. *April* Oikopleura dioica in 4, Acartia Clausi in 1. *May*, Sagitta bipunctata in 2, Calanus finmarchicus in 1. *June*, Fish egg in 1, Crab zoëa in 1, Evadne Nordmanni in 1, Pseudocalanus elongatus in 1, Steenstrupia rubra in 1. *July*, Calanus eggs in 1, Calanus finmarchicus in 1, young fish indet. in 1. *August*, Sagitta bipunctata in 6, Calanus finmarchicus in 2, Podon intermedius in 1, Tomopteris heligolandicus in 1, Temora longicornis in many. *September*, Sagitta bipunctata in 5, Oikopleura dioica in 2, Calanus finmarchicus in 1. *October*, Sagitta bipunctata in 6, Obelia medusa in 1, Pseudocalanus elongatus in 1. *November*, Sagitta bipunctata in several, Paracalanus parvus in 1.

Outer Grounds, 1920–1921, *March*. Oikopleura dioica in 1. *April*, young pilchards in 3. *May*, Sagitta bipunctata in 2. *September*, Sagitta

bipunctata in 7, Calanus nauplii in 1. *October*, Sagitta bipunctata in 1. *November*, Sagitta bipunctata in a few. *December*, Sagitta bipunctata in several. Sagitta is certainly the most frequent food of Obelia. In one large sample from Station E 1 (14 miles S. of Breakwater) in November, 1921, every specimen had one or two Sagitta inside it, and besides these out of over 60 examined over two-thirds contained Sagitta. One sample in August, 1920, from the region of the Knap buoy had all eaten Temora longicornis. A few odd ones had eaten Calanus, Pseudocalanus and Acartia at various times, and once a Crab zoëa, Evadne and Podon. Four contained fish and one a fish egg. The fact that 3 contained young pilchards proclaims them an enemy of the little fish.

PHIALIDIUM sp. (chiefly *P. hemisphericum* Gron., but probably including *P. buskianum* Browne).

Inner Grounds, 1920–1921, *January*. Young Herring in 5, Sprat egg in 1, Sagitta bipunctata in 2. *February*, Onos egg in 1, Sagitta bipunctata in 1. *April*, Oikopleura dioica in 1. *May*, young Whiting in 1, fish egg in 1, Crab zoëa in 4, Gebia larva in 1, Porcellana larva in 1, Sagitta bipunctata in many, Obelia medusæ in many. *June*, Cottus bubalis juv. in 3, Labrus juv. (bergylta type) in 1, fish eggs in 5. Temora longicornis in 1, Calanus eggs in 1, zoëa of Carcinus mænas in 1, Crab zoëa indet. in 2, Acartia Clausi in 2, Pandalus larva in 2, Gebia larva in 1, Hippolyte larva in 1, Sagitta bipunctata in 4. *July*, Labrus juv. (bergylta type) in 1, Blennius ocellaris juv. in 1, young Whiting in 1, young fish indet. in 1, fish eggs indet. in 2, Sagitta bipunctata in 6, Sarsia prolifera in 2, Obelia medusa in 1, Gebia larva in 3, Pandalus larva in 1, Polychast larva in 2. *August*, Sagitta bipunctata in many, Calanus finmarchicus in many, Cosmetira pilosella in 1, fish remains in 1, Sagitta bipunctata and Onos egg in 1. *September*, Acartia Clausi in 4, Calanus finmarchicus in 3, Temora longicornis in 1, Sagitta bipunctata in 1. *October*, Acartia Clausi in 1. *November*, larval spionid in 1, Sagitta bipunctata in many, Obelia medusa in 2, Phialidium sp. in 1, Crab zoëa in 1, Oikopleura dioica in 5, Paracalanus parvus in 1. *December*, Pseudocalanus elongatus in 2, Pseudocalanus elongatus and Sagitta bipunctata in 1, Sagitta bipunctata in many.

Outer Grounds, 1920–1921, *April*. Gobius juv. in 1, Callionymus juv. in 1, young Pilchard in 2, Pseudocalanus elongatus in 1. *May*, Sagitta bipunctata in 7. *June*, Calanus finmarchicus in 1. *July*, Sagitta bipunctata in 6, Calanus eggs (16) in 1, Callionymus eggs in 2, Crab zoëa in 1, Oikopleura dioica in 3, Muggiæa atlantica in 1. *August*, Sagitta bipunctata in several, Calanus finmarchicus in 2. *September*, Sagitta bipunctata in 3, Gobius juv. in 1, Temora longicornis in 1, Oikopleura dioica in 1. *October*, Sagitta bipunctata in many. *November*, Sagitta bipunctata in

many, Crab zoëa in 1. *December*, Crangon larva in 1, Phialidium sp. in several, Sagitta bipunctata in many.

It will be seen from the records given above that Sagitta is certainly the commonest food of Phialidium, in several samples examined containing many specimens almost every one was eating Sagitta. On two occasions they were all eating Obelia medusæ, and once they were all eating Calanus. At other times, out of over 150 examined, over 60 were eating Sagitta. Phialidium, however, also certainly eats young fishes which have been noted from 39 specimens, young pilchards, herrings and sprat eggs being among those eaten. Various Crustacea were in over 30, Medusæ other than Obelia were in 2, Muggiæa in 1, Oikopleura in 10 and Annelids in 6.

In the late autumn of 1921 Sagitta was very abundant, and at that time served specially as food for Phialidium from both Inner and Outer Grounds. The food taken inside and outside differed hardly at all.

From the records in the plunger jar (page 653) it is seen that it is quite natural for Phialidium to catch and eat the young fishes.

Cosmetira pilosella Hartlaub.

Inner Grounds, 1921, *June*. Crab zoëæ in 4, Caligus rapax in 1.

Outer Grounds, 1920–1921, *July*. Lepadogaster gouani juv. in 1, Autolytus sp. in 1, Sagitta bipunctata in 1. *October*, Sagitta bipunctata in 1.

Saphenia gracilis Forbes & Goodsir.

Inner Grounds, 1921, *July*. Gebia larva in 1.

Aglantha digitale Hæckel.

Outer Grounds, 1920, *July*. Calanus finmarchicus in 2.

Aurelia ephyra.

Inner Grounds, 1921, *February*. Larval gastropod in 1. *March*, Gobius juv. in 3, Crab zoëa in 1.

From the plunger jar records (page 650) it is seen that the ephyræ eat many fishes.

Pleurobrachia pileus (Fab.).

Inner Grounds, 1920–1921, *July*. Calanus finmarchicus in 1, Crab zoëa in 1. *October*, Calanus finmarchicus in 2, Calanus finmarchicus and Sagitta bipunctata in 1, Gebia larva in 1.

Outer Grounds, 1920, 1921, *July*. Calanus finmarchicus in many, Calanus finmarchicus with a few Centropages typicus in a few, Porcellana larva in 1, Crab zoëa in 1, Labrus juv. in 1. *August*, Calanus finmarchicus in several, Centropages typicus in 1. *October*, Sagitta bipunctata in 1.

In other years where records were not kept Pleurobrachia was often seen to be eating young fishes, although only one is recorded here. Calanus seems to be the commonest food, and large masses of Pleurobrachia and Calanus often occur together, especially in the outside waters.

Beroë cucumis Fab.

Inner Grounds, 1920–1921, *July*. Pleurobrachia pileus (many) in 2. *October*, Calanus finmarchicus in 1.

Outer Grounds, 1920, *September*. Pseudocalanus elongatus in 1, Pseudocalanus and Podon intermedius in 1, Pseudocalanus elongatus and Centropages typicus in 1.

One of those in July was packed tight with Pleurobrachia.

From these records of the food of the Cœlenterates in general we find that many of them eat young fishes, the worst offender being *Phialidium*. We also find that *Sagitta* is a favourite food both of *Phialidium* and *Obelia*, and is also taken by several other medusæ. These, however, also eat a good many Crustacea, and *Turris pileata* takes more Crustacea than anything else, although it can eat fishes and Cephalopods. *Calanus* is by far the most commonly eaten, other copepods, decapod larvæ and *Podon* and *Evadne* much more rarely and a few annelids. Occasionally other medusæ are taken. *Pleurobrachia* eats more *Calanus* than anything else, but also eats decapod larvæ, other copepods, *Sagitta* and very young fishes. *Beroë* eats Crustacea and *Pleurobrachia*.

CHÆTOGNATHA.

Sagitta bipunctata (Quoy & Gaimard).

Inner Grounds, 1920–1921, *February*. Pseudocalanus elongatus in 2. *September*, Temora longicornis in 1, Sagitta bipunctata in 1. *October*, Sagitta bipunctata in 9, Acartia Clausi in 3, Temora longicornis in 2, Calanus finmarchicus and Pseudocalanus elongatus in 1, copepod remains in 1. *November*, Pseudocalanus elongatus in 1, copepod remains in 4. *December*, Pseudocalanus elongatus in 2.

Outer Grounds, 1920–1921, *February*. Centropages typicus in 1. *August*, Calanus finmarchicus in 1. *September*, Sagitta bipunctata in 1, Calanus finmarchicus in 5, Centropages typicus in 1. *October*, Corycæus anglicus in many. *November*, Sagitta bipunctata in 3. *December*, Sagitta bipunctata in 1, Corycæus anglicus in 3.

Although in the above records copepods form the chief food of Sagitta, in other years they have very frequently been seen eating one another and also feeding on newly hatched herrings.*

* In January, 1922, large hauls of Sagitta came in from outside, nearly all of which had eaten other Sagitta, a few having eaten very young herrings.

PHORONIDEA.

Actinotrocha.

Inner Grounds, 1920–1921, *May*. Many small peridinians, Peridinium brevipes, Tintinnus subulatus, Tintinnopsis beroidea, coccoliths in 1. *June*, many Peridinians in 1. *July*, copepod egg, Peridinium sp. in 1, Skeletonema costatum, peridinian indet. in 1, Skeletonema costatum, Prorocentrum micans, Peridinium ovatum, Goniaulax sp. in 1. *August*, many Peridinians indet, in 3. *October*, several small flagellates in 1. *November*, many peridinians and disc-shaped diatoms, including Peridinium ovatum and Coscinodiscus in 1, Peridinium leonis, Tintinnopsis beroidea, Coscinodiscus Grani in 1.

Outer Grounds, 1920–1921, *April*. Peridinians indet. in 1, Peridinians and Coscinodiscus sp. in 1, Coscinodiscus excentricus, Peridinians indet, and Tintinnopsis sp. in 1. *October*, many small Peridinians in 1, Peridinium depressum and other Peridinians indet. in 1, Peridinium depressum, Peridiniopsis asymmetrica, other small Peridinians indet. in 2. Coscinodiscus radiatus, C. excentricus, Thalassiothrix nitzschioides, Pleurosigma sp., Navicula sp., Cerataulina Bergoni, Peridinium conicum, P. brevipes, many small Peridinians indet. in 1, Calanus egg, Peridinians and diatoms indet. in 1, Coscinodiscus Grani, C. excentricus, Thalassiothrix gravida, Peridinians indet. in 1, Coscinodiscus Granii, Peridinium depressum, Peridiniopsis asymmetrica, other small Peridinians indet. in 1. *November*, Larval bivalve, many Coscinodiscus excentricus, Peridinium conicum, many Peridinians indet. in 1, Coscinodiscus Grani, Peridinium ovatum, several small Peridinians indet. in 1, Dictyocha fibula, Navicula sp., Echinospira, Peridinium conicum, many small Peridinians indet. in 1. *December*, Coscinodiscus sp., Thalassiosira sp., Navicula sp., Peridinium conicum, Peridinium indet., Rhizosolenia Shrubsolei stuck in throat in 1, Coscinodiscus excentricus, C. radiatus, Thalassiosira sp., Peridinium depressum, P. indet. in 1.

It is thus seen that Actinotrocha is essentially a Peridinian eater, although diatoms, tintinnids and other unicellular organisms, besides an occasionally larval mollusk or copepod egg, are also taken, all these being swept into the mouth by the currents set up by the cilia. Diatoms and Peridinians of a rounded shape are most frequently eaten, these probably being most easily swept into the mouth. A very great many organisms can be inside at the same time, the alimentary canal usually being full and chiefly with Peridinians. No difference is apparent in those from Inner and Outer Grounds.

POLYZOA.

Cyphonautes.

The various species were not distinguished.

Inner Grounds, 1921, *May*. Coscinodiscus excentricus, Peridinium sp. in 1. *June*, Thalassiothrix Nitzschioides, Biddulphia sinensis, Eucampia zoodiacus, Pleurosigma sp., Goniaulax spinifera in 1, Navicula sp., Thalassiosira Nordenskiöldii, Goniaulax spinifera, Tintinnopsis beroidea in 1, Nitzschia seriata, Thalassiosira Nordenskiöldii in 1, Tintinnopsis beroidea, Navicula sp., Nitzschia seriata, Peridinium sp. in 1, Nitzschia seriata, Peridinium sp. in 1, Licmophora sp., Goniaulax spinifera in 1.

Outer Grounds, 1921, *April*. Coscinodiscus sp. and green cells in 1, Tintinnopsis sp. and green cells in 1. *December*, Coscinodiscus sp., Navicula sp., coccolith in 1.

Although there are very few records these show that the various Cyphonautes are regular diatom feeders, with occasional peridinians and tintinnids.

ANNELIDA.

Tomopteris heligolandicus Greef.

Inner Grounds, 1921, *June*. Fragments of diatoms in 1.

Outer Grounds, 1921, *October*. Indistinguishable cells in brownish débris in 1, green cells (flagellates ?) and brown débris in 1.

These three were the only specimens seen with anything inside, although many empty specimens occurred. The food is apparently minute and mixed with a clear slime.

Larva of Pœcilochætus sp.

Outer Grounds, 1921, *April*. Slime with bits of diatoms and a gastrula larva in 1, slime with small green cells in 1, slime with indistinguishable débris in 1. *October*, brownish débris indistinguishable in 1.

Here again these were the only specimens with anything inside. Many were quite empty.

Larva of Magelona papillicornis Fr. Müller.

Inner Grounds, 1920–1921, *April*. Larval bivalve in 1. *August*, larval bivalve in many (at least 30%, the rest empty). *September*, larval bivalve in 14. *October*, larval bivalve in 3.

Outer Grounds, 1920–1921, *September*. Larval bivalve in 9. *October*, larval bivalve in many. *November*, larval bivalve in 2.

It is striking that the only food ever seen in the larva of Magelona is larval bivalves, and a very large number have been examined. It is evident that this is the natural food, which it probably catches with its two long tentacles whilst swimming about. The tentacles are not ciliated, and the nature of the food may explain this, as the larval bivalves seem to be too big to be drawn in by ciliary currents. It is more likely that the tentacles encircle the mollusks and draw them into the mouth, as is the case with the Polydora, which were seen to devour young gobies (Lebour, 1920).

Larvæ of TEREBELLA sp.

Two kinds of terebellid larvæ are common in the tow-nets, one with a soft gelatinous tube much wider than the worm, the other with a pipe-like stiff hyaline case, wider at the top than the bottom, open at both ends and sometimes plastered with small organisms, diatoms, coccoliths, or with sponge spicules. Most of the records are from those with the hyaline tubes.

Hyaline Tubes.

Inner Grounds, 1917 and 1921, 1921, *February*. Coscinodiscus excentricus in 1. *March* (1917), Thalassiosira sp. (many) in 2, Thalassiosira sp. (many), Navicula sp. in 1. 1921 *April*, Coscinodiscus sp. in 2, Lauderia borealis in 1.

Outer Grounds, 1921, *April*. Coscinodiscus sp. and green remains in 3. *October*, bits of Biddulphia sinensis, coccoliths in 1, bits of diatoms and coccoliths in 4, bits of diatoms, encysted Peridinians in 1, coccoliths, bits of Pleurosigma sp. and Rhizosolenia Shrubsolei in 1. *November*, Paralia sulcata, Rhizosolenia Shrubsolei, coccoliths in 1.

Gelatinous Tube.

Inner Grounds, 1921, *February*. Navicula sp., Nitzschia closterium, coccoliths in 1. *December*, coccoliths and Navicula sp. in 1.

The larval terebellids are evidently predominantly diatom feeders, coccospheres and peridinians also being eaten.

SPIONID larva.

Two specimens only examined, from the Inner Grounds, one in April, 1917, contained Coscinodiscus and Thalassiosira spp., the other in February, 1921, contained Coscinodiscus excentricus.

POLYNOID larva.

Inner Grounds, 1917 and 1921, *February*. Coscinodiscus excentricus in 1. *April*, Prorocentrum micans, Thalassiosira gravida (many), Skeletonema costatum, coccolith in 1, Coscinodiscus Granii, Thalassiosira gravida in 1, Peridinium pallidum in 1.

NEMERTEA.

Pilidium larva.

Most of these were empty, one from Outer Grounds, *October*, contained small peridinians and one large one indet.

CRUSTACEA.

Evadne Nordmanni Lovén.

Inner Grounds, 1921, *April*. Green cells, probably Phæocystis, in 1.

Outer Grounds, 1921, *March*. Phæocystis spores in 1.

Podon intermedius Lillj.

Soft brown remains with no apparent structure in several from Inner Grounds, *August*, 1921.

Cirripede nauplius.

Inner Grounds, 1921, *January*. Green cells, probably flagellates, in several. *February*, Coscinodiscus sp. in 1. *April*, green cells, probably Phæocystis, in 1.

Calanus finmarchicus (Gunn.).

Inner Grounds, 1920–1921, *January*. Green remains containing pieces of diatoms in 4. *February*, green cells in 1. *June*, remains of Phæocystis in 1. *August*, bits of diatoms, Skeletonema in 1. *November*, Thalassiosira sp. in 1, coccoliths, bits of diatoms in 1.

Outer Grounds, 1921, *April*. Many Thalassiosira sp. in 3, many Thalassiosira and Coscinodiscus sp. in 1, Phæocystis in many, Phæocystis and Peridinium pellucidum in 1, Phæocystis and Thalassiosira in 1, Phæocystis, Thalassiosira sp., bits of Chætoceres and other diatoms in 1. *July*, remains of Rhizosolenia (chiefly R. alata) in many, remains of Rhizosolenia sp., coccoliths and black débris in 1, remains of Rhizosolenia alata, Coscinodiscus radiatus, bits of copepods, green cells in 1, remains of Rhizosolenia alata, bits of copepods, bits of green alga, green cells in 2, remains of Rhizosolenia sp., bits of copepods, green cells in 2, green cells and bits of copepods in 1. *August*, bits of diatoms, flagellates, bits of copepods in 1, bits of copepods, flagellates, Pontosphæra Huxleyi in 1, remains of diatoms in 1, several coccospheres, flagellates, bits of copepods in 1, coccoliths, bits of diatoms, bits of copepods in 1. *October*, many Thalassiosira sp. in many. Many Thalassiosira sp., Coscinodiscus radiatus, bits of Rhizosolenia sp. in 2, many Thalassiosira sp., Coscinodiscus radiatus, Rhizosolenia Shrubsolei, coccolith in 1, many Thalas-

siosira sp., Coscinodiscus radiatus, Dytilium Brightwelli, bits of copepods in 1. *November*, many Paralia sulcata and Thalassiosira sp. in 2, Thalassiosira sp. in 1, Paralia sulcata, Thalassiosira sp., Biddulphia sinensis in 1. *December*, remains of Paracalanus parvin, Paralia sulcata, Coscinodiscus sp. in 1, many Thalassiosira sp. and Coscinodiscus sp. in 1.

Thus in the spring when Phæocystis abounds it serves as food for Calanus, although diatoms are also taken; in two samples nearly all the many Calanus were feeding on green cells, one lot certainly Calanus, the other almost certainly so, also others examined singly contained Phæcocystis. Diatoms form the food of Calanus to a very great extent; in one sample many were feeding on Rhizosolenia alata and Shrubsolei at the time when these were very abundant. In the autumn Thalassiosira was the commonest food. Sometimes bits of copepods were found inside mixed with the other débris. The food from inside and outside was not essentially different. It has been described by Esterley (1916) how copepods eat the minute food, rolling it up in a ball, and they certainly crush the hard shells, such as the diatom shells, very few of which come through whole, except very small valves such as Thalassiosira. Diatoms may be said to be the chief food of Calanus, flagellates probably coming very near; but as they are much more quickly digested it is difficult to identify them and estimate their numbers. In the plunger jar organic débris from the bottom seemed to be the main food of *Calanus*.

Pseudocalanus elongatus Boeck.

Inner Grounds, 1921, *January*. Green remains and bits of diatoms in 4, Coscinodiscus sp. in 1, Coscinodiscus sp. and Paralia sulcata in 4, green remains, Navicula sp., bits of diatoms in 1. *February*, remains of diatoms in 2, green remains and bits of diatoms in 4, Coscinodiscus sp. in 1, Navicula sp. in 1.

Outer Grounds, 1921, *November*. Paralia sulcata, Thalassiosira sp., coccoliths in 1, many Paralia sulcata in 1, coccoliths, bits of diatoms in 1.

These few records show that Pseudocalanus is essentially a diatom feeder.

Paracalanus parvus (Claus).

Inner Grounds, 1921, *November*. Thalassiosira sp. in 4, Thalassiosira sp., Navicula sp. in 1, Thalassiosira sp. and coccoliths in 1, bits of Chætoceros in 1, bits of Chætoceros and diatoms indet. in 1, bits of Coscinodiscus sp. and Rhizosolenia Shrubsolei in 1, bits of diatoms indet. in 4, Thalassiosira sp. and Paralia sulcata in 1. *December*, Thalassiosira sp. in 1.

Paracalanus was only examined in the late autumn and Thalassiosira was its chief food. It seems to feed on much the same as Pseudocalanus.

ACARTIA CLAUSI Giesbrecht.

Inner Grounds, 1921, *January*. Green remains and bits of diatoms in 5. *February*, bits of diatoms in 1. *April*, green remains, probably Phæocystis, in many.

OITHONA SIMILIS Claus.

Inner Grounds, 1921, *January*. Green remains and bits of diatoms.

TEMORA LONGICORNIS (O. F. Müller).

Inner Grounds, 1921, *January*. Remains of Crustacea, probably copepods, in 1. *February*, green cells in 1, Navicula sp., Nitzschia sp., bits of Coscinodiscus excentricus in 1. *April*, green cells, Peridinium sp. in 2, bits of copepods in 2. *May*, remains of diatoms, Paralia sulcata in 1, bits of diatoms, Coscinodiscus sp., bits of diatoms, Navicula sp. in 2, green remains, bits of diatoms, coccoliths in 1.

Outer Grounds, 1921, *March*. Green cells, probably Phæocystis, in 1. *April*, green remains in 1.

Temora is rather a miscellaneous feeder, eating more copepods than Calanus, but also feeding on diatoms and flagellates.

CENTROPAGES TYPICUS Kröyer.

Inner Grounds, 1921, *February*. Thread-like green alga in 1, remains of copepod nauplius, bits of larval mollusk in 1, remains of copepods in 2, remains of copepods and green cells in 1, bits of mollusk shell in 1. *May*, brownish remains, indistinguishable in many, bits of copepods in 1, Paralia sulcata, other diatoms in 1, bits of diatoms, Prorocentrum micans, coccoliths in 1, bits of diatoms, Coscinodiscus radiatus, Tintinnus subulatus (3), coccoliths in 1, bits of diatoms in 1, Navicula sp., Thalassiothrix Nitzschioides, Ceratium tripos, Phæocystis spores in 1.

Outer Grounds, 1921, *March*. Green cells and Thalassiosira gravida in 1. *April*, green cells, probably Phæocystis, in 1, many Thalassiosira in 1, Peridinians indet., green cells in 1. *December*, bits of Coscinodiscus sp. and Paralia sulcata in 1, spores of alga, bits of diatoms in 1.

Again several copepods were eaten, besides flagellates and peridinians, but mostly diatoms.

CORYCÆUS ANGLICUS Lubbock.

Inner Grounds, 1921, *February*. Peridinium sp. (cf. depressum), green remains and bits of diatoms in 1.

Outer Grounds, 1921, *March*. Phæocystis in several. *October*, Navicula sp., small flagellate in 1, Navicula sp., coccoliths in 1, bits of larval mollusks (?), coccoliths in 1, coccoliths and remains of flagellates in 1. *November*, green flagellates in 1. *December*, Navicula sp. in 1.

These are too few records to be of much interest, but flagellates, including coccospheres, seem to be an important food.

Labidocera Wollastoni Lubbock.

Inner Grounds, 1921, *May*. Bits of copepods in 1.

Outer Grounds, 1921, *April*. Green remains. *July*, bits of copepods and diatoms.

Anomalocera Pattersoni Templeton.

Outer Grounds, 1921, *March*. Phæocystis in 2. *July*, Crustacea remains, flagellates (?) in 1, many green cells (flagellates ?) in 2, copepod remains and green cells in 1. *October*, many bits of Harpacticid copepods, probably Euterpina acutifrous, Rhizosolenia Shrubsolei in 1, remains of copepods in 2.

Euterpina acutifrous (Dana).

Outer Grounds, 1921, *November*. Green fluid in several (nothing solid). *December*, bits of copepods, green fluid in 1.

In looking at the food of these copepods, we find some that are certainly typically diatom feeders, such as Pseudocalanus, Paracalanus and Acartia, also Calanus, Centropages and Temora, although they also occasionally eat copepods. On the other hand, although only very few have been examined, Anomalocera and Labidocera seem to eat more copepods; these are probably typically Crustacean feeders, large masses of copepod remains having been found in most of those examined. Harpacticids are known to feed on dead organic matter. Flagellates form a large part of the food of many of the copepods, especially Phæocystis, when it is present in quantities every spring, and from the number of coccoliths found coccosphæres must be eaten largely. Peridinians also form part of the food.

It is striking that disc-shaped diatoms are much eaten by the copepods, Thalassiosira and Coscinodiscus specially. Masses of the siliceous skeletons of these come away from their devourers, either broken or whole valves. Thalassiosira usually comes through in whole valves massed together tightly. It is possible that these are more easily manipulated than long or spiny diatoms such as Biddulphia or Rhizosolenia, which are only occasionally found inside the copepods, although quite as abundant as the others, or more so, in the same hauls. Rhizosolenia was, however, in July, when very abundant, found to be in a large number of Calanus.

Zoëa of CARCINUS MÆNAS (Pennant).

One from the *Inner Grounds*, *February*, 1921, contained masses of broken Coscinodiscus and Skeletonema.

Zoëa of EBALIA sp.

Outer Grounds, 1921, *November*. Crushed diatoms, chiefly Coscinodiscus, in 1, Paralia sulcata, Rhizosoleni Shrubsolei in 1.

CRAB ZOËÆ indet.

Inner Grounds, 1920–1921, *February*. Green cells and bits of diatoms in 1, green remains in 1. *April*, bits of Crustacea in 1. *June*, Rhizosolenia hebetata f. semisperia, Phæocystis, burst egg (?) capsules in 1, Rhizosolenia hebetata f. semisperia in 1, Phæocystis in 1. *December*, many Coscinodiscus in 1.

Outer Grounds, 1921, *October*. Peridinium ovatum, bits of Coscinodiscus in 1.

In these few records diatoms are shown to be the chief food of the crab zoëæ, and more recent records in 1922 agree with this. Coscinodiscus is a great favourite.

CRAB MEGALOPA.

Inner Grounds, 1920, *June*. Chewed decapod larvæ.

PORCELLANA LARVA.

Inner Grounds, 1920–1921, *June*. Green filamentous alga in 3, green spores, probably Phæocystis, in 1, siliceous fragments, probably diatoms, in 1. *July*, coccoliths, bits of diatoms and Peridinians in 1, bits of diatoms and echinoderm larvæ in 1, bits of diatoms (small Naviculoid), bits of Peridinians in 1, bits of Foraminifera, Rhizosolenia and other diatoms in 1, coccoliths, bits of diatoms in 1, Paralia sulcata, bits of other diatoms, coccoliths in 1.

Rather miscellaneous feeders, although diatoms form a large part of the food.

GALATHEA LARVA.

Inner Grounds, 1921, *February*. Coscinodiscus excentricus, green cells in 1, green remains in 1. *March*, bits of larval mollusks in 1. *June*, Calcareous fragments in 1. *November*, masses of bits of Coscinodiscus and Rhizosolenia in 1.

Young GALATHEA.

Outer Grounds, 1921, *July*. Bits of mollusks, Crustacea and diatoms in 1, mud, bits of diatoms, coccoliths in 1.

Homarus vulgaris Milne-Edwards. Young Lobster.

Inner Grounds, 1921, *May*. Remains of larval decapods and copepods in 1. *July*, remains of larval decapods in 1.

Gebia larva.

Inner Grounds, 1921, *June*. Pleurosigma, Asterionella japonica, calcareous fragments, probably larval mollusks, in 6. *December*, many bits of Coscinodiscus in 1.

Pandalus larva (including Nika).

Inner Grounds, 1921, *June*. Calcareous fragments, probably mollusks, in 2, bits of mollusks (?), coccoliths, spines of larval echinoderms in 1.

Outer Grounds, 1921, *November*. Coccoliths, bits of mollusks, Coscinodiscus in 1, coccoliths, bits of mollusks, echinoderm larva spines in 1, Coscinodiscus radiatus, bits of diatoms indet. in 1. *December*, coccoliths in 1.

A Nika larva in the plunger jar ate the débris at the bottom, which consisted of diatoms and much dead organic matter.

Axius larva.

Inner Grounds, 1921, *June*. Bits of diatoms, Nitzschia delicatissima in 1. *July*, bits of diatoms, Navicula sp. in 1. *August*, coccoliths, bits of echinoderm larva spines, diatoms indet.

Eupagurus larva.

Inner Grounds, 1921, *June*. Coccoliths, sand grains, in 1.

Larva of Crangon vulgaris (L.).

Outer Grounds, 1921, *October*. Bits of mollusks, diatoms and coccoliths in 1. *November*, coccoliths, bits of copepods, sand in 1. *December*, Navicula sp., spines of Chætoceros in 1, Paralia, Coscinodiscus in 1, many Paralia in 1, coccoliths and débris indet. in 1, many Nitzschia in 1, Navicula, Pleurosigma in 1.

Chiefly a diatom feeder.

Larva of Ægeon trispinosus (Hailstone).

Inner Grounds, 1921, *July*. Bits of mollusks (?) diatoms, Licmophora sp., bits of Ceratium sp. in 1. *August*, calcareous and siliceous particles, bits of Echinoderm larva spines, diatoms in 1.

Calocaris larva.

Inner Grounds, 1921, *August*. Bits of diatoms in 1.

Outer Grounds, 1921, *August*. Bits of Peridinians and diatoms in 1, Exuviella perforata, coccoliths, bits of Rhizosolenia setigera in 1, Exuviella perforata, bits of diatoms, coccoliths in 1, bits of diatoms in 1, Paralia sulcata, bits of other diatoms in 1, coccoliths, bits of diatoms in 1.

Chiefly diatoms, Peridians and Coccospheres.

Euphausiid larva.

Outer Grounds, 1921, *October*. Bits of mollusks, diatoms and coccoliths in 1, bits of mollusks and coccoliths in 1, coccoliths, bits of Coscinodiscus, Navicula and Paralia in 1. *November*, fine débris with coccoliths in 1.

Nyctipanes Couchii T. Bell. juv.

Outer Grounds, 1921, *October*. Bits of green weed, bits of mollusks and diatoms in 1.

Hyperia sp.

Outer Grounds, 1921, *July*. Copepod remains in 1.

Caprella sp. juv.

Inner Grounds, 1921, *July*. Coccoliths, bits of diatoms and larval mollusks in 1.

These food records for the larval and young Crustacea are too fragmentary to be of much value, but from them we find that diatoms are largely eaten together with other unicellular plankton, and that larval mollusks and echinoderms are also taken. All are crushed up and the hard fragments are found among a greenish brown débris. Except in the case of the lobster and crab megalopa, it is very unusual to find any trace of Crustacea inside these larvæ.

MOLLUSCA.

Echinospira (larva of Lamellaria).

Inner Grounds, 1921, *February*. Coccolith, Navicula sp., bits of diatoms, green cells in 1. *December*, bits of diatoms in 1, bits of diatoms, Surrirella in 1, Navicula, coccoliths in 1.

Outer Grounds, 1921, *November*. Pleurosigma, Coscinodiscus, Thalassiosira in 1. *December*, many small Navicula in 1, Navicula, Pleurosigma, coccoliths in 1.

LARVAL GASTROPOD indet.

Outer Grounds, 1921, *November*. Remains of diatoms, Navicula in 1, Paralia sulcata, Coscinodiscus excentricus, Thalassiosira sp., Navicula sp. in 1, Thalassiosira sp. in 1, much Paralia sulcata in 1.

LIMACINA RETROVERSA auth. (?)

Outer Grounds, 1921, *October*. Paralia sulcata and other diatom remains in 1.

LARVAL BIVALVE indet.

Outer Grounds, 1921, *December*. Navicula, Coscinodiscus, Paralia in 1, Navicula, flagellates in 1, Thalassiosira in 1, coccoliths, Surrirella in 1, Navicula sp. in 1, Coscinodiscus in 1, Paralia in 1.

These few records show that all these pelagic mollusk larvæ and one Pteropod are pre-eminently diatom feeders. Larval mollusks seem to be eaten by various decapod larvæ.

ECHINODERMATA.

OPHIOPLUTEUS.

Inner Grounds, 1921, *August*. Thalassiothrix nitzschioides in 1, small green flagellates in 3, small peridinians indet. in 1.

Outer Grounds, 1921, *July*. Skeletonema costatum in 1.

BIPINNARIA.

Inner Grounds, 1921, *August*. Chætoceros curvisetus in 1.

ECHINOPLUTEUS of ECHINUS MILIARIS L.

Outer Grounds, 1921, *October*. Coccoliths, bits of diatoms, Thalassiosira, Navicula sp. in 1.

By far the greater portion of the echinoderm larvæ examined were empty, and it was very difficult to distinguish food inside them. The few records show, as is already known, that they are diatom feeders, with small flagellates and Peridinians. Echinoderm larvæ are themselves eaten by several of the larval Crustacea.

ENTEROPNEUSTA.

TORNARIA LARVA.

Nearly all the Tornaria larvæ examined were empty, but two contained a faint slime with diatoms.

Outer Grounds, 1921, *October*. Thalassiosira Nordenskiöldii, Navicula (very small) in 1. *November*, Rhizosolenia Stolterfothii in 1.

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On the Food of Young Plaice (*Pleuronectes platessa*).

By

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(*Work from the Lancashire Sea Fisheries Laboratory, Piel, Barrow-in-Furness.*)

THE investigation of the food contents in the stomachs of young fishes was included in the scheme of scientific investigations drawn up and initiated by Professor Herdman for the Lancashire Sea Fisheries Committee nearly thirty years ago. The lengthy series of Annual Reports contain here and there accounts of the observations made on the stomach contents of various Pleuronectidæ captured close inshore, and the pelagic stages of other fishes caught from time to time in the plankton tow-nets. No systematic attempt has, however, been made, in connection with the investigation of the Irish Sea, to determine the food of any particular species of fish during the early part of its life history.

Other observers working in other areas, notably Dr. Marie Lebour at Plymouth, have added very much to our knowledge of the early food of young fishes. Dr. Lebour's reports, published in the *Journal of the Marine Biological Association*, Vols. XI and XII, deal with a very large number of larval and post-larval stages of the more important food fishes caught in the tow-nets and young fish-trawl in Plymouth Sound and beyond.

The present report gives an account of the food contained in the stomachs of young plaice (*Pleuronectes platessa*, Linn.) from a few days after hatching to about five months old. The samples examined in April and May were taken from the spawning pond at Port Erin, Isle of Man, where they had hatched from the pelagic eggs spawned by the adult plaice early in 1921. The later stages examined during May to August represented young plaice hatched in the open sea about the same time as those in the pond, and which had made their way close inshore. They were caught by wading with a push-net lined with mosquito netting. These inshore plaice were caught by my colleague, Mr. W. Birtwistle, on the North Wales, Cheshire, and Isle of Man coasts, and varied from 13 mm. to 87 mm. in length. The fish were preserved in weak formalin, and the food contents were in every case ascertained by dissecting out the alimentary canal with the help of the large Leitz dissecting microscope. The contents of the alimentary canal were well preserved, and there was no difficulty in determining them.

The smallest fish with food taken externally was 8½ mm. in length. The stomachs of any fish less than that size contained only unconsumed yolk. The earliest food of the plaice from the spawning pond proved to be the spores of algæ. These were found in the stomachs of 80% of one sample where external food had been consumed, but none were present in any of the stomachs of young fish over 10 mm. in length. The spores of algæ when newly set free are very motile, and quickly make their way to where the brightest light is. Wherever there are algæ growing at the sea bottom it is pretty certain that, in the spring at least, the surface water, where the young fish are usually found, will be richly supplied with spores. There will be no shortage of food, and the post-larval stages of fishes will not have much difficulty in capturing the spores. The glass aquaria at Piel are very difficult to keep clean in the spring and early summer, owing to the growth of algæ. The water is filtered through duffel bags, and small growing plants of *Ulva*, etc., are kept in the aquaria to provide food for Nudibranchs. From time to time conspicuous clouds of pale green spores are to be seen on the best lighted side of the aquaria. Examined under the microscope they appear as small, oval, almost colourless, and very motile bodies, and are identical with the oval organisms found in the stomachs of the smaller sizes of post-larval plaice from the spawning pond. The feeding on spores of algæ is quickly followed by the capture of Copepoda, consisting of small Harpacticoida, such as *Tigriopus fulvus*, *Mesochra pygmæa*, *Idya gracilis*, etc., with an occasional Copepod and Barnacle nauplius and *Podon*. Although no spores were found in post-larval plaice over 10 mm. in length, there does not appear to be any distinct demarcation in size when the little plaice begin to feed entirely on Harpacticoida. In a sample of 8½ mm. 24% had fed on spores only, and 8% on Harpacticoida. At 9½ mm. 80% had fed on spores and 20% on Harpacticoida. At 10 mm. 76% of the stomachs contained spores only, and 24% had Harpacticoida. No stomach contents of any of the fish examined consisted of a mixture of spores and Harpacticoida. The smallest Harpacticoid found in the stomach of a young plaice 8½ mm. long seemed almost too large and solid for such a small fish to swallow. Dr. Lebour states, " fish caught in the act of swallowing Copepoda always show the tail sticking out of the mouth, so that they are swallowed head first." In some cases they apparently pass rather rapidly through the alimentary system and reach the rectum almost undigested. In these instances the tail setæ of the Copepod were always projecting through the anus to the exterior. In fact, the Copepod seemed so perfect and apparently undigested that one would almost be justified in concluding that it had entered through the anus and not through the mouth ! No diatoms were found in any of the stomachs of the larval and post-larval stages sent to me for examination. While none of my material had fed

on diatoms, it is quite clear that they are present in the pond, at times, in sufficient numbers to form a part of the food supply of the young plaice. Professor Herdman, who examined living fish taken from the pond on April 8th, found diatoms present in the stomachs along with algal spores and exoskeletons of Copepoda. None of the stomachs of the young plaice from the pond contained any trace of Polychæta. Although none of my samples of larvæ suggested that the capture of external food began before the yolk-sac was absorbed, Professor Herdman, from his examination of living and very recently preserved material on the spot, is satisfied that the plaice larvæ begin to feed by taking in solid food through the mouth long before the yolk-sac is exhausted.

The larval and post-larval plaice from the spawning pond at Port Erin were $5\frac{1}{2}$ mm. to $11\frac{1}{2}$ mm. in length. The young plaice caught close inshore with the push-net ranged from 13 mm. to 87 mm. Although the difference between the largest pond example and the smallest inshore specimen only amounted to $1\frac{1}{2}$ mm. in length, there is probably a material difference in their ages. Older examples could have been obtained from the pond by prolonging the collecting. This was not thought desirable, as the shallow-water plaice would be surrounded by a more varied food supply, and there could be no real comparison.

Young plaice hatched naturally in the open sea work their way inshore and are generally found in the very shallow water early in May. They are frequently left behind in the little pools of water on the shore when the tide has gone out and can be caught with a spoon. These plaice are not completely metamorphosed, and are nearly colourless and transparent. All that can be seen of them in a shallow pool are the eyes. When they are disturbed they rise from the sand and swim in an upright position until apparently exhausted, and then settle flat on the sand again. They complete their metamorphoses, increase in size, and finally disappear from the sand and mud flats in August.* The invertebrate fauna of these sand and mud flats is rich and varied. No matter how varied the fauna be, we find that these little plaice select their food from particular groups of the invertebrata. These groups are Worms, Crustacea, Mollusca, and Tunicata. A mixed menu from these groups is not uncommon ; but one finds, however, that they just as frequently limit their diet to one of these groups. It is by no means rare to find them even more than selective to one group. Individuals in a catch are found to have been feeding on one particular species, although it is quite obvious, from the examination of the stomachs of their companions, that species belonging to the same group and also to other groups were present. It does not follow that the species consumed is always abundant in the plankton where the fish

* This applies to the Morecambe Bay region. Further south the metamorphosis and offshore migration occur earlier.

are. One may examine samples of the shallow-water plankton, and even scrapings of the mud, most carefully, and find no trace of the species which sometimes completely fills the stomach and intestines of a young plaice. The late I. C. Thompson many years ago described and figured a new species of Harpacticoid Copepod (*Jonesiella hyænæ*) which was, and is still, far more common in the stomachs of young pleuronectidæ than in a free state. Another point which was noted during the present investigation was the predominance of specimens of the male sex of Cumacea and Amphipoda in the stomachs. The speciographer, whose investigations are confined to the examination of plankton and bottom scrapings, usually finds the female sex is more abundant than the male sex. Sars says the Cumacea, on the whole, are "true bottom forms, though the more agile adult males of some species may be found at times swarming near the surface, especially at night."

In some samples a considerable number of the young plaice dealt with had no trace of food either in the stomach or intestine. A few contained small quantities of sand, probably derived from the broken down tubes of *Pectinaria* after the soft parts of the worm had been digested. The quite empty alimentary systems are difficult to explain, but there must be some solution. It is known that there is a certain amount of regurgitation of the stomach contents by more fully grown fish. Occasionally interesting invertebrata have been obtained in this way from adult plaice recently caught and transferred to our tanks. The stomachs of larval and post-larval stages are easily ruptured and the contents lost. In the metamorphosing and later stages it is probable that the unnatural violence due to capture and sudden immersion in preservative brings on regurgitation of the alimentary system.

The following are the results of the examination of the stomach contents of the various samples examined :—

1. Plaice from Port Erin Spawning Pond.

April 25, 1921.—11 @ 5½ mm., unconsumed yolk. 25 @ 6½ mm., ditto. 7 @ 8½ mm., ditto. 35 @ 9 mm., 31 quite empty, 1 with 1 *Tigriopus*, 1 with 1 Copepod nauplius, 1 with 1 Barnacle nauplius, 1 with 1 *Podon*. 6 @ 9½ mm., 3 quite empty, 2 with 1 *Tigriopus* each, 1 with 1 *Mesochra*.

April 26, 1921.—25 @ 8½ mm., 17 quite empty, 6 with algæ spores, 2 with 1 *Tigriopus* each. 155 @ 9½ mm., 100 quite empty, 45 with algæ spores, 5 with 1 *Mesochra* each, 3 with 1 *Tigriopus* each, 2 with 1 *Laophonte strömi* (male) each. 228 @ 10 mm., 54 empty, 169 with algæ spores, 2 with 1 *Amphiascus* each, 2 with 1 *Tigriopus* each, 1 with 1 Barnacle nauplius.

April 27, 1921.—30 @ 8 mm., all with unconsumed yolk only. 25 @ 9½ mm., 20 with algæ spores, 1 with 3 *Tigriopus*, 1 with 1 *Tigriopus*, 1 with 1 *Idyæa*, 1 with 1 *Amphiascus*,* 1 with 1 *Mesochra* and 1 *Westwoodia*.

A sample of plankton taken from the pond on this date contained a few Medusoid gonophores, a few *Idyæa gracilis*, and 1 Crab zoea.

April 28, 1921.—10 @ 6½ mm., all with unconsumed yolk only. 24 @ 9½ mm., 23 empty, 1 with 1 *Tigriopus*. 28 @ 10 mm., 14 empty, 3 with 1 *Mesochra* each, 1 with 3 *Amphiascus*,† 4 with 1 *Amphiascus*† each, 1 with 3 *Amphiascus*† and 1 *Tigriopus*, 1 with 3 *Amphiascus* and 1 Copepod nauplius, 1 with 1 *Amphiascus*, 1 *Mesochra*, and 1 Copepod nauplius, 1 with 1 *Amphiascus* and 1 Copepod nauplius, 1 with 1 *Westwoodia*, 1 with 1 *Westwoodia*, and 3 Barnacle cypris.

April 30, 1921.—30 @ 8½ mm., 19 empty, 9 with algæ spores, 1 with 3 *Amphiascus*, 1 with 1 *Idyæa*. 17 @ 9½ mm., 14 with algæ spores, 1 with 2 *Amphiascus*, 2 with 1 *Amphiascus* each, in the rectum. 27 @ 10 mm., 17 with algæ spores, 1 with 4 *Idyæa*, 1 with 2 *Idyæa*, 1 with 3 *Idyæa*, and 2 *Amphiascus*, 3 with 1 *Idyæa*, and 2 *Amphiascus* each, 1 with 1 *Idyæa*, 3 *Amphiascus* and 1 *Tigriopus*, 3 with 2 *Tigriopus* each.

May 4, 1921.—21 @ 9½ mm., all empty. 53 @ 10 mm., 17 empty, 27 with algæ spores, 2 with 3 *Tigriopus*, 1 with 1 *Tigriopus*, 1 with 1 *Tigriopus*, 1 *Mesochra* and 1 *Laophonte strömi* (male), 1 with 2 *Idyæa*, 1 with 1 *Idyæa*, 1 with 2 *Idyæa* and 1 *Mesochra*, 1 with 2 *Mesochra*, 1 with 1 Barnacle cypris. 4 @ 11 mm., all empty.

May 6, 1921.—20 @ 9½ mm., 3 empty, 14 with algæ spores, 1 with 3 *Amphiascus* and 1 *Laophonte strömi* (male), 2 with 1 *Amphiascus* each. 21 @ 10 mm., 16 with algæ spores, 1 with 8 *Mesochra*, 2 with 1 *Mesochra* each, 1 with 2 *Mesochra* and 3 *Idyæa*, 1 with 5 *Idyæa* and 1 *Tigriopus*. 39 @ 11–11½ mm., 1 with 10 *Idyæa*, 1 with 15 *Idyæa*, and 1 *Amphiascus*, 1 with 12 *Idyæa* and 1 *Amphiascus*, 3 with 3 *Idyæa* each, 2 with 1 *Idyæa* each, 2 with 1 *Idyæa* and 1 *Amphiascus* each, 1 with 1 *Westwoodia* and 1 *Amphiascus*, 28 with semi-digested Copepods (*Idyæa* and *Amphiascus*) :—

2. Plaice Caught with Push-net Lined with Mosquito Netting.

May 6, 1921, Douglas Bay, Isle of Man.—26 @ 15–25 mm., 12 filled with many young *Pectinaria*, 1 with many *Pectinaria* and 10 *Harpacticus*, 2 with many *Pectinaria* and 4 *Harpacticus* each, 1 with few *Pectinaria* and 18 *Harpacticus*, 1 with few *Pectinaria* and 14 *Harpacticus*, 1 with few

* This copepod was in the rectum with its tail seta projecting through the anus and quite undigested.

† In rectum as before and undigested.

Pectinaria and 3 *Harpacticus*, 3 with few *Pectinaria* and 1 *Harpacticus* each, 1 with 3 *Pectinaria* and 1 *Harpacticus*, 1 with 3 *Pectinaria*, 2 *Harpacticus* and 1 *Laophonte curticauda*, 1 with 8 *Pectinaria*, 3 *Harpacticus* and 1 *Longipedia*, 2 with 3 *Harpacticus* each. 15 @ 26–35 mm., 1 empty, 14 with many young *Pectinaria*. 24 @ 26–41 mm., 1 empty, 19 with many young *Pectinaria*, 2 with many *Pectinaria* and 1 *Bathyporeia* (male) each, 1 with many young *Pectinaria* and 1 *Cuma* (male), 1 with many *Pectinaria* and 1 *Pseudocuma* (male). 12 @ 42–46 mm., 8 with many young *Pectinaria*, 2 with many *Pectinaria* and 1 *Bathyporeia* (male) each, 1 with many young *Pectinaria*, 1 *Bathyporeia* (male) and 6 *Harpacticus*, 1 with 1 *Perioculodes*.

May 12, 1921, New Brighton Shore.—Only samples of food from stomachs sent for examination. These were filled with *Temora*.

May 19, 1921, Hoylake Shore.—1 @ 21 mm., filled with *Temora*, but also contained 1 *Calanus*, 1 *Centropages*, and 2 *Oithona*. 52 @ 16–17 mm., 3 filled with young *Pectinaria* only, 48 filled with *Temora*, and all contained one or two *Calanus*, *Centropages* and *Oithona*, 1 with 17 *Temora*, 1 *Calanus*, 1 *Centropages*, 2 *Oithona* and 1 Barnacle cypris.

May 20, 1921, New Brighton Shore.—71 @ 15–18 mm., 34 with few young *Pectinaria*, many *Temora*, 1 or 2 *Calanus*, *Pseudocalanus* and *Centropages* (1 stomach contained 1 young *Pectinaria*, 30 *Temora*, 4 *Calanus*, 3 *Pseudocalanus* and 2 *Centropages*). 37 with many *Temora*, 1 or 2 *Calanus*, *Pseudocalanus* and *Centropages*. 4 @ 21 mm., 1 with 1 young *Pectinaria*, 30 *Temora*, 1 *Calanus*, 1 *Cythere*, 1 Barnacle nauplius and 1 Barnacle cypris. 1 with 1 young *Pectinaria*, 2 *Temora*, 2 *Calanus* and 1 Barnacle cypris. 1 with many *Temora*, and 1 *Pseudocalanus*, 1 with 5 *Temora* and 1 *Calanus*. 1 @ 24 mm., 90 *Temora* and 1 Barnacle cypris. 1 @ 30 mm., 182 *Temora* and 2 *Pseudocalanus*.

May 21, 1921. From Weir at Mouth of Ogwen River, near Bangor, N. Wales.—34 @ 20–36 mm., 13 empty, 6 filled with young *Pectinaria*, 2 with few young *Pectinaria*, large number of *Laophonte curticauda* and 2 *Temora*, 2 with few young *Pectinaria* and a large number of *Laophonte curticauda*, 1 with few young *Pectinaria*, 3 *Temora* and 2 *Laophonte*, 1 with few young *Pectinaria*, 2 *Temora*, 8 *Laophonte* and 1 *Thalestris*, 1 with 3 young *Pectinaria*, 2 *Temora* and a large number of *Laophonte*, 1 with 1 young *Pectinaria*, 4 *Laophonte* and 1 Barnacle cypris, 5 with 2 young *Pectinaria* and 1 Barnacle cypris, 1 with few young *Pectinaria*, 2 *Temora*, many *Laophonte*, 3 *Corophium*, and 3 Barnacle cypris, 1 with 3 *Laophonte*, 1 *Thalestris* and 1 *Corophium*.

May 29, 1921, Port Erin Bay, I.O.M.—46 @ 15–32 mm., 11 empty, 1 with few young *Pectinaria* and many *Temora*, 17 with a large number of *Temora*, 1 with many *Temora* and few *Pseudocalanus*, 1 with many

Temora, few *Pseudocalanus* and 1 Barnacle cypris, 2 with many *Temora*, few *Pseudocalanus* and *Oikopleura*, 3 with many *Temora*, few *Pseudocalanus* and many *Harpacticus*, 2 with 8 *Temora* each, 1 with many *Temora*, many *Harpacticus*, 2 *Jonesiella* and 2 *Oikopleura*, 1 with many *Temora*, 26 *Harpacticus* and 2 *Jonesiella*, 1 with 8 *Temora* and 20 *Oikopleura*, 2 with many *Temora* and 1 Crab megalopa each, 1 with 12 Crab megalopa, 1 with 1 young *Mysis*, 1 with many *Pseudocalanus*, few *Harpacticus*, and 1 *Oikopleura*.

June 1, 1921, Derby Haven, I.O.M.—34 @ 17–37 mm., 8 empty, 12 filled with young *Pectinaria*, 3 with young *Pectinaria* and 8 *Pseudocuma* (male) each, 1 with young *Pectinaria* and 12 *Pseudocuma* (male), 2 with young *Pectinaria* and 1 *Pseudocuma* (male), 1 with young *Pectinaria* and 3 *Diastylis* (male), 1 with young *Pectinaria*, 1 *Pseudocuma* and 1 *Bathyporeia*, 2 with young *Pectinaria* and 2 *Bathyporeia* (male) each, 2 with young *Pectinaria* and a few *Harpacticus*, 1 with 5 *Pseudocuma*, 1 *Idotea* and 1 *Bathyporeia* (all males), 1 with a large number of *Harpacticus* (all males).

[June 1, 1921, Derby Haven, I.O.M. Flounders (*P. flesus*).—75 @ 16–30 mm., 9 empty, 8 filled with young *Pectinaria*, 52 filled with young *Pectinaria* and each with a few *Harpacticus*, 3 with young *Pectinaria*, a few *Harpacticus* and 2 *Bathyporeia* (male) each, 2 with young *Pectinaria* and a few *Harpacticus* and *Jonesiella*, 1 with a large number of *Harpacticus*.

June 6, 1921, Douglas Bay, I.O.M. Flounders (*P. flesus*).—16 @ 17–30 mm., 2 empty, 2 with many young *Pectinaria*, 12 with many young *Pectinaria* and a few *Harpacticus*.

June 2–6, 1921, New Brighton Shore.—70 @ 13–28 mm., 6 empty, 23 with young *Pectinaria*, *Pseudocalanus*, *Temora*, *Centropages*, *Eurytemora*, *Acartia discaudata*, *Harpacticus* and Barnacle cypris, 41 with Copepoda only as above.

The presence of *Eurytemora* and *Acartia discaudata* in the stomachs of this sample of young plaice indicate that the fish had been feeding in a brackish water area.

June 8, 1921, Ramsey Bay, I.O.M.—48 @ 16–35 mm., 7 empty, 15 with young *Pectinaria*, *Ectinosoma*, *Jonesiella* and *Harpacticus*, 4 with Copepoda only as above, 18 with *Temora*, *Ectinosoma*, *Jonesiella*, *Harpacticus*, and *Diastylis* (two stomachs had each 8 *Diastylis*), 3 with 1 *Bathyporeia* each, and 1 with 1 *Synchelidium*.

June 24, 1921, Hoylake Shore.—1 @ 32 mm., 1 *Bathyporeia* (male). 1 @ 34 mm., many young *Pectinaria*. 16 @ 38 mm., 6 filled with young *Pectinaria*, 2 with young *Pectinaria* and a few *Temora* and *Ectinosoma*,

1 with young *Pectinaria* and 2 *Jonesiella*, 3 with 2 young *Crangon* each, 2 with 2 young *Crangon* each and a few *Ectinosoma* and *Jonesiella*, 1 with 1 *Bathyporeia* (male), 1 *Ectinosoma*, 1 *Jonesiella* and 1 *Cythere*, 1 with 1 *Bathyporeia* and 1 young *Mytilus*. 15 @ 40 mm., 6 with young *Pectinaria*, 1 with *Pectinaria* and 1 young *Crangon*, 1 with *Pectinaria* and a few *Jonesiella*, 3 with 1 young *Crangon* each, 1 with a large number of *Jonesiella*, 1 with 70 *Jonesiella* and 8 *Ectinosoma*, 1 with 8 *Jonesiella* and 2 *Ectinosoma*, 1 with 2 *Bathyporeia* (male), 1 *Cythere* and 2 young *Mytilus*. 27 @ 42–43 mm., 7 filled with young *Pectinaria*, 3 with young *Pectinaria*, 1 *Bathyporeia* (male) and a few *Jonesiella*, 2 with *Pectinaria* and 1 Crab zoea each, 2 with *Pectinaria* and 1 *Corophium* each, 1 with *Pectinaria*, 1 young *Crangon* and 2 young *Mytilus*, 4 with *Pectinaria*, a few *Ectinosoma* and 2 young *Mytilus* each, 3 with 2 *Bathyporeia* (male) each, 1 with 1 *Bathyporeia* (male), 1 young *Crangon* and 1 Crab zoea, 3 with 1 *Crangon* each, 1 with 1 Crab zoea. 9 @ 45 mm., 4 filled with young *Pectinaria*, 1 with *Pectinaria* and 1 *Crangon*, 2 with 3 Crab zoea each, 1 with 2 Crab zoea, 1 young *Crangon*, and 1 *Synchelidium*, 1 with 1 *Bathyporeia* (male). 3 @ 48 mm., all filled with young *Pectinaria*. 5 @ 50–52 mm., 2 filled with young *Pectinaria*, 3 with *Pectinaria* and 1 Crab zoea each.

August 1, 1921, Port Erin Bay, I.O.M.—140 @ 27–87 mm., 32 quite empty, 54 filled with young *Pectinaria*, 15 with *Pectinaria* and from 1–12 *Eurydice*, 2 with *Pectinaria* and a few *Jonesiella*, 1 with *Pectinaria* and a few *Ectinosoma*, 1 with 1 *Pectinaria* and a few *Acartia clausi*, 4 with *Pectinaria* and a few *Ectinosoma*, *Jonesiella* and *Harpacticus*, 1 with *Pectinaria*, 1 *Bathyporeia* (male) and a few *Jonesiella*, 3 with *Pectinaria* and 1 Crab megalopa each, 1 with 1 *Bathyporeia* (male), 1 with 1 *Bathyporeia* and 1 *Diastylis* (males), 2 with 1 *Haustorius* each, 2 with 1 *Schistomysis* each, 1 with 1 *Schistomysis* and 1 *Eurydice*, 1 with 1 *Crangon*, 1 with 2 Crab megalopa, 1 with 2 *Diastylis* (male), 1 with 3 *Diastylis* and a few *Jonesiella*, 1 filled with *Temora*, 1 with many *Ectinosoma*, 4 with *Ectinosoma* and a few *Jonesiella* and *Asellopsis*, 2 with *Ectinosoma* and a few *Jonesiella*, *Asellopsis*, and *Harpacticus*, 4 with *Ectinosoma* and a few *Jonesiella*, 2 with *Ectinosoma* and a few *Harpacticus*, 1 with a large number of *Laophonte curticauda*, 1 with a large number of *Asellopsis* (many males).

Analyses of the Results.

1. Plaice Hatched in the Spawning Pond at Port Erin.

Total number examined	896
Deduct larvæ with unconsumed yolk . . .	83
	813

Food.		No. of Stomachs.	% of Stomachs.
Empty		306	37·64
Algal spores		337	41·45
Copepoda.	*Amphiascus minutus* Claus	57	7·00
	Idyæa gracilis T. Scott	52	6·39
	Tigriopus fulvus Fischer	23	2·81
	Mesochra pygmæa G. O. Sars.	18	2·21
	Laophonte strömi Baird	4	·49
	Westwoodia minuta Claus	4	·49
	"Nauplii"	4	·49
Chadocera.	*Podon intermedium* Lillj.	1	·12
Cirripedia.	"Nauplius"	2	·24
	"Cypris"	2	·24

2. Inshore Plaice Caught with Push-net.

Total number examined		656	
Empty		79	12·04
Vermes.			
	Pectinaria sp.	327	49·84
Crustacea.			
Brachyura,	"Zoea"	10	1·52
	"Megalopa"	7	1·06
Macrura,	*Crangon vulgaris* (Linn.)	17	2·59
Schizopoda,	*Macromysis flexuosa* (Mull.)	1	·15
	Schistomysis ornata (Sars.)	3	·45
Cumacea,	*Cuma scorpoides* (Mont.)	1	·15
	Diastylis rathkei (Kr.)	22	3·35
	Pseudocuma cercaria (V. Ben.)	10	1·52
Isopoda,	*Eurydice pulchra* Leach	16	2·43
	Idotea pelagica Leach	1	·15
Amphipoda,	*Bathyporeia pelagica* Bate	27	4·11
	Haustorius arenarius (Slabber)	2	·30
	Synchelidium haplocheles (Grube.)	2	·30
	Perioculodes longimanus (B. & W.)	1	·15
	Corophium bonellii H. M. Edw.	4	·60
Ostracoda,	*Cythere pellucida* Baird	3	·45

	Food.	No. of Stomachs.	% of Stomachs.
Copepoda (Calanoida),	*Calanus septentrionalis* (H. Goodsir)	124	18·90
	Pseudocalanus elongatus, Boeck.	145	22·10
	Centropages hamatus (Lillj.)	185	28·20
	Temora longicornis (O.F.M.)	249	37·95
	Eurytemora affinis (Poppe)	64	9·75
	Acartia discaudata (Giesb.)	64	9·75
	,, *clausi* Giesb.	1	·15
Copepoda (Cyclopoida),			
	Oithona similis Claus	50	7·62
Copepoda (Harpacticoida),			
	Longipedia coronata Claus	1	·15
	Ectinosoma sarsi Boeck.	66	10·06
	Thompsonula (*Jonesiella*) *hyænæ* (I. C. Thomp.)	67	10·21
	Harpacticus flexus Brady.	133	20·27
	Thalestris longimana Claus	2	·30
	Laophonte curticauda Boeck.	12	1·82
	Asellopsis hispida Brady.	7	1·06
Cirripedia,	"Nauplius"	1	·15
	"Cypris"	35	5·33
Mollusca.			
	Mytilus edulis Linn.	7	1·06
Tunicata.			
	Oikopleura flabellum J. Mull.	5	·75

During the first five months of its life, the young plaice feeds almost entirely on the pelagic and semi-pelagic invertebrate living in the water through which it moves. The areas frequented by the young fish in the summer months contains a very mixed fauna. A tow-netting taken a few yards out from the shore and in not more than two feet of water usually contains truly pelagic forms, such as *Calanus* and *Temora*, along with the bottom living Cumacea, Amphipoda, etc. The young *Pectinaria* would very likely be captured just as they were settling into the sand at the conclusion of their pelagic life.

The results of the examination of two samples of young flounders are included to show that their food is much the same as that of the young plaice of about the same age.

On the Young Stages of *Blennius ocellaris* L., *Blennius pholis* L., and *Blennius gattorugine* L.

By

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With Figures 1–12 in the Text.

THE post-larval blennies taken off Plymouth during recent years have been assigned to three species, namely, *Blennius ocellaris* L., *Bl. pholis* L., and *Bl. gattorugine* L. The process of identification, however, has emphasised the need for the revision of the available descriptions and illustrations of the various stages in development, and I have, therefore, thought it desirable to report on the Plymouth forms and give drawings of them.

BLENNIUS OCELLARIS (12 rays to the pectorals).

Ehrenbaum (2, p. 83) has copied two figures of the young of this species which were originally produced by Holt (5, p. 45), and it is somewhat unfortunate that both need to be commented upon. The first is satisfactory as a representation of the appearance of a larva some time after hatching, but the length is wrongly given as 6·3 mm. Garstang (4, p. 74) explained that this was an error in transcription, and, after a re-examination of Holt's specimens, he found that the total length was 4·6 mm. (varying between 4·55 and 4·65). The second figure, depicting a young fish of 18 mm., which Holt tentatively assigned to *Bl. ocellaris*, was taken exception to by Garstang and the identification questioned, and it must be admitted that it does not portray satisfactorily the characters of the species.

The full account of the rearing of the young from the egg by Garstang (4) was unfortunately not illustrated ; but as several of his original specimens are in the Plymouth collection, I have been able to identify quite readily the more recently captured examples and have made a series of drawings which are given below (Figs. 1 to 4).

BLENNIUS PHOLIS (13 rays to the pectorals).

The Plymouth post-larvæ of corresponding length agree with the description and figure of a specimen of 15·5 mm. taken off Falmouth, which was given by Holt (5, p. 47) and copied by Ehrenbaum (2, p. 81).

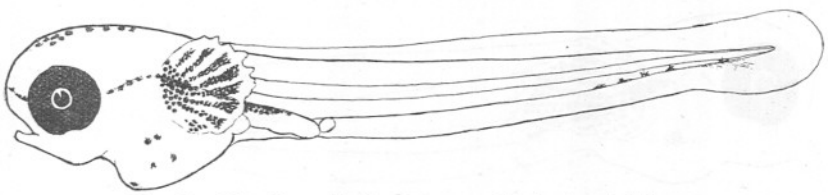

FIG. 1.—*Blennius ocellaris*. 4·6 mm. Newly hatched larva.

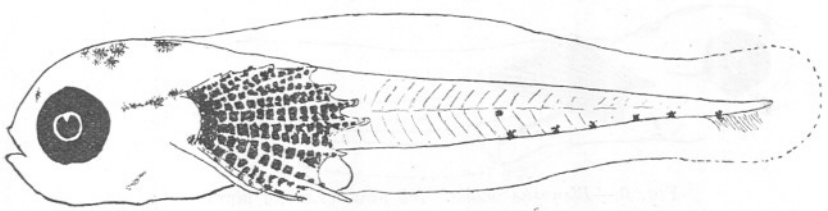

FIG. 2.—*Blennius ocellaris*. 5·0 mm. Y.F.T. 1920.

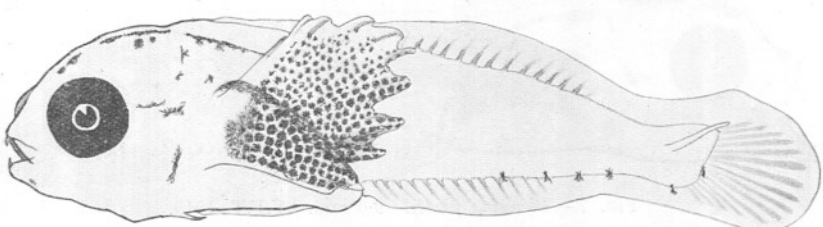

FIG. 3.—*Blennius ocellaris*. 8·5 mm. Y.F.T. 1919.

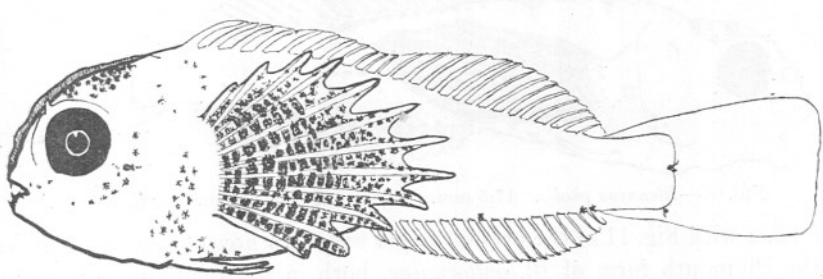

FIG. 4.—*Blennius ocellaris*. 13·25 mm. Y.F.T. 1913. External pigment only.

In the more recent publication of Le Danois (I, p. 161), descriptions and drawings of post-larvæ of 11 mm. and 15 mm. respectively have been presented which cause some difficulty. Le Danois' specimens both exhibit a restricted distribution of pigment on the pectorals, a condition

not shown in the Plymouth forms. Then, again, his two post-larvæ differ greatly from one another in body proportions, whereas the Plymouth examples show no such striking variation. It is really difficult to regard Fig. 313 of Le Danois as representing the post-larva at a similar length of the same species as that figured by Holt, and a comparison of the

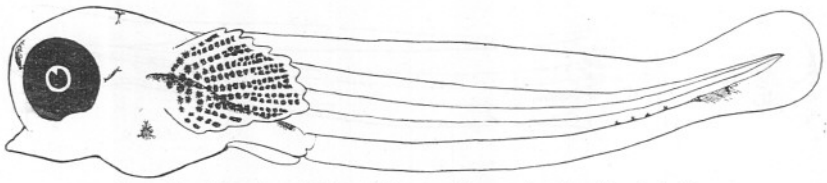

FIG. 5.—*Blennius pholis.* 5·0 mm. Larva shortly after hatching.

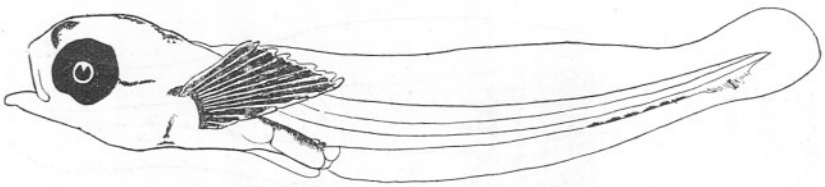

FIG. 6.—*Blennius pholis.* 5·5 mm. End of larval stage.

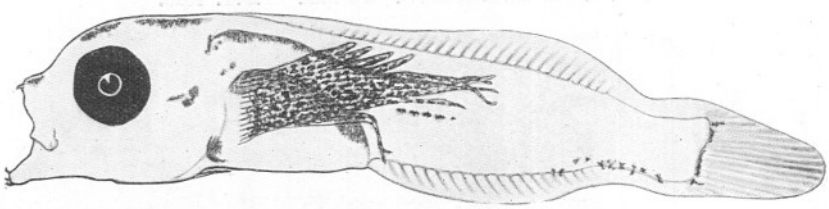

FIG. 7.—*Blennius pholis.* 9·0 mm. Y.F.T. 1919.

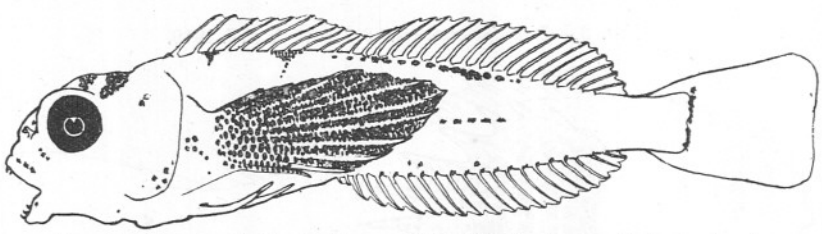

FIG. 8.—*Blennius pholis.* 17·5 mm. Y.F.T. 1914. External pigment only.

former with Fig. 11 of the present paper will show how closely it resembles the Plymouth form of *Bl. gattorugine*, both in the relatively long post-anal part of the body, and in the limited distribution of pigment on the pectorals.

The post-larvæ in general have a body form which is distinct from that of the other species in that it lacks the depth and robustness of *Bl. ocellaris*, and is not greatly attenuated post-anally as in *Bl. gattorugine*. The

pectorals are always deeply pigmented and relatively long. At 9 mm. black pigment is strongly marked along the upper surface of the spinal cord anteriorally, and the commencement of pigment on the exterior surface of the body may be seen along either side on the middle line slightly posterior to the anus (Fig. 7).

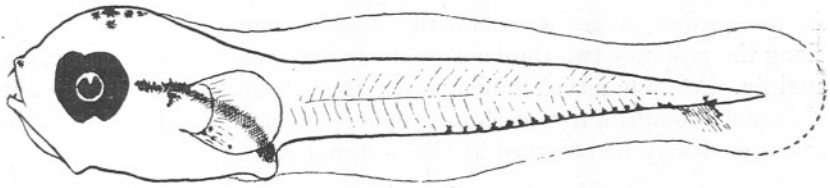

FIG. 9.—*Blennius gattorugine.* 6·0 mm. Y.F.T. 1920.

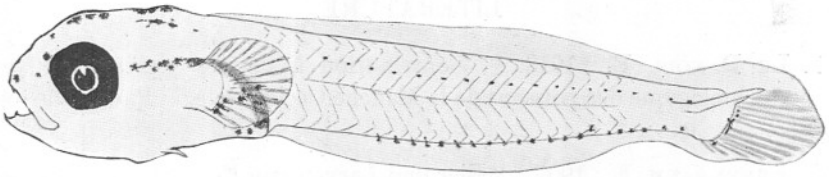

FIG. 10.—*Blennius gattorugine.* 9·0 mm. Y.F.T. 1920.

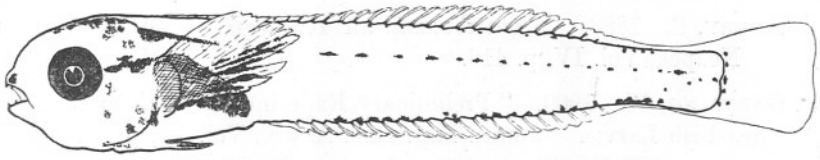

FIG. 11.—*Blennius gattorugine.* 13·0 mm. Y.F.T. 1913.

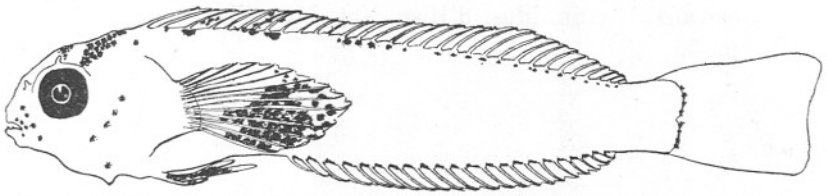

FIG. 12.—*Blennius gattorugine.* 18·5 mm. Y.F.T. 1913. External pigment only.

BLENNIUS GATTORUGINE (14 rays to the pectorals).

Emery (3) has given an account of the later developmental stages of this species with two excellent coloured drawings, and Le Danois (I, p. 120) has figured young fish of 23 mm. The Plymouth specimens of corresponding length agree with these descriptions. The smallest individual in which the rudiments of the tentacles could be observed measured 18 mm., at which size also the foundation of the first two pigment bands across the dorsal fin is already laid.

The post-larvæ are distinguishable from those of the other species on account of the long post-anal length, and the smaller and typically pigmented pectorals. At 6 mm. (Fig. 9) the little pigment which is present on the pectorals is restricted to a few chromatophores at the base of the rays on the lower part of the fin. With ensuing growth this pigment becomes augmented, but is still confined to the postero-ventral region. In the earliest stages a metameric series of chromatophores extends along the posterior two-thirds of the body post-anally at the base of the anal fin. At an observed minimum length of 8 mm. the beginning of an ultimately complete row of chromatophores along the upper side of the spinal cord may be observed at the posterior end.

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On the Post-larvæ of the Wrasses occurring near Plymouth.

By

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Naturalist at the Plymouth Laboratory.

With Figures 1–9 in the Text.

Six species of Wrasses occur in the adult stage in the neighbourhood of Plymouth (Clark 3, p. 217), namely, *Labrus bergylta* Asc., *Labrus mixtus* (L.), *Ctenolabrus rupestris* (L.), *Crenilabrus melops* (L.), *Centrolabrus exoletus* L., and *Julis julis* (L.). The post-larval forms of four of these species have been described by the following authors :—

Labrus bergylta	Le Danois	(4, p. 155).
Labrus mixtus	Allen	(1, p. 223).
Ctenolabrus rupestris	Ehrenbaum	(5, p. 7).
Julis julis	Fage	(6, p. 50).

It is the purpose of the present publication to add descriptions of the post-larvæ of the two remaining species, *Crenilabrus melops*, and *Centrolabrus exoletus*, and to suggest a key for the practical identification of the post-larvæ of the six species, based on the distinct and easily recognisable characters of the pigmentation schemes.

In his report for the year 1914 (1, p. 222), Dr. Allen pointed out that the most numerous of the forms occurring at Plymouth is the one in which the body and the greater part of the tail is covered with many black stellate chromatophores, which, however, cease more or less abruptly behind the anal fin, leaving the hinder end of the tail unpigmented. The post-larvæ of this form were recorded in 1913 (Clark, 2) and in 1914 (Allen, 1) as *Labrus bergylta*, and in 1919 (Clark, 3) under the general heading, "labrid types." Subsequently, however, living post-larvæ of this type have been reared in aquaria until the adult characters could be definitely observed, and it has been found the type actually represents three species, namely, *Labrus bergylta*, *Crenilabrus melops*, and *Centrolabrus exoletus*. A renewed study of the post-larvæ showed that they

could be separated into three groups, each exhibiting characteristic features in their scheme of pigmentation, and ultimately it was found possible to assign the individuals of each group definitely to a particular species.

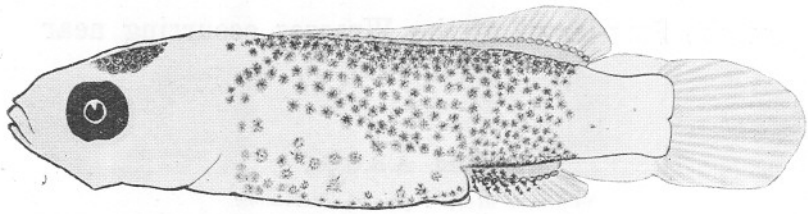


FIG. 1.—*Labrus bergylta.* 8·7 mm. Y.F.T. 1920.

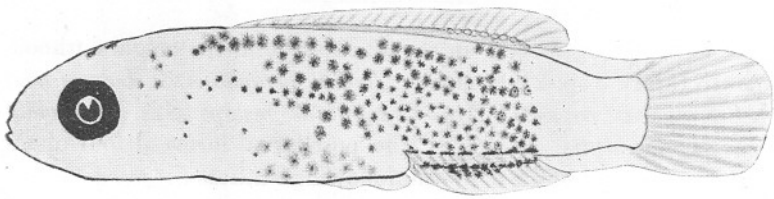


FIG. 2.—*Crenilabrus melops.* 8·6 mm. Y.F.T. 1920.

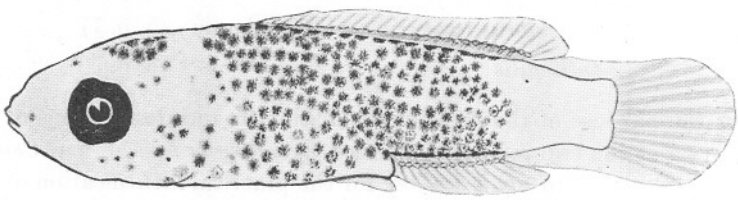
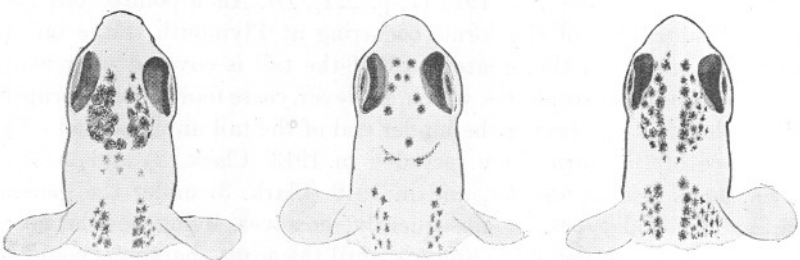


FIG. 3.—*Centrolabrus exoletus.* 8·5 mm. Y.F.T. 1920.



PIGMENTATION ON THE HEAD.

FIG. 4.
Labrus bergylta.
8·7 mm.

FIG. 5.
Crenilabrus melops.
8·6 mm.

FIG. 6.
Centrolabrus exoletus.
8·5 mm.

Post-larvæ of First Group.

These agreed satisfactorily with the descriptions given by Le Danois (4) for *Labrus bergylta*, and it will suffice to note that they are characterised by the presence of two crescent-shaped areas of black stellate chromatophores, disposed longitudinally one on either side of the middle line on the top of the head ; and by the restriction of the black pigment on the anal fin to the anterior portion on the interradial membrane (see Figs. 1 and 4).

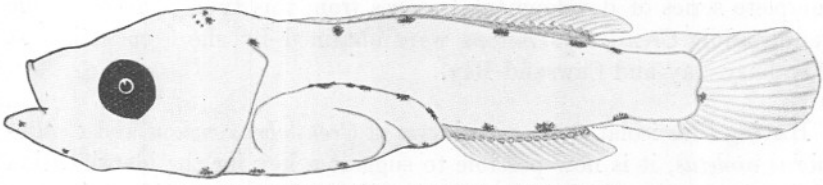


Fig. 7.—*Labrus mixtus*. 9·0 mm. Y.F.T. 1914.

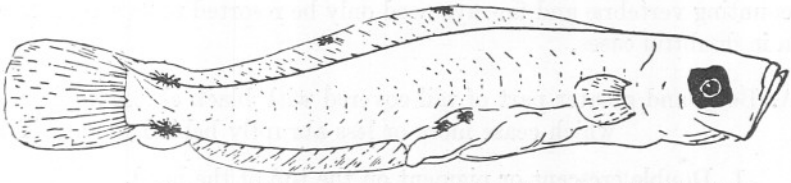


Fig. 8.—*Julis julis*. 8·5 mm. (After Fage, 6, p. 53, Fig. 40.)

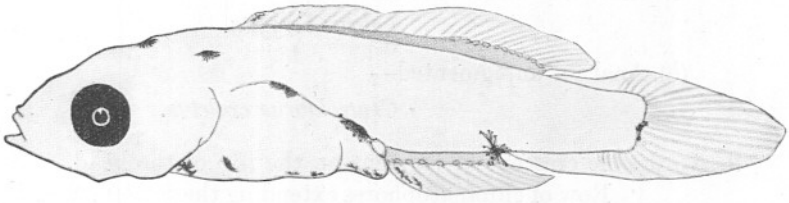


Fig. 9.—*Ctenolabrus rupestris*. 8·5 mm. Y.F.T. 1919.

Post-larvæ of Second Group.

These possess the two crescent-shaped pigment areas referred to in the case of *Labrus bergylta*, but unlike the latter, the anal fin is devoid of pigment (see Figs. 3 and 6). The body pigment generally is dense, and fairly well marked on the abdominal region. On August 26th, 1920, a living post-larva of this type was transferred to an aquarium, where it eventually acquired the characters of a young adult *Centrolabrus exoletus*.

Post-larvæ of Third Group.

The individuals of this group do not present the two crescent-shaped areas on the head, having instead a few scattered chromatophores, and, on the anal fin, a row of black chromatophores extends along its full length at the base of the fin-rays. The body pigment is more regularly arranged, and appears to be only sparsely represented on the abdominal region (see Figs. 2 and 5). Holt's drawing of a post-larva which he records as *Ctenolabrus rupestris* (7, Plate V, Fig. 49) indicates very effectively the appearance of the post-larva of this group. During August, 1920, a complete series of developmental stages from this type to recognisable young adult *Crenilabrus melops* were obtained by shore collecting at Wembury Bay and Cawsand Bay.

Having thus isolated the post-larvæ of *Crenilabrus melops* and *Centrolabrus exoletus*, it is now possible to suggest a key for the identification of the six species, which may prove useful for the more rapid sorting out of a large collection of specimens, so that the somewhat laborious process of counting vertebræ and fin-rays need only be resorted to as a confirmation in doubtful cases.

A. Body and greater part of tail covered with black chromatophores, which cease more or less abruptly behind the anal fin.

1. Double crescent or pigment on the top of the head.

(*a*) Pigment on the anal fin restricted to the anterior portion on the interradial membrane—

Labrus bergylta.

(*b*) Anal fin unpigmented—

Centrolabrus exoletus.

2. No double crescent of pigment on the top of the head. Row of chromatophores extending the full length of the anal fin, at the base of the fin-rays—

Crenilabrus melops.

B. The chromatophores on the body and tail limited in number and characteristically situated.

1. A series of chromatophores along the dorsal and ventral edges of the body, typically five along the base of the dorsal fin, and three along the ventral post-anal portion of the body—

Labrus mixtus (see Fig. 7).

2. A single conspicuous and much-branched chromatophore on the ventral edge of the body post-anally. Pigment entirely absent from the dorsal edge beneath the dorsal fin, or reduced to a single chromatophore immediately above the ventral chromatophore.

(*a*) A chromatophore on each of the 5th and 13th interradial spaces of the dorsal fin—

Julis julis (see Fig. 8).

(*b*) The 5th and 13th interradial spaces of the dorsal fin unpigmented—

Ctenolabrus rupestris (see Fig. 9).

The Plymouth collection of post-larval Wrasses does not include any examples of *Julis julis*, but the other species are well represented. It will be seen from the accompanying table (p. 698) showing the relative frequency of the post-larvæ taken during the years from 1913 onwards, that *Labrus bergylta* and *Labrus mixtus* occur considerably earlier in the year than the others, indicating an earlier spawning period.

Month.	Labrus bergylta.			Labrus mixtus.			Ctenolabrus rupestris.			Centrolabrus exoletus.			Crenilabrus melops.		
	No. of hauls in which Labrids occur. 1913–21	No. of hauls in which species occurs.	Per-centage	No. of hauls in which Labrids occur. 1914–20	No. of hauls in which species occurs.	Per-centage	No. of hauls in which Labrids occur. 1913–20	No. of hauls in which species occurs.	Per-centage	No. of hauls in which Labrids occur. 1913–20	No. of hauls in which species occurs.	Per-centage	No. of hauls in which Labrids occur. 1913–20	No. of hauls in which species occurs.	Per-centage
April	2	2	*100·0*	—	—	—	—	—	—	—	—	—	—	—	—
May	12	10	*83·3*	7	3	*42·9*	7	—	—	7	—	—	7	—	—
June	27	15	*55·6*	17	8	*47·1*	27	7	*25·9*	27	—	—	27	2	*7·4*
July	95	19	*20·0*	64	16	*25·0*	81	33	*40·7*	81	38	*46·9*	81	56	*69·2*
August	40	—	—	23	—	—	33	11	*33·3*	33	15	*45·5*	33	19	*57·6*
September	1913–20 3	—	—	2	—	—	3	1	*33·3*	3	Broken. 1? doubtful.	*33·3* ?	3	1	*33·3*

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On the Manufacture of Drift Bottles.

By

E. W. Nelson,

Scientific Superintendent, Fishery Board for Scotland, formerly Senior Naturalist at the Plymouth Laboratory.

With 3 Figures in the Text.

CONTENTS.

INTRODUCTION

In the spring of 1920 the Ministry of Agriculture and Fisheries approached the Marine Biological Association of the United Kingdom with a view to the Association undertaking the manufacture of a large number of "Drift Bottles," to be used in an extensive research into the resultant movements of the waters of the North Sea.

These "Drift Bottles" were to be of two kinds, viz., one to float on the surface and the other to trail along the bottom. The former type of instrument has been in use from an early date and presents no very

special features, the latter type was originated by Dr. G. P. Bidder and called a "Bottom Trailer." A successful series of experiments was carried out by him in the North Sea with bottles made to his specification.*

The "Bottom Trailer" is a glass bottle containing a printed postcard for the use of the finder and a label which can be read through the glass instructing the finder to "Break the Bottle." The neck of the bottle carries a straight wire tail pointing in the direction of the long axis of the bottle. The bottle is adjusted so as to have a small negative buoyancy in sea-water. When the bottle is dropped overboard it sinks to the bottom, tail first, and when the tip of the tail touches the bottom drifts with the current in that position.

The method used in the manufacture of the original bottles was to adjust them by trial and error in a tank of sea-water of known salinity, and to correct for temperature by adding a small measured length of wire. The bottle, containing a definite make-weight, was placed upside down in the sea-water and shot were added to the bottom until the bottle was almost sinking. After removing the make-weight the postcard and label were inserted, and the neck above the end of the rolled postcard was packed with cotton-wool. The shot were dried, placed on the top of the wool, the bottle was stoppered, the stopper sealed with marine glue, and the tail fitted. The bottle was again placed in the sea-water and wire was added to the neck until the bottle would neither sink nor float. Entering a table with the temperature of the sea-water as argument, a length of wire was found which, when added to the neck, completed the adjustment.

The handling of the bottle in sea-water and the necessary drying was found to be troublesome. Difficulty was also experienced in changes in temperature due to warmth from the hands when adjusting the bottles.

When the Director of the Association placed this matter into my hands, I considered that the required output could not be obtained without great difficulty unless the method of adjustment was improved, and therefore started experiments with this end in view.

As it would seem probable that "Bottom Trailers" may be used to a much greater extent in the near future, it is thought that the methods adopted, and herein described, may be of interest to others.

I am indebted to Mr. E. Ford for the drawing of Figs. 1 and 2, and to the Proprietors of *The Western Evening Herald* for the block of Fig. 3.

* Bidder, G. P., "Conseil Internat. L'Expl. de la Mer." *Rapports et Proc. Verb.* IV, July, 1905, Appendix F, p. 102, Copenhagen, 1905. *Loc. cit.*, VI, November, 1906, Annexe C, p. xxxv, Copenhagen, 1906.

BOTTOM TRAILERS.

Specification.

Before commencing the manufacture of "Bottom Trailers," it is necessary to decide on a standard to which they shall be made. Dr. Bidder, for bottles to be used in the North Sea, finally adopted a negative buoyancy of 1·7 gm. in sea-water of specific gravity 1·02750 at 8° C. It should be noted that it is necessary to fix upon a standard temperature as well as a specific gravity *in situ* (σ_t) since a correction must be applied for the expansion of the bottle itself. Dr. Bidder also adopted a wire tail two feet (610 mm.) long, measured clear of the neck of the bottle.

Since these bottles proved very successful, and no further data were available, it was decided to make this series to the same specification.

It is, therefore, required that the mass of the "Bottom Trailer" shall bear such a relation to its volume at 8° C. that an equal volume of sea-water at the same temperature and of specific gravity 1·02750 shall weigh 1·7 gm. less.

Outline of New Method.

The procedure described below is dependent upon a supply of bottles of which the external volume is constant within certain limits. Bottles such as are sold to the manufacturers of mineral waters are made by blowing molten glass into iron moulds. Several moulds are generally used for any one type of bottle, and these different moulds will, as a rule, slightly differ in size. It was found that bottles from each separate mould could be identified and sorted into batches by a careful examination of the marks left in the surface of the glass by the mould. It would be preferable to arrange with the manufacturers for the whole supply to be made in one definite mould.

Although it was found that the external volume of the bottles chosen was sufficiently constant, their weight differs greatly on account of the varying amounts of glass in each. The actual weight, however, of the original bottles within fairly wide limits is not material.

A supply of suitable bottles having been obtained, the procedure is as follows :—The postcard and label are placed in the bottle with a pad of cotton-wool above, and the whole made up to a given constant weight in air by the addition of shot on top of the cotton-wool. The bottle is then corked, sealed, and fitted with the tail. In this condition the bottle is weighed in air and then immersed in distilled water. With these weights, the temperature of the distilled water, and the weight of a metre of the adjusting wire as arguments, the length of wire to be added to the

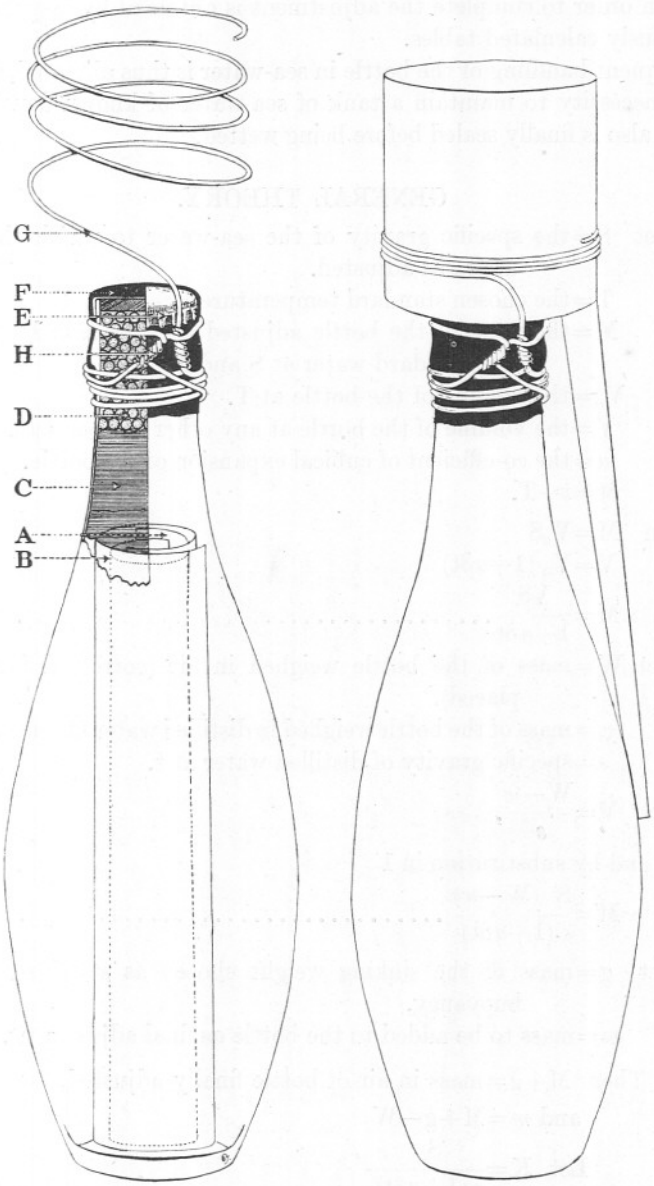

FIG. 1.—A. Rolled postcard.
B. Label.
C. Cotton-wool.
D. Shot.
E. Metal cap.
F. Sealing of marine glue.
G. Tail coiled for adjusting and packing to be straightened before use.
H. Adjusting wire.

FIG. 2.—Shows method of coiling wire.

neck in order to complete the adjustment is obtained by inspection from previously calculated tables.

Frequent handling of the bottle in sea-water is thus obviated, and there is no necessity to maintain a tank of sea-water of known density. The bottle also is finally sealed before being wetted.

GENERAL THEORY.

Let S = the specific gravity of the sea-water to which the bottles are to be adjusted.

T = the chosen standard temperature.

M = the mass of the bottle adjusted to have zero buoyancy in the standard water at S and T.

V_0 = the volume of the bottle at T.

V = the volume of the bottle at any other temperature t.

a = the co-efficient of cubical expansion of the bottle.

$\delta t = t - T$.

Then $M = V_0 S$

$V = V_0 (1 + a\delta t)$

$$M = \frac{VS}{1 + a\delta t} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1).$$

Let W = mass of the bottle weighed in air (corrected for air displaced).

w = mass of the bottle weighed in distilled water at temperature t.

s = specific gravity of distilled water at t.

Then $$V = \frac{W - w}{s},$$

and by substitution in 1

$$M = \frac{S\ (W - w)}{s\ (1 + a\delta t)} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2).$$

Let g = mass of the sinking weight chosen as standard negative buoyancy,

m = mass to be added to the bottle as final adjustment.

Then $M + g$ = mass in air of bottle finally adjusted,

and $m = M + g - W$

Let $$K = \frac{S}{s\ (1 + a\delta t).}$$

Then from (2) $m = K\ (W - w) - W + g \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3).$

Let $K = K_0 + \delta K$

$W = W_0 + \delta W$

$w = w_0 + \delta w$

where K_0, W_0, and w_0 are mean values of K, W, and w.

Then by substitution in (3) and rearranging

$$m=K_0(W_0-w_0)-W_0+g-K_0\delta w+ \quad (=a)$$
$$(K_0-1)\,\delta W+ \quad (=b)$$
$$(W_0-w_0)\,\delta K+ \quad (=c)$$
$$(\delta W-\delta w)\,\delta K \quad \ldots\ldots\ldots\ldots\ldots\ldots (4)$$

or neglecting the last term,

$$m=a+b+c.$$

Let n=length in mm. of 1 gm. of adjusting wire in sea-water at S and T.

L=length in mm. of m gm. of wire.

Then $L=mn$.

Let $n=n_0+\delta n$

where n_0 is a mean value of n.

Then $L=m(n_0+\delta n)$.

Let $A=n_0a$

$B=n_0b$

$C=n_0c$

$D=m\delta n$

Then $L=A+B+C+D$ .....(5).

a form convenient for practical use.

Calculation of Tables.

It was found that the external volume of the bottles was sufficiently constant to permit neglecting the last term of equation (4). Since the method used in preparing the bottles provides that W shall be practically constant δW will always be small. δK depends only on variation in the temperature of the distilled water in which the bottles are weighed and will not be large under ordinary laboratory conditions. The variation in the volume of the bottles will be shown in the factor δw. In equation (5) above, therefore, the term A, which includes the variable δw, can be considered as the main result to which the corrections B, C, and D, should be applied.

Table K.

$$K=\frac{S}{s\ (1+a\delta t)}$$

expanding and neglecting quantities of the second and higher order of magnitudes where less than 0·0000005

$$K=[1+(S-1)]\,[1+(1-s)+(1-s)^2]\,[1-a\delta t]$$
$$=S+(1-s)+(1-s)^2-a\delta t-a\delta t\,(1-s)+(S-1)\,(1-s-a\delta t)$$

Table K has been calculated for the following values of the constants :—

$S = 1{\cdot}02750$

$T = 8{\cdot}0°$ C.

$a = 0{\cdot}000026$

The tabular values $= 1000\ (K-1)$

Taking K_0 as the value of K at 15·5° C., $\delta K \times 10^6$ has also been tabulated.

TABLE K.

t	1000 (K–1)	$\delta K \times 10^6$
8·0° C	27·627	–644
·5	27·647	–624
9·0	27·671	–600
·5	27·695	–576
10·0	27·725	–546
·5	27·758	–513
11·0	27·794	–477
·5	27·834	–437
12·0	27·877	–394
·5	27·912	–348
13·0	27·960	–298
·5	28·026	–245
14·0	28·084	–187
·5	28·143	–128
15·0	28·206	–65
·5	28·271	0
16·0	28·339	68
·5	28·411	140
17·0	28·486	215
·5	28·566	295
18·0	28·646	375
·5	28·729	458
19·0	28·815	544
·5	28·906	635
20·0	28·996	725
·5	29·093	822
21·0	29·190	919
·5	29·290	1019
22·0	29·394	1123

TABLE A.

Find experimentally the mean value of W, the weight in air, and w, the weight in distilled water, of a series of bottles made up to any given sufficient weight, and fitted with tail, etc., as in the finished article.

Then from (2) $M=K(W-w)$

Calculate $M+g$ which will be the mean weight of the correctly adjusted bottle. Allowing 3 gm. for the addition of adjusting wire, $M+g-3$ will be the weight to which the bottle should be made up when ready for final adjustment.

It will be found convenient to choose W_0 so that its apparent value, without correcting for air displaced, will be a whole number. The nearest value to $M+g-3$ which fulfils this condition will therefore be chosen.

Example :—

Mean apparent W	521·82	
Add for air displaced	0·53	
Mean W	522·35	
Mean w	12·45	at 15·5° C.
$\therefore$ W$-w$	509·90	
$K_0(W-w)$	524·3	
$+g$	1·7	
$M+g$	526·0	
-3	3	
$\therefore$ $M+g-3$	523·0	
Therefore let W_0 =	522·53	
which gives apparent W_0 =	522·0	
W_0	522·53	
$W-w$	509·90	
$\therefore$ w_0	12·63	
W_0	522·53	
w_0	12·6	
$\therefore$ W_0-w_0	509·93.	

From equation (4) and (5) above :—

$$A=n_0a=n_0[K_0(W_0-w_0)-W_0+g-K_0\delta w]$$

Let $l=n_0[K_0(W_0-w_0)-W_0+g]$

Calculate l.*

* For value of n_0 see p. 705.

Calculate $n_0K_0\delta w$ for each value of δw differing by 0·1 gm.
Calculate $A=l-n_0K_0\delta w$
Tabulate A with w to 0·1 gm. as argument.

Example :—

$$\begin{aligned} n_0 &= 79{\cdot}66 \\ K_0 &= 1{\cdot}028271 \\ W_0 &= 522{\cdot}53 \\ w_0 &= 12{\cdot}6 \\ g &= 1{\cdot}7 \end{aligned}$$

$$\begin{aligned} \text{If } \quad w &= 10{\cdot}6 \\ \delta w &= 10{\cdot}6-12{\cdot}6 \\ &= -2{\cdot}0 \end{aligned}$$

$$\begin{aligned} \text{Then } \quad l &= 280{\cdot}08 \\ n_0K_0\delta w &= -163{\cdot}82 \\ \text{and } A=l-n_0K_0\delta w &= 443{\cdot}90 \end{aligned}$$

With $w=10{\cdot}6$ gm. as argument, Table A would give 444 mm. as the length of adjusting wire to be added.

It has been found that a range of 10 gm. in the value of w is sufficient for Table A. As w increases, A decreases ; the upper limit of the table should be where the value of A is about −150 mm.

Table B.

Calculate $B=n_0(K_0-1)\delta W$.
Tabulate B with apparent W as argument for values of δW differing by 1 gm.

Example :—

$$W=526{\cdot}0 \text{ gm.}$$

Constants as above :—

$$W_0=522{\cdot}53$$

Correction for air displaced ·53

Apparent $W_0=522{\cdot}00$

$$\begin{aligned} \text{Therefore } \delta W &= +4{\cdot}0 \\ \text{and } n_0(K_0-1)\times 4 &= 9{\cdot}0 \end{aligned}$$

Table C.

Calculate $C=n_0(W_0-w_0)\delta K$

Tabulate C with temperature of water in which bottle was weighed as argument for values of δt differing by 0·5° C.

Example :—

$t=16{\cdot}5°$

δK (page 706) $= 0{\cdot}000140$

$\therefore\ n_0(W_0-w_0)\delta K = 6$

Table D.

Let d = weight of 1 m. of copper wire in air

Then $0{\cdot}8841\,d$ = weight of 1 m. of copper wire in sea-water under standard conditions.

Therefore $$n=\frac{1000}{0{\cdot}8841\,d}$$

Determine a mean value for d by weighing a considerable measured length of the wire to be used, the corresponding value of n gives n_0.

Calculate the values of n corresponding to values of d differing by 0·1 gm. and hence δn from $\delta n=n-n_0$.

Calculate the value of m corresponding to values of mn_0 (=A+B+C) for each whole 100 mm.

Calculate $m\delta n$ from values thus obtained and tabulate with d and A+B+C as arguments.

Example :—

mean $d=$ 14·2

therefore $n_0=$ 79·66

If $d=$ 14·0

$n=$ 80·8

and $\delta n=$ 1·14

If $mn_0=500$

$m=$ 6·29

therefore $m\delta n=$ 7

the tabular value under $d=$ 14·0 and A+B+C=500.

MATERIALS USED.

In order that success may be obtained in using this method of manufacture, it is essential that the ratio of the volume to the mass of the bottle after being sealed should fall within certain fairly restricted limits. Care should be taken, therefore, to obtain materials which are as uniform as possible, and to adopt a definite routine in using them.

The following details of the materials used in the manufacture of this batch of " drift bottles " are given as a general guide.

Bottles. " 10 oz. Flat Bottom, Crown Mouth, Sodas," obtained from Messrs. Kilner Bros., London, who, in order that the external volume should be as constant as possible, kindly arranged that the bottles should as far as possible be made in one particular mould. On the average these bottles have an external volume of 510 cc. and weigh 480 gm. If the ratio of volume to weight is too small, a large number of rejects for excessive weight will be obtained, if too large an unnecessary amount of shot will be used.

Corks. " Crown Corks " are circular discs of tinned sheet-iron, with edges bent over and crenelated. On the inside is a circular piece of sheet cork. The corks are put on the bottles by means of a machine which is made for this purpose.*

This type of sealing is very largely used for Mineral waters.

Sealing. Marine Glue, No. 2 brand, made by Alfred Jeffrey and Co., London. In order to render the sealing less liable to be chipped, a small quantity, about 1%, of tallow is added to the melted glue.

Wire. Good quality copper wire used both for making the tails and for finally adjusting the bottles. This wire is technically known as " Soft Drawn " British Wire, Gauge No. 17, diameter 1·42 mm., weight 14·2 gm. per metre. To obtain uniformity in weight for length a large quantity should be obtained from one drawing.

Shot. Ordinary No. 4 lead shot weighing about 6 to 1 gramme.

Cards. In each box containing twenty-five of each sort of drifter, two postcards are placed for the use of the officer who liberates the bottles. One is addressed to the Ministry of Agriculture and Fisheries and the other to the Marine Biological Association. The other side is printed thus :—

* We are indebted to Messrs. " The Plymouth Breweries " for the loan of a corking machine.

INTERNATIONAL FISHERY INVESTIGATIONS.

Name of Lightship ?
Name and Rank of Officer-in-Charge ?
Year and Date of Liberation of Bottles ?
Day and Time of Liberation ?
Bottles put out ?

Surface numbers.....to.....inclusive.

Bottom numbers.....to.....inclusive.

Remarks, if any ?

Both kinds of drifters contain a label printed in large black type on a red ground :—

BREAK THE BOTTLE.

CASSEZ LA BOUTEILLE.

BREK DE FLESCH.

5692

BRECHEN DIE FLASCHE.

SLAA FLASKEN ITU.

The fronts of the postcards used in both the surface and bottom drifters are the same :—

POST CARD.

BREVKORT—POSTKARTE—BRIEVKAART.

No stamp required from British ports.

Reward / Récompense / Belooning / Belohnung / Belonning — the equivalent of ONE SHILLING (English).

Please fill up blanks on the back of this card and post it.
Prière de répondre aux questions sur l'autre face de cette carte, et de la remettre en poste.
Wees zoo goed dese kaart intevullen en aan de post overgeven.
Bitte die nötigen Einträge an der Rückseite zu machen, und die Kart der Post zu übergeben.
Hav den Godhed at besvare Sporgsmaalene paa Bagsiden af dette Kortet og send det med Posten.

THE FISHERIES SECRETARY,
Ministry of Agriculture
and Fisheries,
43, Parliament Street,
LONDON, S.W. 1,
ENGLAND.

The back of the postcard placed in a bottom trailer :—

INTERNATIONAL FISHERY INVESTIGATIONS. CARD NO. 5692 B.

Where was this bottle found ? 20
Où a-t-on trouvé cette bouteille ?
Waar werd deze flesch gevonden ?
Wo würde die Flasche gefunden ?
Hvor blev denne Flaske funden ?

Date when found
A quelle date l'a-t-on trouvée ?
Op welken dag
Datum des Fundes
Hvilken Dato blev den funden ?

Was it taken in a trawl ?
S'est-elle trouvée dans un chalut ?
Werd ze in een schrob-net gevonden ?
Würde sie in ein Trawl-nets erhalten ?
Blev den funden i et Trawl eller Snurrevaad ?

At what depth ?
A quelle profondeur ?
Welke diepte ?
Aus welcher Tiefe ?
I hvilken Dybdi ?

What length of wire tail had it ?
Quelle est la longueur du fil de métal ?
Hoe lang was de drood aan de flesche ?
Wie lang war der freie Draht an der Flasche ?
Hvad Længde havde Slæbetrossen ?

Name of Ship
Nom du vaisseau
Scheeps naam
Namen des Schiffes
Skibets Navn

Port and number of Ship
Port et numéro du vaisseau
Haven en nommer van het schip
Heimathaven und Nummer des Schiffes
Skibets havn og Nummer

Name and address of finder
Nom et adresse de celui qui la trouve
Naam en adres van den vinder
Namen und Adresse des Finders
Finderens Navn og Adresse

The back of the postcard placed in a surface drifter :—

INTERNATIONAL FISHERY INVESTIGATIONS. CARD NO. 5692 S.

Where was this bottle found ? 20
Où a-t-on trouvé cette bouteille ?
Waar werd deze flesch gevonden ?
Wo würde die Flasche gefunden ?
Hvor blev denne Flaske funden ?

Date when found .. ..
A quelle date l'a-t-on trouvée ?
Op welke dagteekening ..
Datum des Fundes .. ..
Hvilken Dato blev den funden ?

Name and address of finder ..
Nom et adresse de celui qui la trouve
Naam en adres van den vinder
Namen und Adresse des Finders
Finderens Navn og Adresse ..

DETAILED PROCEDURE.

The following gives, in detail, the procedure adopted by us in making "Bottom Trailers." The weights mentioned apply to the particular type of bottle used and are only given to simplify the explanation.

1. The bottles are sorted into three batches :—over 500 gm. are discarded, 460 gm. to 500 gm. are used for "bottom trailers," and under 460 gm. are used for surface drifters. This sorting is easily and quickly done by means of two pairs of pan scales, each weighed to one limit, viz., 500 gm. and 460 gm.

2. The sorted bottles are cleaned with a bottle brush and duster.

3. The postcard is rolled on a length of glass tubing, the label is rolled on top of the postcard, and both are slipped off the tube into the bottle.

4. The neck of the bottle above the rolled postcard is loosely filled with cotton wool.

5. The bottle is placed on a balance weighted to 506 gm. and shot added until equilibrium is reached. The type of balance employed is that used in banks for the weighing of bullion. A small tray fixed in the chains suspending the pan on which the bottle is placed will be found convenient for receiving the shot, and a tin funnel fitting over the neck facilitates the introduction of the shot into the bottle. This funnel should be fitted with a small wedge-shaped ring of cork to prevent shot catching on the glass lip. If necessary, the cotton wool should be pressed down with a length of wire to leave just sufficient room for the shot.

6. The Crown Cork is fitted with the machine.

7. The neck of the bottle is dipped twice into melted marine glue ; the first dip to just cover the metal part of the cork, the second to immerse the neck for about a depth of 1 in. (25 mm.). The bottle is twisted round rapidly for a few seconds and dipped into cold water to harden the glue.

8. A wire tail, previously cut to a fixed length allowing sufficient to go round the neck of the bottle, is fitted to the neck by twisting up the short end so as to leave 2 feet clear beyond the stopper.* The tail is made into a coil of about 2 ins. diameter by twisting it loosely round a short cylinder of wood fitted with a handle, which is held in the same hand as the bottle (Fig. 2). This method of coiling the tail leaves the neck free to receive the adjusting wire to be added subsequently.

* Care should be taken not to strain the wire when twisting so as to make the tail liable to break off.

9. The bottle is weighed in air to the nearest 0·5 gm. and the weight, W, recorded in the notebook against the serial number.

10. The bottle is weighed to the nearest 0·05 gm. immersed in distilled water and the weight, *w*, and temperature, t, are noted.

A considerable volume of water is used so as to avoid rapid changes in temperature. The bottle is held upside down in the water by means of a copper ring, which is slung from the balance by means of three short copper wire stays and above them waterproof silk.

11. One metre of the copper wire used for the final adjustment is weighed to 0·1 gm. and the weight, *d*, noted. This measurement is only made occasionally as a check, say after every twenty bottles adjusted.

12. The tables are entered with *w*, W, t, and *d*, as arguments, and the resultants added together (A+B+C+D) give the length of wire in millimetres to be used as the final adjustment.

13. A length of wire equal to the sum of the resultants is measured along a metre rule, cut off, and twisted round the neck of the bottle, giving extra security to the tail. If the sum happens to be a minus quantity, a result which should be avoided as much as possible, this length, if small, is cut off from the end of the tail. In no case has more than 150 mm. been so cut off. The bottle is now finally adjusted and only requires the tail to be straightened before liberation.

SURFACE DRIFTERS.

These instruments are required to float on the surface, submerged as much as possible to reduce windage, but with sufficient buoyancy to ensure that they will not sink in water of the lowest density into which they may drift. Any bottle with sufficient displacement for its weight is suitable.

Our experience with " bottom trailers " proved that the external volume of a given batch of bottles is practically constant. It was decided, therefore, to make up any one type of bottle to be used as a " surface drifter " to a given fixed weight. It was also decided to make the " surface drifters " with an average positive buoyancy of 3 gm.

The following example will show how the weight to which the bottle should be loaded before sealing is calculated :—

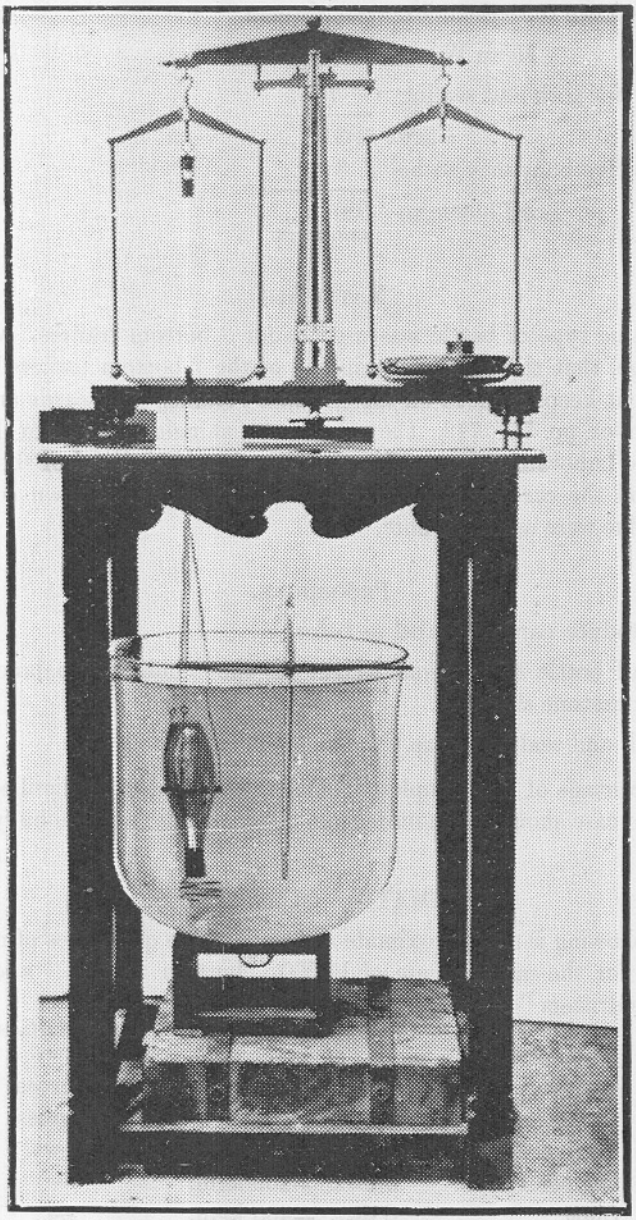

FIG. 3.—Apparatus used for weighing a "bottom trailer" immersed in water.

A series of bottles are overloaded, corked, sealed, and weighed in air and water as in the case of " bottom trailers."

Mean $W-w$	=509·9 gm.
Allow for cork and sealing	7·0 ,,
=say	503 ,,
Positive buoyancy to be	3 ,,
∴ Load bottle to	500 ,,

Materials.

The same type of bottle was used as for " bottom trailers," with this advantage, that the constant weighing of the " bottom trailers " during adjustment keeps a check on the external volume. The cards and labels are shown on pp. 711, 712. Clean limestone shingle taken from the beach below the Laboratory was used to load the bottles instead of shot, which would soil the cards unless waxed into the bottom. Crown corks and marine glue were used as before.

Procedure.

1. Introduce card and label as before.
2. Load bottle to required weight on the " bullion " balance, using limestone shingle.
3. Cork and seal as before.

A percentage of the completed bottles were tested in distilled water, and in no case should have more than a very small negative buoyancy.

ESTIMATED QUANTITIES.

The following is an approximate estimate of the quantities of materials required for the manufacture of 10,000 " bottom trailers " and 10,000 " surface drifters " :—

Material.	10,000 Bottom Trailers.	10,000 Surface Drifters.
Bottles	75 gross=10,800	75 gross
Corks	75 gross	75 gross
Marine Glue	1 cwt. = 50 kgm.	1 cwt.
Copper Wire	3 cwt. =150 kgm.	—
Shot	8 cwt. =400 kgm.	—
Cotton wool	32 lb. = 15 kgm.	—

One man, with a boy to help him, can on the average make from fifty to seventy of each sort of bottle in a day of seven hours.

The Hydrogen Ion Concentration of Sea Water in its Biological Relations.

By

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Head of the Department of General Physiology at the Plymouth Laboratory.

With 1 Chart.

CONTENTS.

INTRODUCTION.

It has long been known that sea water is alkaline and numerous determinations of its alkalinity have been made. The method adopted was the usual one for mixtures of carbonates and bicarbonates, or some modification of it. Those waters which give no colour with phenolphthalein contain bicarbonate only, but for the most part ocean waters have a small amount of carbonate also. Owing to the presence of larger amounts of carbonates and bicarbonates the reaction of sea water is more stable than that of rain or river water, inasmuch as it has a greater alkaline reserve which acts as a "buffer." The significance of this has been pointed out by Moore, Prideaux, and Herdman (1915) and by other workers. The measurement of alkalinity was carried out by the abovenamed using N/100 hydrochloric acid and titrating to the end points with phenol phthalein and methyl orange. The results are recorded in cubic centimetres of centinormal acid per 100 c.c. of sea water; this is convenient as it is what is measured directly, but others adopt the perhaps more rational notation of milligram equivalents of hydroxyl per litre (Buch, 1914). One cubic centimetre of N/100 acid per 100 c.c. corresponds to 0·1 milligram equivalent per litre. Some workers on fresh waters, Birge and Juday (1911) for example, consider water as acid if it contains more carbon dioxide than that sufficient to convert the carbonate into bicarbonate, and titrate back to a pink with phenolphthalein. Their acid water is, however, still alkaline to methyl orange.

The advent of the methods of measuring the hydrogen ion concentration of water removed all such ambiguities, and it is very convenient to have one scale on which are measured all reactions, whether acid or alkaline. This may be done by giving the hydrogen ion concentration in grams per litre, symbolised as C_H or $[H^+]$, or by using the expression pH, which is defined as the logarithm of the reciprocal of the hydrogen ion concentration expressed in grams per litre. For a discussion of the advantages of this method of stating acidity reference may be made to Clark (1920). But to render what follows more intelligible to those biologists who are familiar neither with the methods by which hydrogen ion concentrations are determined nor with the terminology adopted, a brief account of the subject is given here.

HYDROGEN ION CONCENTRATION AND ACIDITY.

It has been found that the behaviour of enzymes and of living organisms towards acid or alkaline media is not correctly shown by titration, which gives a measure of the total replaceable hydrogen (or hydroxyl), not of

the concentration of the hydrogen ion effective at any instant. It is by the latter that the action on the organism is governed, as well as that on enzymes and the solubility of various salts. For example, equal volumes of N/100 hydrochloric and acetic acids require identical amounts of alkali for their neutralization, but the former solution has a far greater concentration of hydrogen ions than has the latter, since in dilute solution hydrochloric acid is almost completely ionized, whereas acetic is not; it is accordingly said to be a weaker acid. If it is assumed that the ionisation of hydrochloric acid is complete at this dilution, viz., N/100, its concentration in terms of gram ions is C_H or $[H^+]=1\times10^{-2}$ grams per litre.

This may be written $[H^+]=\frac{1}{10^2}$ or $pH=\log\frac{1}{[H^+]}=-\log[H^+]=2$.

It may at first sight appear to be both cumbersome and unnatural to use such an expression as pH, viz., $-\log[H^+]$, to denote hydrogen ion concentration, but in practice it is extremely convenient, and gives simple numerical values which are easily remembered. Moreover, since an increase of unity in a pH value denotes a decrease to one-tenth in the hydrogen ion concentration, it is obvious that for any graphical presentation of results difficulties arise when the changes are of the order of 10^{-3} or 10^{-6}, yet these are quite common in researches on enzyme action. For certain purposes, however, it is at times convenient to use C_H values. To convert the $-\log[H^+]$ or pH values into C_H or $[H^+]$ values use may be made of semi-logarithmic paper, as pointed out by Roaf (1920). The first decimal points of the pH values are marked off from right to left as abscissæ, the C_H values from 0·1 to 1·0 being ordinates against the logarithmic rulings. A diagonal being drawn, the C_H value of any pH value may be read off, the whole number of the latter being the negative power of ten, by which the C_H value must be multiplied. Thus, for example, with pH4·50 it is found that 0·50 on the abscissa corresponds to 0·32 on the ordinate; the C_H value is then $0\cdot32\times10^{-4}$. Conversion tables are, however, given by Clark, also at shorter intervals by Schmidt and Hoagland (1919). A further reason for using $-\log[H^+]$ values will be given later on.

C_{OH} is used by Helland-Hansen (1914) and Gaarder (1917), but while it has perhaps some advantages of a local nature—if only sea water were being considered—its use is highly objectionable, as it necessitates the use of two scales and cuts off research on sea water from that on plant and animal tissues, which are often acid. It is true, of course, that as hydrogen ions increase, hydroxyl ions decrease, the product being constant, but for that very reason it is more convenient to adhere to the original use of pH values and C_H values, especially as the pH value of pure water is

not as yet known with any great precision. All C_{OH} values require recalculation for any alteration in the value accepted for pure water.

Thus while measurements of titratable alkalinity are of value as giving useful information and a measure of the buffer action of water between any selected hydrogen ion concentrations, yet the latter alone is capable of showing the reaction correctly, for a total alkalinity, equivalent say to one cubic centimetre of centi-normal acid, due to sodium hydroxide, is at a very different pH value from the same alkalinity due to a carbonate or bicarbonate. The titration value is, however, of much importance, as upon it the maintenance or otherwise of an approximately constant pH value depends.

BUFFER ACTION.

If alkali is added to acid, after a certain amount has been run in, a neutral solution is obtained. With a strong acid, such as hydrochloric, the addition of the alkali in successive portions results in a progressive diminution in the hydrogen ion concentration, as may be seen from the fact that the strong acid was largely ionised at the start. But with a weak acid the neutralization of the hydrogen ions existing at any instant results in a new equilibrium being attained by the remaining undissociated molecules of the acid, which become ionised to the same percentage as before. The alteration in the hydrogen ion concentration is therefore much less over a considerable range. If the results are plotted with pH values as ordinates and cubic centimetres of alkali as abscissæ, the slope of the curve will be less steep for the weak acid than for the strong, except near the neutral point. Curves illustrating this are given by Clark (1920), and in the older literature. A weak acid thus has a considerable "buffer action" in preventing rapid alterations in pH values, and the same is true of weak bases.

THE DETERMINATION OF HYDROGEN ION CONCENTRATIONS.

From theoretical considerations Nernst developed an equation connecting the electromotive force of a concentration cell with the concentration of its ions. Platinum black deposited on platinum and immersed in a solution through which pure hydrogen is bubbled constitutes a hydrogen electrode. On the assumption that in very dilute solutions the ions obey the gas laws, it has been shown that the potential of the hydrogen electrode changes with the concentration of hydrogen ions as follows :—

$$dE=\frac{RT}{nF}\frac{dP}{P},$$ which on integration becomes

$$E=\frac{RT}{nF}\log_e P+A,$$ where A is an integration constant, R

the gas constant, T the absolute temperature, n the valency of the ion, and F the faraday or quantity of electricity carried by one gram equivalent of the ion, and $\log_e P$ is the natural logarithm of the partial pressure due to hydrogen ions. Now if two such hydrogen electrodes are connected to form a concentration cell, the electromotive force developed is equal to $\frac{RT}{nF}\log_e\frac{C}{C^1}$, where C and C^1 are the concentrations of hydrogen ion, since the ratio of the pressures may be taken as equal to the ratio of the concentrations.

In practice such a hydrogen electrode is connected by an inverted U-tube arrangement containing a solution of potassium chloride, with a calomel electrode, and the electromotive force of this cell is measured by means of a potentiometer, with all due precautions. The constants being evaluated, use is then made of the following equation to determine the hydrogen ion concentration :—

$$\left.\frac{\text{E.M.F. (obs.)}-\text{E.M.F. (of normal hydrogen and calomel electrode cell)}}{0{\cdot}0001983T}\right\}=\log\frac{1}{[H^+]}=\text{pH}.$$

Using an N/10 KCl—calomel electrode, this becomes at 25° C. :—

$$\text{E.M.F. (obs.)}-0{\cdot}336=0{\cdot}0591\log\frac{1}{[H^+]}=0{\cdot}0591\text{pH}.$$

The fact that the term $\log\frac{1}{[H^+]}$ is thus directly determined is another reason for expressing hydrogen ion concentrations in pH values.

Full proofs of these equations and directions as to technique may be found in " The Determination of Hydrogen Ions," by Clark.

The method is used as the fundamental one for determining pH values. By its means buffer solutions of accurately reproducible pH values may be standardised for use in colorimetric determinations by means of indicators.

NEUTRALITY.

It has not been explained as yet what is meant by a neutral solution or neutrality. Pure water dissociates primarily into hydrogen and hydroxyl ions, and the product of the ionic concentrations is a constant at constant temperature, namely :—

$$[H^+]\times[OH^-]=k[H_2O]=K.$$

Since the ions are produced by pure water in equal numbers, the concentration is the same for both when reckoned in gram-equivalents. A

solution is accordingly termed neutral when the hydrogen and hydroxyl ions are present in equivalent amounts, as in pure water. With an acid solution the hydrogen ion is in excess, and the hydroxyl is correspondingly reduced, since the product is constant. In alkaline solutions hydroxyl ions preponderate, but it is possible and convenient to state the reaction of the solution in terms of hydrogen ion concentration rather than in terms of the hydroxyl. One scale, the pH or $-\log H$, is therefore obtained instead of two, based upon the concentrations in pure water as a starting point for both. Before the use of pH values became general it was however proposed by Walker and Kay (1912) to express the acidity or alkalinity of natural waters in terms of specific values, taking pure water at the same temperature as unity. More recently Wherry (1919) has re-introduced these specific values for work on soil reaction. Thus, taking neutrality at pH7, sea water, which is near pH8, would be of specific alkalinity 10, containing ten times the concentration of hydroxyl ions. The writer considers that such specific values are unnecessary, though in a general way they undoubtedly serve to give a readily grasped conception of the condition of a liquid. To adopt these specific terms would be to destroy the value of having one scale. The reasons have been given fully by Clark (1920) and have been again urged (1921) in reply to a further paper by Wherry and Adams (1921).

THE HYDROGEN ION CONCENTRATION OF PURE WATER.

It is of interest to determine the hydrogen ion concentration of pure water, but this is attended by many experimental difficulties. Those due to the solution of minute traces of glass may be avoided by the use of silica or platinum vessels, but the absorption of carbon dioxide is still a trouble. It is not possible to determine the pH value by the potentiometer on account of the great internal resistance of the hydrogen electrode half-cell when made up with pure water. The various methods adopted have recently been reviewed by Beans and Oakes (1920). They are as follows :—

1. By deduction from measurements of the electromotive force of concentration cells made up with dilute solutions.

2. By conductivity methods, giving the ionic mobilities of hydrogen and hydroxyl ions in very dilute solutions, these values are considered to be valid for pure water ; the conductivity of the latter is then measured, and from it and the ionic mobilities the concentration of the ions is calculated. This is the method used by Kohbrausch and Heydweiller (1894), and is the only one of those already mentioned which is based upon an examination of pure water ; but even then use is made of data derived

from solutions. For pure water these workers found the hydrogen ion concentration at 26° C. to be $1{\cdot}10\times10^{-7}$ or pH6·96. The value varies with the absolute temperature, as may be seen from the equation previously given. Michaelis (1914) gave for pure water at 10° C., pH7·10; at 22° C., pH7·00; and at 28° C., pH6·90.

3. By methods based on the hydrolytic dissociation of salts.

4. By measurements of the rates of certain reactions.

5. By the use of indicators, which give definite tints at known concentrations.

All these methods agree in giving values which are not very far from pH7·1. Method (5) is used by the writer to test the purity of freshly boiled distilled water and to test glass apparatus for solubility. As shown by its behaviour to bromthymol blue and phenol red, and compared with Clark's standard solutions, the purest, freshly boiled, thrice distilled water which has been prepared here gave pH7·10 to 7·05 at about 15° C. Boiling to remove carbon dioxide in certain types of "hard" glass tube may, however, give any value from about pH8 upwards, so every tube has to be tested and re-tested after use with alkaline precipitates. If used in the cold such "hard" glass tubes are safe for a time, whereas soft glass tubes are not. Pure water boiled in a soft glass test tube is at about pH9 or over. Once the indicator has been added the liquid is no longer pure water, hence method (5) is open to objection; but if due precautions are taken in making up the indicator, and if only a very minute amount, say, two drops of a 0·02 per cent solution, is added to 10 c.c. of liquid, the error from this source is slight. Precautions are necessary on account of the fact that pure water has a negligible buffer action.

An entirely new method has been introduced by Beans and Oakes (1920). In this the hydrogen electrode is set up with the purest water, and the cell $Hg\,|HgCl.KCl\,|KCl\,|H_2O\,|H_2$ is used to charge a condenser of one microfarad capacity, which it does in three to five minutes, whereas a cell of lower internal resistance in which dilute acid is substituted for pure water only requires an instant. Now, the quantity of electricity stored is equal to the product of the E.M.F. of the cell and the capacity of the condenser, viz. $Q=EC$. When discharged through a ballistic galvanometer a deflection d_1 is obtained when the pure water hydrogen electrode is used. Using a standard cell of known voltage a deflection d_2 is shown. Now, since d is proportional to Q, and $Q=EC$, therefore

$$d_1/d_2=\frac{E_1C}{E_2C} \text{ or } \frac{E_1}{E_2}=d_1/d_2;$$

and, since E_2 is known, E_1 may be found and

the pH value calculated as in the potentiometer method. The value arrived at for pure water at 25° C. is pH7·91 or $[H^+]=1{\cdot}23\times10^{-8}$. This differs considerably from the results previously obtained, but the method seems very simple and direct. From the biological standpoint it is not of great importance to ascertain this constant precisely, as pure water is never found in nature, and its reaction is not necessarily the most favourable for living cells, which can, in different species, tolerate a large range of acidity and alkalinity, a reaction suitable for one being possibly fatal for another.

The uncertainty as to the precise pH value of pure water is another reason against taking this quantity as a unit and using specific acidity or alkalinity values. Furthermore, since pure water is never found in nature, and is most difficult both to prepare and to keep, its use as a standard has no practical utility. Rain water, at about 10° C., examined almost immediately after its fall has been found to be at pH5·9—6·0 at Plymouth and Malvern. It is presumably in equilibrium with the carbon dioxide of the air. On standing corked for about two months at room temperature in a " hard " glass tube the rain water changed to about pH6·4, owing to the solubility of constituents of the glass. These traces of alkali become evident at once and neutralise the carbonic acid, thus altering the reaction of this unbuffered liquid.

THE COLORIMETRIC METHOD OF DETERMINING HYDROGEN ION CONCENTRATIONS.

In addition to the electrical methods already mentioned the colorimetric method, introduced by Friedenthal (1904) and Salm (1904), is available. It is based upon the potentiometer method as a standard, the latter being used to determine the pH values of the buffer solutions, made up to be at convenient intervals on the pH scale. Measured amounts of various indicators are added to these solutions. The electrical method is also used to check sources of error such as those due to proteins and salts, which cause the indicators to give readings higher or lower than the true values.

The colorimetric method was improved and extensively tested by Sörensen (1909), who introduced new indicators, and eliminated those liable to mislead. More brilliant water-soluble indicators of the sulphone phthalein series were introduced by Clark and Lubs (1917), as well as standard buffer solutions, having certain advantages over those of Sörensen. The indicators and standard solutions used in this research are those introduced by Clark and Lubs, also some due to McClendon. A full account of the method has been given by Clark (1920) and by Cole (1920). Clark gives a coloured chart which can be purchased separ-

ately, and is a great aid in approximate work or in field work without standard tubes. In most cases the colours are faithful renderings of those in the standard tubes, with the indicator in the specified amount.

The intervals in the Clark and Lubs series are uniformly pH0·2 from pH1·2 to pH10·0. For sea water McClendon (1917) has given a very useful series at intervals of pH0·05. These are made up corrected for salt error at the normality of chloride usually found in sea water, corrections being given for slight differences in salinity. In work of this nature care should be taken to check and cross-check the solutions and indicators, both when freshly made up and after storing. It is always advisable to use two or more indicators in orienting experiments.

HYDROGEN ION CONCENTRATION OF SEA AND ESTUARINE WATERS.

The earliest determinations were those of Ringer (1908) by means of the electrical method. Samples were obtained and brought to the Laboratory after storage for some time, when it was found that the pH values lay between 8·24 and 7·86. The waters tested were from the Zuider Zee, North Sea, and Bömmelfjord. The values appear fairly normal, as viewed in the light of subsequent work, but are open to three sources of error, firstly owing to the sweeping out of carbon dioxide by hydrogen the values tend to be too high numerically, viz. more alkaline, though precautions were taken to minimise this error. Hasselbach (1910, 1911) and other workers have introduced modified apparatus and methods whereby the errors owing to loss of carbon dioxide from liquids are greatly reduced. Secondly, on account of the evolution of carbon dioxide by bacteria and plankton organisms the pH value tends to be lowered on keeping. Thirdly, owing to the possible giving off of alkali by the glass, especially in new bottles, the values may be high. It should be noted that ground glass stoppers and the ground portion of bottle necks are particularly liable to give off alkali, as may be seen in indicator bottles. It is advisable, therefore, to remove indicators with pipettes in preference to pouring them out.

The colorimetric method was then applied to sea water by Sörensen and Palitzsch (1910) as a development of the perfection of the method by Sörensen (1909). These workers (1910) also introduced α-naphtholphthalein as a useful indicator for the range covered by sea water. This along with phenolphthalein gave reliable results, when compared with the electrometric method so as to allow for errors due to the action of the neutral salts on the indicators. The values at first obtained, which vary from pH6·6 to 8·6 are, as pointed out by the authors, vitiated by storage.

Palitzsch continued the work in the *Thor* expedition (1911, 1912), and his numerous determinations on freshly taken samples are the first really reliable measurements over extended areas. In the western end of the Baltic, in the Sound, Skagerak, and south of the North Sea the surface water was between pH8·00 and 8·05. Off Scotland and the Faroe Is. it varied from 8·08–8·22. Further south, off the coast of Portugal, it rose to 8·25. In the Mediterranean it was usually above 8·22, being as much as 8·27 in the eastern end. In the Sea of Marmora, Bosphorus, and Black Sea it was close to 8·35. These values are, of course, obtained by interpolation between the standard buffer solutions which, as given by Sörensen for the range, are at irregular intervals from pH0·06 to over pH0·3. It is thus a matter of opinion whether a shade differs by pH0·03 or 0·04 from a standard, but if the sample is kept and tested with fresh indicator it is easy to see whether the next sample is nearer or further from the standard. Neither phenolphthalein nor α-naphtholphthalein remain constant in tint if kept for a day, ever when protected from the carbon dioxide of the air.

Palitzsch further studied the relation of pH to depth, and found that there is always a noticeable, though not very great, diminution from the surface downwards. In the North Sea and Atlantic there is as a rule no colour with phenolphthalein below 400 metres, with this indicator a colour is given above pH8·06 with salinity $35^{o}/_{oo}$. It may be mentioned that the salt error is 0·21 and 0·16 at $35^{o}/_{oo}$ and $20^{o}/_{oo}$ respectively, and for α-naphtholphthalein 0·22 and 0·17 ; these amounts have to be subtracted from the pH value as found. The following figures given by Palitzsch are of interest. He does not mention the possibility of seasonal changes in pH values, so the dates are only recorded indirectly. The Faroe Is. results were, however, obtained in May and June, and the Mediterranean results between June and September inclusive.

Depth in metres.	N. Sea, E. of Faroe Is. pH	Atlantic, W. of Portugal. pH	Mediterranean, between Sardinia and Italy. pH	Black Sea. pH
0	8·13	8·22	8·23	8·34
100	8·09	8·13	8·21	7·86
400	8·03	8·04	8·19	7·53†
1000	7·98*	8·01	8·14	7·26
2000	—	7·95	8·09	—
3200	—	—	8·07	—

The decrease in alkalinity with depth is clearly shown by the above figures. In the light of more recent work the extension of the region of high alkalinity into the deeper layers of the Mediterranean water may

* 700 metres. † H_2S below 180 metres.

be interpreted as due to the measurements being made later in the season. Palitzsch also looked for a relation between increase in alkalinity and increase in oxygen content of the water. He notes, for example, that Vaag Fjord (Faroe Is.) water was very rich in oxygen and of high alkalinity pH8·22–8·24. In general a region of high pH value is also high in oxygen, and low pH values are found in regions where oxygen is in reduced amount, or even absent, as in the Black Sea ; this results in sulphuretted hydrogen accumulating, as the sulphur is no longer oxidised to sulphate. The relation is not, however, one which holds always, for excess of carbon dioxide, over and above the amount in equilibrium with the air, is produced by the respiration of plants and animals. Photosynthesis, on the other hand, breaks up the oxide and sets free oxygen, which may also be obtained direct from the air, especially where vertical currents mix the water. Owing to the buffer action of the bi-carbonate, the carbon dioxide content of sea water cannot fluctuate as rapidly as can the oxygen, so water of low oxygen content may have its supply renewed from the air before its carbon dioxide content has come into true equilibrium with that of the air. The high alkalinity of the water in Vaag Fjord was probably due to the action of the coastal algal belt, or to a local abundance of algal plankton. Palitzsch further notes that at the level of the Murray Firth the sea water twenty miles out rose to pH8·15 to 8·18.

Palitzsch (1915, 1916) also introduced mixtures of borax and boric acid for preparing buffer solutions for work with sea water. These are at intervals of pH0·06–0·10 for the most part.

Following up this work Helland-Hansen (1914) investigated the Atlantic waters from the west of the Hebrides, viz. about 8–32° W. and between 54°–60° N. His interesting data, illustrated by graphs, bring out clearly the general correspondence between oxygen concentration and hydrogen ion concentration, or in hydroxyl ion, as this author prefers to state his results. He found that a surface alkalinity of pH8·22, converted from C_{OH} values by Gaarder's tables (1917), rose at 20 metres or so to 8·26, thereafter falling to pH8·00 at about 100 metres and to 7·95 at 500 metres ; with slight rises, not exceeding pH8·01 at 1200 metres, the value pH7·95 was maintained to 2000 metres. These figures are from the most westerly station, but those from the most easterly follow much the same trend, the graphs intersecting five times. The measurements were all made in July, 1913.

During 1911 and 1912 Buch (1914) carried out a series of determinations of alkalinity, carbon dioxide, and hydrogen ion concentration in the Pojowiek (or Pojovik) as part of an elaborate hydrographic survey of this slightly brackish diverticulum of the Gulf of Finland. It is about thirty kilometres long and in breadth from about one-fifth to three

kilometres. The observations were made from August to August, in November, March, and June. The greatest acidity, pH6·53, was found in melted ice water in March, and the greatest surface acidity, pH6·90, was found at the same time in fresh water, salinity 0·07 parts per thousand. As a general rule the more saline bottom water is also more alkaline than the surface water; the relation is, however, frequently reversed in the summer. At this season, in addition to occasional very high values for acidity near the bottom, such as pH6·75 and 6·86, it is often found that, as in the ocean, the surface water is the more alkaline although less saline. For example, with salinity increasing from 5 (surface) to 6°/$_{oo}$ (bottom) pH values decrease from 7·90 and 7·86 at 0 and 10 metres respectively to 7·65 and 7·57 at 20 and 27 metres. Furthermore, in the August measurements exceptionally high alkalinity values are found, pH being greater than 8·45 in one case, with salinity only 0·73°/$_{oo}$ and above 8·37 in another with 3·64°/$_{oo}$. These figures were obtained at different stations at the surface and 4·5 metres.

The much wider range of hydrogen ion concentration as compared with sea water is very striking. Apparently the low salinity of the water denotes also a great diminution in the magnesium carbonate and bi-carbonate, which act as a buffer mixture in the sea; owing to the lower solubility of the corresponding calcium salts the amounts brought down in the fresh water are so small as quickly to undergo change due to photo-synthesis and respiration even in large masses of water. These interesting results are quite in keeping with the alkalinity determinations of Birge and Juday (1911) on the waters of the inland lakes of Wisconsin, but these workers did not determine hydrogen ion concentrations. Similar changes due to photosynthesis are recorded by Chambers (1912).

A further investigation was carried out from 1912–14 by Buch (1917) upon the same variables in the Baltic. The very comprehensive tables given are, as before, illustrated by charts. As a general rule the surface water is more alkaline than the bottom water, but the minimum alkalinity, though usually, is not always at the bottom. Particularly noticeable are the high values found in Gulf of Finland in summer, surface samples giv ng pH8·30–8·34 at the eastern end as against pH7·85 in the sea halfway to Sweden. The change with depth is often very noticeable, thus in May in the Gulf the warmer surface water at 5° C. and 4·5°/$_{oo}$ salinity is at pH8·34, 8·27, and 7·93 at 0, 10, and 15 metres, while the colder water 1° to 2° at salinity 6°/$_{oo}$ decreases from pH7·51 to 7·29 from 20 to 47 metres. In October in the same region the deep water is slightly over 2°, and the salinity slightly higher than before, 7°/$_{oo}$, while the surface water is at 11° and 4·9°/$_{oo}$, yet the reaction has altered to pH7·90 at the surface and at 50 metres to 7·57. These changes are illustrations of

the effect of photosynthesis upon large masses of water, as is made even clearer by the fact that the high pH values for surface water were accompanied by an oxygen content of 110% of saturation, whereas the lower October value corresponded to 92%.

From 1912 onwards special attention was given by Moore and his co-workers to the changes in alkalinity of sea water induced by photosynthetic activity of plankton and attached algæ. Thus Moore, Prideaux, and Herdman (1915) showed for sea water the existence of seasonal alterations in this medium such as had been noticed in fresh water by Birge and Juday (1911) and by Chambers (1912). They moreover showed that an epidemic of ulceration of the skin occurring in the Port Erin fish hatchery was due to the exceptionally high alkalinity of the water occasioned by the presence of numerous monocellular green flagellates and algæ. The values obtained varied from pH8·10 in December to 8·37 in May for the water of the Irish Sea. The original paper is readily accessible and should be consulted for details and computations of the magnitude of the crop of plankton algæ indicated as minimum values by the changes in alkalinity throughout the year. Calculating from the observed alteration in carbon dioxide content per litre and assuming that owing to efficient mixing this change affects the water to 100 metres, as seems justifiable in the open sea from Palitzsch's work, it is shown that a crop of about two tons of organic matter, dry weight, must result per acre, or at least ten tons of moist plant. These authors emphasize the value of the buffer effect of sea water and showed that about 1–3 c.c. of centi-normal hydrochloric are required to neutralise sea water, 100 c.c., to phenolphthalein and a further 21–24 c.c. to neutralise it to methyl orange. The figures give a measure of the buffer action of the sea water between the limits of the colour changes of these indicators, namely, approximately pH7·6 (allowing for the action of neutral salts) and pH4 (Prideaux 1919).

The alkalinity of the sea off the Norwegian coast and of the fjords has been the subject of a lengthy paper by Gaarder (1917). The results are recorded in " hydroxyl numbers," namely, the number of gram-equivalents of hydroxyl ion per litre multiplied by 10^7, but conversion tables to pH values are given. The relationship between alkalinity and oxygen content is well brought out, as is also the seasonal variation in these two factors. Thus in the figures given for Mofjord the water is saturated with oxygen at 10 metres in October, but by June the saturation limit has sunk to 18 metres, after which it rises slowly again. Supersaturation (as measured at 760 mm. pressure) to the extent of 10 per cent occurs from March to September or October at a depth of 10 metres. It is also in May and June, and at a depth of 10 metres, that the maximum alkalinity occurs, a hydroxyl number of 14 being reached, corresponding to pH8·29.

whereas in the winter months at 10 metres the alkalinity is pH7·98. At the depth of 60 metres there is little variation from pH7·3 throughout the year.

Most noticeable of all are the figures for certain shut off basins and fjords, such as Inderöpollen, in which early in September, 1914, the water down to 2 metres was from 100 to 102 per cent saturated with oxygen while from 3 to 4 metres it was 188 per cent, 97 per cent at 7 metres and only 23 at 10 metres. Corresponding to these it was found that at 0—2 metres the alkalinity was pH8·43, at 3—4 metres 8·49 or over, at 7 metres 8·01, and at 10 metres 7·4 or less. The existence of sulphuretted hydrogen is recorded in some of the isolated basins with no free oxygen in the depths.

Some measurements are also given by Nansen (1915) of the pH values, determined with Palitzsch's mixtures, of the Spitzbergen waters. These were carried out during the cruise of the *Veslemöy* in July and August, 1912. They are of interest as being the only ones on very cold salt water, 4·8° to —0·55°. As might be expected the values are mostly low, pH7·94 to 8·08. Some remarkably high values are also given, pH8·25–8·19. The foregoing are all surface measurements. With the higher values the gradient of change with depth is steeper than in warmer seas, a decrease from pH8·24 at the surface to 8·07 at 50 metres being recorded.

McClendon (1916) and his co-workers Magoon, Gault, and Mulholland carried out a series of electrical measurements on sea water at various pressures of carbon dioxide (1916, 2, and 3 ; 1917, 1, and 2). In addition a new set of carefully standardised buffer solutions was prepared, at intervals of pH0 05 over the required range. These were made up with the stable sulphone phthalein indicators in sealed tubes. The data and graphs given are of permanent value to all workers in this field. McClendon drew attention to the small amount of residual buffer action found in natural sea water after removal of the carbonates and bicarbonates, the volatile buffers. These non-volatile buffers are of obscure identity, but minute amounts of boric acid added to artificial sea water give similar quantitative action, that with 0·0008 molecular boric acid being nearest to the Tortugas water, which must not be taken to mean that the water contains this amount.

McClendon (1917, 1918) also drew attention to the fact that all the sea water he examined was supersaturated with calcium carbonate and would lose some of it if shaken with calcite or aragonite crystals. He showed that, for example, in the Marquesas lagoon the removal of carbon dioxide by plants had raised the water to pH8·46 and a precipitate of the carbonate was coming down and incrusting the eel-grass.

The changes in the sea were also considered by McClendon (1918) in their biological aspect. The general relationship found between hydrogen

ion concentration and oxygen content found by Palitzsch and other workers was confirmed and illustrated by many examples. Thus during the day the surface water to the east of Loggerhead Key rose usually by pH0·03 to 0·08, pH8·20 being a very usual morning value with pH8·26 in the evening, the temperature being about 27–30° C. The depth at this station is not given, but it was well within the six fathom line. Closely similar results were obtained in a tank of sea water in the open, pH8·18 being obtained at 5·40 a.m. and 8·26 at 4 p.m. in a series of readings throughout twenty-four hours. During this time the oxygen varied from 3·1 to 5·3 c.c. per litre, reduced to N.T. and P. The rise in temperature from 28·0–35·2° C. lessened the solubility of oxygen and so resulted in more passing out of solution than would have been the case at the minimum temperature. Details are given of the action of the algæ symbiotic with corals, actinians and species of bottom medusæ in evolving oxygen in excess during daylight.

The surface water between Tortugas and New York was found by McClendon to vary from pH8·16–8·23, omitting certain values which are obviously not comparable, such as 8·46 in Marquesas lagoon. The measurements were made about the beginning of August. In the open sea far from land there does not appear to be any appreciable diurnal change in pH.

Mayer (1919) studied the water of the Pacific from Fiji to Honolulu, Samoa, and San Francisco. He found an average temperature of 27·5°, pH8·22 and pressure of carbon dioxide 3·15 ten thousandths of an atmosphere and concludes that the latter is slightly above that of the atmosphere in general. He, however, noticed that the water drifting in a westerly direction was at about pH8·23, back currents moving eastwards being at pH8·10 to 8·18, which he considers to indicate an upwelling of water from 200 to 400 metres roughly. Low values, such as pH7·85 at 10·5° C., about 50 miles off the coast of San Francisco and Vancouver point to a similar upwelling. In general McClendon and Mayer agree that surface water falls pH0·01 per degree fall in temperature, but this does not apply to water which has recently come up from a considerable depth.

Mayer also pointed out that the cold water of the shore current between Nova Scotia and Florida is relatively acid pH7·9 to 8·1 in winter as compared with Gulf Stream water, found by McClendon to be about pH8·2. But it must be added that the latter is a summer measurement, so this difference is quite probably only apparent.

Previously Mayer (1916) studied the solution of limestone in relation to the theory of coral atoll formation and the carbon dioxide content of sea water. This equilibrium was investigated by Henderson and Cohn (1916) as well as by McClendon.

More recently Michael (1921) has sought to correlate the abundance of

phytoplankton off the coast of Southern California with the upwelling of deep oceanic water rich in carbon dioxide and—it is suggested—in nutritive salts. These plankton estimations were made at sea, but the water was examined in July, 1919, at 8 a.m. daily at various points along the shore. There is a distinct connection between R, the reduction of temperature below that normal for the latitude, and the pH value found; thus at La Jolla, R=0·3°–1·1°, pH8·15–8·20; at Summerland, R=1·8°–4·3°, pH7·80–8·10; at Point Hueneme, R=3·8°–9·9°, pH7·60–7·95, and at Arguello, R=8·1°–10·6° and pH7·55–7·80.

HYDROGEN ION CONCENTRATION PRODUCED BY CONSTITUENTS OF SEA WATER.

Haas (1916) examined the effect of the addition of alkali to sea water upon its hydrogen ion concentration. It had been shown by Moore, Prideaux, and Herdman (1915) that the earlier view that the alkalinity of sea water was in the main due to calcium salts was incorrect, because on boiling magnesium carbonate or hydroxide and very little calcium was deposited. Haas, however, found that the addition of 0·05 c.c. of approximately 2·5 N. sodium hydroxide at 21° C. to 24 c.c. of sea water raised it from pH7·9 to 10·1, thereafter little or no rise occurred while magnesium hydroxide was being precipitated, but the value subsequently rose, but not sharply, to pH12·0 to 12·7 with precipitation of calcium hydroxide. This explains why it is that magnesium hydroxide is precipitated first on boiling. It has been found by the writer that a solution of magnesium oxide, solid being in excess, gave in the presence of the carbon dioxide of the air pH9·9–10·0, and pH10·0 when boiled to remove carbon dioxide, using thymol phthalein and Clark and Lubs (1917) standards. Calcium hydroxide gave over pH10 and barium hydroxide well over this value. Thus it is seen that the hydroxyl ion concentration producible by calcium hydroxide is greater than that arising from magnesium hydroxide, yet owing to the lower solubility of calcium carbonate and its greater resistance to hydrolysis, the limiting pH value for the calcium salt is below that for the magnesium salt. The solubility of calcium carbonate in water free from carbon dioxide is given by Schloesing (1872) as 0·0131 grams per litre, or when in equilibrium with the carbon dioxide of the air, viz. about 3 parts per 10,000, it is 0·0646 grams at 16° C. (Cameron and Briggs, 1901). Ruppin (1909) calculates the hydroxyl ion concentrations to be $1{\cdot}05\times10^{-5}$ and 17×10^{-7} respectively, which on conversion into pH values give 9·01 and 8·37. Wells (1915) gives the solubility of calcite in contact with the atmosphere as varying from 81 to 70, 61, 52, 44, 38 parts per million as the temperature rises from 0°–50° in 10° steps, and it is probable that a similar relation obtains for mag-

nesium. These large changes in solubility are occasioned by the driving off of carbon dioxide as the temperature rises ; accordingly in a mixture of saturated calcium carbonate with the bicarbonate, in equilibrium with air, it appears that the ratio of carbonate to bicarbonate will rise with the temperature, for the former either remains constant or increases slightly while the latter decreases greatly. Thus neglecting the effect of other salts in the sea the pH value should be greater in the warmer regions, as appears to be the case, except in so far as results are affected by active photosynthesis.

Thus, as calculated, no solution containing only calcium carbonate can exceed pH9·01. A direct determination starting with calcite crystals and boiling to remove carbon dioxide gave a result very close to pH9·0 with Clark and Lubs buffer mixtures.

With magnesium carbonate and magnesite, however, a value as high as pH10·0 was obtained, so as this is identical with the maximum value for magnesium hydroxide it appears that the solution must be saturated with respect to this salt also, while the solution which is saturated with calcium carbonate does not become sufficiently rich in the corresponding hydroxide to give its maximum value, which lies well over pH10.

Thus neglecting the action of neutral salts in altering ionisation and solubility it is evident that pH9·0 would be the maximum attainable in sea water if no magnesium were present, whereas since magnesium is present as well as calcium a value up to pH10·0 might be reached. The action of neutral salts may, however, be shown to have an effect qualitatively ; to study the quantitative effects salt by salt in a mixture such as sea water would be very difficult.

Kahlbaum's magnesium chloride, tested and found neutral, was added to magnesium carbonate and boiled, when it was found that no colour was obtained with thymol phthalein and a tint corresponding to pH9·2 with thymol blue. Thus the solubility of the carbonate is lowered by the common ion sufficiently to depress the maximum from pH10·0. Similar results were obtained on adding magnesium sulphate. Again, on boiling solid calcium carbonate with solid calcium sulphate, cooling and testing, pH8·0 or near it was reached, instead of pH9·0.

Sodium carbonate could of course produce much higher pH values, but in presence of the carbonates of calcium and magnesium the immediate result would be the precipitation of these just as Haas found with sodium hydroxide. Moreover, carbonic acid in presence of the chlorides and sulphates of sodium cannot produce sodium carbonate. By insolating Ulva in sea water the value pH9·7 was attained and fresh water on Staddon Heights overlooking Plymouth Sound was raised to pH9·7 a so by insolation with the algæ found in it. Fresh inland water could not be got to surpass pH9·0 (Atkins, 1922).

VARIATIONS IN HYDROGEN ION CONCENTRATION IN REGARD TO HEALTH AND MOVEMENTS OF FISHES.

Within the last few years several researches have been carried out to test the sensitiveness of fishes to variations in the hydrogen ion concentration of the sea. The earliest work in this line appears to be that of Moore, Prideaux, and Herdman (1915) who correlated the development of a disease in plaice in the fish hatchery at Port Erin with the unusually great alkalinity of the water, which occasioned the appearance of ulcerated areas on the skin. They traced the increase in alkalinity to the action of very numerous unicellular algæ and a green flagellate infusorian.

Shelford and Powers (1915), continuing the work of the former on the reactions of fishes to dissolved gases, showed how sensitive are herring and other marine fish to increase in the carbon dioxide content of the sea, also to traces of hydrogen sulphide. They drew special attention to the influence of such factors upon the eggs and young fish. This work was continued for fresh water fish by Wells (1915), who pointed out the bearing that the work of Birge and Juday (1911), upon the alkalinity of the Illinois lakes in relation to algal photosynthesis, must have upon the movements of fishes. The subject has been further pursued by Shelford (1918), Hall (1918), Powers (1920, 1921) with the result that fishes have been proved capable of detecting very small changes in hydrogen ion concentration, the active migratory fishes being in this respect far more sensitive than those which normally rest on or near the bottom. Powers has also traced the limits within which various fish were found in Puget Sound and its neighbourhood. Thus herring were only once found in water with a pH above 7·90 and they were never found in water with a pH below 7·71. The greatest number of herring were observed in water at pH7·76–7·73. On the other hand, salmon smolt were found only between pH7·98 and 8·08. It must not, however, be left out of consideration that large numbers of fish themselves modify the hydrogen ion concentration by their respiration, and the water in a shoal must be less alkaline than the sea water in general. These shoal fish are presumably accustomed to this somewhat stuffy sea-atmosphere, and so it appears probable that if separated from the shoal a herring by making for a region of lower pH would be led back to the shoal. For the many interesting details the original papers should be consulted. Powers (1922) has further studied the respiration of fishes and correlated the hydrogen ion concentration of the sea water with their ability to utilize low concentrations of dissolved oxygen.

RELATION OF SOME PHYSICAL AND CHEMICAL FACTORS TO ALGAL DISTRIBUTION.

The work of Gail (1918, 1919) has brought to light even more definitely the factors governing the distribution of *Fucus evanescens* Ag. in the Puget Sound. It was shown that desiccation prevented growth on gravel, by killing the young plants, which also failed to grow when planted more than three decimetres below the surface of the water, ultimately dying from the deficiency in illumination. Furthermore, well-grown plants receiving less than one-fourth the normal daylight darkened in colour and died. This factor therefore limits the vertical distribution.

Gail also showed that the growth of both sporelings and larger plants of *F. evanescens* is almost completely inhibited in sea water having a pH value above 8·6, it is also very much inhibited in water above pH8·4. With a temperature above 24° C., pH8·6 proved fatal. In the other direction there is very considerable inhibition below pH7·2, and above 24° C. at pH7·0 the plants die. It was observed that under conditions otherwise favourable, *F. evanescens* was absent where much Ulva was present, as the latter causes the water to have too high pH values. The absence of the plant from tide pools is attributed to this and to the great variations in temperature.

LINES OF RESEARCH INDICATED.

Preliminary experiments and a study of the literature which has been summarised in the foregoing pages showed the desirability of obtaining information on the following :—

1. The hydrogen ion concentration produced by the constituents of sea water. This has already been discussed briefly.
2. The hydrogen ion concentration of the water of the aquarium tanks as compared with that of Plymouth Sound and the town fresh water supply.
3. The relation of the Sound water to that of the tide pools.
4. The relation of the Sound water to that over seaweeds in shallow parts.
5. The variation of the Sound water with tide and time of day.
6. The relation of the hydrogen ion concentration, salinity and temperature of the Sound water to the corresponding values in the open sea.

7. Factors affecting the hydrogen ion concentration of the open sea, such as sunlight and photosynthesis, respiration, aeration, temperature, and salinity.

8. Correlation of the hydrogen ion concentration and seasonal variations in the marine fauna and flora.

Before considering these questions an account must be given of the methods adopted in the colorimetric estimations.

COLORIMETRIC AND TITRIMETRIC DETERMINATIONS OF THE HYDROGEN ION CONCENTRATION AND THE LIMITS OF ACCURACY.

Standard solutions at intervals of pH0·2 were made up according to the directions given by Clark and Lubs. The indicator used for the work on sea water was cresol red, but for the more alkaline water produced by insolation with algæ thymol blue must be used. For the more acid water of the tanks phenol red is also of service. As previously mentioned, Sörensen and Palitzsch found α-naphtholphthalein and phenolphthalein suitable for work on sea water, and gave the corrections for salt error. According to Palitzsch the latter gives no perceptible pink tinge below pH8·06 at $35°/_{oo}$ salinity or pH8·27 with fresh water. Prideaux (1919) has pointed out that in titrating up to the bicarbonate limit, pH8·4–8·2, a trace of colour should be left in the solution when this is fresh water, but in sea water the bicarbonate equilibrium point, when in contact with the carbon dioxide of the air, is pH7·6 or about 7·8, allowing for the fact that it appears higher owing to the salt present. The last trace of colour should be removed when titrating sea water with acid.

It has been the writer's custom to titrate 100 c.c. of sea water in a conical flask of hard glass, in the cold, immediately upon the withdrawal of the sample from the sea. Alcoholic phenolphthalein was used, and enough added to give a light pink, usually forty drops of the solution. It was found that small differences in the quantity of indicator were unimportant. The titration was made by running in N/100 sulphuric acid till the last faint trace of lavender-pink vanished, the liquid being examined from the side, namely, through a thickness of about nine centimetres. The amount required was usually between two and three cubic centimetres. The limit of accuracy is apparently due to the fatigue of the colour sensitiveness of the eye to red. Titration with N/100 acid is always a difficult matter when accuracy to 0·1 c.c. is aimed at, and under the conditions met with at sea this accuracy could not be relied upon. From the winter of 1921 onwards the plan was adopted of testing the pH value

of the liquid, when colourless to phenolphthalein, by adding cresol red, and the figures for added acid were accepted as correct and truly comparable only when the liquid after titration was at pH7·85, as shown by cresol red without correction for salt error. On correction this figure becomes pH7·67, which is close to the equilibrium point given by Prideaux, pH7·6. This may be illustrated by examples from the L and E1 series (described further on) obtained on December 13th on water, in this case, taken on 12th. The pH values were estimated to pH0·05, the standards being at pH0·2 intervals. To save repetition the table also shows the sea temperatures, salinity and pH values corrected for salt error according to McClendon's standards as determined immediately the water was drawn up. The standards are at intervals of pH0·05, estimations to pH0·01.

December 12th :—

Samples.	Temperature. °C.	Salinity $^0/_{00}$.	pH corrected.	c.c. of N/100 H_2SO_4 per 100 c.c. sea water.	pH of liquid after titration, uncorrected.
L1 surface	10·6	33·12*	8·01	1·57	7·85
L2 ,,	11·4	34·34	8·07	2·04	7·85
L3 ,,	11·8	34·99	8·11	(2·63)	7·70
L4 ,,	12·3	35·35	8·13	2·43	7·85
L5 ,,	12·6	35·39*	8·14	(3·10)	7·60
L6 ,,	12·8	35·44	8·14	2·62	7·85
E1 ,,	12·95	35·40	8·14	2·83	7·85
E1, 25 metres	13·15	35·41	8·14	(3·01)	7·75
E1, 70 metres	13·11	35·42	8·12	2·71	7·85

High water at 3.20 a.m., L1 taken 9.30 a.m. ; E1 at 12.30 noon. The salt error correction for the figures in the last column is pH0·18 to be subtracted.

It is evident that L3, L5, and E1, 25 metres were overshot in titration. The water in this case became more alkaline as the harbour was left and the open sea entered. Inspection of the pH values for the sea water and of the titration results shows that the latter are less regular than the former, as it is easier to compare the intensity of colour of tubes that are a good red than to be sure that an absolutely colourless condition is attained and not overshot.

By comparing the mean values obtained for L6 and E1 with those for L1, it is seen that over this range, pH0·14, the addition of 0·08 c.c., N/100, H_2SO_4 to 100 c.c. sea water produces a change of pH0·01. This is an approximate value and is open to the objection that the titrations were carried out on the day following the pH determinations.

* Duplicate determinations, mean value.

It may be objected, however, that since the titration values represent quantities of acid added and the pH values are —logH the two are not comparable. Over a small range it is, however, allowable to assume an inverse proportionality between differences in pH and C_H values, as may be seen from the following figures from Schmidt and Hoagland's tables:—

pH	C_H	
8·40(2)	0·396	
8·500	0·322	Half-way value in each case, but the
8·60(5)	0·248	C_H value is equivalent to pH8·49.

Thus the value judged to be half-way by tint, assuming no error, is in reality not pH8·50, but 8·49, since the colour change is proportional to C_H. The error, however, is only +pH0·01, and with intervals of pH0·05, as in McClendon's series, becomes altogether negligible. It is in any case within the error of judgment using Clark's standards.

Fresh determinations made by adding exactly 1·00 and 2·00 c.c. of N/100 acid to 100 c.c. sea water at pH8·07 showed that here 0·11 c.c. corresponds to a change of pH0·01. Over the limited range pH8·14 to 7·98 it may accordingly be taken that 0·1 c.c. corresponds to pH0·01 with the sea water at 35°/$_{oo}$ salinity. With water of lesser salinity. the buffer action would be smaller, namely, the change due to the same quantity of acid would be greater.

By adding a very little methyl orange and titrating the liquid rendered colourless to phenolphthalein a measure of the bicarbonate is obtained. The sum of the two titrations affords a measure of the buffer action of the sea water. Since only slight differences in salinity were encountered titration with methyl orange was abandoned after a while, the results always being the same within the limits of error. The average of eleven determinations gave 24·65 c.c. as the amount of N/100 sulphuric acid. According to Prideaux (1919) 1 to 2 c.c. should be subtracted from this figure, as with salts present the result is somewhat high. Since, however, about 2 c.c. is required to neutralise to phenolphthalein, the total required is 26·6 c.c., or correcting, as suggested by Prideaux, the hydroxyl present amounts to about 2·46 milli-equivalents per litre at the time these determinations were made, July, in the English Channel, when the salinity was 35·2°/$_{oo}$. The values given by Schloesing, 2·48, Dittmar, 2 41, Moore, Herdman, and Prideaux, 2·36–2·50, average 2·44 in the Irish Sea from November to July, do not appear to include such an allowance, so the writer's results are in reality higher, a different end point being reached in the titrations. Methyl orange ceases to be red beyond pH3·8, so this value is just reached in the titrations, or about pH4·0 correcting for salt error. By adding 30 c.c. of N/100 acid to 100 c.c.

of sea water initially at pH8·0 corrected for salt error, it was found that pH3·7 corrected was reached at a temperature of about 12° C. and with the minimum possible agitation of the liquid, so that carbon dioxide should not be lost. This 30 c.c. becomes 28 c.c. when corrected, a result noticeably higher than the value found in summer by titration, the addition of 25 c.c., however, left the liquid at pH4·4 corrected, to methyl red, methyl orange being, of course, yellow at this stage. The precise magnitude of the buffer effect depends, therefore, on the conditions of titration to a very marked degree. On plotting a graph pH values as ordinates and c.c. of N/100 acid as abscissæ a steep curve concave to the ordinates was obtained up to 5·0 c.c. and pH6·9 corrected for salt error, from that to 20 c.c. at pH5·4 the curve was almost a straight line with a much less steep slope, or the graph up to 25 c.c. at pH4·4 might be regarded as a second shallow concave curve with a straight line from 25 c.c. to 30 c.c. The end-point chosen, therefore, affects the result very largely.

The estimation of carbon dioxide by titration in respiration measurements, as carried out by Moore and his co-workers (1912, 1913), has been severely criticized by Morgulis and Fuller (1916). The method is admittedly subject to errors in titration as already noted. But for measurements of small differences it is certainly more accurate than these criticisms appear to indicate, as is shown by the uniformity of the results for sea water. Passing in considerable quantities of the gas and then estimating the amount recovered, as Morgulis and Fuller did, is not a permissible procedure as a criterion ; under these conditions the liquid readily parts with the gas during titration ; but with sea water more alkaline than bicarbonate the alkalinity is being measured and no carbon dioxide is free ; beyond the bicarbonate point small amounts of the gas are free, but in the experiments dealt with here this was chosen as the end-point ; as the bicarbonate point is left and pH7 is approached errors due to loss of gas become more serious, as was noticed in studying the titration —pH curve in the acid direction.

From these results it follows that titration is not as accurate in determining the condition of the sea as is the colorimetric determination of pH values by McClendon's standards. These were made up and marked, giving the corrected pH value for the salinity and amount of indicator added.

McClendon (1917) records values for sea water 0·4N, 0·5N, and 0·6N with respect to chlorine ions, the salt error increasing by pH0·05 for each decimal place. The sea water off Plymouth Sound and the adjacent parts of the English Channel usually lies between salinity 35·0 and 35·3‰, equivalent to 0·546 to 0·550 normality with respect to chloride. Using 0·5 c.c. indicator with 10 c.c. sea water these values become 0·520 and 0·524N. The salt error is accordingly for both pH0·01 more than

for 0·50N sea water. In marking the standard tubes this correction was introduced, pH8·20 being marked pH8·19. In making these comparisons, working to the second decimal place, it is essential to allow sufficient time to elapse for the freshly drawn water to attain the temperature of the standard tubes in the cabin, since the pH value varies inversely as the absolute temperature, the neutral point altering from pH7·10 to 7·00, with a rise from 16° to 22° C. Accordingly in winter the samples, when freshly drawn, always appear more alkaline than they are in reality, since they are colder than the standard tubes.

Furthermore, owing to the lessened solubility of carbon dioxide and the increased dissociation pressure of this gas in bicarbonates, rise of temperature leads to a new, but slowly attained, equilibrium. The alteration according to McClendon amounts, for sea water, to pH0·01 decrease per degree fall in temperature. Thus water at pH8·22 at 16° C. would at 10° C. be at pH8·16. This change is superposed upon that in the water itself, which alters by pH0·10 in the opposite sense, pH7·10 becoming at 10° C. pH7·20. Since, however, the water in the standard tubes also changes with temperature the latter alteration is automatically corrected in the observations, provided the two tubes are at the same temperature.

This temperature co-efficient, given by McClendon as pH0·01 decrease per degree fall in temperature, will be considered later in regard to the seasonal changes.

It may be added that a tube made up with indicator and buffer at pH8·14 on November 8th and kept in a test tube of hard glass, with a rubber cap, remained quite unaltered ; this was shown by examination on February 28th against buffer from the standard bottle, waxed internally and never opened between the dates mentioned. The standard tube at pH8·16 was noticeably darker, so it may be considered that the estimations are correct to pH0·01. In comparing these tubes it is best to hold them slanting against ruled white paper, with blue lines. Differences in tint then become more easy to detect than with the plain white paper usually recommended.

For colorimetric work the sulphone phthalein indicators are much superior to the phthaleins, inasmuch as their solutions are far more stable. Standard tubes made up with phenol- and α-naphtholphthaleins were found to show marked diminutions in intensity of colour in less than a day, and thymol phthalein, though a most useful indicator for the region of pH10, fades in less than an hour. In contrast to this the stability of the sulphone phthaleins is remarkable ; for one thing, being more soluble, they do not tend to precipitate. No trace of fading has as yet been observed in tubes made up with *o*-cresol sulphone phthalein four months ago. These were kept in the dark and the stock solutions before mixing

were saturated with toluene. If toluene is not added this indicator does fade slowly in intensity, even though its tint remains almost unaltered. The change appears to be due to bacterial action upon the indicator. In this respect phenol red and brom thymol blue seem to be somewhat more resistant. The fading of cresol red was, however, scarcely perceptible in ten days in warm summer weather. No change whatever could be detected in two days. In sea water the fading is more rapid, but quite apart from this there is a marked change in tint, also owing to the production of carbon dioxide by the bacteria acting upon the organic matter in the sea water. With sea water at pH8·1–8·2 it is, however, sometimes useful to use α-naphtholphthalein when comparing two closely similar samples, as in this region it shows a decided change in tint, whereas the change in cresol red is mainly one in intensity. The layers sometimes found in the sea in summer may thus be differentiated with greater certainty.

For some months much use was made of a Duboscq colorimeter for comparing water samples. It was found that exposure of the standard solution in one of the tubes for a working day had no measurable effect upon its pH value as tested against a freshly drawn portion. Such colorimeter comparisons are accurate only when the indicator change is one of intensity rather than of tint, namely, a one colour change. For this reason phenolphthalein was used, but abandoned as its low solubility leads to alterations in the standard inside a few hours. It is permissible to use cresol red over a narrow range, for samples slightly above or below the nearest standard; but when pH8·4 is matched in intensity against pH8·2 there is always a visible difference in tint. Working by intensity change rather than by alteration in tint is always open to the error due to small differences in the quantity of indicator added; it was found necessary to reject a number of measurements owing to this, since it was ascertained that if the pipette delivering the indicator drop by drop was not held vertically the size of the drops altered slightly, from 27 to 25 per c.c.; the greater the departure from the vertical the larger the drops, up to an increase of roughly 50% with an almost horizontal pipette. In rough weather it was impossible to avoid some inclination, so further work was done by adding 0·50 c.c. of indicator from a pipette delivering 1·00 c.c. from a length of 145 mm. With these precautions it was found possible to measure out fresh portions and obtain agreement to within 0·1 mm. on the scale of the colorimeter, taking the mean of four adjustments. This accuracy is not surpassed by successive sets of readings on the same solution. In making these readings the most accurate method seems to be to adjust the standard to a fixed depth, 12·5 mm. was used, and to screw the plunger in the sample till the tint is just perceptibly darker than the standard; then to reverse and screw till it is just a shade

lighter, after this to screw back half-way again, by judgment. This method minimises error due to fatigue of red-sensitiveness of the eye, lessens eye strain generally, and is more rapid than trying to adjust to exact equality. In the next measurement of the series the sample is adjusted from too light to too dark and half-way back. It was found that when one tube was at or near 12·5 mm. depth a difference of pH0·01 with McClendon's standards corresponded to a scale reading of 0·1 mm. between the limits pH8·07–8·23.

The determinations of pH values of sea water made with the Clark and Lubs standards prior to November, 1921, were corrected for salt error by comparison with the McClendon series with cresol red. Thus a sample of salinity 35°/$_{oo}$, judged to be pH8·40 by the former, lay between the tubes at pH8·24 and 8·19, and was taken to be at pH8·22.

Similarly the pH8·20 Clark and Lubs standard was found to be pH8·02 with McClendon's series. These results show that the salt error is pH0·18, for cresol red with these borate solutions. It would be slightly larger with Palitzsch's mixtures, as the Clark and Lubs standards have M/5 KCl with the boric acid. There appears to be no appreciable inaccuracy in taking the salt error to be the same in the other sulphone phthaleins, namely, in the phenol and thymol homologues ; McClendon (1917) considers this to be true for these three, and probably all of the series.

THE HYDROGEN ION CONCENTRATION OF THE WATER OF THE AQUARIUM TANKS.

The tanks are filled with salt water, with the exception of one which is fresh. There are two main salt water reservoirs of about 225 cubic metres capacity each. These are filled by pumping up from the Sound at high spring tide, when the salinity is not below 35°/$_{oo}$ as a rule, but inspection of salinities for the Sound shows that this value is not often reached. Pumping is only carried out when the water is clear, and it is allowed to settle before introducing into the circulation. The reservoirs are used week about to supply the tanks, the water being delivered in numerous small jets which carry down much air. This bubbling of air also serves to mix the water and to remove carbon dioxide. The circulation is stopped twice a day for three hours, but a cessation for five hours is accompanied by distress to the fishes. Water is pumped in only at long intervals, about half a reservoir being changed every six months. Small quantities are added at irregular intervals to make good losses.

When the pH values of the various tanks are determined the uniformity and constancy of the values are somewhat surprising. The variations are,

however, considerably greater than in the sea and the tanks are always much more acid. The reservoirs were constantly at pH7·6 from April to July. In July, owing doubtless to the higher temperature and lessened solubility of carbon dioxide with increased dissociation pressure, the west reservoir water rose to pH7·65. Owing to the intake of sea water the east reservoir was then at pH8·0. The sea water at the slip near the intake was at pH8·27–8·32 during this period. There is thus a very considerable difference, pH0·6–0·7 between the water of the Sound and of the reservoirs. Taking McClendon's values for 20° C. this corresponds to a change in the carbon dioxide pressure from 0·16 mm. to 1·00 mm., the concentration of free carbon dioxide is accordingly more than six times as great in the reservoirs as in the Sound water inshore.

The water in the tanks on April 12th was found to vary between pH7·57 and 7·27. On 28th they were mostly between pH7·62 and 7·45. One was down to 7·32 and three tanks contaminated with prawns, which died after having been brought in from the sea, were at pH7·2–7·05. The figures for 28th correspond to a pressure of 1·0 to 1·7 mm. in the tanks generally, and of 3·7 to 5·7 mm. in the contaminated tanks. The water in the latter stank, although circulation was in progress. One of two *Sepia* died and the tanks were cleaned out, after which the pH value rose to 7·3 and then became normal.

In another instance two tanks occupied, A by conger eels, etc., and B by poor cod, turbot, etc., were both at pH7·45. Owing to a crack appearing in A most of the occupants were transferred to B. The pH value fell to 7·32 and remained constant at that for several days. This corresponds to a carbon dioxide pressure of 2·6 mm. or 30·4 parts per 10,000, about nine times the normal amount in sea water. The temporary cessation of the circulation did not appear to produce any alteration. It was noticed that the respiration of some of the fish seemed to be laboured, in spite of the numerous bubbles of air carried deep into the tank with the inflowing water. Two turbot in this condition respired at the rate of 30–32, whereas two breathing easily showed 23–24 per minute, the temperature being about 13° C. In January it was noticed that the turbot were all respiring easily at 13–15 per minute in water at pH7·62 at 9·5° C. The changes in the respiration rate of fish is probably worthy of attention, as the oxidation in their tissues must decrease with falling temperature. Furthermore, even when oxygen is abundant, since its quantity in sea water varies independently of the pH value—though in a general way an increase in one denotes a decrease in the other—it is probable that a low pH value may stimulate the fish to respire more rapidly. [See Kanitz (1915) for the temperature effect, and Powers (1922) for the variation of respiration with change in hydrogen ion concentration.] The constancy of the reservoirs was puzzling in view of the fact that water from the tanks

flowed back into them. Prideaux (1919) has pointed out that in sea water very dilute bicarbonate solutions in equilibrium with the air are at pH7·6, corrected for salt error, instead of at pH8·2 as in fresh water. It was at first thought that this explained the constancy of the reservoirs. Both McClendon (1917) and Prideaux state that the equilibrium value for sea water in contact with the air is close to pH8·1, so it is not to be expected that surface water will normally be much below this. On reducing water from the Sound to about pH7·5 by breathing into it, and then shaking with laboratory air in a test tube a value pH8·02 was reached, the initial value having been pH8·06. Similarly tank water at a little below pH7·6 was raised to about pH8·0 by agitation in a test tube. Both tubes were slightly warmed to hasten the expulsion of gas, but were cooled before determining the final pH values. Thus it is clear that the figure found for the reservoirs is due to the fact that conditions in this considerable mass of water are sufficiently constant to maintain a constant pH value.

The higher values found in the sea, pH8·2 and over at times, as against pH8·1, the equilibrium value, are due to the photosynthetic action of plants in removing carbon dioxide, but in addition the equilibrium value will shift somewhat with temperature, rising with increase of temperature. For this pH0·01 is taken as shown by McClendon.

The only algæ which flourish in the main aquarium tanks are certain minute species ; these are not at all easy to identify, and may be abnormal forms ; they include, what seems to be *Callithamnion Rothii*, Lyngb., *Polysiphonia sp.* (?) *urceolata*. Grev., or the smaller but very similar *P. pulvinata* Spreng., also *Sphacelaria sp.*, and *Cladophora* (?) *fracta* Kg., with Naviculoid ensheathed diatoms. These appear only in the tanks next the southern windows. A tank in the yard close to the north side of the building was covered with " railway green " glass. In it larger red algæ were established and its pH value was identical with that of the reservoirs. An adjacent uncovered open air tank contained green algæ and was evolving oxygen bubbles. This was at pH8·4, namely, more alkaline than the water of the Sound, but like a tide pool.

It is clear, therefore, that if it is required more closely to approximate to the pH value of the sea in autumn it will be necessary to aerate with air partly deprived of carbon dioxide. With air altogether deprived of this gas the carbonate limit, almost reached in very active photosynthesis, might be attained in time—were it not for the respiration of the animals. This is injuriously alkaline for organisms. Less thorough aeration with CO_2 free air would doubtless attain the same result, or untreated air would bring the water to about pH8·0–8·1.

It was noticed that a newly made cemented shallow tank was at pH8·2, the same as the sea. The addition of lime in correct proportions might

also be made to the reservoirs to regulate the reaction. Inadequate illumination appears to be the most important factor in limiting the growth of brown and green algæ judging from Gail's work.

As a result of the examination of the aquarium tanks it may be concluded that water is suitable for many fish and invertebrates if the circulation and aeration are sufficient to maintain it close to pH7·6. For the more delicate organisms attempts must be made more nearly to maintain a reaction close to pH8·2.

It was found that small Ctenophores and medusæ, which die in the tanks, lived for several days in jars of sea water, provided very few organisms were present. The reason that the tow-net jars become malodorous and contain only dead animals inside a day or less, is that the oxygen supply becomes exhausted and the carbon dioxide so increased as to have poisonous effects. For example, a jar rich in *Phæocystis globosa*, collected at 2 p.m. on April 26th and examined at 1 p.m. on 27th, was found to have changed from pH8·2, that of the sea where it was taken, to pH7·0, and this in spite of the fact that photosynthesis should have been possible in the jar on deck for some hours after the sample was obtained.

It was also observed that jars containing *Ulva sp.* brought up and placed in sunlight rapidly became more alkaline owing to photosynthesis. The limit reached was pH9·77 or 9·95 uncorrected for salt error with thymol phthalein. If too many algæ were present, however, such as in a closely packed mixture of green, brown, and red varieties, the pot developed a stench and the reaction became as acid as pH6·4, whereas a single *Ulva* plant might live for months in such a jar.

The only fresh water tank was found to be at pH7·25 in April, and pH7·4 a fortnight later, the tap water being then at pH7·0. Probably the presence of limestone pebbles tends slightly to increase the pH value. In January the value was pH7·05, that of the tap being pH6·8. The range, at least pH0·35, is greater than in the sea water tanks, as the buffer action of fresh water is very slight. The conditions in fresh water will be considered elsewhere.

THE WATER OF THE SOUND AT VARIOUS TIMES COMPARED WITH THAT OF TIDE POOLS AND LOW WATER *LAMINARIA* BELTS.

It was found that the Sound water at low tide in April was at pH8·27, whereas when taken from very shallow water over *Laminaria digitata* it was at pH8·42. A rock pool exposed for about two hours was also at pH8·42. This contained an abundance of green algæ, *Ulva sp.* A similar pool higher up exposed for about four hours was at pH8·47. This was on April 25th, but on May 10th water over *Laminaria* in the

same place with one hour flood tide was at pH8·07, while the Sound water both then and at full tide was at pH8·3, the temperature being 13° C. Water in among densely packed algæ may, therefore, fluctuate considerably, photosynthesis rendering it more alkaline and decomposition with its attendant abundance of bacteria and protozoa working in the opposite way.

Again, on July 11th, at 3 p.m., in full sunlight one of the exposed rock pools was found to be at pH8·57, 21·4° C., and took 3·7 c.c. N/100H_2SO_4 per 100 c.c. with phenolphthalein. High tide was at noon, the Sound water at the west slip was then at pH8·32, and took 2·0 c.c. for neutralisation, the temperature being 18·7°. When the pool was examined the Sound water was unchanged save for a rise in temperature to 19·2° in the shallow water by the west slip.

It may be stated that in the open sea there are no perceptible daily variations in pH value, but small changes may be noted close to the shore in the Sound.

		High water.		
pH, values taken.	July 26th. 8 a.m.	July 28th. 11.30 a.m. t=17·8° C. c.c.	Dec. 16th. 6.20 a.m. t=11·7° C.	Feb. 1st. 8 a.m. t=9·4° C.
10 a.m.	8·18	8·22 (0·73)	8·06	8·06
Noon	—	8·23 (0·67)	8·02	—
3 p.m.	8·28	8·24 (0·73)	8·02	8·04
5 p.m.	—	8·33 (1·12)	8·02	—

Such changes appear to be due to the effect of the photosynthesis of algæ on the shallow water, which at times may counterbalance the influence of a greater quantity of organic suspended matter as compared with open sea water. The July days were sunny, the December day completely overcast. The figures in brackets are c.c. of acid per 100 c.c. sea water as before. Some uncertainty exists as to the absolute values for pH found in July since the solutions had been made up some time and not—as later on—preserved with toluene. They appear considerably too high for the titration results, which, however, are unusually low.

THE RELATION OF THE WATER OF THE SOUND TO THAT OF THE OPEN SEA.

Since certain fish pass every year from fresh to salt water and return again to the rivers it becomes of interest to ascertain whether there are any differences in the water, as one approaches the shore, which may possibly serve as a guide to them. It may be stated at once that such differences have been found, but no proof has been adduced that they are adequate to direct the migration of the fish.

Moore, Prideaux, and Herdman, in discussing alkalinity determinations, state that " the degree of photosynthesis and the corresponding weight of ocean crop is probably much more abundant nearer to the littoral. It is frequently observable in the table, that water taken along shore is more alkaline than that taken from on board a vessel three to five miles from shore. Observations at great distances out at sea during the various seasons are most desirable, but difficult to obtain."

Since the calculations upon the annual crop of the sea made by these workers is based upon the magnitude of these photosynthetic changes it is important to determine the variations in alkalinity as precisely as possible.

Accordingly for such work carried out from this Laboratory stations were selected as follows :—

Station.	Situation.
L1	In the fairway of Plymouth Sound below the Laboratory near the Mallard buoy. Lat. 50° 22′ N., Long. 4° 08′ W.
L2	In the fairway between the western extremity of the Breakwater and the Cornish coast north of Cawsand.
L3	Off Rame Hd., on the line between the Breakwater Lighthouse and the Eddystone.
L4	Half-way between Rame Hd. and the Eddystone. Lat. 50° 15′ N., 4° 13′ W.
L5	Eddystone, 10 miles S. 42° W. from Breakwater Lighthouse.
L6	Half-way between the Eddystone and the International Station, E1, viz. 5 miles on a S.W. course. Lat. 50° 06′ N., Long. 4° 20′ W.
E1	Ten miles S.W. from the Eddystone. Lat. 50° 02′ N., Long. 4° 22′ W. Depth 40 fathoms. Bottom samples 70 metres.

This series gives a line of 22–23 miles, with estuary water, coastal water, and sea water at nearly as great depth as may be attained in the Channel. Fresh water effects and the action of coastal algæ should be shown if of sufficient magnitude.

The table already given on p. 737 for results obtained on samples taken on December 12th points to an increase in pH value as the Sound and coastal zone are left. Even when the values are corrected to the extent of pH0·01 per degree it is seen that there is a well-marked difference ; the correction is added to the samples drawn in the colder regions to render them truly comparable as regards the photosynthetic change with those further from the land. This difference is not accounted for by any change in salt error, since the latter only amounts to pH0·05 for a difference of 0·1 in chloride normality, namely, an alteration of salinity of close on 20 per cent, and the necessary small corrections have been

introduced for this source. Similar results are given quite consistently by other autumn and winter determinations as shown in the following tables :—

Surface samples.	pH corrected. July 4th.	July 26th.	c.c. of N/100 H_2SO_4 per 100 c.c. sea water. July 4th.
L1	8·29	8·27	2·6
L2	8·27	8·24	2·5
L3	8·27	8·22	2·5
L4	8·23	8·22	2·6
L5	—	8·20	—
L6	—	—	—
E1 (July 2nd)	8·17	—	2·0

High water at 4.20 a.m. on 4th, L1 at 10 a.m. inward trip.
,, ,, ,, 9.10 a.m. ,, 26th, L1 ,, 9.20 a.m. ,, L5 at 6.20 a.m.
E1, salinity 35·23%, temp. 14·8°. Temp. about 16° on 26th.

Surface samples.	Temp. °C. Aug. 12.	Aug.15	Aug.22.	Salinity ‰.* Aug.12.	Aug.15.	pH cor'ted. Aug.12.	c.c. of N/100H_2SO_4 per. 100 c.c. sea water Aug.12.	Aug.15.	Aug.22.
L1	16·2	15·9	15·6	34·6*	—	8·16	1·6	1·9	2·2
L2	15·5	15·7	15·2	35·0	35·0	8·25	2·5	2·2	2·5
L3	15·3	15·7	15·1	35·1	35·1	8·23	2·6	2·3	2·6
L4	15·5	15·8	15·0	35·1	35·2	—	2·1	2·4	2·7
L5	15·3	15·5	—	35·3*	35·2	8·22	2·1	2·4	—
L6	15·5	15·5	—	35·1*	35·2	8·24	2·4	2·2	—
E1	16·1	16·2	—	35·1	35·2*	8·27	2·3	2·4	—

High water.	L1 taken.	E1	Date.
Noon	8.30 a.m.	Noon	12th
3.30 p.m.	2.30 p.m.	5 p.m.	15th
7.40 a.m.	10.0 a.m.	—	22nd

Surface samples.	Temp. °C. Sept. 1st.	Sept. 6th.	pH corrected. Sept. 1st.	c.c. of N/100H_2SO_4 per 100 c.c. sea water. Sept. 1st.	Sept. 6th.
L1	15·9	16·3	8·22	2·9	2·5
L2	15·8	15·7	8·23	2·9	2·5
L3	15·6	16·0	8·22	2·9	2·7
L4	15·6	15·9	8·24	3·3	2·6
L5	15·6	15·6	8·24	3·3	2·5
L6	—	16·3	—	—	2·8
E1	—	16·4	—	—	3·0

High water at 5 a.m., L1 taken at 8.30 a.m. Examined next day, September 2nd.

High water at 8 20 a.m., L1 taken at 11 a.m., E1 at 6 a.m. on inward trip. Titrations made next day.

* Denotes duplicate titrations. Salinity results given to the nearest decimal place.

September 15th :—

Surface samples.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100 H_2SO_4 per 100 c.c. sea water.
L1	16·3	33·7*	8·20	—
L2	16·1	34·8	8·25	—
L3	15·9	34·9	8·23	—
L4	15·7	35·1	8·23	—
L5	15·5	35·2	8·25	—
L6	15·8	35·2*	8·25	—
E1	15·8	35·1	8·25	—

High water at 4.30 a.m., L1 taken at 9.30 a.m., E1 at 2 p.m.

October 18th :—

L1	16·0	35·0*	—	—
L2	16·0	35·2	—	—
L3	16·0	35·2	—	2·1†
L4	16·0	35·2	—	—
L5	15·9	35·4*	—	2·3†
L6	15·7	35·3	—	—
E1	15·6	35·3*	—	2·2†
$E1_1$	15·6	35·3	—	—

High water at 5.40 a.m., L1 taken at 8.30 a.m., E1 at 11 a.m., and 1 p.m.

November 9th :—

Surface Samples.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100H_2SO_4 per 100 c.c. sea water
L1	11·6	35·0	8·07	—
L2	13·0	35·2	8·16	2·4
L3	14·0	35·3	8·18	2·4
L4	14·3	35·3	8·22	—
L5	14·4	35·4*	8·23	—
L6	14·3	35·4*	8·23	2·9
E1	14·3	35·4	8·22	2·7
E1′	15·0	35·4*	8·23	—

High water at midnight, L1 at 8.30 a.m., E1 at 11.15 a.m.

The duplicate E1 was taken after the completion of a depth series in the water bottle. The other samples were from the bucket.

For December results see previous table p. 737.

* Denotes duplicate titrations. Salinity results given to the nearest decimal place.

† Not titrated till three days after drawing.

January 11th :—

Surface samples.	Temperature °C.	Salinity $^0/_{00}$.	pH corrected.	c.c. of $N/100 H_2SO_4$ per 100 c.c. sea water.
L1	10·0	30·69*	8·05	—
L2	10·1	32·25	8·09	—
L3	10·3	33·97	8·11	—
L4	10·3	34·67	8·11	—
L5	11·1	35·23	8·13	—
L6	11·2	35·26	8·13	—
E1	11·2	35·36	8·14	—

High water at 3.30 a.m., L1 at 9.45 a.m., E1 at 12.15, noon.

On January 10th water at the east slip was at pH8·07 at high water.

February 6th :—

Surface Samples.	Temperature °C.	Salinity $^0/_{00}$.	pH corrected.
L1	8·2	31·02*	8·12
L2	8·8	33·04	8·12
L3	8·9	34·93	8·11
L4	9·2	35·16	8·13
L5	9·5	35·17*	8·14
L6	9·9	35·33	8·14
E1	9·9	35·33	8·14

High water 8.10 a.m., L1 at 10 a.m.

March 29th :—

Surface Samples.	Temperature °C.	Salinity $^0/_{00}$.	pH corrected.
L1	8·0	33·46*	8·12
L2	8·4	34·37	8·14
L3	8·6	34·94	8·14
L4	8·7	34·97	8·14
L5	8·9	35·16	8·15
L6	9·1	33·23*	8·14
E1	9·7	35·35	8·16

High water at 6.20 a.m., L1 at 10.15 a.m.

The less saline estuary waters were corrected for salt error by adding the pH values as shown below to the values indicated on the standard tubes, the marking of which has been explained previously.

* Denotes duplicate titrations.

Salinity ‰.	Chloride.	Chloride normality.	Chloride normality, with indicator.	pH correction added.
35·30	19·54	0·550	0·524	0·00
35·01	19·38	0·546	0·520	0·00
33·71	18·66	0·526	0·496	0·01
32·25	17·85	0·503	0·480	0·02
30·72	17·00	0·479	0·456	0·03

It was unfortunately necessary to reject a large number of careful determinations made with the colorimeter, owing to the variation in the size of the drops of indicator added. This has rather spoiled the series. With the exception of the July measurements, however, the titrations and pH values recorded and all the rejected series were in agreement in showing a decidedly lower alkalinity at L1 than at L2 and stations further out. The July values show a gradient in the reverse direction. It will be of interest to see what results obtained earlier in the summer are like. The McClendon series was used from November 9th inclusive onwards, so greater reliance is to be placed on the values obtained with standards at pH0·05 rather than on those with the Clark and Lubs standards at pH0·2 intervals. Since the tubes of a series were kept and compared with each other there is not, however, much risk of their order of magnitude being erroneous.

The salinity naturally varies more at L1 than at the other stations, and the dilution was greatest in the wet winter months. Calculating the range as a percentage of the maximum value the figures are L1, 12·3 ; L2, 8·3 ; L3, 3·2 ; L4, 1·9 ; L5, L6, and E1, 0·6–0·7 per cent. A variation in salinity, of course, affects the titration results for alkali, but not the pH values unless sufficiently marked to alter the salt error.

The titration results are more readily compared when tabulated together as follows :—

C.c. of N/100 H_2SO_4 to neutralise 100 c.c. sea water to phenolphthalein.

Date.	L1.	L2.	L3.	L4	L5.	L6.	E1.
July 4th	2·6	2·5	2·5	2·6	—	—	2·0
Aug. 12th	1·6	2·5	2·6	2·1	2·1	2·4	2·3
Aug. 15th	1·9	2·2	2·3	2·4	2·4	2·2	2·4
Aug. 22nd	2·2	2·5	2·6	2·7	—	—	—
Sept. 1st	2·9	2·9	2·9	3·3	3 3	—	—
Sept. 6th	2·5	2·5	2·7	2·6	2·5	2·8	3·0
Oct. 18th	—	—	2·1	—	2·3	—	2·2
Nov. 9th	—	2·4	2·4	—	—	2·9	2·7
Dec. 12th	1·6	2·0	—	2·4	—	2·6	2·8

The lowest values shown are those for the first part of August and in December, the highest in September.

The extremes are grouped together below :—

	Max. c.c.	Min. c.c.	Difference. c.c.	Max. °C.	Min. °C.	Difference. °C.
L1	2·9	1·6	1·3	16·3	10·0	6·3
L2	2·9	2·0	0·9	16·1	10·1	6·0
L3	2·9	2·1	0·8	16·0	10·3	5·7
L4	3·3	2·1	1·2	16·0	10·3	5·7
L5	3·3	2·1	1·2	15·9	11·1	4·8
L6	2·9	2·2	0·7	16·3	11·2	5·1
E1	3·0	2·0	1·0	16·4	11·2	5·2
	3·1	2·1	1·0	16·15	10·95	5·2

Mean of open sea values (in square).

The open sea values give a range of 1·0 c.c. from July to December, the temperature range corresponding being 5·2° C. Since over the range studied 0·1 c.c. corresponds to pH0·01 and the latter change is brought about by alteration in bicarbonate equilibrium due to 1° C., it would be necessary to correct the total alteration in alkalinity, 1·0 c.c. for this, to arrive at a measure of the amount due to photosynthesis. Thus part of the fall in alkalinity and pH value which occurs in winter is a pure temperature phenomenon, more carbon dioxide being retained in the colder water. Almost equally great changes occur, however, between August and September, though the water was slightly warmer in the latter month. It appears, therefore, that 1·00 c.c. of N/100 acid may be taken as a reliable minimum value for the photosynthetic changes occurring in sea water between July and December inclusive. That it is only a minimum value is evident from the fact that two processes are at work in opposite directions, namely, photosynthesis and plant and animal respiration. In addition to this, since the sea has a higher pH value than that attained by its water when in equilibrium with the carbon dioxide of the air, it is steadily absorbing this gas, which tends to lower the pH value. The additional aeration taking place in stormy weather also lowers the pH value, as will be mentioned later on. This furnishes the surface of the sea with carbon dioxide more rapidly than does the respiration of the plankton.

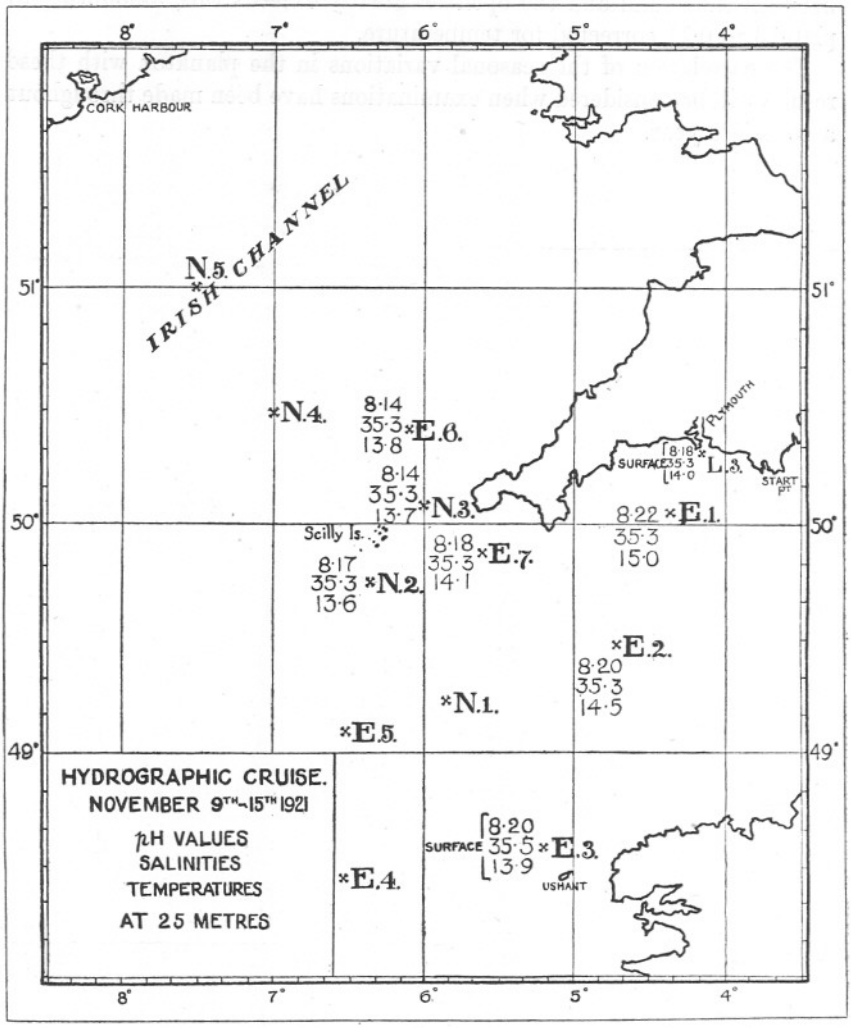
The value 1·0 c.c. of N/100 acid per 100 c.c. sea water corresponds to 0·44 milligrams of carbon dioxide, namely, 4·4 mgrm. per litre, or 1·2 mgrm. expressed as carbon. This amount converted into carbohydrates throughout a large volume of water yields a considerable quantity. The discussion of this will be resumed after considering the results of other stations at sea.

Taking into account only the pH values obtained with the short range, pH0·05 standards and tubes carefully selected for uniformity of bore, it may be seen that the change between November 9th and January 11th amounts to pH0·11 to 0·09 at L4 to E1; the fall in temperature amounted to 4° C., so correcting for this the alteration due to respiration and more complete aeration was at least pH0·07 to 0·05. The difference between the Sound and the open sea greatly exceeds this, amounting to pH0·13 to 0·11 corrected for temperature.

The correlation of the seasonal variations in the plankton with these results will be considered when examinations have been made throughout a complete year.

THE HYDROGEN ION CONCENTRATION OF OPEN SEA WATER AND ITS VARIATION WITH DEPTH AND SEASON.

The stations studied were all in comparatively shallow water, the greatest depth to which the water bottle could safely be lowered being 100 metres.



The alkalinity titrations and pH values were determined, some on the s.s. *Oithona*, but the majority on the s.s. *Salpa* in the course of the routine hydrographic cruises at the seasons arranged by international agreement.

The station E1 is studied monthly, the others five times a year as far

as weather permits, namely, early in February, about the time of maximum density as judged by E1, again six weeks later, in the middle of March, and twelve weeks later, at the end of April, also in June and November. The salinity determinations given in this paper were made in the usual way at the Government Laboratory, London. The June cruise had to be postponed till the second of July. As shown in the accompanying map the stations E1, E2, E3 lie on an approximately S.W. course from Plymouth to Ushant; N1 and N2 are on a line joining Ushant and Cork Harbour, N2 being south of the Bishop Light, Scillies. (The completion of the line, N4 and N5, has been undertaken by the Irish Fisheries Dept.) N3 lies near the Longships, 6° W. between Cornwall and the Scillies; E6 is about twenty miles further north, and E7 is S.E. from the Wolf Light off the Lizard. Of these it may be said that N1 and E2 are at any rate well removed from the effect of the shore algæ upon the hydrogen ion concentration, since the latter is about fifty miles from the coast of Brittany and Devon, though only thirty-five miles from the nearest Cornish headland. N1 is even further out, sixty miles from the nearest point of the French coast, and fifty-three from Cornwall.

The tables which follow give the monthly readings for E1 at the various depths and the readings for the others at longer intervals.

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100H_2SO_4 per 100 c.c. sea water.
E1, April 26th :—				
0	10·18	35·12*	8·24	—
30	9·83	35·11	8·22	—
70	9·82	35·15	8·22	—
E1, July 2nd :—				
0	14·79	35·23	8·17	2·0
30	13·20	35·17	8·12	1·8
70	12·51	35·18	8·12	1·7
E1, August 12th :—				
0	16·13	—	—	2·3
5	16·17	35·10	—	2·3
10	16·13	35·11	—	2·6
15	16·12	35·10	—	2·6
20	15·46	35·14	—	2·2
25	13·34	35·16	—	2·2
30	13·32	35·19	—	—
40	13·30	35·19	—	2·1
50	13·28	35·19	—	2·1
60	13·28	35·17	—	—
70	13·28	35·13	—	2·0

* Denotes duplicate titrations.

E1, September 15th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100 H_2SO_4 per 100 c.c. sea water.
0	15·80	35·13	8 25	—
5	15·76	35·15	8·26	—
10	15·70	35 12	8·25	—
15	15·64	35·13	8·24	—
20	15·55	35·15	8·25	—
25	14·95	35·15	8·25	—
30	14·06	35·16	8·25	—
40	13·86	35·15*	8·24	—
50	13·80	35·14	8·23	—
60	13 80	35·14	8·23	—
70	13·80	35·10	8·22	—

pH determined on following day.

E1, October 18th :—

0	15·55	35·29*	—	2·2
5	15·51	35·33	—	2·9
10	15·50	35·25	—	—
15	15·46	35·31	—	2·6
20	15·46	35·25	—	—
25	15·44	35·30	—	2·6
30	15·43	35·22*	—	—
40	15·42	35·26	—	2·5
50	15·40	35·20	—	—
60	15·38	35·26	—	—
70	15·34	35·26	—	—

Titrations for alkalinity made three days after taking sample.

E1, November 9th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.
0	14·96	35·37*	8·23
5	15·00	35·33	8·23
10	14·96	35·33	8·23
15	15·00	35·41	8·23
20	14·99	35·32	8·23
25	14·96	35·29*	8·22
30	14·98	35·33	8·21
40	14·95	35·32	8·21
50	14·96	35·35	8·21
60	14·96	35·38	8·21
70	14·98	35·28	8·20

* Denotes duplicate titrations.

E1, December 12th :—

Depth in metres.	Temperature °C.	Salinity ‰	pH corrected.
0	12·95	35·40	8·14
5	13·13	35·41	8·14
10	13·13	35·42	8·14
15	13·15	35·41	8·14
20	13·15	35·42	8·14
25	13·15	35·41	8·14
30	13·12	35·40	8·14
40	13·12	35·41	8·14
50	13·13	35·41	8·14
60	13·12	35·40	8·14
70	13·11	35·42	8·12

E1, January 11th, 1922 :—

0	11·24	35·36	8·14
10	11·31	35·36	8·14
20	11·32	35·36	8·14
30	11·32	35·35	8·14
60	11·35	35·36	8·13
70	11·38	35·34*	8·13

E1, February 6th :—

0	9·9	35·33	8·14
5	10·51	35·31	8·14
25	10·52	35·33	8 14
70	10·50	35·33	8·14

E1, February 11th :—

0	9·9	35·36	8·11†

E1, March 29 :—

0	9·7	35·35	8·16
5	9·64	35·28	8·16
10	9·64	35·32*	8·16
25	9·62	35·27	8·16
70	9·62	—	8·16

* Denotes duplicate titrations.

† Examined on 16th, drop of pH0·03 due to keeping, probably in view of identical values for January and February.

E2, July 2nd, 1921 :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100 H_2SO_4 per 100 c.c. sea water.
0	14·98	35·14	8·17	1·9
40	12·31	35·13	8·12	1·7
95	12·30	35·20	8·12	1·7

E2, September 5th :—

0	14·68	—	8·2	2·7
5	14·55	—	—	2·8
10	14·54	—	—	2·9
15	14·50	—	—	2·9
20	14·46	—	—	2·8
25	14·46	—	—	2·7
30	14·40	—	—	2·9
40	14·40	—	—	2·7
50	14·40	—	—	2·8
60	14·40	—	—	2·8
70	14·41	—	—	2·8
80	14·45	—	—	2·7

E2, November 9th :—

0	14·45	35·26	8·20	—
5	14·45	35·28	8·20	2·2
10	14·48	35·32*	8·20	—
15	14·50	35·29	8·20	—
20	14·48	35·29	8·20	—
25	14·50	—	8·20	—
30	14·49	35·40	8·20	—
40	14·50	35·29	8·20	—
50	14·53	35·39*	8·20	—
60	14·53	35·29	8·20	—
70	14·52	35·36	8·16	—
85	14·50	35·29	8·16	—

Titration made two days after taking sample when it was at pH8·16.

E2, February 11th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.
0	10·5	35·36	8·12
5	10·64	35·42	8·14
25	10·62	35·41	8·14
50	10·62	35·45	8·14
85	10·62	35·39	8·14

pH determinations made on 16th, results possibly low by pH0·03, probably less.

* Denotes duplicate titrations.

E2, March 29th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.
0	10·1	35·45*	8·17
25	9·90	35·46*	8·17
85	9·90	35·38*	8 17

E3, July 2nd, 1921 :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100H_2SO_4 per 100 c.c. sea water.
0	14·39	35·22	8·18	2·0
40	12·82	35·27	8·17	2·0
100	12·81	35·35	8·17	2·0

E3, November 10th :—

0	13·9	35·48	8·20	2·5

E3, February 11th :—

0	10·7	35·42	8·11†	—
5	10·76	35·42	8·13	—
25	10·79	35·45	8·13	—
50	10·80	35·44	8·13	—
100	10·80	35·42	8·14	—

pH determinations made on 16th, results possibly low by pH0·03, probably less.

E3, March 30th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.
0	9·9	35·41	8·17
25	10·20	35·41	8·17
100	10·16	35·38	8·17

N1, July 3rd :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100H_2SO_4 per 100 c.c. sea water.
0	15·50	35·29	8·22	2·2
40	11·10	35·22	8·17	2·0
100	11·10	35·25	8·17	2·2

N1, February 12th, 1922 :—

0	10·5	35·37	8·09†	—

* Denotes duplicate titrations. † Examined on 16th.

N1, March 30th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100 H_2SO_4 per 100 c.c. sea water.
0	9·6	35·25	8·16	—
25	9·80	35·21	8·16	—
100	9·83	35·25	8·16	—

N2, Ju'y 3rd :—

0	16·88	35·26	8·22	2·3
27	14·12	·22	8·22	2·2
32	12·07	·16	8·17	2·1
40	11·65	·17	8·17	2·0
95	10·90	·11	8·16	2·0

The break between 27 and 32 metres was examined for pH with α-naphtholphthalein and phenolphthalein as well as the usual cresol red. The difference was estimated at pH0·05 with the first and last named, and pH0·03 with the second. The difference between top and bottom was judged to be pH0·06 with α-naphtholphthalein, and pH0·05 with the other two indicators.

N2, November 12th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100 H_2SO_4 per 100 c.c. sea water.
0	13·43	35·28	8·14	—
5	·52	·32	8·14	—
10	·55	·33*	8·16	—
15	·55	·30*	8·16	—
20	·56	·26	8·17	—
25	·57	·28	8·17	—
30	·56	·26	8·17	—
40	·52	·24	8·17	—
50	·52	·24	8·17	—
60	·45	·25	8·16	—
75	·30	·31*	8·16	—
90	·32	·26	8·16	—

N2, February 12th, 1922 :—

0	10·2	35·21	8·12†	—
5	10·23	35·23	8·14	—
25	10·25	35·20	8·15	—
85	10·26	35·20	8·14	—

* Denotes duplicate titrations.

† Examined on 16th, results possibly low by pH0·03, probably less.

N2, March 30th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.
0	9·5	35·20	8·15
85	9·37	35·19	8·15

N3, November 12th :—

0	13·70	35·32	8·14
25	13·67	35·26	8·14
65	13·68	35·29	8·14

The usual intermediate depths were done, but gave identical readings for temperature and pH.

N3, February 12th, 1922 :—

0	9·4	35·23	8·10

pH determined on 16th.

N3, March 30th :—

0	9·7	35·22*	8·15
65	9·35	35·17	8·15

E6, November 13th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100 H_2SO_4 per 100 c.c. sea water.
0	13·80	35·28	8·14	—
30	13·83	35·28	8·14	—
83	13·83	35·27	8·14	—

E6, March 30th :—

0	9·1	35·17	8·14	—
70	9·30	35·16	8·14	—

E7, July 4th :—

0	15·90	35·24	8·25	2·30
40	14·28	35·28	8·17	2·28
45	13·27	35·26	8·21	2·22
70	11·98	35·28	8·17	—

Taken in order 70, 40, 45, 0 metres. Note undulating upper surface of warmer more alkaline layer. Values for pH at 40 and 70 metres identical with two indicators, for 45 metres pH0·02 to 0·04 lower than surface.

* Denotes duplicate titrations.

E7, November 12th :—

Depth in metres.	Temperature °C.	Salinity ‰.	pH corrected.	c.c. of N/100 H_2SO_4 per 100 c.c. sea water.
0	14·10	35·25	8·13	2·1
5	·12	·27	8·15	—
10	·10	·23*	8·16	—
15	·10	·26	8·16	—
20	·12	·27	8·18	—
25	·12	·25	8·18	—
30	·12	·26	8·18	—
40	·10	·24*	8·18	—
50	·10	·25	8·16	—
60	·12	·26	8·15	—
70	·15	·28	8·15	2·6 ?
E7, February 12th, 1922 :—				
0	9·9	35·29	8·10†	—
5	10·10	35·26	8·12	—
25	10·18	35·31	8·14	—
65	10·18	35·28	8·14	—

The results obtained for pH values and alkalinity titrations show that there is normally a decrease of pH0·05 to 0·01 from top to bottom, and about 0·3 c.c. diminution in the amount of acid required to neutralise to phenolphthalein. This is quite in agreement with the results obtained by Palitzsch. Sometimes, however, the water at 5–10 metres appears to be considerably more alkaline than at the surface, cp. E1, August 12th, October 18th, and possibly E2, September 5th. Other instances in which this was thought to occur have not been recorded, owing to the previously mentioned uncertainty in the colorimeter work. However, Helland-Hansen and Gaarder have obtained similar results with masses of water in a comparatively undisturbed condition, subjected to the action of plankton algæ in sunlight.

Sometimes, however, the surface water is undoubtedly less alkaline than the deeper layers at 20–40 metres, beyond which the gradient is in the normal direction. This is illustrated by the results of the cruise of November 9th. Here E1 and E2 were almost uniform in temperature from top to bottom, but showed normal gradients in pH value, from pH8·23 to 8·20 and pH8·20 to 8·16 respectively. E3, surface, was also at pH8·20, but it was impossible to obtain the usual series of deep water samples as the weather became too rough. It was necessary to run to Falmouth for shelter, and on emerging early on November 12th a great change was observed.

* Denotes duplicate titrations. † Determined on 16th.

The stations were taken in the order E7, N2, N3, and E6. Station E7 had a reversed gradient, pH8·13 to 8·18, followed by a normal gradient, pH8·18 to 8·15. N2 gave similar results, the gradients in both cases being less steep. Neither N3 nor E6 showed any gradient at all. These changes appear to be due to the aeration of the surface layers of the sea, resulting in absorption of carbon dioxide, and a marked lowering in the pH value. Owing to the mixing the less alkaline water is carried down and decreases the alkalinity of the deeper layers by an amount depending on the extent of the mixing. The February 11th cruise showed similar reversed gradients in pH values.

With regard to the seasonal photosynthetic changes in alkalinity, the results given by the mid-channel station, E2 for July and September, show as mean values 1·8 c.c. and 2·8 c.c. acid per 100 c.c. sea water for neutralisation. These give as difference 1·0 c.c., which is just what the mean maximum and minimum values for L4–E1 inclusive gave, though the actual values were higher, 2·1 c.c. to 3·1 c.c. The probability is that the variation is even greater when the whole year is considered. The figures obtained from the July and November cruises are not helpful in judging this change as the autumn alkalinity maximum has been passed apparently, though the pH values for E1 were still high. Attempts will be made to obtain data bearing on the vernal maximum which Moore, Herdman, and Prideaux showed to be so important.

The value 1·0 c.c., however, corresponds as previously explained to 4·4 milligrams per litre of carbon dioxide, or 1·2 mgrm. expressed as carbon. This is a well-established minimum value for the open sea in the English Channel between July and December. For the harbour water a larger change, up to 1·3 c.c., was noted. Taking the open sea figure, 1·2 mgrm. of carbon, since a sugar such as dextrose contains 40 per cent of this element, the amount suffices to provide 3 mgrms. per litre of dextrose, or slightly more than that amount of starch. This is equivalent to 3 grams of dextrose per cubic metre of water, or taking the depth of the Channel off Plymouth as 80 metres the total amount of carbohydrate expressed as dextrose synthesised in the column of water with a base of one square metre is 240 grams. Taking the depth as 83·3 metres, which is probably as accurate as 80 metres,* the amount synthesised is 250 grams, or one kilogram per four square metres. Over an area of one square kilometre this amounts to 250,000 kilograms.

Were photosynthesis to remain uniform and respiration in abeyance it is obvious that there would be a vast accumulation of carbohydrate in the sea. As it is the amount present is an equilibrium between the production due to photosynthesis and the destruction by plant and animal respiration. Under conditions which limit respiration, viz. by ensuring

* The depths at E1, E2 and E7 are respectively 74, 94 and 78 metres, mean 82.

the absence of animal life and in the presence of abundant supplies of necessary salts, as for example in a pure culture of diatoms, the amount of carbohydrate photosynthesised, as judged by the alkalinity, is greatly in excess of that actually found in the sea ; in good illumination the water of the culture is maintained in a strongly alkaline condition, at about pH9·4. This, however, is abnormal. The conditions in nature are regulated by the illumination, for with increasing sunlight in these latitudes the effects of photosynthesis becomes noticeable in the altered reaction of the water in spring. It might be expected, therefore, that on the score of more intense illumination, life ought to be maintained in greater abundance in the warmer than in the colder regions of the oceans. The reverse is by some alleged to be the case. The action of Brandt's denitrifying bacteria has been invoked to explain this, the small amount of nitrate present being considered to act as a limiting factor. It has, however, been shown by Moore, Whitley, and Webster (1920) that algæ may themselves fix gaseous nitrogen, or that bacteria normally occurring on their surfaces can do so in sunlight. The more intense illumination of the equatorial oceans ought therefore to be more favourable to plant development, and accordingly to the animal life which depends upon the former. Against this, however, is the fact that in summer the polar regions have a longer period of daylight, but this is counterbalanced by a shortened illumination in winter.

On considering Blackman's work on limiting factors and the quantitative results obtained by Miss Matthaei (1905), it is seen that an increase of temperature may effect a very decided increase in the rate of photosynthesis. If, however, the light intensity is low, a point is reached beyond which a further temperature rise, even when not injurious, is unaccompanied by any rise in assimilation. The amount of light energy available is insufficient to decompose carbon dioxide at a rate in excess of that already attained at the lower temperature. From the results obtained, when light was not the limiting factor, Kanitz (1915) has shown that the van't Hoff rule holds approximately, the ratio being 2·40 per 10° C. rise from 0°–10° and 2·12 from 10°–20°. For respiration similar coefficients are given, so it is only when light is a limiting factor that increase of temperature is unattended by increase in assimilation as well as in respiration. It appears that in the sea light must often be a limiting factor at anything more than a relatively small depth. Under such conditions the full effect of increase in temperature would be exerted in increasing respiration, whilst assimilation would not increase. Accordingly the amount of carbohydrate actually in the sea would diminish. It seems to be quite possible that the ultimate limit to the weight of living organisms found in unit—a large unit—volume of water at any mean temperature depends upon the quantity of carbohydrate available in that

volume ; the level at which the latter stands must be regulated by the relative rates of photosynthesis and respiration. Since the temperature only fell 3° C. between August and December it is scarcely necessary to correct this factor, but the full correction, assuming the temperature extremes to correspond with the alkalinity extremes, is 0·3 c.c., reducing the result by 30 per cent.

In order the more easily to compare the pH values of the various stations at different seasons the results for the surface, or in some cases for 5 metres, have been tabulated together. In reviewing these it must again be pointed out that, as previously explained, there may be a small error in the results prior to November, 1921, and the determinations are less accurate, being interpolated over a larger range, pH0·2 instead of pH0·05. The values at any time, the July cruise for example, are believed to be strictly comparable *inter se*.

pH values of the surface sea water at the stations, 1921–22.

	E1	E2	E3	N1	N2	N3	E6	E7
April	8·24	—	—	—	—	—	—	—
July	8·17	8·17	8·18	8·22	8·22	—	—	8·25
August	8·27	—	—	—	—	—	—	—
Sept.	8·25	—	—	—	—	—	—	—
Nov.	8·23	8·20	8·20	—	8·14	8·14	8·14	8·13
Dec.	8·14	—	—	—	—	—	—	—
Jan.	8·14	—	—	—	—	—	—	—
Feb.	8·14	8·12*	8·13*	8·09*	8·12*	8·10*	—	8·10*
Range	0·13	0·08	0·07	0·13	0·10	0·04	—	0·15

As far as these incomplete data permit of the drawing of any conclusions it appears that the more westerly stations have a higher pH value than the more easterly in July. The reverse is the case in November, though the effects of the storm seem to have modified the latter results very considerably as already noted. In February, even allowing for the necessary correction, there appears to be a slight decrease in a westerly direction.

With regard to the seasonal range in pH values, owing to the limited number of observations only those for E1 should be considered as truly representative, the value pH0·13 which results is equivalent to 1·3 c.c. of acid per 100 c.c. of sea water. This is somewhat reduced if corrected for the change in the equilibrium brought about by alteration in temperature, a difference between August and December of 3° C. brings the figure to pH0·10 as due to the photosynthetic changes alone, this is equivalent to 1·0 c.c. of acid.

* These determinations were made on 16th, samples taken on 11th and 12th. The corresponding E1 sample gave pH8·11, but one on 6th and examined at once gave pH8·14, so it appears that there was a small decrease, not exceeding, and in some cases not as much as, pH0·03, owing to the samples having been stored for three or four days.

SUMMARY.

The experimental results obtained seem to warrant the following conclusions :—

(1) The salt error of cresol red for sea water of salinity 35 $^{0}/_{00}$ is pH0·18 when determined with Clark and Lubs standard borate buffer mixtures, and compared with McClendon's set as corrected by his potentiometer measurements. When tightly closed with rubber caps tubes of McClendon's series mixed with cresol red were found to have undergone no measurable change in four months, using toluene as a preservative. The measurements can detect a change of pH0·01, and are believed to be accurate to plus or minus pH0·01. To attain this accuracy the indicator added must be measured with exactness, not by drops.

(2) Sea water may become as alkaline as pH9·7 as a result of very active photosynthesis. This it does in virtue of the presence of magnesium salts, since the limiting pH value of magnesium carbonate is pH10·0, the same as for magnesium hydroxide ; calcium carbonate in the form of pure calcite gives as a limiting value pH9·0.

(3) The salt-water tanks of the aquarium are always less alkaline than the water of Plymouth Sound, the values being respectively pH7·6 and 8·1 on an average. Any lowering of the value for the tanks below pH7·6 denotes abnormal excess of carbon dioxide, and when the decrease amounts to pH0·3, viz. a fall to pH7·3, symptoms of distress may appear among the fishes ; water at pH7·1 is definitely foul and evil smelling. The water round rotting sea weed in a jar may be as acid as pH6·4. Agitation of water from the tanks with air results in the removal of carbon dioxide, and a value close to pH8·0 is attained.

(4) The water of the Sound varies slightly with the state of the tide, a drop of pH0·05 may be observed between high and low water. Over *Laminaria* in shallow water, through which the tops of the algæ appear, the water may be pH0·15 more alkaline than the general mass of water. Rock pools in summer may be at least as much as pH0·25 more alkaline than the Sound water, and up to 3·7 c.c. of N/100 acid may be required per 100 c.c. for neutralisation to phenolphthalein ; they may be over 2·5° C. warmer.

(5) During July the water of the Sound is definitely more alkaline than that of the open sea, a difference estimated at pH0·12 having been observed. From August to January the Sound water is less alkaline than that of the sea, the latter being from pH0·02 to 0·16 greater. These

results are confirmed by the figures given by titration. The pH values are considered more reliable than the latter, since with a carefully measured volume of indicator it is unlikely that the error will be as great as pH0·02, more probably pH0·01, with the McClendon standards at pH0·05 intervals ; the order in a series will always be correct if tubes of uniform bore are compared at the same temperature. On the other hand, it is quite possible to have fluctuating errors of 0·2 c.c. in each titration, which corresponds approximately to pH0·02 over the range studied, using N/100 H_2SO_4 and 100 c.c. of sea water with phenolphthalein.

The lowest value observed for the Sound water was pH8·01 at low water in December. The highest was pH8·29 in July. From 1·6 to 2·9 c.c. of N/100 acid are required to neutralise to phenolphthalein. These figures relate only to the period, July to March.

(6) In the open sea between July and December the pH value varies from pH8·27 to 8·14, the April figure being pH8·24. In the second six months of the year titration as before varied between 2·0 and 3·3 c.c., the mean minimum and maximum being 2·1 and 3·1 c.c. around Station E1. The decrease with depth amounts to about pH0·03, or 0·3 c.c. from surface to seventy metres. Occasionally the change is quite abrupt, within five metres, more usually it is gradual. A storm may reverse the normal gradient by mixing atmospheric carbon dioxide with the surface water. The range at E2 is also 1·0 c.c.

(7) Calculating on the basis of an alteration in titration of 1·0 c.c. of N/100 acid and taking the mean depth of the Channel off Cornwall as 83·3 metres an approximate value, the minimum amount of carbohydrate, as dextrose, photosynthesised is one kilogram per four square metres of surface, or 250,000 kilograms per square kilometre between July and December. It is suggested that the limiting factor for the amount of living matter present in any considerable area may be the amount of light available as energy for photosynthesis. The thorough mixing of the water, as shown by the pH measurements, makes it permissible to calculate the seasonal changes as from bottom to surface, rather than in the surface layers where they originate. These results are subject to a correction for alteration in alkalinity due to rise of temperature ; this as a maximum cannot exceed 0·3 c.c., viz. reducing the values found by 30 per cent. The pH range for E1 was observed to be 0·13, or corrected for the part possibly due to the establishment of a temperature change equilibrium, and it is doubtful that equilibrium is reached, the range is pH0·10. This corresponds to 1·0 c.c. of acid, so the figure 250,000 kilograms given above may stand as an approximate value.

(8) In order that the conditions under which marine animals and plants exist at the various sites examined may be the more readily compared

the follow ng data have been tabulated. The values are, of course, not necessarily the actual maximum and minimum values that occur at the places named.

	pH	Temperature °C.	Salinity ‰
Aquarium tanks, normal	7·6 –7·45	18·9–9·1	36·14–36·85
,, ,, abnormal	8·0 –7·05	—	—
Rock pool	8·57–8·01	21·4–8·2	—
Shallow water	8·42–8·01	19·2–8·2	—
L1, Plymouth Sound	8·29–8·01	16·3–8·2	35·00–30·69
L2 ,, Breakwater	8·27–8·07	16·1–8·8	35·17–32·25
E1, about 20 miles out to sea from Breakwater	8·27–8·14	16·2–9·9	35·40–35·13

The ranges of the above data are shown below.

	pH	Temperature °C.	Salinity ‰
Aquarium, normal	0·15	9·8	—
,, abnormal	0·95	—	—
Rock pool	0·56	13·2	—
Shallow water	0·41	11·0	—
L1	0·28	8·1	4·31
L2	0·20	7·3	2·92
E1	0·13	6·3	0·27

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The Respirable Organic Matter of Sea Water.

By

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With 1 Figure in the Text.

THE photosynthetic activity of algæ results in the conversion of carbon dioxide into carbohydrates. These are available for respiration, and within a limited time all, or almost all, the photosynthetic products of one year are broken down again into carbon dioxide and water through the action of plants, animals and bacteria.

The removal of carbonic acid tends to lower the hydrogen ion concentration of the sea water, namely to increase its pH value. Conversely production of the acid lowers the pH value. By exposing a plant of *Ulva* to sunlight in a jar of sea water a reaction of pH9·7 may be reached, whereas algæ decomposing in a jar in the dark may through bacterial action yield so much carbonic acid as to render the water as acid as pH6·4.

It has long been known that when making accurate determinations of the hydrogen ion concentration of sea water, it is necessary to examine it almost immediately; if this is not done the pH values obtained are too low. It might be thought that this was due to absorption of carbon dioxide from the air. Sea water does absorb this gas if above about pH8·1, for it is in equilibrium with the concentration in the atmosphere at close to pH8·1; consequently sea water in spring or autumn at pH8·2 takes it up. This would not, however, account for the fact that on keeping the water may fall to below pH8. Furthermore, samples are stored either in bottles almost completely filled and tightly stoppered, or in test tubes closed by rubber caps, which greatly reduces the amount of carbon dioxide that can gain access.

The source of the additional carbon dioxide which reveals itself in the lowering of the pH value during storage is accordingly to be sought in the respiration of the plankton algæ and animal organisms, and ultimately, after their death and disintegration, in bacterial action. Bacteria also act upon the waste products of the animal organisms, so it appears

rational to conclude that any sample of sea water which decreases to a marked extent in pH value when stored is rich in respirable organic matter. If stored till the pH value no longer falls the decrease from the original value may be taken as a measure of this in the volume of water examined. It is, of course, possible that resistent spores, etc., may still remain, so the amount indicated is probably less than that given by a method based upon complete oxidation by a powerful oxidising agent such as alkaline permanganate. The method, however, gives results which are comparable *inter se* and is of great delicacy. Moreover, it is a very rapid one as far as actual time consumed in working is concerned, for it is only necessary to store the test tubes already used for the colorimetric determination of the pH values. Since, however, even the very stable indicator cresol red fades slowly under these conditions, though not when toluene is added as an antiseptic, it is more accurate to store the sample in its bottle as obtained, and to examine it afresh in the usual way as described in detail in an accompanying paper on "The hydrogen ion concentration of sea water in its biological relations." Obviously the chance presence of a single relatively large organism may give a result which is erroneous for the water as a whole, but as a rule the contents of a sample bottle, about 170 c.c., seem to be truly representative.

On standing a scum develops on the surface of the tubes which show a decrease in pH value, but it is absent in those which remain constant.

To render the method quantitative it is necessary to ascertain the change in pH value, over the particular range studied, which the addition of a given amount of carbonic acid produces. For convenience dilute sulphuric acid was used, from which the equivalent quantities of carbonic acid were calculated.

It was found that the addition of 1·00 c.c. of N/100 acid to 100 c.c. of sea water at pH8·06 changed the latter to pH7·97. It was not possible to study the change in the less acid region as standards of the same degree of accuracy had not been prepared, but it is hoped to prepare them shortly. It may be taken, therefore, that over this range, and without much error slightly outside it, the addition of 0·1 c.c. of N/100 acid per 100 c.c. of sea water produces a change of pH0·01. To change water from about pH8·2 to the limit for titration with phenolphthalein, viz. about pH7·6, requires only about 2·5–3·0 c.c. of acid, since the higher decimal places of a logarithmic scale are close together, as on a slide rule. Over this more extended range the value is 1·0 c.c. to pH0·2 roughly, viz. about 0·1 c.c. for a change of pH0·02.

Now, since the molecular weight of carbonic acid is 62, and the bicarbonate stage is the limit with phenolphthalein as indicator, M/100*

* When titrating to the bicarbonate stage, carbonic acid behaves as monobasic, hence M/100 is the same as N/100.

carbonic acid contains 0·62 grams per litre, or 0·12 grams of carbon. The combined carbon in 1 c.c. is therefore 0·00012 grams, or 12×10^{-6} grams per 0·1 c.c. But 0·1 c.c. of N/100 acid added to 100 c.c. of sea water at pH8·06 produces a lowering of pH0·01. This is equivalent to adding 0·001 c.c. to each cubic centimetre of sea water, viz. 12×10^{-8} grams of carbon when converted into carbonic acid. A change of pH0·01 in 10 c.c. of sea water under examination corresponds therefore to the addition of 12×10^{-7} grams of carbon dioxide, reckoned as carbon, which is derived from respiratory action when taking place in nature. Some of the more recent measurements were made to $\pm$pH0·01, but the earlier ones were not as accurate, though probably to within $\pm$pH0·05 in a series, or to $\pm$pH0·1 in tubes in which the cresol red had faded through bacterial action.

Water samples were taken for examination at the usual stations, the L series running out from Plymouth Sound to beyond the Eddystone Lighthouse. These and E1 were examined monthly, other stations at longer intervals, as explained in " The hydrogen ion concentration of sea water in its biological relations."

It may be said at once, that in the L series the change on keeping was usually most rapid and greatest at L1, and decreased as the land was left. On looking at the tubes in a row the alterations were very striking, the tubes showing a blue-pink tint at pH8·2 and a yellowish colour at pH7·4.

The changes in the different samples of the depth series at E1 were also obvious, especially during the summer months when there did not appear to be much vertical mixing as indicated by these results and the temperatures.

The samples were tinted with indicator and examined in a colorimeter tube, after which each was returned to its own tube and closed with a rubber cap. Owing possibly to a larger amount of bacterial infection and to the acquisition of traces of dust, it was found that samples which had been examined in the colorimeter changed on keeping more rapidly and to a greater extent than those examined in the test tube only. As this was not noticed for some considerable time the results recorded here are not to be considered as absolute values, but they seem to be comparable. The initial values were in all cases close to pH8·2, but as precise determinations to the second place of decimals they had to be rejected on account of an uncertainty in the colorimeter measurements, as explained in another communication.

Samples of August 12th, originally at pH8·2 approximately. Samples taken noon to 2 p.m.

Station and depth.	pH Aug. 20.	pH Aug. 25.	pH Sept. 2.	Change from pH 8·2 on Sept. 2.	Temperature and salinity of Aug. 12.	
L1, surface	8·0	7·7	7·6	0·6	16·2°	34·61*
L2 ,,	7·9	7·65	7·6	0·6	15·4°	35·01
L3 ,,	8·0	7·75	7·7	0·5	15·3°	35·09
L4 ,,	7·9	7·7	7·7	0·5	15·5°	35·14
L5 ,,	8·0	7·9	7·85	0·35	15·2°	35·31*
L6 ,,	7·95	7·8	7·5	0·7	15·5°	35·10*
E1 ,,	8·2	7·75	7·65	0·55	16·13°	—
E1, 5 metres	8·2	7·95	7·9	0·3	16·17°	35·10
E1, 10 ,,	8·3	8·2	8·15	0·05	16·13°	35·11*
E1, 15 ,,	8·25	7·9	7·8	0·4	16·12°	35·10
E1, 20 ,,	7·9	7·65	7·65	0·55	15·46°	35·14
E1, 25 ,,	8·2	8·05	7·95	0·25	13·34°	35·16
E1, 30 ,,	8·25	8·1	8·0	0·2	13·32°	35·19
E1, 40 ,,	8·25	8·15	8·1	0·1	13·30°	35·19
E1, 50 ,,	8·25	8·15	8·1	0·1	13·28°	35·19
E1, 60 ,,	8·2	8·15	7·95	0·25	13·28°	35·17
E1, 70 ,,	8·2	7·85	7·8	0·4	13·28°	35·13

It was considered that by September 2nd the tubes would change no further, which is only approximately true in all probability. A number of similar determinations were made, but owing to the uncertainty introduced by pouring into the colorimeter these have been held over pending further work.

The results of August 12th are shown graphically in the accompanying figure, as are also those of November 9th, for Station E1 in both cases. The November results appear to be the more reliable, as the samples were measured directly into the test tubes and never withdrawn. Moreover, since the initial values were accurately known, and the McClendon series of standard tubes was used, the results are more accurate. The tubes which changed most, however, were compared with the old standards and are not quite as accurate. The fading of the indicator introduces a small error too. For this reason further work will be done on samples stored without indicator.

Viewing the figures here recorded, and those held back, it is seen that in a general way the surface values tend to be higher than those at five and ten metres. There is often, but not always, a notable increase in the amount of change shown between about 20–25 metres. After this low values are obtained down to about 60 metres, where a rise is found, which increases towards the lowest reading, 70 metres ; the bottom was actually at 74 metres, but the difference has to be allowed on account of the motion of the ship for fear of damaging the water bottle.

* Signifies duplicate titrations.

pH values of sea water from E1, taken November 9th, 11–1 p.m. and examined at intervals.

Depth in metres.	Nov. 9 pH.	Nov. 15 pH.	Feb. 8. pH.	March 3. pH.	Maximum alteration in pH.	Temperature and salinity of water when drawn.	
0_1	8·22	8·03	7·97	7·97	0·25	14·31°	35·35°/$_{oo}$
0_2	8·23	8·03	8·13	8·09	0·20	14·96°	35·37*
5	8·23	8·09	7·9	7·9	0·33	15·00°	35·33
10	8·23	8·06	8·14	8·12	0·17	14·96°	35·33
15	8·23	8·06	8·08	8·08	0·17	15·00°	35·41
20	8·23	8·16	7·7	7·7	0·53	14·99°	35·32
25	8·22	8·14	7·7	7·7	0·52	14·96°	35·29*
30	8·21	8·08	8·13	8·08	0·13	14·98°	35·33
40	8·21	8·09	8·12	8·06	0·15	14·95°	35·32
50	8·21	8·09	8·12	8·04	0·17	14·96°	35·35
60	8·21	8·15	8·01	8·00	0·21	14·96°	35·38
70	8·20	8·08	8·01	8·01	0·19	14·98°	35·28

On examining the foregoing tables it becomes clear that salinity changes have no connection with the alterations in pH value on keeping. The same is true for temperature changes in the column of water.

When the changes in pH value are scrutinised it is seen that they amount to pH0·05 as a minimum and pH0·7 as a maximum, or in the depth series at E1, pH0·55 as a maximum. The range corresponds approximately with 0·5 c.c. to 2·5 c.c. of N/100 acid per 100 c.c. of sea water, or 5 to 25 c.c. per litre. These quantities are equivalent to 0·0006 to 0·003 grams of carbon per litre, or if calculated to the form of carbohydrate, which contains 40 per cent of carbon, to 0·0015 to 0·0075 grams per litre. The amount due to photosynthesis during the second half of the year, as shown in the accompanying paper, is as a minimum 3 milligrams per litre.

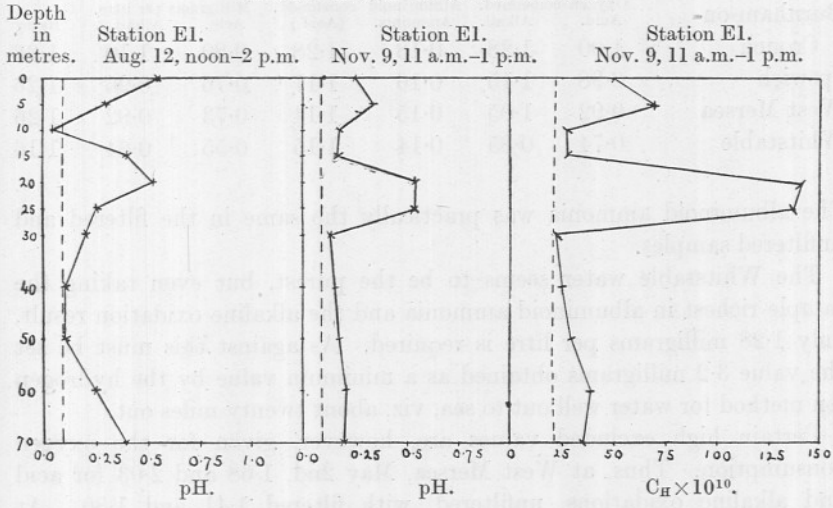
This corresponds to an alteration of 1·00 c.c. in the amount of acid required to bring 100 c.c. of sea water to the bicarbonate stage, or very approximately to pH0·1 at around pH8 and to pH0·2 at about pH7·7. Were there an exact proportionality between the change in pH or C_H values and the volume of acid added it would be permissible to compare the areas of the curves shown in the figures by the whole and dotted lines. Since, however, the change is more rapid at the values below pH8, the right-hand areas of the whole line curves do not exactly correspond with equal increments of added acid. Pending the preparation of a curve showing, more accurately than has been done so far, the relation between the two it is permissible to say that almost all the samples show a greater change on storing than could be accounted for by the decomposition of

* Signifies duplicate titrations.

three milligrams per litre of a hexose sugar. In fact, the amount seems to be between once and a half and twice as great, probably nearer the former value. For complete oxidation three milligrams per litre of a hexose would require 3·2 milligrams of oxygen.

Organic matter in water is usually estimated by means of permanganate solutions; the solutions used are sometimes acid, sometimes alkaline, and the temperatures and times of experiment are also diverse. The results are, therefore, only strictly comparable if carried out under identical conditions. Furthermore, carbon in nitrogenous compounds

CHANGES IN SEA WATER ON STORING.



Decrease in alkalinity in terms of pH. Ditto in terms of C_H.

The dotted line shows the change produced by adding 1·00 c.c. of $N/100 H_2SO_4$ to sea water, at pH8·06, 100 c.c.

Area of curve (Aug.)	Area of curve (Nov.)	Area of curve (Nov.)
660 units.	580 units.	1,240 units.

Area of dotted curve, 280 units.....630 units.

is more resistant to oxidation than that in carbonaceous organic matter. The official method of the American Public Health Association specifies boiling for thirty minutes with an acid solution. The methods used in the Government Chemist's Laboratory, London, are (a) treatment in an acid solution for four hours at 80° F. and (b) boiling for ten minutes in an alkaline solution. It is customary to use 100 c.c. of the sample, or a convenient amount diluted to that volume, and permanganate of which 1 c.c. is equivalent to 0·1 milligram of available oxygen. The writer is indebted to Dr. J. H. Orton for drawing his attention to certain unpublished results of analyses carried out by the Government Chemist on

samples taken from estuarine oyster beds by Dr. W. Wallace, by whose courtesy the following estimations are quoted, since no direct determinations of the oxygen consumed by organic matter in sea water at Station E1 have as yet been made.

The results are shown in the following table, based on averages for seventy-six samples, and excluding twelve with abnormally high results. The samples were of surface water, taken at various times throughout the year.

	Unfiltered Samples. Milligrams per litre.			Ratio, Oxygen consumed. (Alkali.) Oxygen consumed. (Acid.)	Filtered Samples. Milligrams per litre.		
Burnham-on-	Oxygen consumed. Acid.	Alkali.	Albuminoid Ammonia.		Acid.	Alkali.	Ratio.
Crouch	1·00	1·28	0·18	1·28	0·80	1·10	1·38
Ipswich	0·98	1·15	0·16	1·17	0·76	0·96	1·26
West Mersea	0·92	1·05	0·15	1·14	0·73	0·92	1·26
Whitstable	0·74	0·85	0·14	1·15	0·55	0·64	1·16

The albuminoid ammonia was practically the same in the filtered and unfiltered samples.

The Whitstable water seems to be the purest, but even taking the sample richest in albuminoid ammonia and the alkaline oxidation result, only 1·28 milligrams per litre is required. As against this must be set the value 3·2 milligrams obtained as a minimum value by the hydrogen ion method for water well out to sea, viz. about twenty miles out.

Certain high excluded values are, however, given for the oxygen consumption. Thus, at West Mersea, May 2nd, 1·68 and 2·03 for acid and alkaline oxidations, unfiltered, with filtered 1·41 and 1·80. At Whitstable, March 12th, 2·98 and 3·13, with filtered 1·36 and 1·50. At Burnham, February 20th, 4·44 and 5·61, with filtered 3·94 and 5·14. It might be thought curious that the results for unfiltered water should be so little higher than those for filtered, but many plankton organisms, such as minute diatoms and flagellates, pass through ordinary filter paper. It may be noted that even at E1 with water close to pH8·2 a fall to pH7·7 was observed, corresponding to about 2·5 c.c. of acid, to 7·5 mgrms. of a hexose, and consequently to 8·0 mgrms. of oxygen.

There is certainly a considerable discrepancy between the results for oxygen consumed as determined by the permanganate methods, and by changes in pH values. The latter have been calculated on the assumption that a hexose is oxidised, and accordingly are minimum values, for fats and other carbonaceous substances, except organic acids, are less highly oxygenated than hexoses.

In the absence of direct comparisons on the same sample of water, it would be unwise to go in detail into the causes of the discrepancy,

but it appears that the permanganate results may be too low owing to the fact that they correctly indicate the relative magnitudes of the quantities of oxidisable organic matter in the water at the time the estimation is made. These amounts are certainly not those in the water when the bottles were filled, if, as is usually the case with samples sent away for analysis, several days necessarily elapse, and possibly several weeks, between collection and estimation. This is shown by the fall in the pH value which, in the case of the L series for August 12th, as recorded previously, decreased by as much as pH0·3 in eight days or pH0·5 in thirteen days, after which it changed but little. Analysis after this period would obviously show only the oxygen consumption of the matter which could not be completely oxidised by the organisms present and under natural conditions. It is, in fact, comparable to estimating a solution of glucose sown with yeast after several days' fermentation and taking this as a measure of the original concentration. More accurate results would most likely be obtained by the permanganate method applied to fresh samples, or by adding the amounts indicated by the fall in pH value to those for the residual matter as found by permanganate after storing.

In any case it seems that the very considerable differences shown by water from various stations and at different depths at the same place are to be taken as a measure of the amount of the minute plankton present. For this reason the storing of a bottle of water seems preferable to the storing of 10 c.c., as the bottle will contain copepods, which are unlikely to be drawn up into the pipette, yet three or four per bottle increase the amount of organic matter considerably. It is hoped that the matter may be investigated more fully.

Since the foregoing paragraphs were written analyses of organic matter in sea water made by Raben (Wissensch. Meeresuntersuch., XI, Kiel, 1910, 111–117) have come to the writer's notice. The sea water was filtered through a Berckefeld filter and was analysed at once, also two days later, the latter appear to give lower values within the limits of error. The results are given in organically combined carbon, which may be converted into hexose by the factor 2·5. For the Kiel Ford values of 13·9–11·4 milligrams per litre of carbon were obtained, for a Baltic Sea station 3·0 mgrms. This corresponds to 7·5 mgrms. of a hexose, agreeing well with some of the E1 figures, although for completely filtered water.

SUMMARY.

1. On storing, sea water suffers a decrease in pH value. The amount of change varies from that produced by adding 1·0 c.c. of N/100 acid to 100 c.c. of sea water, up to that due to adding about 2·5–3·0 c.c. The decrease is due to the production of carbonic acid by organisms.

2. The change corresponding to 1·0 c.c. of acid, as above, is equivalent to that produced by the complete oxidation of 3 milligrams per litre of a hexose sugar, which requires 3·2 mgrms. per litre of oxygen. This is the minimum value, from 8·0–9·6 mgrms. corresponds to the higher values of acid. Figures for oxygen consumption by estuarine waters, which are available for an approximate comparison, show that 1·5 mgrms. of oxygen is not often exceeded in estimations by means of alkaline permanganate. The highest of this series is 5·6 mgrms. It is suggested that the different results given by the two methods are due to the fact that respiratory changes taking place in the water during storage set free much of the organically combined carbon before the oxidation by permanganate has been started. Determinations on freshly drawn filtered sea water give, according to Raben, 7·5 mgrms. of hexose or 8·0 mgrms. of oxygen consumed.

3. It is probable that the change in pH value on storing indicates the amount of plankton present, at any rate when sewerage contamination is negligible. It appears that water near the surface, at 20–25 metres and sometimes at the bottom, 70 metres, is particularly subject to change during storage. Four cases out of seven showed marked decreases in pH value at 20–25 metres, and two others exhibited the change to a less marked degree.

4. The total amount of carbon, reckoned as hexose, which is set free during storage by respiration in sea water at E1 is about twice that photosynthesised between July and December, taking for this the minimum value 3 mgrms. per litre, namely a total of 6 mgrms. per litre. Considering the column of water from bottom to surface, this is equivalent to about 500,000 kilograms per square kilometre in the English Channel off Plymouth.

Di Brom Thymol Sulhpone Phthalein as a Reagent for Determining the Hydrogen Ion Concentration of Living Cells.

By

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In work of a physiological or cytological nature it is desirable to ascertain whether the processes and appearance of the dead cell are truly representative of the condition in the living. This is especially true when dealing with the hydrogen ion concentration in a small organism which is of necessity accompanied by a considerable quantity of the medium in which it lives. Haas (1916, 4) accordingly ascertained the reaction of various coloured plant cells, such as those of petals with naturally occurring anthocyan indicators by noting the tints, and changes in tint, seen in living cells and comparing these with the colours given when the various anthocyans were added to buffer solutions of known concentration.

Harvey (1911, 1914), too, sought for indicators suitable for use in living tissues, and made use of neutral red, which had previously been of service to Bethe (1909) in similar work on permeability. Finding it impossible to stain living cells with any other dye which would act as an indicator for acid, Harvey (1915) studied penetration of acids in the tissues of a holothurian *Stichopus ananas*, certain cells of which have a dark red pigment which becomes orange in dilute acid, N/1000 to N/500 HCl, viz. at about pH3.

Extensive use has been made by Crozier (1912–1919) of the blue pigment found in a Bermudan Nudibranch, *Chromodoris Zebra*, which changes from blue to pink at pH5·6 in presence of sea salts as found naturally, at $^5/_8$ M total concentration. From the fact that the animal is normally blue it may be concluded that its reaction is less acid than pH5·6.

Certain sponges were also found by Crozier to show indicator changes from yellow to blue, from scarlet to brown-yellow and from colourless to green as acidity decreased in the respective varieties. Similar properties were exhibited by the pigments of a colonial hydrozoan and of various holothurians. He concludes that the tissues of marine animals are in

general more acid than the surrounding sea water, since the pigments appeared in the animals studied to denote reactions lying between pH6·0 and pH7·6.

Heidenhain (1907) and Ehrlich (1910) have reviewed the subject of the staining of the living cell, and the last edition of Lee (1921) mentions no addition to their lists of useful reagents for this purpose.

By comparing these lists with those given by Clark (1920) as indicators selected by Sörensen and other workers, the following are found to combine both properties :—

Substance.	Range pH.	Notes.
Methyl violet, 6B.	0·1–3·2	Slight penetrating power
Methyl orange	3·1–4·4	
Congo red	3–5	Rejected by Sörensen as indicator
Lackmus (lacmoid)	4·4–6·2	
Neutral red	6·8–8·0	Penetrates very rapidly. Has also a blue-red change in 2N to N acid
Cyanin	7–8	
Hæmatoxylin	0–1·0 and 6·0–11·0	
Alizarin	10·1–12·1	Very unreliable indicator

For work on living cells some substances in the list may be suitable as stains, but not as indicators, since they are outside the range to be studied. Neutral red is, however, available, and its changes in the Clark and Lubs standard buffer mixtures were studied. From pH6·6–7·0 it gives a good clear red, decreasing in intensity ; at pH7·2–7·4 it is reddish, at pH7·6 it is a dirty reddish, and at pH8·0 it is orange-red, beyond which it becomes more yellowish. Thus it may be seen that around the neutral point the changes are not such as to enable one to judge accurately the decimal points of the pH values in a tissue, though pH8 may be distinguished from the neighbourhood of pH7. That is to say, it is possible to judge whether the reaction of a cell or tissue is closely the same as that of sea water, pH8·2 approximately, or whether it has a reaction which is perceptibly different. That the latter is the case may be seen at once when organisms such as *Pleurobrachia pileus*, *Clytia Johnstoni*, medusa stage, and *Tiara pileata* are placed in dilute neutral red in sea water. Even when so dilute as to be imperceptible in a green glass jar the stain is taken up with remarkable rapidity, so that in five or ten minutes the bases of the tentacles, the canals and other structures are shown up with beautiful vividness in red. The tint appears to be in the neighbourhood of pH7, but beyond that it is not possible to judge. Lightly stained specimens remained actively motile in jars in the laboratory for three days, more deeply

stained specimens lived actively for a day, so the reaction indicated is that of the normal state.

In order to define the reaction more precisely the newer indicators selected by Clark and Lubs were tried. No trace of di-ethyl red was taken up even from a bright yellow solution, nor was cresol red able to penetrate when colouring the water deeply. *Pleurobrachia* and *Clytia* lived for two days in the former stain and one in the latter without showing any effects of toxic action.

Clytia was tried also in brom thymol blue, drops of a 0·02 per cent solution being added to sea water to produce a faint but quite perceptible blue tint. The indicator penetrated slowly and the manubrium, ocelli, and bases of the tentacles became a light green, judged to be pH6·6. The specimen was lost in course of transference to fresh sea water to observe the general staining. Experiments were continued on *Tiara*. When in light blue indicator for sixteen hours no trace of colour was taken up, as judged by examination in sea water without indicator. On increasing the indicator till a deep blue was produced and allowing a further sixteen hours to elapse the specimen was seen to be light yellowish green pH6·4, or possibly 6·2, in the circular canal of the mantle and in the tentacles, which were still actively motile. The umbrella was a good light blue, pH7·2 or over, but as the indicator changes only in intensity from this onwards to pH7·6, it was not possible to affirm that the reaction was not more alkaline than pH7·2. Experience with neutral red though shows that the reaction is nearer pH7 than pH8. It is possible that the mantle was pathologically permeable when blue, as medusæ may be observed pulsating even when the mantle has largely disintegrated.

Brom cresol purple was also tried, as a stain for *Vorticella*, but though non-toxic in the concentrations used it failed to penetrate. Neutral red, however, stained it deeply, the colour indicating pH7, or probably rather less. These deeply stained specimens were actively motile after sixteen hours, when observations were discontinued.

SUMMARY.

1. Brom thymol blue may be used in dilute solution for ascertaining the hydrogen ion concentration of certain marine organisms. It penetrates slowly, but the stained portions remain actively motile, so its toxic action does not appear to be great at the dilutions found serviceable.

2. The animals studied gave values from pH6·2 to about pH7·5, though possibly the more alkaline end of the range may be pathological. About pH0·2 should be subtracted from these figures for neutral salt error. The sea water used was initially at pH8·2, corrected.

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The Hydrogen Ion Concentration of the Cells of some Marine Algæ.

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In a previous paper (Atkins, 1922) reasons were given to show how desirable it is to have precise information concerning the reaction of plant cells, as reaction regulates enzyme action. Details were also given of the application of the colorimetric method to such determinations, using either sections or drops of expressed sap. The standard solutions were those of Clark and Lubs (1917). *Intra vitam* staining has also been of service in certain cases.

The tissues of land plants are of very variable reaction from species to species and from tissue to tissue ; they are rarely alkaline, up to pH8, but may be as acid as pH1·4 ; more generally, however, they are in the neighbourhood of pH5–6 ; habitat probably has a large influence on the reaction, though liming slightly acid soil has only a very small effect, if any, upon the reaction of the sap of the crop (Kappen, 1918 ; Truog and Meacham, 1919 ; Clevenger, 1919). But little work appears to have been done upon the acidity of algal cells. Estimations of total acidity were made by L. Clark (1916) by titrating expressed sap with N/50 sodium hydroxide, diluting the sap with neutral sea water, four volumes to one of sap. Phenolphthalein and alizarin were used as indicators, thus with the first the pH value to give a pink colour was about pH8·1, correcting for the salt error, and with the second about pH9. The latter is, however, a thoroughly unreliable indicator. These titrations give a measure of the buffer action of the saps to an arbitrary alkaline limit, but not of the pH values. Stated in percentages of normal acid, fairly usual values seen to be 0·3–0·7 per cent. Values as low as 0·1 and as high as 3·1 per cent were, however, reached. It is possible that the sap expressed was not in every instance truly representative of that in the cells, but the results of Dixon and Atkins (Atkins, 1916) on *Ascophyllum nodosum* show that the error from this source is probably very small, though large in many land plants. It may be added that the low molecular weight, 35–45, for the expressed sap of the above-mentioned alga on April 6th, shows that its osmotic pressure is chiefly due to salts, at least at this

season. Since its freezing-point depression is slightly greater than that of sea water around it, 1·988° as against 1·870°, the salt error for colorimetric estimation of pH values may be taken as 0·18, the sea water freezing-point corresponding to a salinity of 34·6°/$_{oo}$.

The only measurements of the pH value in algal cells which the writer has as yet found in the literature are those of Crozier (1919) on *Valonia macrophysa*, from the vacuole of a single cell of which several cubic centimetres of liquid may be obtained. Fifty measurements showed that the reaction varied from pH5·0–6·7, average 5·9, though the water around was at pH8·1–8·3, and even in an aquarium jar as high as pH9·5. Values such as pH7·0–8·0 when found were always accompanied by the presence of sulphate in the sap, which, as demonstrated by Wodehouse (1917), is an indication of a pathological increase in permeability. Crozier does not mention having corrected for salt error. In the absence of evidence as to the agent causing the osmotic pressure in the species or genus, this correction is a doubtful one, for the preliminary results of Lapicque (1921) indicate that the ratio of chloride to soluble carbohydrate in the sap varies with the season, the latter increasing in the summer.

In the measurements which follow care was taken to wash the thallus rapidly in changes of fresh water before crushing, or to wash and excise portions of the interior with a stainless steel knife. The values given are not corrected for salt error, for which, however, at least pH0·1 should be subtracted, possibly pH0·18. All the plants were growing on the beach below the Laboratory in sea water at about pH8·2, towards the end of April.

Alga.	Where examined.	pH.
Laminaria digitata	Interior of stem of holdfast, cut out	7·3
,, ,,	Frond, crushed	7·3
L. saccharina	Interior of stem of holdfast, cut out	7·3
,, ,,	Young thallus	7·2–7·3
Fucus platycarpus	Medullary tissue of receptacle, cut out	7·2
Himanthalia lorea	Liquid from disc, 3·5 cm. diam., perfectly clear and colourless	6·9
,, ,,	Strap, near disc	6·6
,, ,,	Strap, near end	6·6
Nitophyllum sp.	Thallus	7·3
Ceramium rubrum	Large plant, crushed	7·2
Ulva latissima	Thallus crushed	7·0

The plants appeared quite healthy and were examined within a few hours of having been collected.

Were it possible to get indicators into living cells the pH values could be ascertained without any suspicion that the death of the cells had

appreciably altered them. Unfortunately most of the indicators fail to penetrate, save neutral red, and to a much lesser degree brom thymol blue, as mentioned in an accompanying note.

On examining in watch-glasses on white paper, neutral red was found to give a good clear red diminishing in intensity from pH6·6–7·0 ; from pH7·2–7·4 it was reddish, at pH7·6 a dirty reddish and orange-red at pH8·0. Owing to the fact that it not only penetrates plant and animal cells, but accumulates in the living cell, it is of especial value as an indicator, the more so as its salt and protein errors have been shown to be low.

The cells of the filament of the diatom *Skeletonema costatum* were rapidly penetrated, a good red colour being produced. This indicates a pH value in the region of pH7, or a more acid value. The colour changes to orange in dead cells, indicating that these are in reaction close to that of the sea water. It was observed that, if a large drop of the indicator, dissolved in fresh water, was added to the sea water around the diatom filament under microscopic observation, the cells swelled up and the valves or frustules suddenly popped apart in succession along the filament. This illustrates well the behaviour of a plant cell in a hypotonic solution, for ordinarily the distension is limited by the resistance of the cellulose wall.

Similarly in *Ulva* the cells become a pink with this indicator, and with *Enteromorpha* dilute neutral red causes a faint pink coloration, which collects and becomes intensified into a reddish purple in two to eight granules per cell. With stronger concentrations the permeability of the cell is evidently altered, for the cell becomes orange or brick-red at first, denoting a pH value of about, or over, 7·5. Then the colour collects in the granules, which still show the red-purple of acidity. On the death of the cells the processes are reversed, and the dead cells no longer retain the indicator in greater concentration than the external sea water. It appears that this selective absorption may be due to the fact that neutral red only dissolves in sea water, which is alkaline, with difficulty, whereas it is many times more soluble in distilled water, which is acid, like plant sap.

Ceramium rubrum was also observed to give a good red with neutral red, indicating a reaction of about pH7 or less in the sap. Dead cells gave tints with brom cresol purple and brom thymol blue, indicating about pH6·8 ; these indicators do not penetrate the living cells. The observations are, however, probably not quite as accurate as those with phenol red and the crushed plant, which showed pH7·2. Quite possibly the plants differed, as did *Valonia*, from plant to plant.

SUMMARY.

The measurements recorded for marine algæ of various groups show that the reaction of the sap is in most cases almost neutral, and in no case is the sap of the pronounced acid character met with in many land plants. This being so it follows that the enzymes concerned in the metabolism of these algæ must be quite different from those which effect corresponding changes in land plants, as may be seen on referring to the optimum pH values for various enzymes quoted in the writer's previous paper on the reaction of plant cells (1922).

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* References to be found in Clark's (1920) list have been omitted here.

The Influence upon Algal Cells of an Alteration in the Hydrogen Ion Concentration of Sea Water.

By

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When studying the hydrogen ion concentration of algal cells it was thought desirable to see whether an increase in the alkalinity of the water had any influence upon the reaction of the cell sap. With normal sea water at pH8·2 the algæ examined were at about pH7. When subjected to an approximate correction for salt error *Ulva latissima* L., was at pH6·9 and *Ceramium rubrum* at pH7·1.

It had previously been ascertained that when *Ulva* was exposed to sunlight in a jar of sea water it abstracted so much carbonic acid as to render the water as alkaline as pH9·7. This seemed a suitable method for increasing the alkalinity with the least possible change in the constituents of the sea water. Accordingly specimens of *Ceramium* growing on *Fucus serratus* were gathered and one was placed in a glass jar of sea water at pH8·2 ; *Ulva* was arranged round and over it in a single sheet so that no portion of the *Ceramium* was exposed directly to sunlight. The water rose to 27° C. when insolated. *Ulva* may be kept for a month or more with daily exposure to intense sunlight and remains uninjured.

Before exposure the cells of *Ulva*, *Ceramium*, and of *Vorticellæ* epiphytic on the latter, stained very rapidly with neutral red, which tinges sea water faintly yellow, but showed no trace of colour with brom cresol purple. The latter gives its full blue in sea water, and ceases to be blue about pH6, changing to clear yellow at pH5·4. Since the cells are at about pH7 the colour should have shown up as blue if any had penetrated. Normal cells of these algæ are therefore permeable to neutral red and impermeable to brom cresol purple.

After twenty-five minutes, water drawn from round the *Ceramium* was found to be at pH9·4, giving a good blue with thymolphthalein and with thymol sulphone phthalein. On examining under the microscope it was seen that the *Vorticellæ* were still motile. These and the *Ceramium* cells behaved as before towards the two indicators, and were therefore still normal.

The plant, together with an unstained control, was then arranged

behind the *Ulva* screen as before and was examined again at 6.15 p.m., after two and a half hours. Owing to the less intense illumination the water was then at pH9·2. The *Vorticellæ* were still all quite motile, though stained deep red from previous exposure to neutral red. These and the *Ceramium* were kept in sea water at pH8·2 tinged blue with brom cresol purple, and no penetration was observed inside ten minutes.

On keeping in the dusk for three hours it was noticed that some of the *Vorticellæ* were dead on the two pieces of *Ceramium* exposed to high alkalinity, but most of them were active, as were all on the unexposed control. The exposed pieces of *Ceramium* were now stained blue at the tips and adjacent cortical cells, whereas the unexposed control remained perfectly colourless. On changing the water the blue colour diffused out to a considerable extent. It is clear that sea water at pH9·4 has had an effect upon the permeability of the cells.

On allowing to stand overnight with very dilute neutral red the control *Ceramium* was stained throughout, whereas the treated pieces were nearly colourless at the tips, and in many of the cortical cells of the main axis, the large central cells of which were, however, stained deeply as in the control. The *Vorticellæ* were now nearly all dead and colourless in the treated pieces, but quite active though deeply stained in the control. On placing in brom cresol purple the treated portions, which were only slightly tinged with neutral red, became blue, but not as intense a colour as before. Scarcely any stain was visible in the *Vorticellæ*.

Thus the alteration in permeability induced by water at pH9·4 has not the nature of a reversible change, at least with the duration of treatment tried, viz. two and a half hours as a minimum. It has sufficed to render the cells quite permeable to the normally non-penetrating brom cresol purple and to permit the diffusion outwards of neutral red; in other words, it has killed the *Ceramium* cells which were in direct contact with the water, and the *Vorticellæ* though more resistant died also.

There is accordingly a difference between its action upon the plant and animal cells, and even between the cells at the tips of the *Ceramium* branches and those further down, which are of greater age. A physiological axial gradient is seen to exist. Such gradients have been studied by Child (1916 onwards) in a series of papers on hydrozoa and algæ, and it has been demonstrated, by means of potassium permanganate acting on the former, that the rate and amount of reduction decrease basipetally. The more actively reducing portions are those most easily oxidised, and they are apparently the most sensitive to the action of alkalis also.

As already mentioned, *Ulva* can endure even direct exposure to sunlight and the resulting high alkalinity and remain living and healthy for considerable periods. It seems therefore that the distribution of these plants on the sea-shore must be to some extent limited by their capability

to survive changes in alkalinity. Even though in rock-pools the writer has failed to observe a higher value than pH8·57, higher values may probably be found, and in fresh water where the buffer action is very slight pH9·0 has been observed in a pond. In any case Gail (1919) has shown that *Fucus evanescens* never grows in tide pools or where *Ulva* is found in any considerable quantity, and furthermore that the growth of young plants is very much inhibited in sea water at pH8·4, and almost completely ceases at pH8·6.

Considerable changes in pH value may be observed within a few minutes in the water close to the filament of an insolated green alga. In fresh water a good blue, indicating about pH9, was observed around such a filament while the general colour of the indicator, thymol blue, was yellow as given by pH8·2 or less. These localised changes in pH value would suffice, for instance, to make it impossible for a sensitive epiphyte to develop in such a situation.

SUMMARY.

Sea water originally at pH8·2 became as alkaline as pH9·4 by the photosynthetic action of *Ulva* in removing carbonic acid. This degree of alkalinity did not prove injurious to *Ulva*, but exposure to it for two and a half hours at 27° C. sufficed to increase the permeability of the superficial cells of *Ceramium rubrum* irreversibly and fatally. Similar though less rapid changes brought about the death of numerous *Vorticellæ* epiphytic on *Ceramium*. The criteria for living cells were permeability to neutral red and impermeability to brom cresol purple. Dead cells do not retain neutral red specifically and are permeable to brom cresol purple.

It is suggested that the above facts have a bearing upon the distribution of these and similar algæ upon the shore.

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The Preparation of Permanently Non-acid Formalin for Preserving Calcareous Specimens.

By

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Attention was directed by the Plankton Committee to the desirability of using neutral formalin for preserving calcareous and other specimens likely to be injured by the acidity usually found in the ordinary preparation.

Chamberlain, in *Methods of Plant Histology*, 1915 Ed., recommends the distillation of the formaldehyde solution after addition of sodium bicarbonate. The solution thus obtained is said to develop acidity on standing.

Lee, in the *Microtomists' Vade-mecum*, 1921 Ed., recommends neutralising "by saturation with magnesium carbonate or sodium carbonate." Of the two magnesium carbonate may appear preferable, since it only dissolves appreciably as acidity is developed and does not render the liquid very strongly alkaline, though it may reach pH10, whereas the sodium salt in slight excess gives a much more pronounced alkaline reaction, which is likely to damage the soft parts of specimens since it makes the skin very slimy. Against the use of the magnesium carbonate must be set the fact that being a solid it is liable to settle out and is not attacked more readily than the calcareous plankton. Sodium bicarbonate added to the acid formalin in excess has the advantages that it acts readily and uniformly, since it is in solution, and it gives no undue alkalinity, the value being about pH8·2, which is close to that of sea water. On the other hand, additional carbon dioxide, set free in neutralising the formalin, remains in solution and the alkalinity tends to fall below pH8. Thus there will be a slight solvent action upon the calcium carbonate of the organisms.

It appears that this can be obviated by the use of borax. This in M/20 solution is no more alkaline than pH9·24, which is unlikely to cause trouble through being too alkaline. Neither concentration nor dilution within wide limits alter this appreciably. The neutralisation of a little formic or other acid merely results in the liberation of boric acid and slightly lowers the pH value, which, however, only reaches pH8·2 when

approximately half the boric acid has been set free. It is not, therefore, till this, the bicarbonate, stage has been passed that there will be any appreciable amount of hydrogen ion to attack the specimens. About five to ten grams of borax per litre should be added, or considerably less may be required. It need not be weighed out, but the resulting solution should be tested by adding phenolphthalein, when the appearance of a good red colour denotes a reaction of pH8·5–9·0. The tint may be compared with that given by the borax alone in water. With thymol blue a slaty blue colour is shown at pH9 ; this is a good region in which to have the reaction of the formalin. Strong formalin should be diluted in the test tube before adding the indicator.

That the quantities of acid found in fresh and stored formalin are by no means negligible may be seen by titrating with sodium hydroxide. A sample of 30 per cent formaldehyde was found to contain 0·51 grams of acid per litre, calculated as formic acid. The carboy 40 per cent formaldehyde was at pH2·8. Dilute "5 per cent formalin," viz. that percentage of the laboratory stock 30 per cent formaldehyde, after being stored with fish for fifteen years was at pH5·0, and its titratable acid amounted to 1·26 grams per litre. This solution had a considerable buffer action while a precipitate appeared in the solution, probably an organic product derived from the fish. The acid of the "5 per cent formalin" should only be 0·025 grams calculating from its original strength and the dilution.

By the help of indicators formalin may be observed to produce acid in quite a short time. About two litres of 30 per cent formaldehyde were neutralised and brought to slight alkalinity, pH8·0, by means of sodium hydroxide. One portion stood in the warmth and light of a south window, the other under the bench in darkness. The condition after various times is shown below.

Time in days from Aug. 9.	In dark. pH.	In sunlight and warmth. pH.
0	8·0	8·0
20	7·0	5·9
56	6·4	5·2

The acidity of the latter is accordingly more than ten times as great as that of the former. This appears to prove that light as well as heat has been concerned in effecting the change, for were the mean temperature of the insolated solution as much as 10° C. above that of the other, it would only result in the rate of production of acid being doubled or trebled, and the temperature difference is certainly an overestimate.

This development of free acidity is in contrast to the behaviour of 5 per cent formalin rendered alkaline by borax, so that it was at pH8·8–9·0.

After insolation for thirty-six days from August 29th, it was at pH8·95, showing that it had remained constant in reaction, nor was there any appreciable change in a further period of six months. Since 5 per cent formalin was found to have developed about 1·26 grams of acid in fifteen years and borax neutralises almost one-fourth of its weight of formic acid it is evident that an amount of 5 grams per litre would avoid acidity for over this period, or longer with a solution kept in the dark.

The distillation of 40 per cent formaldehyde was also tried to test the acidity of the product. When the liquid at pH2·8 was distilled through a Liebig condenser the distillate was also at pH2·8.

On adding sodium bicarbonate in excess, so that the liquid was at about pH8·2, and distilling, the first portions that came over were at pH5, the greater bulk at pH2·8. The residue in the flask was at pH5.

Similarly formalin distilled with excess of sodium hydroxide became acid and gave an acid distillate. With magnesium carbonate in excess and renewed when dissolved the liquid in the flask was maintained at pH8–9, but even then the distillate was very uniformly at pH4·4, due in part probably to carbonic acid.

In this connection attention may be drawn to the results obtained by R. K. S. Lim (Proc. Physiol. Soc. in J. Physiol., **53**, ciii, 1920), who showed that fixation in formalin of mucous membranes bearing goblet cells, resulted in an abnormal appearance through discharge of the mucinogen. With formalin neutralised by means of sodium hydroxide this does not occur.

SUMMARY.

Formalin, which is permanently non-acid and only slightly alkaline, close to pH9, may be prepared by the addition of borax till a good red colour is shown with phenolphthalein, or a slaty blue with thymol blue, when added to the diluted formalin.

Distillation of formalin from solid magnesium carbonate gives an acid product, which is at pH4·4.

Formalin neutralised with sodium hydroxide becomes acid on standing, the change being more rapid in sunlight than in the dark. A reaction of pH5·2 was reached in fifty-six days in light, the initial value being pH8·0. Commercial formalin, " 40 per cent," may be as acid as pH2·8.

Plymouth Peridinians.

By

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I. DIPLOPSALIS LENTICULA AND ITS RELATIVES.

With Figures 1–20, Table and Diagram.

Much discussion has recently taken place over the peridinian described by Bergh (1882) as *Diplopsalis lenticula*, and certainly three distinct species have severally been designated as the original. As it is a name frequently to be found in the International Plankton lists, it is important that we should be clear as to which species we are noting, and as recent research at Plymouth has shown that all the three are common in the district it seems worth while to investigate them once more. The question is interesting also from the point of view of phylogeny, as in these species we have close relatives of *Peridinium* proper which seem to indicate the method of evolution of that genus from simpler forms with fewer plates, and from this point of view alone they are well worth study.

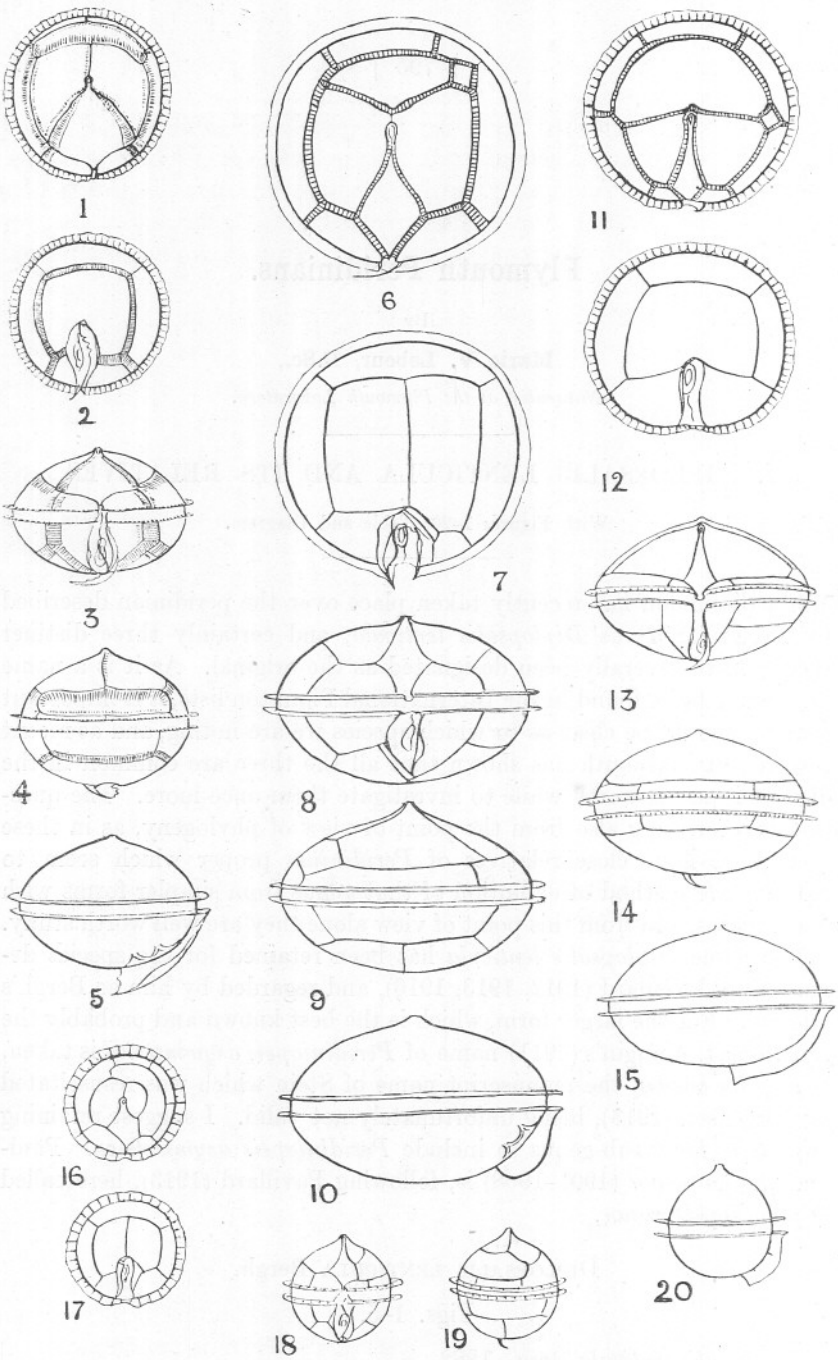
The name *Diplopsalis lenticula* has been retained for the species described by Pavillard (1912, 1913, 1916), and regarded by him as Bergh's original. For the larger form, which is the best known and probably the commonest, Mangin's (1911) name of *Peridiniopsis asymmetrica* is taken, *Diplopelta bomba*, the manuscript name of Stein which was resuscitated by Jörgensen (1913), being unfortunately not valid. I suggest retaining *Diplopelta* for a sub-genus to include *Peridiniopsis asymmetrica*. Paulsen's *forma minor* (1907–1908) is, following Pavillard (1913), here called *Diplopeltopsis minor*.

Diplopsalis lenticula Bergh.

Figs. 1–5.

Diplopsalis lenticula Berg., 1882.
Stein, 1883 (Plate IX, Fig. 1).
Pavillard, 1912, 1913, 1916.

The following description is taken from the Plymouth specimens:



EXPLANATION OF FIGURES 1–20.

1–5. *Diplopsalis lenticula* Bergh. Tow-nets, 2.5.21. Eddystone-Rame. Breadth 40 μ.

6–10. *Peridiniopsis* (*Diplopelta*) *asymmetrica* Mangin. Tow-nets 2.5.21. Eddystone–Rame. Breadth 66 μ.

11–15. *Diplopeltopsis minor* (Paulsen). Tow-nets, 28.2.21. Breakwater. Breadth 52 μ.

16–20. *Peridiniopsis rotundata* n.sp. Tow-nets, 9.6.21. Breakwater.

1, 6, 11, 16. Epitheca from top. 2, 7, 12, 17. Hypotheca from bottom.

3, 8, 13, 18. Ventral view. 4, 9, 14, 19. Dorsal view. 5, 10, 15, 20. Side view.

In Fig. 9 the central precingular is erroneously figured as one plate where it should be divided into two (3″ and 4″).

Cell lenticular, depressed, epi- and hypotheca the same shape. Apex ending in a very inconspicuous projection, on which opens the apical pore. Cell contour almost circular. Left longitudinal list conspicuous, reaching almost to the centre of the hypotheca, bent across the ventral area, its margin more or less denticulate. Right longitudinal list inconspicuous. Flagellar pore oval, on the left lower corner of the ventral area, more or less hidden by the list. Girdle not displaced, indented. Transverse list supported by very fine spines. Cell contents pink. Large pusule apparatus. Theca very finely punctuate. Sutures broadly striated, except in young forms. Epitheca with 3 apicals meeting in the centre, a fourth plate having the position of an intercalary occupying a large part of the dorsal area, 6 precingulars, the 2 connected with the dorsal plate very narrow and inconspicuous. Hypotheca with 5 postcingulars and only 1 large apical. Plate formulæ,* 4′0a 6″ 5‴ 1⁗ or 3′1a 6″ 5‴ 1⁗. Probably the latter. Reproduction by sporulation, 2 spores emerging from the cell between the epi- and hypotheca.

Bergh's specimens varied between 29 μ and 34 μ in length, and 40 μ to 43 μ in breadth. Pavillard's ca. 25 μ long, 45 μ broad. The Plymouth specimens vary from 33 μ to 56 μ across, the most usual breadth being from 40 to 45 μ. One with 2 spores inside measured 53 μ across, each spore measuring ca. 39 μ across. The usual shape is flattened, but some from Plymouth were much rounder; these, however, were very rare.

It occurred frequently in Plymouth Sound from the region of the Breakwater, but also further out between Rame and the Mewstone and round about the Eddystone. Once (July 4th, 1921) it occurred further out, at Station E 7, Bishop's Lighthouse, Scilly Is., bearing N. by W. 8 miles (magnetic), from the surface over 40 fathoms, and once from Station N 2 (July 3rd, 1921), Wolf Rock bearing N.W. 8 miles (magnetic), at a depth of 30 metres, at the line of a rapid change of temperature, 11° (warmer water 14° above). Its natural habitat appears to be inshore, and it is most probably to be regarded as a neritic and surface form. It occurred from May to August in the Plymouth district (see table). It was found in the Clyde off Millport, inshore, from just off Keppel Pier, in August, 1921, and at Cullercoats, Northumberland, off the rocks at low water in September, 1921.

The present records are the first from the International area if we except that of Bergh. Pavillard records it from the Lake of Thau and from the Mediterranean, and in a letter (1921) tells me he has found it at Roscoff. Stein gives no special locality for this particular figure, which came either from the Baltic or from the Mediterranean.

Bergh's specimens agree with this description as to size, shape, colour

* Kofoid's (1909) system of numbering the plates is used throughout this paper.

and form and size of the left longitudinal list. The plate arrangement was not described by him. Stein (1883, Plate IX, Fig. 1) figures what he describes as a young specimen of *Diplopsalis lenticula* having 3 apicals and 5 precingulars. Although he does not figure nor describe the hypotheca of this form, the epitheca shows it to be almost certainly the species described above, the dorsal end-plate and the division of the corresponding precingular into 2 not being shown. It is extremely easy to make such a mistake, as these 2 precingulars are very narrow and in certain positions seem blended with the dorsal plate. Pavillard (1912), having rediscovered this species in the Lake of Thau, describes the dorsal plate, but not the 2 corresponding precingulars. Later (1913) he believes the dorsal plate to be non-existent. His latest description (1916) from Mediterranean specimens, gives 3 apicals, 5 precingulars, 5 postcingulars and 1 antapical, and he regards this species as identical with Bergh's, and Stein's Fig. 1, Plate IX. Through the kindness of Professor Pavillard I have been able to examine some of his Mediterranean specimens, and find the plates to be identical in number and position with those from Plymouth, the first apical being rather larger and the longitudinal list slightly more conspicuous. Moreover, Professor Pavillard tells me he has found specimens similar to those at Plymouth at Roscoff, and regards them as the same species, though possibly a variety of the Mediterranean form.

I agree with Pavillard that this species is more like Bergh's than either of the other two. The most important reason given against it is the fact that it has not as yet been found again in the locality from which Bergh's originals came (Skaggerak), whereas *Diplopeltopsis minor* is common there, and *Peridiniopsis asymmetrica* also occurs (recorded by Cleve-Euler (1917) as *Diplopsalis lenticula* Bergh, f. typica). As, however, in this last year it has been found both from the Clyde and from the Northumberland coast, it seems not unlikely that it may still be found further north and in the Skaggerak, when all difficulties would disappear.

Ostenfeld's (1901 and 1908) species, *Diplopsalis caspica*, has the same plate formula as *Diplopsalis lenticula* as given above, and certainly belongs to the same genus. The fact brought out that the genus *Diplopsalis* has 6 precingulars brings it into touch more easily with other closely related and already known forms, and the affinities are more easily seen.

Peridiniopsis (Diplopelta) asymmetrica Mangin.

Figs. 6–10.

Diplopsalis lenticula Stein (Plate VIII, Figs. 12, 13, 14 and Plate IX).

Figs. 2, 3 (?) and 4.
Schütt, 1895.
Paulsen, 1907, 1908, p. 35, Fig. 44.
Meunier, 1919, in part.

Peridiniopsis asymmetrica Mangin, 1911 (b.c.), 1913.

Peridinium lenticula Paulsen, 1912, in part.

Diplopelta bomba Jörgensen, 1913.
Pavillard, 1913, 1916.

Preperidinium asymmetricum Mangin, 1913.

Peridinium asymmetricum Ostenfeld, 1915.

This is the largest, the best known and most widely distributed of the 3 forms in question. Mangin (1911b) completely elucidated its structure, interpreting its plate formula as 4′1a 6″ 5‴ 2⁗, the small anterior intercalary being normal to the species. Cell rather more depressed than *Diplopsalis lenticula*, but similar in shape and nearly always larger. Left longitudinal list projecting but only reaching about half-way across the radius of the cell, edge smooth, not bent over the flagellar pore. Flagellar pore on the left near the hind end of the list. Right longitudinal list inconspicuous. Girdle not displaced, indented. Transverse lists not supported by spines. Cell contents pink. Large pusule apparatus. Theca distinctly punctuate. Sutures striated, except in the very young forms. Epitheca with 4 end-plates, only numbers 1, 2 and 4 reaching to the apical pore, number 3 meeting 2 and 4 behind the apex. A very small anterior intercalary on the left side between apicals 2 and 3. 6 precingulars. Hypotheca with 5 postcingulars and 2 antapicals. A small plate, called by Mangin a supplementary postequatorial, is probably, as Pavillard suggests (1913), part of the ventral area. Reproduction as in *Diplopsalis*. Usual diameter about 80 μ. Small specimens have, however, been found in the Plymouth district measuring 50–66 μ. One, 84 μ across, was the largest seen and contained 2 spores. Mangin mentions 89 μ as the extreme size.

Mangin (1911c) recorded a spherical form, which he designates as *V. spherica*, and he also found a dextral variety having the plates reversed, and a variety with 3 antapicals, both the last very rare.

All the Plymouth specimens conform to the type, and agree very well with those from other localities.

It is difficult to be sure of the distribution of this species, owing to the fact that in the early records we do not know with which species we are dealing. We have, however, the following fully established records: North Sea (Northumberland coast), Swedish and Norwegian Seas, Clyde,

Irish Sea, Atlantic Ocean, English Channel, Flemish coast, Brittany coast, Mediterranean, Baltic and Boeten Straits, Calebes. It is thus widely distributed. The Plymouth records show it present from May to November, abundant in the Sound round the Breakwater and also outside from Rame and the Mewstone to beyond the Eddystone. It occurs in much the same places as *Diplopsalis*, but more frequently outside, and altogether it seems to be more abundant. This may be, however, that it is caught more frequently in the very fine nets.

Most of Stein's figures refer to this species (Plate IX, Figs. 3 and 4; Plate VIII, Figs. 12–14; Plate IX, Fig. 2, with no small intercalary and only 5 precingulars is possibly erroneous), and all are designated *Diplopsalis lenticula*. Stein has in his MSS. notebook, as he tells us, called them *Diplopelta bomba*, but on identifying them with Bergh's species gave this up in favour of *Diplopsalis lenticula*. Schütt's (1895) figures also almost certainly belong to this species, although Fig. 50_5 agrees with Stein's Fig. 2 in having only 4 end-plates and 5 precingulars. If this be correct it must apply to another species not since rediscovered. Paulsen's (1907 and 1908) description of the larger form almost certainly refers to this species, although he takes his figure of the epitheca from Stein's Fig. 2 and describes it with 5 precingulars. Later, however (1912), he identifies it with Mangin's *Peridiniopsis asymmetrica*, which has 6 precingulars and a small left anterior intercalary. Jörgensen (1913) gives it Stein's name of *Diplopelta bomba*, in which he is followed by Pavillard. Ostenfeld (1915) relegates it with all its relatives to the genus *Peridinium*. Meunier (1919), uniting it with Paulsen's *forma minor*, regards it once more as *Diplopsalis lenticula*.

It is probably this large species which is recorded in most of the International Plankton lists. Paulsen (1912), in his summary in which he gives 4 maps of the distribution of *Peridinium lenticula* (by which name he covers both *Peridiniopsis asymmetrica* and *Diplopeltopsis minor*) in the International area, says: "It is possible that this species must be divided into two, viz. *P. lenticula* and *P. minus*, the first named being oceanic and the second neritic. They may, however, be forms of the same species and in the following they will be treated as such. They have not been separated in the Bulletins, so it is impossible to say where one and where the other has been found, they must therefore be regarded as a unity, and only in a few cases suggestions can be made as to which of them we are dealing with." After discussing the records the suggestion is made that it is probably the form *minor* that is recorded from the English Channel. As a matter of fact, as is here shown, all three species occur, but probably *Peridiniopsis asymmetrica* is the commonest, at any rate in the tow-nets. Gran (1915) records this form and the next together as *Diplopsalis lenticula*, but definitely states that it is a collective name.

DIPLOPELTOPSIS MINOR (Paulsen) Pavillard.

Figs. 11–15.

Diplopsalis lenticula forma minor Paulsen, 1907, p. 9, Fig. 9; 1908, p. 36, Fig. 45.

Diplopsalis lenticula Meunier, 1910.

Diplopsalis sphærica Meunier, 1910.

Peridinium lenticulatum Mangin, 1911a.

Peridinium Paulseni Mangin, 1911b.

Peridinium Meunierii Pavillard, July, 1912.

Peridinium lenticula, neritic form (=*Peridinium minus*), Paulsen, Oct., 1912.

Diplopsalis lenticula Jörgensen, 1913.

Diplopeltopsis minor Pavillard, July, 1913.

Peridinium lenticula Ostenfeld, 1915.

Diplopsalis lenticula Meunier, 1919, in part.

This little species, first described by Paulsen (1907), from the Zuider Zee, Western Baltic, Kattegat and in great masses from a fjord in the Faröes as *forma minor* of *Diplopsalis lenticula* is regarded by the Scandinavian authorities as the true *Diplopsalis lenticula*, of Bergh. Certainly its dimensions and general shape approximate to that form, but the left longitudinal list is much narrower and less conspicuous than in the form described and figured by Bergh; also it is quite smooth at the edge, not denticulated, nor curved over the flagellar pore. Moreover, it does not reach nearly to the centre of the hypotheca. The most important reason for its being regarded as Bergh's original is that so far it is the species known to occur commonly in the region investigated by him (Skaggerak). As Pavillard has discovered a species much more like the original, and it is here shown that this species occurs in the English Channel, in the Clyde and in the North Sea (off the Northumberland coast), it is not impossible that it may still be found in the Skaggerak. It seems more likely to be Bergh's form than the present one, and certainly agrees with Stein's figure (Plate IX, Fig. 1), whereas *Diplopeltopsis minor* is not figured at all by him.

Diplopeltopsis minor has the cell of the same shape as *Diplopsalis lenticula*. Cell contour almost circular. Left longitudinal list narrow, not denticulate, reaching not so far as the centre of the hypotheca, usually of the same width for its whole length, not bent over the flagellar pore. Right longitudinal list inconspicuous. Girdle not displaced, slightly indented. Transverse list supported by very fine spines, sometimes hardly perceptible. Cell contents pink. Large pusule apparatus. Theca

very finely punctuate. Sutures striated in the older specimens. Epitheca with 4 end-plates meeting in the centre, the first very narrow, the third occupying more than half the apical area. A small anterior intercalary plate on the left side. Seven precingulars. Hypotheca with 5 postcingulars and 1 large antapical. Plate formula 4′1a 7″ 5‴ 1⁗ or 3′2a 7″ 5‴ 1⁗. Reproduction as in *Diplopsalis*.

Paulsen's original specimens measured 52 μ to 56 μ across, Mangin's 35 μ to 49 μ. The Plymouth specimens ranged from 28 μ to 45 μ across, usually over 40 μ.

As in the case of *Diplopelta* it is difficult to be sure of the distribution of this species, because we do not know with which species we are dealing in the earlier records. It is known to be more neritic than *Peridiniopsis asymmetrica*, although occasionally found right out at sea in the present records. On the other hand, it is the only one of the three found in an estuarine situation (up the River Tamar).

Records of its distribution which are certain are the following : Zuider Zee, Western Baltic, Skaggerak, Kattegat, Belt Sea, fjord in the Faröes, coast of Brittany, English Channel, Barents and Kara Sea. It is thus the most northerly species of the three forms, the Brittany coast being so far its most southerly record. It is found in the Plymouth district from February to November, but only very rarely between April and October. It seems to be chiefly an Autumn and early Spring form. It occurs most frequently near the shore, but also was found outside in the Rame-Eddystone region, and on one occasion at Station E 1, 14 miles from land. All records are from the surface.

	Feb.		*April.*					*May.*				*June.*						
	16	28	1	12	14	19	26	2	13	20	27	2	6	13	17	23	27	29
	1	1	1	0	0	0	0	0	1	1	0	1	1	1	1	1	1	1
Diplopsalis lenticula								×			×	×		×		×	×	×
Peridiniopsis asymmetrica								×	×	×		×	×		×	×	×	×
Diplopeltopsis minor	×	×	×	×	×	×	×										×	

	July.									*August.*						*Oct.*	
	1	3	4	7	11	12	15	21	26	2	4	8	9	15	19	22	10	11
	1	0	0	1	1	1	1	0	0	1	1	1	0	1	1	0	0	0
Diplopsalis lenticula	×	×	×	×		×		×	×		×		×					
Peridiniopsis asymmetrica			×	×	×	×			×	×		×		×	×	×	×	×
Diplopeltopsis minor				×			×						×					

	October.					*November.*						*December.*						
	13	17	19	24	31	1	3	7	17	24	29	1	5	6	7	9	12	19
	0	0	0	0	0	0	0	1	0	1	0	0	0	0	0	0	0	1
Diplopsalis lenticula																		
Peridiniopsis asymmetrica	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×
Diplopeltopsis minor					×		×						×		×	×	×	

Table showing distribution of *Diplopsalis lenticula*, *Peridiniopsis asymmetrica* and *Diplopeltopsis minor* in 1921 (September excepted).
1=inner grounds from the shore to Rame all over the Sound.
0=outer grounds outside Rame.

In the table the records are divided into those from the Sound and up the Tamar, as far as Rame (marked 1 for " inside records " in the table) and those from outside the sound from Rame Head and on a level with it outwards (marked 0 for " outside records " in the table).

Mangin records a reversed form having the anterior intercalary on the right, and a symmetrical variety with 2 anterior intercalaries, 1 right and 1 left. The symmetrical form is also figured by Meunier (1913) as *Diplopsalis lenticula*, and a spherical form called by him *Diplopsalis spherica* and afterwards united with *D. lenticula*. He gives, however (1913), only 5 precingular plates, following Paulsen in his original description, but later (1919) he describes it as having 6, the seventh having probably been overlooked. Amongst the Plymouth specimens is a very flat variety, having the first apical extremely narrow, which is very like Meunier's figure, but with the asymmetrical intercalary. Also almost spherical forms were found. The longitudinal list is also variable, but never reaches the dimensions, nor has the form which is present in *Diplopsalis lenticula*.

We thus have the 3 species in the Plymouth district, each of which has been regarded as the original *Diplopsalis lenticula* of Bergh. Giving the preference to Pavillard's species one goes upon its evident resemblance in size and form, as Bergh described no plates ; but besides this resemblance its presence in the English Channel, and also in the Clyde and North Sea off the Northumberland coast, makes it much more probable that it may be found in Bergh's original locality. The fact that it had not before been found further North and West than the Mediterranean seemed to put it out of account in the International lists, and it was inferred that all records referred either to the large form *Peridiniopsis asymmetrica* or the *forma minor* of Paulsen, *Diplopeltopsis minor*, see Paulsen (1912). The latter being the species commonly found where Bergh obtained his specimens most of the Scandinavian authorities (Paulsen, Ostenfeld, Jörgensen) believe it to be the original. As we can only go by its colour, size and shape, it is impossible to be sure ; but the fact that the left longitudinal list is short and narrow and without denticulations at its edge, and also does not reach nearly so far as the centre of the hypotheca, makes it correspond with the original description less exactly than Pavillard's species. Mangin, who regards *Peridiniopsis asymmetrica* as the original of Bergh, is not supported by others except when it is regarded as one species with *Diplopeltopsis minor*. It agrees only in colour and shape with Bergh's species, and as it is very much larger and as the plates distinctly differ from both the smaller forms it does not seem likely that it is identical with either.

The fact brought forward by Pavillard (1913) that Stein figured two distinct species as *Diplopsalis lenticula* is evidently quite correct, and as Stein was the first to figure any plate structure, strictly speaking one of his forms must be kept as *Diplopsalis lenticula*. It seems only natural that we should take the smaller form which corresponds more with Bergh's species, and as this is seen to be similar to Pavillard's from the Mediterranean and also to the Plymouth specimens, it is right that we should regard this as *Diplopsalis lenticula* rather than the other very much larger form, or the smaller *Diplopeltopsis minor*, which was not figured by Stein at all.

Whilst investigating these species many small peridinians were found, which in their size, shape and colour resemble *Diplopsalis pillula* Ostenfeld (1908) and *Diplopsalis minima* Mangin (1911c), but on closer enquiry all were found to differ and a surprising variety of plate arrangement was found. On account of their small size it was not always possible to be certain of the plates, although they are all closely related to *Diplopsalis* and its near allies. One species, however, was carefully investigated and found to be unlike any other in the fact that there was a very large dorsal plate, anterior intercalary or third apical, which curved round to the front and touched apical 1, the latter instead of being diamond-shaped has on the left a third side, so that in shape (but not in its relation to precingular 2) it resembles the first apical in the section Metaperidinium (Jörgensen) of *Peridinium*. The plate formula is 4′0a 6″ (?) 5‴ 2⁗, or 3′1a 6″ (?) 5‴ 2⁗, the number of precingulars being apparently 6, but their complete elucidation was so difficult that on the left side there is a slight doubt as to their number. Bearing this in mind, it will be taken as 6, lacking further observations, and it would thus belong to the genus *Peridiniopsis*. I propose for it the name *Peridiniopsis rotunda*, and give herewith a short description of it :—

PERIDINIOPSIS ROTUNDA n. sp.

Figs. 16–20.

Cell almost spherical, sometimes slightly depressed, sometimes higher than broad. Left longitudinal list conspicuous but not reaching to the centre of the hypotheca, edge smooth. Girdle broad, not displaced, slightly indented. Transverse list supported by conspicuous spines set far apart. Cell contents pink. Large pusule apparatus. Sutures in older specimens broadly striated. Cell diameter 22 μ to 28 μ. Epitheca with 3 apicals meeting at the centre, a fourth plate, third apical or anterior intercalary, having a dorsal position and coming round to the front on the left side, joins the first apical which has a third straight side

where it is in contact with this dorsal plate. 6 (?) precingulars. Hypotheca with 5 postcingulars and 2 antapicals. Plate formula 4′0a (or 3′1a) 6″ (?) 5‴ 2⁗.

Distribution in all parts of Plymouth Sound and occasionally outside in centrifuged water samples.

It is interesting that the outline of this species might easily represent Ostenfeld's *Diplopsalis pillula*. The shape and size are almost identical. It is therefore absolutely untrustworthy to go by the outline only. Van Breemen's specimen from the Aral Sea (1905) might just as well represent this species (see Paulsen, 1908, p. 37).

Other very small forms with outline exceedingly like *Diplopsalis pillula* and *D. minima* and the present species showed the plate structure of *Diplopsalis lenticula* and *Diplopeltopsis minor*, and were apparently young forms of these. At least three more of similar size and shape were found, but with different plate structure. They have not yet been fully worked out. Owing to the fact that *Diplopsalis pillula* and *D. minima* are exceedingly small and have not been recorded by other observers, it is difficult to be quite certain of their plate structure. It seems almost possible that they may be found to have 6 precingular plates instead of 5, as these can be so narrow dorsally that they are easily overlooked. It is noteworthy that they both have 2 antapicals. For want of further knowledge of their structure these two are left out of account in the following survey.

There is no doubt about the fact that in these species we have a very large variety in the number of plates, and it seems probable that amongst them we have forms comparable to those from whence sprang *Peridinium*. All the investigators seem to be agreed that they are closely related to *Peridinium* and form a series leading up to it. Are they to be regarded as primitive or degenerate forms ? It seems to be the prevailing opinion that they are more primitive than *Peridinium*. All are symmetrical cells, lenticular to spherical in shape, girdle not displaced, with the left longitudinal list more or less produced, pink in colour with a large pusule apparatus, and with fewer plates than *Peridinium*.

At the present time, besides *Diplopsalis*, *Peridiniopsis* and *Diplopeltopsis*, there are species known which are closely related to *Peridinium*, some with only 2 anterior intercalaries (*Peridinium monospinium*, *P. minutum*), which have been separated as *Archæperidinium* Jörgensen, and with only one anterior intercalary, separated as *Diplopsalopsis* Meunier, Pavillard emend (*P. orbiculare*). These are all recognised by Pavillard as genera in his very interesting survey of *Diplopsalis* and its related genera (1913), and Paulsen (1907–1908) had already arranged them in order although regarding them all as *Peridinium*. Jörgensen (1913), whilst separating them into genera, suggests that they

might just as well be regarded as sub-genera, stating that it entirely depends on the variability, if any, of the plates. Mangin (1911) regards the groups as based too much on variable plates, as he shows that the intercalaries may vary from 1 to 2 in *Diplopeltopsis minor* and the antapicals from 2 to 3 in *Peridiniopsis asymmetrica*. It is quite true that the intercalaries may vary from 1 to 2 in a species, for besides Mangin's and Meunier's examples the present writer has found a specimen of *Peridinium orbiculare* with a small right anterior intercalary in addition to the usual left one. The variation from 2 antapicals to 3 is, however, surely to be regarded as a sport rather than a true variety, as *Peridinium* always has 2, and in all the thousands of examples examined of *Peridinium* and its near relatives this is the first to be recorded with 3 antapicals.

The genera so far proposed have the following plate formulæ (as usually designated) :—

Diplopsalis, 4′0a (or 3′1a) 6″ 5‴ 1⁗.

Peridiniopsis, 4′0a 6″ 5‴ 2⁗.

Diplopelta, 4′1a 6″ 5‴ 2⁗.

Diplopeltopsis, 4′1a 7″ 5‴ 1⁗.

Diplopsalopsis, 4′1a 7″ 5‴ 2⁗.

Archæperidinium, 4′2a 7″ 5‴ 2⁗.

Peridinium, 4′3a 7″ 5‴ 2⁗.

I should suggest that starting with *Diplopsalis* (which has 4 plates in the epitheca besides the 6 intercalaries, 5 postcingulars and 1 antapical) we have a forward movement in three directions, one of which leads to *Peridinium* proper :—

Firstly, through *Peridiniopsis* to *Diplopelta* where the antapical first splits as in *Peridiniopsis*, then a small left intercalary appears as in *Diplopelta*. In this case the so-called third apical of *Diplopelta* is apparently homologous with the dorsal plate (intercalary or third apical) in *Diplopsalis*.

Secondly, through *Diplopeltopsis* to *Diplopsalopsis* where the precingulars increase to 7, a left intercalary appears as in *Diplopeltopsis*, and then the apical splits as in *Diplopsalopsis*.

Thirdly, through the so-called *Diplopsalis acuta* Entz fil and *Archæperidinium* to *Peridinium* proper, in which while there is still only one antapical a small apical appears anterior to the large dorsal plate. In this fresh-water species* Entz (1904) describes this dorsal plate splitting

* So far only this fresh-water species is known with this particular plate formula.

into 2 and so leading up to *Archæperidinium*, in which there is a small third apical and 2 anterior intercalaries, the antapicals now being 2, and finally *Peridinium* proper where there are 3 anterior intercalaries.

In this scheme I have purposely taken away *Diplopsalopsis* (type *D.*

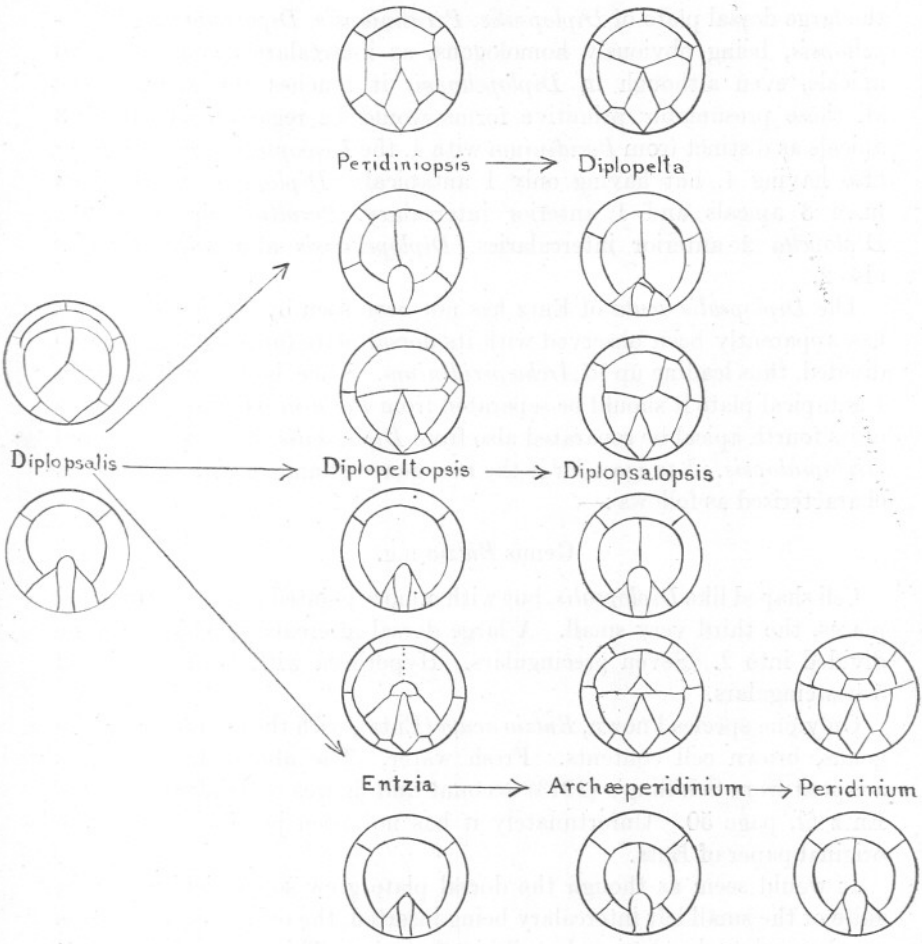


DIAGRAM ILLUSTRATING THE SUGGESTED RELATIONSHIP OF *DIPLOPSALIS* WITH THE ALLIED GENERA

orbiculare) from the direct line to *Peridinium*, because the dorsal plates appear to be homologous with the dorsals of *Diplopeltopsis*, *Peridiniopsis* and *Diplopelta*, and with the large anterior intercalary of the *Diplopsalis acuta* of Entz, and thus also homologous with the 2 anterior intercalaries of *Archæperidinium*. If this be so then the third apical in the *Diplopsalis acuta* of Entz and of *Archæperidinium* and of

Peridinium proper must be of later origin than the 2 intercalaries, and the so-called third apical of *Diplopsalopsis orbiculare* is not homologous with the third apical of *Peridinium*. For this reason it would be advisable to remove *Diplopsalopsis* from *Peridinium*, as it is apparently out of the direct line although closely related. Also, it would be advisable to regard the large dorsal plate of *Diplopsalis*, *Peridiniopsis*, *Diplopelta* and *Diplopeltopsis*, being obviously homologous, as intercalaries and not third apicals, even although in *Diplopeltopsis* it touches the apex. Thus all these presumably primitive forms would be regarded as having 3 apicals as distinct from *Peridinium* with 4, the *Diplopsalis acuta* of Entz also having 4, but having only 1 antapical. *Diplopsalis* would then have 3 apicals and 1 anterior intercalary, *Peridiniopsis* the same, *Diplopelta* 2 anterior intercalaries, *Diplopeltopsis* and *Diplopsalopsis* also 2.

The *Diplopsalis acuta* of Entz has not been seen by the writer, but it has apparently been observed with its dorsal plate (anterior intercalary) divided, thus leading up to *Archæperidinium*. Since, however, it has only 1 antapical plate it should be separated from *Peridinium*, and on account of its fourth apical be separated also from *Diplopsalis*, *Diplopeltopsis* and *Diplopsalopsis*. I suggest for it the new generic name of *Entzia*, which is characterised as follows :—

Genus *Entzia* n.g.

Cell shaped like *Diplopsalis*, but with a more pointed apex. Four apical plates, the third very small. A large dorsal intercalary, which may be divided into 2. Seven precingulars. Hypotheca with 1 antapical and 5 postcingulars.

Only one species known, *Entzia acuta* (Entz), with the characters of the genus, brown cell contents. Fresh water. The above description is adapted from Schilling's (1913) account and figures of *Diplopsalis acuta* Entz fil, page 50. Unfortunately it has not been possible to see the original paper of Entz.

It would seem as though the dorsal plate grew so unwieldy that, in spite of the small left intercalary being inserted, the only way to progress was by a plate developing dorsally at the apex. This once accomplished the line to *Peridinium* was well established.

The above is a suggestion only, but assuming this theory to be correct then *Diplopsalopsis* must be removed from *Peridinium* unless we make the genus, already overweighted, to include all these primitive forms. Even Mangin's *Preperidinium*, proposed for these last, does not meet the case, as it removes *Archæperidinium* from *Peridinium*, and places it amongst forms obviously not so closely related. I should suggest regarding the intercalaries as variable within the genus, the apicals, ant-

apicals and precingulars as constant generic features. In this way *Archæperidinium* would be a sub-genus of *Peridinium*, *Diplopelta* a sub-genus of *Peridiniopsis*, and *Diplopsalis*, *Diplopeltopsis*, *Diplopsalopsis* and *Entzia* would be separate genera, thus :—

Genus DIPLOPSALIS Bergh, plate formula 3′1a 6″ 5‴ 1⁗.

Diplopsalis lenticula Bergh.

Diplopsalis caspica Ostenfeld.

Genus PERIDINIOPSIS Lemmermann, plate formula 3′1–2a 6″ 5‴ 2⁗.

Sub-genus PERIDINIOPSIS, plate formula 3′1a 6″ 5‴ 2⁗.

Peridiniopsis Borgei Lemmermann.

Peridiniopsis rotunda n. sp.

Sub-genus DIPLOPELTA Jörgensen, plate formula 3′2a 6″ 5‴ 2⁗.

Peridiniopsis (*Diplopelta*) *asymmetrica* Mangin.

Genus DIPLOPELTOPSIS Pavillard, plate formula 3′2a 7″ 5‴ 1⁗.

Diplopeltopsis minor (Paulsen).

Genus DIPLOPSALOPSIS Meunier, plate formula 3′2a 7″ 5‴ 2⁗.

Diplopsalopsis orbiculare (Paulsen).

Genus ENTZIA n.g., plate formula 4′1–2a 7″ 5‴ 1⁗.

Entzia acuta (Entz fil).

Genus PERIDINIUM Ehrenberg, plate formula 4′2–3a 7″ 5‴ 2⁗.

Sub-genus PERIDINIUM, plate formula 4′3a 7″ 5‴ 2⁗.

Peridinium depressum Bailey, etc.

Sub-genus ARCHÆPERIDINIUM Jörgensen, plate formula 4′2a 7″ 5‴ 2⁗.

Peridinium (Archæperidinium) monospinum (Paulsen).

" " minutum Kofoid.

" " excentricum Paulsen.

" " thorianum Paulsen.

Several fresh-water species are here omitted, not being known to the writer.

I have added *Peridinium excentricum* to the sub-genus *Archæperidinium*; the plates have the same number, and owing to the greater growth of the second anterior intercalary the fourth precingular is pushed to the left side. It certainly belongs to the section Orthoperidinum as Jörgensen (1913) suggests, and Pavillard (1916) confirms. *Peridinium thorianum* also seems to belong to *Archæperidinium*.

This brings us to the fact, noted by Jörgensen, that all these forms here

discussed which lead to Peridinium belong to the section Orthoperidinium. If *Diplopsalis* is a primitive form then we can trace a definite progress to *Peridinum*, section Orthoperidinium. All the forms considered being of the Orthoperidinium type, therefore unless *Peridinium* had two separate origins Orthoperidinium must be more primitive than Metaperidinium. Barrows (1918), in his very interesting memoir on the significance of skeletal variation in the genus *Peridinium*, is of the opinion that Metaperidinium is the more primitive, his chief reason being that the precingulars 1 and 7 are probably of later origin than the others, as they are of smaller size, and in Metaperidinium they are smaller than in Orthoperidinium, owing to the left-hand extra face, and in Paraperidinium both are smaller, owing to the two extra faces, the first apical being a hexagon. Paraperidinium is thus in his opinion the most primitive, Orthoperidinium the most advanced. Now Barrows himself assumes that *Peridinium* arose from a form with fewer plates. We do not at present know of any form with a first apical shaped like Para- or Metaperidinium with only 5 precingulars from which a *Peridinium* could be derived, but we have *Diplopeltopsis* and *Entzia* with the precingulars 1, 2, 3, 5, 6, 7, shaped like the 6 in *Diplopsalis* and *Peridiniopsis*, with those touching the first apical quite as small as any in Metaperidinium. There seems little doubt here that the fourth apical has been formed between 3 and 5, the original 3 and 4. I should suggest, therefore, that it is more likely that the fourth precingular is the latest of the precingulars in *Peridinium* and not the first and seventh, and that Orthoperidinium is more primitive than Metaperidinium. The fact that in the several species which I have named *Peridiniopsis rotunda* we have a first apical of the same shape as in Metaperidinium is interesting, and suggests a further field for enquiry.

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II. EXUVIELLA PERFORATA GRAN FROM THE ENGLISH CHANNEL.

With Figures 1–9.

In centrifuging samples of sea water from Plymouth Sound and some of the stations outside, some interesting forms of *Exuviella* were found. These are all minute, the largest measuring 22 μ in length, and consequently are easily overlooked, although one at least is abundant during the summer months. This species, which is the commonest *Exuviella* in the Plymouth district, has been identified as *Exuviella perforata* Gran, described by him from the North Sea (1915), and is a new record for the English Channel. As it appears to be very little known and its structure is unusual in several particulars, the following notes are not without interest.

In 1915 Gran (page 99, Fig. 7) describes and figures *Exuviella perforata*, a new species from the North Sea at a depth of 0–20 metres, occurring in the eastern part with a density of 100–760 specimens per litre. It is roundish oval or nearly circular in shape, measuring 22·5–25 μ long and 21–22·5 μ wide, with a broad girdle and thick shell (thickness of cell 14–17 μ). The character, however, that gives it peculiar distinction is the depression in the centre of each valve, as Gran puts it, "with a sharply confined point-shaped perforated deepening in the centre." This separates it from any previously known species of *Exuviella*. The cell contents owing to contraction could not be described in detail, but in a footnote it is stated that living specimens from Arendal in March, 1914, had brown chromatophores. Cleve-Euler (1917) records an *Exuviella*, which he says may be *E. perforata* Gran, as occurring frequently in the Skaggerak, from 0 to 100 metres.

Schiller (1918) describes several new species of *Exuviella* from the Adriatic and Gulf of Naples. Of these *Exuviella bisimpressa* (page 258,

Fig. 11) appears to agree very well with Gran's description and figures. He shows, however, a distinct striation of the girdle, and did not see for certain any perforation in the central depression, although he thinks there appears to be a pore present. Special stress is laid on the twisted flagellar pore, the opening of which is described as being directed sideways. Schiller found three yellow-brown chromatophores in his species. Length 22–27 μ, breadth 18–21 μ. Found in the Adriatic from May to June from 0 to 20 metres, maximum of 180 in a haul at 20 metres; and in August and September from 10 to 150 metres, maximum of 170 at 150 metres. This was the only *Exuviella* found below 100 metres.

There seems no reason to separate the two species, therefore Gran's name must be kept: *Exuviella perforata* Gran=*Exuviella bisimpressa* Schiller.

In the Summer of 1921 a small *Exuviella* agreeing well with Gran's species was found commonly in centrifuged water samples and very fine tow-nets from Plymouth Sound and the surrounding waters. Although casually noticed in former years it was not properly investigated and usually apparently overlooked. The dimpling-in of the centre of each valve characterises the species and when cleared each valve is seen to have this cone-shaped indentation, which occurs exactly in the same position as the pyrenoid described from other species of *Exuviella* (Klebs 1912, Pouchet 1883), where there is no such indentation in the valve. No pyrenoid, however, was observed in the present species. The indentation appears in surface view as a small dark spot in the centre of the valve. No perforation was actually seen, although it is possible that one may be present.

The valves come close together in the centre of the girdle zone as figured by Schiller. This zone may or may not be striated. In the smaller forms, ca 19 $\mu \times 16$ μ, it usually appears to be smooth; in the larger, presumably older forms, ca 22 $\mu \times 19$ μ, it is distinctly striated. The margin of each valve is ornamented with one row of poroids, occurring inside the striations when these are present. This ornamentation is not mentioned by Schiller, but is indicated by faint dots in Gran's lower left-hand figure. It seems to be characteristic of the species, but once a small form occurred, 16 $\mu \times 14$ μ (Plymouth Sound, 2.6.21), which was destitute of these markings and was further abnormal in that one valve had its cone-like structure facing outwards instead of inwards, and anteriorly there were two distinct but very minute spine-like projections in the flagellar region of the same valve (Figs. 5 and 6). This abnormal form is here regarded as belonging to *Exuviella perforata*, although further investigation into more specimens may show it to be a different species.

An interesting feature in *Exuviella perforata* is the structure of each

valve in the region of the exit of the flagella. In all my specimens one valve was excavated at the anterior end as though a bite had been taken out of it; the other, which bore vestiges of very minute spines, was provided with a projection fitting into the excavation in its companion valve. This projection was perforated by 2 pores, presumably for the exit of the 2 flagella (Figs. 3 and 4). Such a structure has not, so far as I know, been described in any *Exuviella*, and no mention of it is made by Gran and Schiller.

The living specimens showed two somewhat irregular bright yellow-brown or almost completely yellow chromatophores, each one pressed against the inner surface of the valve face, leaving a space in the central part of the cell surrounded by the girdle area. Three chromatophores, such as Schiller describes, were never observed (Figs. 1 and 2).

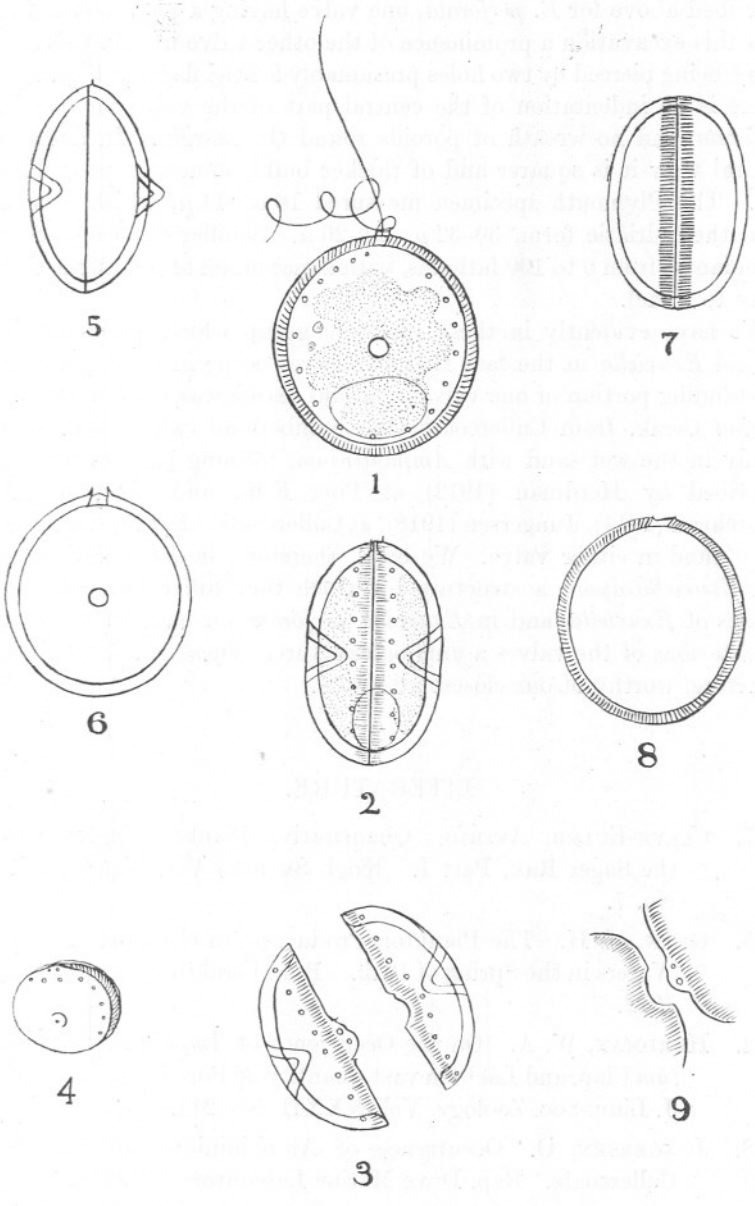
The longitudinal flagellum was held straight out in front, and the transverse flagellum moved rapidly horizontally. The healthy cell moved spirally forward with an occasional backward jerk. The nucleus which stained as a typical peridinian nucleus was situated at the hind end of the cell. The theca stained a faint blue-violet with chlorzinc-iodine.

Only once were more than two chromatophores seen, and in that case division was apparently taking place. The cell, 19 μ long, contained two very much smaller valves and four chromatophores. Further observations on this dividing form are unfortunately lacking.

This little *Exuviella* is widely distributed, and occurs abundantly from June to October, and sparingly in the winter. From close inshore in the region of the Breakwater it extends to beyond the Sound, round by the Mewstone and Eddystone, and was also found at Stations E 1 (14 miles from the Breakwater), E 2 (half-way between Plymouth and Ushant) and E 7 (ca 22 miles south of the Lizard). It is, therefore, well distributed on the west side of the Channel. Most of the samples examined were from the surface, but in some of the water-bottle samples it occurred at 5, 10 and 20 fathoms. It was found inside some of the planktonic animals examined for food, such as copepods, larval decapods and tintinnids.

Assuming that we are correct in regarding the Plymouth species as identical with *Exuviella perforata* of Gran and *Exuviella bisimpressa* of Schiller, we now have its distribution known still further, ranging from the Adriatic and English Channel to the North Sea and the Skaggerak, and further investigation will probably find it in other localities.

Once only from below the Laboratory, 16.6.21 (surface), another species of *Exuviella* occurred. This is probably the *Exuviella apora* of Schiller (1918, p. 258, Fig. 12). It differs from it, however, in having the margins striated. The valves anteriorly have exactly the same structure as is



FIGURES 1–9.

1. Exuviella perforata, valve view
2. ,, ,, ventral view.
3. ,, ,, anterior end of disarticulated valves.
4. Exuviella perforata, side view showing anterior end.
5. Exuviella perforata, abnormal form.
6. ,, ,, ,, ,,
7. Exuviella apora, ventral view.
8. ,, ,, valve view.
9. ,, ,, anterior ends of valves.

described above for *E. perforata*, one valve having a piece out of it and into this excavation a prominence of the other valve fitting, this prominence being pierced by two holes presumably for the flagella (Figs. 7 to 9). There is no indentation of the central part of the valve face, as in *E. perforata*, and no wreath of poroids round the margin. In dorsal and ventral view it is squarer and of thicker build, somewhat resembling a nut. The Plymouth specimen measured 16 $\mu \times 14\ \mu$, which is smaller than the Adriatic form, 30–32 $\mu \times 21$–26 μ. Schiller's species occurred abundantly from 0 to 100 fathoms, with a maximum of 860 at the surface (May to June).

We have evidently in these species a group which differs from the typical *Exuviella* in the fact that two pores occur in front piercing an outstanding portion of one valve. Careful search was made in *Exuviella marina* Cienk. from Cullercoats, Northumberland, which occurs commonly in the wet sand with *Amphidinium*, forming patches with it as described by Herdman (1912) at Port Erin, and Storrow (1913), Whitehead (1914), Jungersen (1918), at Cullercoats ; but no similar pores were found in either valve. We have, therefore, in *Exuviella perforata* and *Exuviella apora* a structure by which they differ from all typical species of *Exuviella*, and in *Exuviella perforata* we have in the central indentations of the valves a unique structure. These small species are, therefore, worthy of our closest attention.

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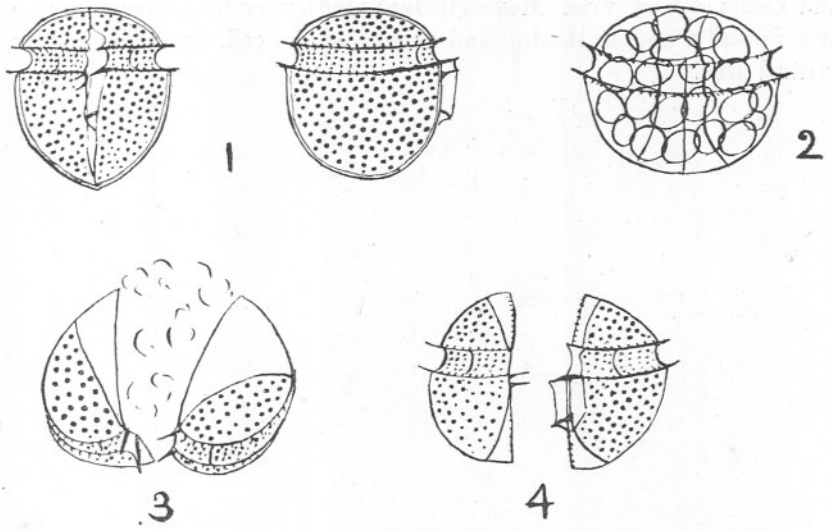
III. A NEW SPECIES OF PHALACROMA.

With Figs. 1–4.

In centrifuged water and in very fine tow-nets there sometimes occurred an extremely small species of *Phalacroma*, which appears to be new. Six specimens in all were found in May, July and August, 1920 and 1921, from Plymouth Sound, in the region of the Breakwater, Cawsand Bay,



FIGURES 1–4. *Phalacroma pulchella* n.sp.

1. Normal individual, ventral and side views.
2. Megacytic form, showing refractive contents of cell.
3. Megacytic form with contents bursting out after treatment with eau de Javelle—seen from above.
4. Separated valves of megacytic form, ventral view.

1 from v. fine tow-net, Cawsand Bay, 5.8.20; 2–4 from Eddystone, W. ½ mile, 9.8.21.

Knap Buoy, Eddystone Grounds and once from the Yealm estuary. It is smaller than any known species of Phalacroma, is perfectly colourless, and measures from 21 μ to 33 μ in length. The cell is full of large refractive bodies (Fig. 2), similar to those often seen in *Dinophysis rotundata*. When not about to divide (Fig. 1) it is slightly flattened from side to

side, but it is more usual to see megacytic forms getting ready for division when there is a large median area added, so that the cell is broader than long. This median part is without sculpture and is thrown off after division. I propose for this little species the name *Phalacroma pulchella*, of which the following is a diagnosis :—

Phalacroma pulchella n.sp.

Cell not much compressed, almost circular in side view, with conspicuous epitheca and broad girdle. Left longitudinal membrane reaching about half-way down the hypotheca. Theca covered with small poroids, which are continued on to the girdle. Transverse lists supported by fine spines. Longitudinal groove extending for a short way along the epitheca. Cell contents colourless, usually with large refractive bodies. Length 0·021–0·033 mm. Habitat Plymouth Sound, Eddystone Grounds and Yealm estuary, rare. Megacytic forms with very broad unsculptured area dorsally, and at the top and bottom of the cell, narrowing at the ventral area.

The Genus *Ilyanthus*, Forbes.

By

T. A. Stephenson, M.Sc.

With Figures 1–3 in the Text.

THE genus *Ilyanthus* was erected by Forbes (1840) for *I. scoticus*; little is known about that species, and its anatomy is undescribed. There is no evidence that any subsequently described species is really a genuine *Ilyanthus*, and the genus-name can only be used provisionally for others until *I. scoticus* can be dissected. Meanwhile the genus-name must be reserved for *I. mitchellii*, the main subject of this paper. Gosse described this species from a specimen which he saw in 1853 (see *Actinologia*, p. 232); it must have been in poor condition or semi-contracted, and his figure (Pl. 8, Fig. 6) is quite misleading. Andres erected (1883, p. 462) a genus *Mesacmæa* for his *Ilyanthus stellatus* of 1880, and, as I hope to show below, *M. stellata* is either specifically or at any rate generically identical with *I. mitchellii*; so that *Mesacmæa* now becomes a synonym; if it should prove, later on, when *I. scoticus* can be examined, that *I. mitchellii* does not really agree with it, i.e. is no *Ilyanthus*, then the name *Mesacmæa* can be revived for *mitchellii* and *stellatus*. Carlgren (Actiniaria of the Danish Ingolf—Expedition, 1921) has given a few anatomical details of *Me acmæa*, gained from notes by Andres in his possession; they agree with my description of *I. mitchellii* as far as they go, but there are not enough of them to make a full understanding of the genus possible. In Andres' 1883 monograph will be found a description of another species, *I. partenopeus* (p. 459). This has been anatomically examined by Simon (1892) and Faurot (1895), and it differs completely from *mitchellii* (and *stellatus*). I have referred to it elsewhere (*Q.J.M.S.*, Vol. 65, 1921, pp. 518, 521, etc.) and have endeavoured to show that it is not only not an *Ilyanthus*, but cannot even remain in the same family as *I. mitchellii*, being in structural grade more like an *Anemonia* which had taken to a burrowing life.

I have been acquainted with *I. mitchellii* for some time, and it is a form unique among anemones and bristling with problems, so I feel that some account of it should appear without further delay, since it has not been sufficiently described. As I shall not be able to work out the finer detail of its anatomy for some time, I now give a description

of its external characters and of a few of its habits, with such anatomical data as are needed to make these intelligible; even now, many points must be left untouched. Some young larvæ of it, which would enable us to trace its development, are badly wanted, and we can hardly hope to clear up its anatomy until these are forthcoming. My experience of the species is as follows: I found a specimen, probably of *I. mitchellii*, in an Irish collection of anemones; and Dr. Allen very kindly lent me some Plymouth specimens of it (preserved) for comparison, one of which I was able to dissect. I also, through the kindness of Mr. Chadwick, received a living specimen, which was moribund, from Port Erin in 1919: this also I dissected. Finally, in 1921, I received two beautiful and healthy living specimens from Plymouth, and these have formed the best of the material for this paper. There are some points in the anatomy which had puzzled me, and upon which preserved specimens could throw no light; but the living specimens, from their manner of holding their tentacles, and their clear display of the arrangement of the latter, solved at least some of these difficulties. As far as we know hitherto, Plymouth is the British headquarters of the species, though it is not of frequent occurrence even there. Mr. Smith tells me that it is obtained outside the Sound, off the Mewstone. Near the Eddystone and further west towards Looe some of the specimens seem to live in clean shelly gravel, and these come to hand naked as in Gosse's figure; others found on fine sandy ground invariably have the column covered by an incrustation which is not very easily removed. He also informs me that a specimen in one of the Plymouth tanks attaches itself firmly at the bottom of a glass dish containing it, and expands above the sand.

DESCRIPTION OF SPECIMENS.

I. The two Living Specimens from Plymouth.

Body.—Form variable, short and broad like a turnip, in contraction, more elongated in expansion, but never (so far as I have seen) becoming vermiform like *Peachia* and *Halcampa*. Physa not distinctly marked off from scapus. Scapus rather corrugated, the mesenteries showing through at the lower end; above, the corrugations take the form of little horizontal projecting shelves of skin, which are rather a noticeable feature; they are not, I think, merely contraction-wrinkles, but are correlated with the presence of a good deal of rough "cuticle," some of which is easily removed; there are also bits of shell and gravel attached to the scapus. Its flesh is brownish purplish flesh-colour, the "cuticle" grey-brown. The upper end of the body is marked off as a narrow, smooth, more cream-coloured capitulum, and in expansion this forms a marked collar, which projects beyond the upper part of the scapus,

leaving a very short " neck " between it and the tentacles (only visible in full expansion) ; this " neck " shows the mesenterial insertions through its wall ; upon it, below each third-cycle tentacle, is a darker patch of colour, sending a line round either side of the tentacle-base ; *between* the bases of every two third-cycle tentacles there runs out a little dark line from the back of the base of an inner tentacle, bounded by two pale ones.

Disc and Tentacles.—In moribund individuals the tentacles may be short and thick, but in these healthy ones, though of considerable thickness, they are long, graceful, and tapering. They can be retracted with a jerk. There are three very clearly marked cycles of them, running 7, 11, 18=36. The fact that there are seven primaries, instead of the usual six of Actinians, is no casual and accidental individual feature, but regular and specific ; and, moreover, these seven are always (so far as my experience goes) held so that they point inwards, in fact they interlace and form a sort of pent-house over the mouth, springing back elastically into that position if one tries to pull them away—they seem rather stiff, and unable to spread outwards at all. Further, *only one of the seven is a directive-tentacle*, although there are two pairs of directives, the other directive-tentacle being a member of the second cycle. The seven primaries divide the others up into seven groups, which, starting from the primary directive-tentacle, run 3.5.5.3.5.5.3, the central group of three containing the secondary directive-tentacle ; the groups of three each contain one secondary and two tertiary tentacles, those of five comprising two secondaries and three tertiaries. The manner of holding the tentacles during life is most interesting. The primaries, as stated, invariably cover the mouth (Fig. 1) ; in daylight they form a rather untidy knot, at night they stretch out more gracefully and cross each other in the middle as shown in the figure. In daylight the animal lies buried up to the disc, and the secondaries and tertiaries lie spread abroad, more or less flat upon the sand, in seven distinct radiating groups. As far as one can tell, the primary tentacles are the shortest, the secondaries intermediate, and the tertiaries longest—as one expects in such a form. At night the upper end of the body is raised above the sand, the collar projects strongly, and the tentacles assume a stiffer and less flaccid habit. In my specimens those of the smaller one usually maintained seven radial groups rather decidedly (cf. Andres' figures of *I. stellatus*, Fig. 38 and Pl. IX, Fig. 5) ; those of the larger one typically spread themselves in such a way that all the secondaries pointed upwards and outwards in a regular ring, the tertiaries, in marked contrast, turning outwards and downwards, so that their tips touched the sand. I may mention that these creatures would not bury themselves, but expanded fully only when I buried them. They lived well in captivity, and ex-

panded permanently save when irritated, and then did not retract for long. The colouration of disc and tentacles is complicated, and would be easier to represent in a drawing than in words ; I hope to be able to illustrate the species fully, at a later date. The colours and markings form an intricate and beautiful pattern, softly shaded, and comprising varying tints of straw-colour, light purplish grey, browner shades, purplish browns, and so on, which vary to some extent according to the degree of distension of the tissues. The most striking point about the tentacles in general is that there is a dark median stripe running longitudinally down the oral face of each, from the tip to about half-way down ; besides this their oral faces are marked, roughly speaking, by a series of alternately light and dark transverse marks, which, towards the tentacle-base, are more or less V-shaped, the V pointing towards the mouth. No two V's on any one tentacle are quite alike, and there are differences between the patterns of the three cycles of tentacles, further complicating details coming in about the tentacle bases ; but in so unique a species it seems superfluous to write down every detail, and that will be done better in a figure later on. The aboral faces of the tentacles are not much marked, save in the case of the primaries, where the *backs* of the tentacles are, apparently, permanently exposed to the light, and here there is a certain amount of pattern—quite an unusual feature. The most striking feature in the colouration of the larger specimen was the conspicuous marking out of the directive-tentacles by large patches of opaque white, quite distinguishing them from the rest. The primary directive-tentacle was white at the base, on its back ; the secondary directive-tentacle was white over most of its basal part, at least on the inner face. In the smaller specimen the secondary directive-tentacle was almost wholly dull purple, the primary being a little purple at the base, and having a white aboral stripe.

The disc is also patterned. The reddish mouth is surrounded by a pale ring, followed by a rather narrow dark one, which is made up of a series of dark marks on the inter-radii, these being darker on the tertiary inter-radii than on the others ; the former break into the pale ring as little points, giving it a star-like effect. Next there comes a broader pale ring, all the inter-radii sharing in its formation. The tertiary inter-radii have no other markings till the base of the tentacle is reached, where there is a pale diamond, save that their outer parts are less pale than the inner. The primary and secondary inter-radii have a dark transverse mark shading off at the four corners into the radii, and this is farther from the mouth on the secondaries than on the primaries ; between this mark and the tentacle base is a pale roughly triangular mark, much larger for the secondaries than for the primaries.

The above details of colour are compiled from the two specimens, there

being differences of detail but the same general plan in both ; the larger had the colours less well defined than the smaller.

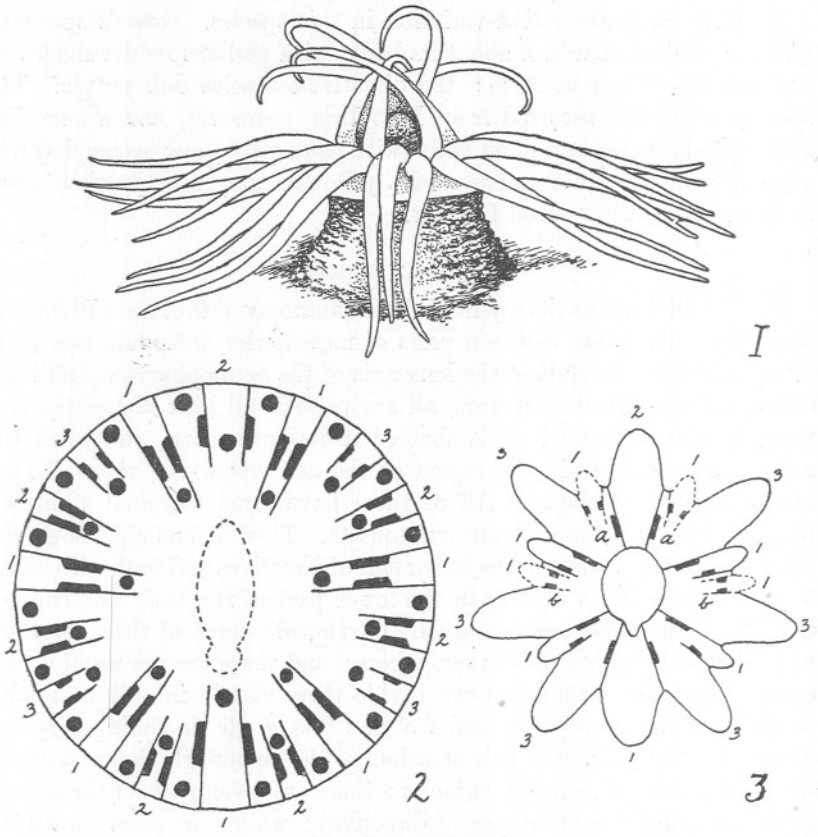
II. Variation.

There is evidently colour-variation in this species. Gosse's specimen had a good deal of scarlet about its body, with perhaps redder shades on the disc also, than in mine ; the directive-tentacles dull purple. The specimen which I received from Port Erin, moreover, had a beautiful softly vivid, orange-vermilion body (with paler collar) and actinopharynx, general colours of disc and tentacles yellowish grey and purplish grey. It had no " cuticle " when I saw it.

III. Anatomy.

First I will describe the essentials in the anatomy of the larger Plymouth specimen. There are eighteen pairs of mesenteries, including two pairs of directives at the ends of the long axis of the actinopharynx ; all have strong circumscribed retractors, all are perfect, all bear filaments (save perhaps a single one which is aborted in its lower part), and when the animal is cut across in the region of the actinopharynx, they all look about equally developed. All of them have large marginal stomata ; but I could not be sure about oralstomata. There is one siphonoglyphe only, and it corresponds to the larger pair of directives and to the directive-tentacle of the inner cycle. In the lower part of the body one can see that the mesenteries are not equally developed ; some of them, or their muscles, reach farther down than others ; and there are the usual differences in breadth ; thus one can divide them clearly into three grades or cycles. There are seven pairs of the first grade, including *only one* of the directive pairs (the pair attached to the siphonoglyphe) and corresponding to the seven inner tentacles ; there are seven pairs of the second grade, including the other pair of directives ; while four pairs constitute the third grade ; the eleven tentacles of cycle 2 belong to the endocoels (7+4) of grades 2 and 3 of the mesenteries, while the 18 tertiary tentacles correspond to the 18 exocoels. In other words, the arrangements of mesenteries and tentacles exactly correspond, and are symmetrical about the long axis of the throat ; but while the tentacles run 7, 11, 18, the mesenteries run 7 p., 7 p., 4 p. (7+4=11), the remaining 18 tentacles being exocoelic. The accompanying Fig. 2 shows a diagrammatic transverse section taken below the level of the actinopharynx, which is dotted in simply to show the relationships of the mesenteries to the siphonoglyph ; it would not actually appear in such a section. All three mesenterial grades are fertile, grade 3 being but sparsely so, grade 2 intermediate, grade 1 richly fertile ; the grade 2 directives are well supplied. Ciliated streaks are present on the filaments.

I think it may be assumed that the type of structure above described is specific and not an individual freak. It is perfectly regular. The Port Erin specimen agreed exactly with what I have recorded for the Plymouth



Ilyanthus mitchellii.

Fig. 1.—Sketch of a living specimen with most of its body buried, showing some of the characteristics of its way of holding the tentacles. This is the smaller Plymouth specimen, perhaps a little enlarged.

Fig. 2.—Diagrammatic transverse section below the level of the actinopharynx. See text. The relations of the tentacles to the mesenteries are shown by including black spots for the tentacles.

Fig. 3.—Reconstruction designed to illustrate the theory proposed in the text, with regard to the development of the species. In this figure the numbers indicate the cycles to which the tentacles will eventually belong. In Fig 2 numbers refer to the mesentery-cycles.

specimen, as far as I could get details from it; also, another Plymouth specimen I dissected some time ago would, so far as I remember, agree also in essentials, though I have not the details at hand. The second of my living Plymouth examples, having the same arrangement of tentacles as the one dissected, is bound to have the same arrangement

of mesenteries, for this is a case in which the one can be deduced from the other. Moreover, in Andres' specimens of *I. stellatus*, there was the same state of affairs—seven inner tentacles bent over the mouth, and seven bunches of outer ones in two cycles. In his examples they ran 3.5.3.3.3.5.3 or 3.3.5.3.5.3.3, giving two tentacles less on each side than in mine ; but that is only because his individuals were less fully developed ; I have seen cases myself with fewer than 36 tentacles, one, I believe, having 28 only. Deducing the arrangement of mesenteries in Andres' examples from that of their tentacles it is evident that the third-grade mesenterial pairs can appear first in *either* the asulcar or in the lateral members of the six main exocoels which lie nearest the siphonoglyphe.

DISCUSSION.

I. Ilyanthus and Mesacmæa.

I do not think anyone who reads Andres' account and looks at his figures can doubt that these two genera are identical (using the name *Ilyanthus* as applying to *I. mitchellii*). The unique arrangement of tentacles that occurs in both, and the unique way of holding them, are too striking resemblances to be overlooked ; and they are borne out by almost everything else. There may be colour-differences, but only such as one expects between British and Mediterranean varieties, and even among our British specimens there are such differences : and there are also several resemblances, such as the axial stripe on the oral faces of the tentacles (rather an important detail), and so on. I am inclined to think that not only are the two genera identical (so much I think is certain), but very likely the species *mitchellii* and *stellatus* also. There is certainly one curious difference between them—in *stellatus* the tentacles of the middle series seem to be a little longer than the others ; but, as the number of tentacles seems to show that these specimens were not quite fully grown, I am not sure that it is impossible for the tertiary tentacles to be considered as being a little short of their eventual length ; they might in the end outgrow the secondaries—although the probable order of tentacle-succession in this genus might not fit in with that idea. But the specific identity of the two is of minor interest, it is the generic identity which is of most importance.

II. Problems connected with Ilyanthus.

Is it possible to make suggestions which will help us to understand the curious state of affairs in *Ilyanthus?* We cannot know anything for certain until its development has been worked out, and it is, of course, always quite possible that the larvæ when examined will reveal something

quite unexpected ; but it does seem worth while to make at least some attempt to think out, quite tentatively, a conceivable explanation. It is possible to think of various schemes by which the result might have been obtained, but as far as I have been able to carry my study, the following seems the likeliest.

In the first place, if *Ilyanthus* is to be compared, for enlightenment, with any other genus, the form indicated seems to be *Peachia*, which, although very different from *Ilyanthus* and less strange, is at any rate probably one of its nearest relatives. In the development of *Peachia* there is the usual 8-rayed Edwardsia-stage, and later on a 12-rayed stage with six pairs of mesenteries. The eight tentacles of the Edwardsia-stage, however, give way to the twelve of the next stage, in a way differing from that of the more usual anemones, the added four arising in connection with the four lateral primary endocoels, so that of the eight original tentacles, six become exocoelic, the other two being the directive-tentacles ; and the exocoelic six are persistently larger than the endocoelic ; just as in the adult *Ilyanthus* the exocoelic tentacles are the largest. It seems that in thinking of *Ilyanthus* it would be unwise to suppose that it had no Edwardsia-stage and no 12-rayed stage ; that would be stretching the case too widely away from the normal, without evidence for doing so. The safest plan to follow, perhaps, would be to postulate for it both these stages ; also, it seems a fairly probable supposition that it develops its tentacles according to the *Peachia* plan. By a slight modification of the history of *Peachia*, moreover, the state of affairs exhibited by *Ilyanthus* can be obtained. Let us suppose that early in the 12-rayed stage, for some reason, two pairs of secondary mesenteries (which should in a normal case come later) appear in two of the exocoels, and grow so fast that they outstrip the asulcar directives, which tend to be rather backward. They bring with them two endocoelic tentacles ; and it may be that both they and their tentacles grow so strongly that they take their places in the first cycle ; while the asulcar directives themselves are relegated to a second grade of size as compared with the interlopers, and their tentacles to the second cycle. So much being admitted, we have accounted for the 7-rayed condition just as it is actually represented in the adult, and the subsequent development offers no difficulty. All this would regularise itself later on, assuming adult proportions, as it does in any anemone. It may be that my suggestion is rather bizarre, but not more so than certain other suggestions which have actually proved correct, in the cases of Endocoelactidæ and of *Tealia*. In *Tealia*, in fact, the 10-rayed adult condition *does* arise from an earlier 6-rayed state, by the precocious growth of four secondary mesenterial pairs which assume primary rank ; and *Ilyanthus* may only be a more extreme case of the same sort of thing.

There is one important point which my suggestion has so far left untouched—in *which* pair of exocoels do the precocious mesenteries appear ? We may perhaps dismiss the sulcar exocoels as being too far away from the scene of action ; and if the sulcar lateral primary pairs (as the fact that they are, in the adult, the most fully developed of all, seems to suggest) in the adult include the sulco-lateral couple of the larva, it is impossible for the precocious pairs to have appeared in the sulcar exocoels. We have, therefore, to choose between the asulcar and the lateral exocoels. In *Peachia*, *Eloactis*, and *Haloclava* there is a backwardness or growth-atrophy about the asulcar region : no metacnemes are formed there, the asulcar directives tend to be less developed than the sulcar, and there is no asulcar siphonoglyphe ; in this *Ilyanthus* resembles them. If the reduction of the asulcar directives, etc., indicate a general (relative) growth-reduction in the asulcar region, precocious or quickly growing mesenteries would be unlikely to develop in that very area, and it would seem more natural to expect them in the lateral exocoels, in a position lateral to the long axis of the actinopharynx, and the region in which, according to the adult, most of the mesentery-formation has taken place. This is actually the spot in which the first of the precocious pairs in *Tealia* make their appearance. This would seem to involve a growth stage at which two endocoelic tentacles, destined to belong to the same cycle, would be adjacent to each other on each side of the axis, until a new exocoelic tentacle came between them. For most of the above suggestions in favour of the lateral exocoels I am indebted to Dr. Gemmill, and agree with him in preferring them, though I do not think we need rule out the other possibilities altogether in the present state of our knowledge. In Fig. 3 I have tried to illustrate both the likelier possibilities ; in this figure the continuous lines represent the actual state of affairs in a *Peachia* larva ; the dotted portions represent the suggested additions for *Ilyanthus ;* but *both* alternatives are included to economise space. Of course, only *one* of the alternatives would actually take place, *either* the pairs a, a would be the precocious ones, *or* the pairs b, b, there would not really be four pairs involved.

Finally, it may be of interest to make a brief comparison of *Peachia* and *Ilyanthus*. I cannot feel that they need separate families (any more than *Tealia* does by reason of its decamery) ; they fit in well enough with the genera *Eloactis* and *Haloclava* (and probably also *Harenactis*), and together constitute the true Ilyanthidæ ; each of the genera is peculiar in some way. *Peachia* is unique, as well as *Ilyanthus* (though not so markedly unusual), by virtue of its conchula ; and its mesenterial and tentacular arrangements are also peculiar. It has twelve tentacles and twenty mesenteries ; *Ilyanthus* has up to thirty-six tentacles and thirty-six mesenteries (sometimes more ?), and no conchula ; *Peachia* has six

primary mesenterial pairs, including both pairs of directives, *Ilyanthus* seven primary pairs, including *one* of the pairs of directives only ; *Peachia* carries its tentacles, when alive, in an ordinary way, *Ilyanthus* in quite an unusual way. *Peachia* shares with *Ilyanthus* a single siphonoglyphe and a predominance of the sulcar directives over the asulcar, and also its general form and that of its tentacles. It is becoming evident, from the observations of Elmhirst (*The Zoologist*, Jan., 1915, p. 3) and others, that *Peachia* is at least to a considerable extent a current-feeder, and does not make a great deal of use of its tentacles. About *Ilyanthus* in this connection we so far know nothing. It typically keeps its mouth raised on a very steep little cone or spout, within the tent formed for it by the inner tentacles (Fig. 1).

A Contribution towards the Life History of *Parorchis acanthus* Nicoll, a Trematode in the Herring Gull.

By

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AND

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With 1 Figure in the Text.

Parorchis acanthus is a Trematode described by Nicoll (1907) from the bursa Fabricii and rectum of the Herring Gull, *Larus argentatus;* in 1906 he first described it as *Zeugorchis acanthus.* Originally found at St. Andrews, it is now known to be common at Millport and also occurs in gulls from the Northumberland coast, and probably is abundant elsewhere. Nicoll found it once in the Common Gull, *Larus canus.* In 1907 (Lebour, 1907) one of the present writers described a cercaria occurring in rediæ from *Purpura lapillus.* This was named *Cercaria purpuræ* sp. inq., and afterwards identified with young stages of *Parorchis acanthus* in the Herring Gull (Lebour, 1914). It was thus shown that the first host of this Trematode is *Purpura lapillus* and the final host the Herring Gull, but the intermediate host was unknown. Localities for the cercaria were Loch Ryan, Wigtownshire; Budle Bay, Fenham Flats and Cullercoats in Northumberland; Robin Hood's Bay in Yorkshire; and Millport. Adults occur at St. Andrews, Northumberland coast and Millport, as already stated.

The Cercaria appeared to be closely related to *Echinostomum* cercariæ, and it was suggested (Lebour, 1914) that the intermediate host would probably be some marine bivalve, as it is often so with *Echinostomum* (e.g. *E. secundum* in *Mytilus edulis* and *Cardium edule*, *E. leptosomum* in *Scrobicularia tenuis*). This suggestion now proves to be correct, and we are able to show that the intermediate host of *Parorchis acanthus* is *Mytilus edulis* or *Cardium edule.*

On August 13th, 1921, at the Millport Station some larval cercariæ were noticed by Elmhirst swimming in a small glass aquarium, which contained a collection of *Purpura lapillus*, *Cardium edule* and *Mytilus edulis.* These cercariæ, which swam actively by a strong side to side lash-

ing of the tail, appearing to the naked eye rather like Chironomid larvæ, were identified as *Cercaria purpuræ*. The *Cardium* and *Mytilus* were kept alive, and at the same time other specimens of *Mytilus* were put in another aquarium and some cercariæ from the infected *Purpura* added.

On September 4th, 1921, these *Cardium* and *Mytilus* from the first aquarium were carefully examined, with the result that the *Cardium* (one specimen only) contained two distinct species of Trematodes encysted in its tissues; the more abundant species, which occurred in the foot, was *Echinostomum secundum*, the other which occurred both in the foot and mantle, but not so abundantly, was undoubtedly *Cercaria purpuræ*. Two specimens of *Mytilus* also contained the *Cercaria purpuræ* in the mantle, more abundantly than Cardium, but no *Echinostomum*. *Parorchis acanthus* is such a very distinct species that there is no mistaking it, and there seems no doubt at all that we have now found in *Cardium edule* and *Mytilus edulis* the intermediate hosts of *Parorchis acanthus*. The life history is this :—

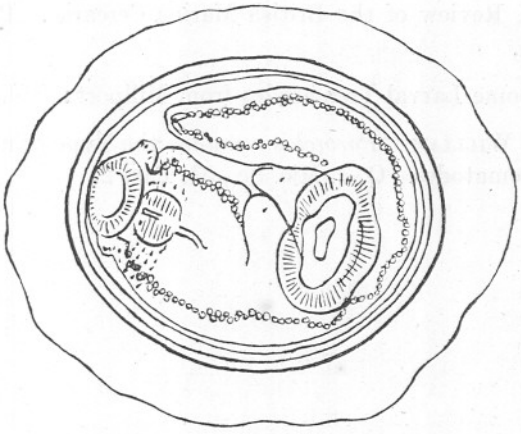
First Host.	Intermediate Host.	Final Host.
Purpura lapillus.	Cardium edule.	Larus argentatus.
	Mytilus edulis.	Larus canus.

The *Mytilus* from the second aquarium were also examined, but were not infected, possibly because the cercariæ removed from the *Purpura* were not quite ready to enter their second stage.

The cysts differ from those of *Echinostomum* in their shape, one side being flattened and the other convex, the outer sheath being drawn out into a rim, giving the cyst the appearance of a hat (Fig. 1), not unlike the egg capsule of *Littorina littorea*. The outer sheath is very tough and thick, ca 0·012 mm., and of a brownish colour. The cysts of *Echinostomum* being ordinarily rounded or oval-shaped, they are at once distinguished from these. The normal habitat for these cysts of *Parorchis* seems to be the mantle; and the flat side was, in most cases, lying nearest the shell.

Seen from above the cyst is roundish oval, measuring without the margin from 0·24 mm. to 0·28 mm. long by 0·20 mm. to 0·22 mm. broad. The wall consists of two parts, the outer brownish tough sheath, to which is attached the flattened margin, and an inner perfectly transparent sheath, which is very thin walled. In this latter sheath lies the larva curled up (Fig. 2). Through the cyst wall can be seen the thick spines set on the body, the main excretory ducts leading to the excretory vesicle, the oral and ventral suckers and the pharynx. The worm when pressed out of the cyst is like the older free-swimming cercaria from *Purpura*, but

without the tail, and also clearly resembles the young stages of *Parorchis acanthus* in the Herring Gull. There can be no doubt that we have here the same species. The suckers are slightly larger than in the tailed form : tailed form, oral sucker 0·06 mm., ventral sucker 0·099 mm. ; encysted form, oral sucker 0·08 mm., ventral sucker 0·10–0·14 mm. In the small forms from the gull both suckers have grown and in the adult they are still larger, the ventral nearly twice as great as the oral.



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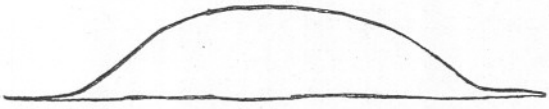

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Fig. 1. Cyst in side view.
Fig. 2. Cyst from above with contained Trematode.

It is to be noted that no younger stages than the rediæ were ever seen in the *Purpura*, and this is interesting in the light of recent observations made on a species of *Echinostomum* (*E. revolutum*) by Johnson (1920), who finds that the miracidium develops into a mother redia, and this gives rise to the daughter rediæ. It is very probable that the same takes place in *Parorchis*, and that the rediæ so commonly found are the daughter rediæ developed from mother rediæ and that no sporocyst occurs.

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Note on the Occurrence of *Echinus esculentus* above Low-tide Mark on the Cornish Coast.

By

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When collecting on the shores in the Plymouth area, while a member of the Easter Class at the Marine Biological Laboratory, Plymouth, in 1921, I was struck by the absence above low-water mark of *Echinus esculentus*, as contrasted with its abundance in this zone at Port Erin in the Isle of Man; for there, as Chadwick states, "it may be collected by hand on the beach, and on the ruined breakwater at low-water of spring tides." There is no record of this species except below tide-marks in the "Plymouth Marine Invertebrate Fauna" list (*Journ. Marine Biological Association*, Vol. VII, 1904).

In 1849 W. P. Cocks, in the *Trans. Penzance Natural History and Antiquarian Society* ("List of Echinodermata procured in Falmouth and Neighbourhood from 1843 to 1849"), recorded the occurrence of this species thus:—

> "*Echinus sphæra.*—Trawl refuse; common: young specimen found attached to stones, low-water mark."

Later (1887–8), in the same journal, G. F. Tregelles, in a paper on "Echinodermata of Mount's Bay," makes the following statement of the species of the genus Echinus: "The commonest and largest is *E. esculentus* (Pennant) [*E. sphæra* Forbes], which literally swarms off this coast at all depths. It is brought in by trawlers; it is found in crab-pots, into which it climbs laboriously after bait; the seaweed gatherers obtain them in from one to two fathoms of water. The Mousehole fishermen call them 'zarts,' doubtless an old Cornish word."

The Mousehole fishermen have told me that "zarts" are often found exposed at low-water on the shore of the islet lying off the village and also on the shore of the mainland opposite this island. Accordingly I took advantage of the spring tide of April 23rd, 1921, to visit the island at low-water, when I found two large specimens of *E. esculentus* exposed on the rocks in sheltered positions on the landward shore. The greatest diameter of one was 11·3 cms. in length, while its height measured 8·9 cms. The corresponding measurements of the other specimen were

11·5 cms. and 8·7 cms. respectively. The spines of both were tipped with green. Both were ripe females and discharged their eggs in captivity.

On September 4th of the same year, the tide being a good one, I found another large specimen of the species near low-water mark on the shore of the mainland sheltered by the island, and two other specimens were brought to me that had been found on this shore on the previous day. All were large, healthy specimens.

It should be mentioned that both these shores are sheltered on the west by the mainland, while the island acts as a breakwater on the east. This sheltered position may be connected with the occurrence of *Echinus esculentus* above low-tide mark in this locality. However, it is rather its apparent absence from the beach fauna elsewhere in Cornwall that calls for explanation. Two alternatives suggest themselves. Either the climatic conditions of Cornwall or the Cornish seas, as contrasted with those of the Isle of Man, are unsuitable to the occurrence of "zarts" in the tidal zone; or the general climatic conditions are suitable, and, except in the case of Mousehole, local conditions are unfavourable.

The solution of the problem would be made more possible by knowledge of the occurrence or absence of this sea-urchin between tide-marks at other parts of the British coast.

Marine Biological Association of the United Kingdom.

Report of the Council, 1921.

The Council and Officers.

Four ordinary meetings of the Council were held during the year, at which the average attendance was fourteen. A formal inspection of the Laboratory was carried out on behalf of the Council by Dr. G. P. Bidder, the other members of the Committee appointed for the purpose being prevented from travelling to Plymouth by a threatened railway strike. The thanks of the Association are due to the Council of the Royal Society, in whose rooms the meetings have been held.

Meeting of Representatives of Marine Biological Stations.

A meeting of representatives of Marine Biological Stations, called together by the Development Commissioners, was held at the Plymouth Laboratory on March 18th and 19th under the Chairmanship of Mr. W. B. Hardy, Secretary of the Royal Society, Chairman of the Advisory Committee on Fishery Research. The various researches being carried out at the Laboratory were explained by the members of the scientific staff, and the meeting expressed its satisfaction with the character of the investigations which were being undertaken and the methods by which the problems were being attacked.

The Plymouth Laboratory.

The new Allen Building was taken into use at the beginning of the year, and has proved to be in every way satisfactory for the purposes for which it was designed. The additional working space has been a great convenience, and the rooms added to the library on the top floor of the west wing of the main building are most valuable and convenient. The old fish laboratory on the ground floor of the east wing has been fitted up as a physiological laboratory, and with the small chemical laboratory in front gives adequate accommodation for the commencement

of the physiological work, pending the carrying out of the full plans for the new building.

Electric light has been installed in all the laboratories, and electric power cables have been brought to the building, with a view to the use of electricity for circulating the sea-water in the aquarium tanks.

Practically £1000 has been received for the sale of specimens for museums, teaching purposes and research work, and, as in former years, a quantity of nets and gear has been provided for various scientific expeditions.

The Boats.

A steam-drifter, which has been named *Salpa*, was purchased from the Admiralty for the sum of £3500, and a sum of £1716 was spent in reconditioning her and providing her with trawling gear. The vessel, which was built of wood at Lowestoft, by Messrs. S. Richards and Co., in 1918, is 88 ft. long between perpendiculars, beam 19.9 ft., depth 9.5 ft. and draught 11 ft. Her gross tonnage is 94.6 and registered tonnage 41.44. The engines, built by Messrs. Wm. Beardmore and Co., are triple expansion, with cylinders 9½ in., 15½ in. and 26 in., by 18-in. stroke, and with indicated horse-power 270. The pressure on the boiler is 180 lb. per sq. in., and speed 9¼ knots. Permanent accommodation for the naturalists and laboratory accommodation have not yet been fitted in the vessel.

The *Salpa* reached Plymouth towards the end of June, and has since been working regularly. She is a powerful sea-boat and is much more capable of facing rough weather than the *Oithona*.

The latter vessel was in commission from March to June. The Council is anxious to sell her, but up to the present no satisfactory offer has been made.

The sailing boat *Anton Dohrn* has been used for work in Plymouth Sound. It is proposed to buy a motor boat for shallow-water collecting.

The Staff.

Dr. W. R. G. Atkins, O.B.E., M.A., F.I.C., of Trinity College, Dublin, has been appointed head of the Department of General Physiology. Dr. Atkins commenced work at the Laboratory in February.

Mr. E. W. Nelson left Plymouth in July, having been appointed Scientific Superintendent of the Scottish Fishery Board.

Mr. H. W. Harvey, B.A., formerly of Downing College, Cambridge, has been appointed administrative and hydrographical assistant, and joined the staff at the beginning of November.

Miss Worsnop, from the University of Leeds, has assisted Dr. Orton

with his special investigations on oysters, and Mr. J. R. Baker, of Oxford, acted as temporary assistant naturalist during the summer vacation.

Occupation of Tables.

The following naturalists have occupied tables at the Plymouth Laboratory during the year :—

J. R. Baker, Oxford (Quantitative Distribution of Benthic Animals).
Miss Bargmann, London (Elasmobranchs).
Miss L. Batten, Bristol (Algæ).
Dr. G. P. Bidder, Cambridge (Sponges).
Miss D. R. Crofts, London (Digitiform appendix in Elasmobranchs).
W. de Morgan, Plymouth (Protozoa).
J. S. Dunkerly, Glasgow, Ray Lankester Investigator (Fish Myxosporidia).
Dr. I. Frost, Birmingham (Artificial Parthenogenesis).
Prof. W. Garstang, Leeds (Echinospira).
Miss S. Garstang, Oxford (Echinoderm Larvæ).
Prof. E. S. Goodrich, F.R.S., Oxford (Nervous System of Fishes).
Dr. Helen Goodrich, Oxford (Parasitic Protozoa).
C. R. Harington, Edinburgh (Teredo).
E. G. Matthews, Cambridge (Marine Bacteria).
H. G. Newth, Birmingham (Echinoderms).
F. A. Potts, Cambridge (Teredo).
A. D. Ritchie, Manchester (Medusæ).
Mrs. E. W. Sexton, Plymouth, Ray Lankester Investigator (Gammarus).
C. C. Stockman, Harvard and Cambridge (Anemones).
Lieut.-Colonel H. J. Walton, I.M.S. (Plankton).
Miss E. Worsnop, Plymouth (Oysters).
J. F. G. Wheeler, Bristol (Postlarval Fishes).

The Easter Vacation Course in Marine Biology was held this year, conducted by Dr. J. H. Orton, and was attended by twenty-five students from Oxford, Cambridge, London and Edinburgh.

Mr. E. W. Shann also brought a class of nine students from Oundle School for practical work.

General Work at the Plymouth Laboratory.

Work on the Life Histories of fishes has been continued by Mr. Clark on the same lines as previously, but the material has been collected from a much wider area, which now includes the Hydrographical Stations E1, E2, N1, N2 and E7. So far, the results show that the western entrance to the English Channel is an intensive spawning area for mackerel and for pilchard and the Plymouth area for whiting. The use of the pelagic trawl during the winter months may furnish some interesting data, though October, November and December have been comparatively poor in the production of young fish—*Gadus luscus* (the pout) excepted.

A short paper has been prepared on the Gurnards, which will include the identification of the early young of four species—*Trigla gurnardus* (grey gurnard), *T. cuculus* (red gurnard), *T. hirundo* (tub gurnard), *T. lineata* (streaked gurnard), two of which, the red and the streaked gurnard, were hitherto unknown in the young stage.

Much time has been given to a study of the eggs and early young of the rays and skates, which occur in the neighbourhood of Plymouth. A complete series of embryos of two species, *Raia clavata* (Thornback) and *Raia brachyura* (Blonde), has been secured by rearing the eggs in the Laboratory tanks. Special features in this work have been the methods of attachment of the egg capsules, the aeration of the egg, the orientation of the embryo, the persistence of the branchial filaments to the end of the embryonic period, the reabsorption of the tail and the period of incubation. The characters of the newly hatched rays have been determined, up to the present, for four species—*R. clavata*, *R. maculata* (Homelyn), *R. brachyura* and *R. naevus* (Cuckoo ray). The descriptions of these and of the later stages are being put forward with the suggestion that other species may be similarly treated, so that the uncertainty in the synonymy of many of the rays and skates, which has been due, in no small measure, to the large amount of variation in the young stages, may at least be partially cleared up and leave the field more open for a systematic study of the adults.

Nine species, six rays and three skates, occur more or less frequently at Plymouth, and these will form the nucleus of a systematic and biological study of the genus *Raia*, though material is being collected also from more distant localities.

Mr. E. Ford has completed and published in the Journal a paper on the life histories of dogfishes, which includes the results on the rate of development of the embryos of the spur-dogfish (*Squalus acanthias*), referred to in last year's report. Since the *Salpa* has been in commission, good catches of *Squalus acanthias* have been made with the large Otter Trawl, so that it has been possible for the studies to be continued. Evidence relating to the growth both of embryos and adults, and to the number and size of embryos has been collected. Mr. Ford has also directed attention to the study of the food of fishes. The stomach contents of fishes captured by the *Salpa* have been examined and recorded.

Dr. Orton has continued his investigation into the cause or causes of the mortality in oysters during the summer of 1920. These researches, originally undertaken for a conjoint committee of the Development Commission, the Ministry of Agriculture and Fisheries and the Oyster Planters' Association, have been continued mainly under the Marine Biological Association, but with the co-operation of the Ministry of Agriculture and Fisheries, with a view to testing as completely as possible

the value of some possible factors, indicated from the earlier investigations, which may have caused the mortality.

The work on rate of growth and breeding in marine animals in relation to the temperature factor has been continued by Dr. Orton, and includes an experiment carried out at Spitzbergen, with the object of investigating the rate of growth of marine animals under conditions approaching those obtaining in Arctic regions.

At the request of the Ministry of Agriculture and Fisheries the Association has undertaken to co-operate with the French and Irish Fishery Departments in a hydrographical investigation of the area to the south-west of the British Isles. A station (E1) south of the Eddystone is being worked monthly by the *Salpa*, temperatures and water samples being taken at different depths. Five times a year a cruise lasting about three days is being undertaken from Plymouth to Ushant (Stations E1, E2 and E3), and then northwards to a station at the mouth of the Bristol Channel (Stations N1, N2, N3 and E6). The work was commenced in April, 1921, by Mr. Nelson, and has been continued by Dr. Atkins and Mr. Harvey. Dr. Atkins has determined the hydrogen-ion concentrations of all the samples of sea-water on board the *Salpa* as soon as they were taken, and samples have been sent to the Government Chemical Laboratory in London, where the salinity has been determined. The results are being sent to the Bureau of the International Council, where they will be co-ordinated with those obtained by the Irish to the westward and the French to the south-west.

Dr. Lebour has been occupied with work on the plankton of the Plymouth area, and has this year devoted special attention to a study of the food actually eaten by the more important planktonic organisms. On account of the difficulty of the investigation comparatively few data have as yet been obtained, but some interesting facts have emerged, showing that certain species keep to special kinds of food, even sometimes to only one kind. A preliminary paper on this subject is nearly ready for publication.

Dr. Lebour has also examined very fine tow-nettings and centrifuged samples of sea-water for Peridinians. This group was systematically investigated and much new information gained both as to undescribed species and as to the structure of those already known. The much discussed *Diplopsalis lenticula* was specially attended to, with the interesting result that all the three forms attributed to that species were found, including Pavillard's Mediterranean form, which is for the first time recorded for north European waters. A paper on this subject is ready for publication, together with a short account of *Exuviella perforata* of Gran, now found to be common in the district, and a new species of *Phalacroma*.

The microplankton from the water-samples taken during the hydrographical cruises has also been examined and recorded.

A paper on Regeneration and Reproduction of the Syllid *Procerastea*, by Dr. Allen, has been published in the *Transactions* of the Royal Society. It is shown that in addition to the usual method of sexual reproduction, this worm reproduces asexually, by fragmentation followed by regeneration at both ends of the fragments. The fragmentation takes place in a definite way, and regeneration of anterior segments continues until the original segments come to occupy exactly the same position in the regenerated worm as they had occupied in the parent.

Work on Mendelian inheritance in the Amphipod *Gammarus chevreuxi* has been continued by Mrs. Sexton. In a paper published in the Journal, jointly with Mr. J. S. Huxley, it is shown that in certain strains of this Amphipod used in the experiments animals occur which are intersexes. Certain specimens which are shown to be female intersexes are described in detail. On reaching maturity these female intersexes usually resemble normal females more or less closely, but gradually come to resemble males more and more nearly, and grow to a very large size.

In continuation of this work, Mrs. Sexton has since made an accurate study, with drawings, of each moult of *Gammarus chevreuxi* from the time of hatching until maturity is reached for males and females, as well as for an intersex.

Mr. C. R. Harington, who is carrying out investigations for the committee of the Institute of Civil Engineers, which is making a special study of the deterioration of structures exposed to sea-action, has spent some time at the Laboratory continuing his experiments with *Teredo*, and has made some interesting observations, which will shortly be published, on the habits of the larva of that mollusc.

Physiological Laboratory.

The Laboratory has been equipped with apparatus of general utility for chemical and physiological research, and is now available for a limited number of outside workers.

Dr. W. R. G. Atkins has been engaged in work upon hydrogen-ion concentration in connection with various biological problems, and in its seasonal changes in salt and fresh water. These are of importance in relation to the balance between plant and animal life in the sea, since photosynthesis tends to increase the alkalinity. There are also changes in hydrogen-ion concentration as an estuary is approached, these being more marked than alteration in salinity.

It has been ascertained that algal tissues are much less alkaline than

is the sea-water, and, further, that active photosynthesis due to intense insolation may so increase the alkalinity of the water as to kill algæ, even when not directly exposed to the sun.

It has long been known that, when stored, sea-water increases in hydrogen-ion concentration. This has been made the basis of a method for ascertaining the carbon in organic matter in the water, including suspended matter. Evidence has been obtained that in settled summer weather this is high near the surface and at the bottom, also in a layer of water usually found at about 25 metres. The method is one of great delicacy, and much time has been spent eliminating sources of error. There are indications that water richest in organic matter is also richest in young fish.

The magnitude and rate of increase of the hydrogen-ion concentration in formalin solutions have also been studied, with regard to the preservation of calcareous specimens. Dr. Atkins finds that the best method for preparing non-acid formalin is to add borax to it till the dilute solution gives a good red with phenolphthalein. It is then slightly alkaline, and owing to the buffer action of the borax it will remain with but little change for a long period even in full sunlight.

Published Memoirs.

The following papers, the outcome of work done at the Laboratory, have been published elsewhere than in the Journal of the Association :—

Allen, E. J. *Regeneration and Reproduction of the Syllid Procerastea.* Phil. Trans. Roy. Soc., B. Vol. CCXI, pp. 131–177.

Atkins, W. R. G. *The Differentiation of Boiled and Unboiled Water.* "Nature," Vol. CVIII, 1921, p. 339.

Dunkerly, J. S. *Nuclear Division in the Dinoflagellate, Oxyrrhis marina, Duj.* Proc. Roy. Phys. Soc., Edin., Vol. XX, 1921, pp. 217–220.

Dunkerly, J. S. *Rhabdamoeba marina gen. n. et sp. n.* Proc. Roy. Phys. Soc., Edin., Vol. XX, 1921, pp. 220–221.

Dunkerly, J. S. *Fish Myxosporidia from Plymouth.* Parasitology, Vol. XII, 1921, pp. 328–353.

Fox, H. M. *An Investigation into the Cause of the Spontaneous Aggregation of Flagellates and into the Reactions of Flagellates to Dissolved Oxygen,* Parts I and II. Journ. Gen. Physiol., Vol. III, 1921, pp. 483–511.

Fox, H. M. *Methods of Studying the Respiratory Exchange in small Aquatic Organisms, with Particular Reference to the Use of Flagellates as an Indicator of Oxygen Consumption.* Journ. Gen. Physiol., Vol. III, 1921, pp. 565–573.

Goodrich, E. S., and Pixell Goodrich, H. L. M. *Gonospora minchinii n. sp., a Gregarine inhabiting the Egg of Arenicola.* Quart. Journ. Micr. Sci., Vol. LXV, 1920, pp. 157–162.

HUXLEY, J. S. *Further Studies on Restitution-bodies and free Tissue-culture in Sycon.* Quart. Journ. Micr. Sci., Vol. LXV, 1921, pp. 293–322.

LEIGH-SHARPE, W. H. *The Comparative Morphology of the Secondary Sexual Characters of Elasmobranch Fishes.* Journ. Morph., Vol. XXXIV, 1920, pp. 245–265.

ORTON, J. H. *The Production of living Clavellina Zooids in Winter by Experiment.* "NATURE," Vol. CVII, 1921, p. 75.

ORTON, J. H. *Sex-change in the Native Oyster (O. edulis).* "NATURE," Vol. CVII, 1921, p. 586.

ORTON, J. H. *Is Bisexuality in Animals a Function of Motion?* "NATURE," Vol. CVIII, 1921, p. 145.

ORTON, J. H. *Sex-manifestations and Motion in Molluscs.* "NATURE," Vol. CVIII, 1921, pp. 303–304.

PIXELL GOODRICH, H. L. M. *The Spore of Thelohania.* Arch. Zool. Exp. et Gen., Vol. LIX, Notes and Revue, No. 1, 1920, pp. 17–19.

The Library.

A commencement has been made in getting together the more important journals and books dealing with General Physiology, and several useful series, including the *Journal of Physiology* from 1903, *Biochemical Journal* from 1906, *American Journal of Physiology* from 1909, *Journal of Biological Chemistry* from 1905, *Pflüger's Archiv* from 1910, etc., have been added to our shelves.

The thanks of the Association are again due to numerous Government Departments, Universities, and other Institutions at home and abroad for copies of books and current numbers of periodicals presented to the Library. Thanks are due also to those authors who have sent reprints of their papers to the Library.

Donations and Receipts.

The receipts for the year include a grant from H.M. Treasury of £1000, a grant from the Development Fund, through the Ministry of Agriculture and Fisheries, of £8500 and a Capital Grant of £5831, one from the Fishmongers' Company, £600, and one from the Royal Society, £60. In addition to these grants there have been received Donations (£11), Special Donations to the Steamer, Building and Electrical Funds (£350), Annual Subscriptions (£119), Composition Fees (£63), Rent of Tables in the Laboratory, including £25 from the University of London and £20 from the Trustees of the Ray Lankester Fund (£120), Sale of Specimens (£1000), and Admission to Tank Room (£254).

Vice-Presidents, Officers, and Council.

The following is the list of gentlemen proposed by the Council for election for the year 1922–23 :—

President.

Sir E. Ray Lankester, K.C.B., LL.D., F.R.S.

Vice-Presidents.

The Duke of Bedford, K.G.

The Earl of Stradbroke, C.V.O., C.B.

Viscount Astor.

Lord Montagu of Beaulieu.

The Right Hon. Sir A. J. Balfour, K.G., M.P., F.R.S.

The Right Hon. Sir Arthur Griffiths-Boscawen, M.P.

The Right Hon. Austen Chamberlain, M.P.

G. A. Boulenger, Esq., F.R.S.

Sir Arthur Steel-Maitland, Bart., M.P.

Prof. W. C. McIntosh, F.R.S.

COUNCIL.

Elected Members.

L. A. Borradaile, Esq.

W. T. Calman, Esq., D.Sc., F.R.S.

H. H. Dale, Esq., C.B.E., M.D., F.R.S.

G. P. Farran, Esq.

Prof. F. W. Gamble, D.Sc., F.R.S.

Prof. J. Stanley Gardiner, F.R.S.

J. Gray, Esq., M.A.

Sir Sidney F. Harmer, K.B.E., Sc.D., F.R.S.

Julian S. Huxley, Esq.

Prof. F. W. Keeble, Sc.D., F.R.S.

H. G. Maurice, Esq., C.B.

T. H. Riches, Esq.

J. A. Robertson, Esq.

Prof. D'Arcy W. Thompson, C.B., F.R.S.

Chairman of Council.

Sir Arthur E. Shipley, G.B.E., D.Sc., F.R.S.

Hon. Treasurer.

George Evans, Esq.

Hon. Secretary.

E. J. Allen, Esq., D.Sc., F.R.S.,
The Laboratory, Citadel Hill, Plymouth.

The following Governors are also members of Council :—

G. P. Bidder, Esq., Sc.D.

E. T. Browne, Esq.

Lothian D. Nicholson, Esq. (Prime Warden of the Fishmongers' Company).

W. T. Brand, Esq. (Fishmongers' Company).

George Evans, Esq. (Fishmongers' Company).

E. H. Chapman, Esq. (Fishmongers' Company).

Lothian D. Nicholson, Esq. (Fishmongers' Company).

Major Nigel O. Walker, O.B.E. (Fishmongers' Company).

Prof. G. C. Bourne, D.Sc., F.R.S. (Oxford University).

Sir Arthur E. Shipley, G.B.E., D.Sc., F.R.S. (Cambridge University).

Sir W. A. Herdman, C.B.E., D.Sc., F.R.S. (British Association).

Dr. *Statement of Receipts and Payments for*

GENERAL

	£ s. d.	£ s. d.
To Balance from Last Year :—		
Cash at Bankers	502 12 1	
Cash in hand	75 5 11	577 18 0
,, Current Receipts :—		
H.M. Treasury	1,000 0 0	
,, Ministry of Agriculture and Fisheries :—		
Grant from Development Fund	8,500 0 0	
The Worshipful Company of Fishmongers	600 0 0	
,, Royal Society :—		
Grant from Gore Fund	60 0 0	
Annual Subscriptions	119 3 5	
Rent of Tables (Ray Lankester Trustees, £20; University of London, £25; various, £75 5s.)	120 5 0	
Interest on Investments	14 12 8	10,414 1 1
,, Extraordinary Receipts :—		
Donations	11 15 6	
Composition Fees	63 0 0	74 15 6
,, Laboratory, Boats and Sundry Receipts :—		
Sale of Apparatus	539 13 3	
,, ,, Specimens	999 17 10	
,, ,, Nets and Gear	308 8 9	
,, ,, Fish 60 3 6		
Less Expenses 6 10 0	53 13 6	
Sundries	5 0 0	1,906 13 4

The Association's Bankers hold on its behalf:—
£410 14s. 8d. New Zealand 4% Stock, 1943–63.
£500 0s. 0d. War Savings Certificates.
£78 9s. 4d. 4% War Loan, 1929–42 Registered Stock, 4%.

£12,973 7 11

SPECIAL

	£ s. d.	£ s. d.
To Balance from Last Year :—		
On Deposit at Bank	626 6 0	
,, Current Account	50 0 0	676 6 0
,, Ministry of Agriculture and Fisheries :—		
Grant from Development Fund		5,831 0 0
,, Donations		350 19 0
,, Interest on Deposit		7 14 4
,, Balance :—		
Amount due to General Fund		692 5 3
		£7,558 4 7

the Year ending 31*st December*, 1921. **Cr.**

FUND.

	£ s. d.	£ s. d.
By Salaries and Wages—		
Director	800 0 0	
Naturalists	1,837 10 0	
Assistant Naturalists	222 9 4	
Physiologist	649 0 8	
Hydrographer	325 0 0	
Salaries and Wages, Laboratory	1,433 6 1	
,, ,, Boats	1,172 4 11	6,439 11 0
,, Travelling Expenses		106 13 5
,, Library	285 8 2	
Less Sales	2 2 5	283 5 9
,, Journal	294 10 6	
Less Sales	28 9 6	266 1 0
,, Buildings and Public Tank Room :—		
Gas, Water, and Coal	334 17 0	
Stocking Tanks and Feeding	17 14 8	
Maintenance and Renewals	512 3 1	
Rent, Rates, Taxes, and Insurance 123 19 4		
Less Income Tax Refunded 13 19 3	110 0 1	
	974 14 10	
Less Admission to Tank Room	254 7 11	720 6 11
,, Laboratory, Boats, and Sundry Expenses :—		
Glass, Apparatus, and Chemicals	605 6 0	
,, ,, ,, Physiological	336 0 7	
Purchase of Specimens	63 11 10	
Maintenance and Renewal of Boats, Nets, etc.	935 6 1	
Coal and Water for Steamer	490 1 6	
Insurance, Boats	333 5 6	
Boat Hire and Collecting Expenses	31 10 11	
Stationery, Office Expenses, Carriage, Printing, etc.	396 8 9	3,191 11 2
,, Interest on Loan		8 19 5
,, Pension		14 10 0
,, Repayment of Loan to Coutts		500 0 0
,, Balance :—		
Cash at Bankers	738 3 7	
Cash in hand	12 0 5	
Due from Special Fund	692 5 3	1,442 9 3
		£12,973 7 11

FUND.

	£ s. d.	£ s. d.
By Purchase of Steamer	3,500 0 0	
Add Cost of Reconditioning (net)	1,716 18 11	5,216 18 11
,, Expenditure on Building Extension		1,990 7 1
,, Purchase of Books for Physiological Library		350 18 7
		£7,558 4 7

Examined and found correct,

(Signed) N. E. WATERHOUSE.
W. T. BRAND.
C. TATE REGAN.

3 *Frederick's Place,*
Old Jewry, London, E.C. 2.
26*th January*, 1922.

List of Annual Subscriptions

Paid during the Year 1921.

	£	s.	d.
W. M. Aders, Esq.	1	1	0
E. J. Allen, Esq., D.SC., F.R.S.	1	1	0
G. L. Alward, Esq.	1	1	0
J. H. Ashworth, Esq., D.SC., F.R.S.	1	1	0
J. R. Baker, Esq.	1	1	0
Prof. W. Bateson, F.R.S.	1	1	0
Prof. W. M. Bayliss, D.SC., F.R.S.	1	1	0
W. J. Bazely, Esq.	1	1	0
Lieut.-Col. T. T. Behrens	1	1	0
Col. H. F. Bidder	1	1	0
Mrs. M. G. Bidder	1	1	0
Birkbeck College	1	1	0
H. M. Blundell, Esq.	1	1	0
L. A. Borrodaile, Esq.	1	1	0
Prof. G. C. Bourne, F.R.S. (1921 and 1922)	2	2	0
Col. Henry Bowles	1	1	0
Sir John Rose Bradford, K.C.M.G., F.R.S.	1	1	0
Brighton Public Library (1921 and 1922)	2	2	0
H. H. Brindley, Esq.	1	1	0
Mrs. E. T. Browne	1	1	0
R. H. Burne, Esq.	1	1	0
L. W. Byrne, Esq.	1	1	0
W. T. Calman, Esq., D.SC., F.R.S.	1	1	0
H. Graham Cannon, Esq.	1	1	0
Prof. Chas. Chilton	1	1	0
J. Clark, Esq., D.SC.	1	1	0
L. R. Crawshay, Esq.	1	1	0
G. S. R. Kitson Clark, Esq.	1	1	0
J. Omer Cooper, Esq.	1	1	0
J. F. Coonan, Esq.	1	1	0
Commander G. L. C. Damant, R.N.	1	1	0
Prof. O. V. Darbishire	1	1	0
Monsieur J. Delphy		10	5
W. De Morgan, Esq.	1	1	0
Prof. A. Dendy, F.R.S.	1	1	0
G. Despott, Esq.	1	1	0
F. A. Dixey, Esq.	1	1	0
C. Clifford Dobell, Esq., F.R.S.	1	1	0
F. Martin Duncan, Esq.	1	1	0
J. S. Dunkerly, Esq.	1	1	0
Howard Dunn, Esq.	1	1	0
Carried forward	44	12	5

	£	s.	d.
Brought forward	44	12	5
Major E. V. Elwes	1	1	0
George Evans, Esq.	1	1	0
G. Herbert Fowler, Esq., PH.D.	1	1	0
H. M. Fox, Esq. (1920–1922)	3	3	0
Prof. F. W. Gamble, F.R.S.	1	1	0
Prof. J. Stanley Gardiner, F.R.S.	2	2	0
Prof. E. S. Goodrich, F.R.S.	1	1	0
J. Gray, Esq., M.A.	1	1	0
Sir Eustace Gurney	1	1	0
Wilfred Hall, Esq. (1920 and 1921)	2	2	0
Prof. W. D. Halliburton, F.R.S.	1	1	0
H. Bertram Harding, Esq.	1	1	0
Prof. S. G. Hickson, D.SC., F.R.S.	1	1	0
Prof. J. P. Hill, F.R.S.	1	1	0
W. T. Hillier, Esq., M.R.C.S.	1	1	0
T. V. Hodgson, Esq.	1	1	0
W. E. Hoyle, Esq., D.SC.	1	1	0
P. Hoyte, Esq.	1	1	0
J. S. Huxley, Esq.	1	1	0
R. Kirkpatrick, Esq.	1	1	0
J. J. Lister, Esq., F.R.S.	1	1	0
Prof. E. W. MacBride, F.R.S.	1	1	0
W. N. McClean, Esq.	1	1	0
Stanilaus Makovski, Esq.	1	1	0
D. J. Matthews, Esq.	1	1	0
E. C. Matthews, Esq.	1	1	0
J. H. Midgley, Esq.	1	1	0
W. S. Millard, Esq.	1	1	0
P. Chalmers Mitchell, Esq., C.B.E., D.SC., F.R.S.	1	1	0
Rev. Canon A. Morford	1	1	0
H. G. Newth, Esq.	1	1	0
Chas. Oldham, Esq.	1	1	0
Enrique Pascual, Esq., O.B.E. (1920 and 1921)	2	2	0
Plymouth Corporation (Museum Committee)	1	1	0
Plymouth Education Authority	1	1	0
Port of Plymouth Incorporated Chamber of Commerce	1	1	0
W. P. Pycraft, Esq.	1	1	0
W. H. St. Quintin, Esq.	1	1	0
Major G. Raymond	1	1	0
C. Tate Regan, Esq., F.R.S.	3	3	0
J. A. Robertson, Esq.	1	1	0
J. T. Saunders, Esq., M.A.	1	1	0
R. E. Savage, Esq.	1	1	0
R. F. Scharff, Esq., PH.D.	1	1	0
F. W. Schiller, Esq.	1	1	0
Edgar Schuster, Esq., D.SC.	1	1	0
W. L. Sclater, Esq.	1	1	0
L. E. Sexton, Esq. (1921 and 1922)	2	2	0
Carried forward	103	8	5

	£	s.	d.
Brought forward . .	103	8	5
Miss Lilian Sheldon	1	1	0
Sir Arthur E. Shipley, G.B.E., D.SC., F.R.S. . . .	3	3	0
Lieut.-Commander R. Spry	1	1	0
W. E. Stoneman, Esq.	1	1	0
Sir H. F. Thompson, Bart.	1	1	0
Sir John T. Thornycroft, F.R.S.	1	1	0
Torquay Natural History Society	1	1	0
Arthur W. Waters, Esq.	1	1	0
A. T. Watson, Esq.	1	1	0
Mrs. Weldon	1	1	0
W. A. Willes, Esq.	1	1	0
R. Winckworth, Esq.	1	1	0
R. H. Worth, Esq.	1	1	0
Total . . .	£119	3	5

Special Donations for Electrical Installation and Building Fund.

1921	£	s.	d.
Colonel H. F. Bidder	1	1	0
British Association	200	0	0
F. W. Harmer, Esq.	100	0	0
T. B. Harmer, Esq.	25	0	0
The Royal Society	10	0	0
Sir G. Sims Woodhead, K.B.E., F.R.S.	10	10	0
Total . . .	346	11	0

OBJECTS

OF THE

Marine Biological Association

OF THE UNITED KINGDOM.

THE ASSOCIATION was founded at a Meeting called for the purpose in March, 1884, and held in the Rooms of the Royal Society of London.

The late Professor HUXLEY, at that time President of the Royal Society, took the chair, and amongst the speakers in support of the project were the late Duke of ARGYLL, the late Sir LYON PLAYFAIR, the late Lord AVEBURY, the late Sir JOSEPH HOOKER, the late Dr. CARPENTER, the late Dr. GÜNTHER, the late Lord DALHOUSIE, the late Professor MOSELEY, the late Mr. ROMANES, and Sir E. RAY LANKESTER.

The Association owes its existence and its present satisfactory condition to a combination of scientific naturalists, and of gentlemen who, from philanthropic or practical reasons, are specially interested in the great sea fisheries of the United Kingdom. It is universally admitted that our knowledge of the habits and conditions of life of sea fishes is very small, and insufficient to enable either the practical fisherman or the Legislature to take measures calculated to ensure to the country the greatest return from the "harvest of the sea." Naturalists are, on the other hand, anxious to push further our knowledge of marine life and its conditions. Hence the Association has erected at Plymouth a thoroughly efficient Laboratory, where naturalists may study the history of marine animals and plants in general, and where researches on food-fishes and molluscs may be carried out with the best appliances.

The Laboratory and its fittings were completed in June, 1888, at a cost of some £12,000. Since that time investigations, practical and scientific, have been constantly pursued at Plymouth. Practical investigations upon matters connected with sea-fishing are carried on under the direction of the Council; in addition, naturalists from England and from abroad have come to the Laboratory, to carry on their own independent researches, and have made valuable additions to zoological and botanical science, at the expense of a small rent for the use of a working table in the Laboratory and other appliances. The number of naturalists who can be employed by the Association in special investigations on fishery questions, and definitely retained for the purpose of carrying on those researches throughout the year, must depend on the funds subscribed by private individuals and public bodies for the purpose. The first charges on the revenue of the Association are the working of the sea-water circulation in the tanks, stocking the tanks with fish and feeding the latter, the payment of servants and fishermen, the hire and maintenance of fishing-boats, and the salary of the Resident Director and Staff. At the commencement of this number will be found the names of the gentlemen on the Staff.

The purpose of the Association is to aid at the same time both science and industry. It is national in character and constitution, and its affairs are conducted by a representative Council, by an Honorary Secretary and an Honorary Treasurer, without any charge upon its funds, so that the whole of the subscriptions and donations received are devoted absolutely to the support of the Laboratory and the prosecution of researches by aid of its appliances. The reader is referred to page 4 of the Cover for information as to membership of the Association.

CONTENTS OF NEW SERIES, Vol. XII., No. 4.

NOTICE.

The Council of the Marine Biological Association wish it to be understood that they do not accept responsibility for statements published in this Journal excepting when those statements are contained in an official report of the Council.

TERMS OF MEMBERSHIP.

		£	s.	d.
Annual Members . . .	per annum	1	1	0
Life Members . . .	Composition Fee	15	15	0
Founders		100	0	0
Governors		500	0	0

Members of the Association have the following rights and privileges: they elect annually the Officers and Council; they receive the Journal of the Association free by post; they are admitted to view the Laboratory at Plymouth, and may introduce friends with them; they have the first claim to rent a place in the Laboratory for research, with use of tanks, boats, &c.; and have access to the books in the Library at Plymouth.

All correspondence should be addressed to the Director, The Laboratory Plymouth.