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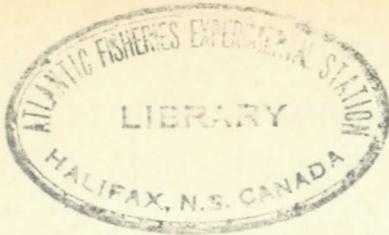
Title

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AN INVESTIGATION OF THE AUTUMN MAXIMUM

OF DIATOM GROWTH AT ST. ANDREWS, N. B.

During the summer of 1924 an attempt was made to grow in special cultures such genera of diatoms as were found in sea-water taken at any convenient time of day from just beyond the breakwater at the Atlantic Biological Station at St. Andrews, N.B. The purpose of the investigation was to find an explanation for the remarkable maximum which diatoms reach in their growth during the autumn in that locality. The fact that the common white jelly fish, Aurelia aurita (Fabricius) is also abundant, reaching a maximum during the late summer, and then rapidly disintegrating and practically disappearing, offered a possible source of increased food supply for diatom growth.

It was observed during the Canadian Expedition to the Strait of Belle Isle in 1923, by Prof. A. G. Huntsman who suggested the problem that the cold waters of the Labrador current were quite barren in fish and food for fish but contained great quantities of jelly fish which died and decomposed when they reached the warmer waters near Newfoundland. The warm water too, seemed to lack fish food but the mixed water resulting from the two was exceedingly rich in diatoms.

Cultures, therefore, enriched with varied amounts of disintegrating jelly fish were set up in the laboratory where the effect of different physiological conditions could be carefully observed and noted at regular intervals.

To identify the various genera of diatoms which had been taken in previous years with No. 5 plankton net at Station No. 6, in the St. Croix River near the Biological Station and preserved, those that were found from day to day in the sea-water, and those that began to appear in the cultures, was an exacting and difficult task. A careful study of descriptions, drawings and photographs to be found in standard works on the subject was made and with the aid of a valuable collection of named slides, made and donated for use at the Biological Station by the late Prof. L. W. Bailey, the writer was able to identify most of the more common genera and species. Special reference was made to the various publications of Prof. Bailey on the diatoms of New Brunswick and Eastern Canada mentioned in the bibliography and to the two papers numbers IV and V published by Miss Clara B. Fritz in "Contributions to Canadian Biology 1918-20."

In the first place, an attempt was made to ascertain just when the fall diatoms reached a period of maximum growth. Collections of plankton had been made in a systematic way at Station No. 6 since 1918 and a careful examination of the preserved

material and records for the contents of No. 5 net was made. No definite conclusion as to the precise dates of maxima could be reached as experience shows that No. 5 net does not satisfactorily obtain diatom tows, a much finer one being required. If small diatoms were abundant in the water they might pass easily through the net and the measurement of the preserved material be misleading. If long spindle-shaped forms were abundant, as was often the case during October, the first specimens caught might clog the meshes of the net and the resulting haul be very large. Such seemed to be the case in fall of 1923 when an exceedingly large amount of diatom material was obtained in all the tows reaching a maximum during the first week of November. The long thread-like form which composed most of each haul is referred to by Prof. Bailey in "An Annotated Catalogue of the Diatoms of Canada," 1924 as Thalassiothrix Frauenfeldii, Grun, or as disputed by McKay to be a Thalassionema. The following table shows the dates of the maxima as nearly as they could be found by a rough estimate of the volume in each tow taken with No. 5 plankton net by measuring the height in centimetres of the material in cylindrical jars of the same size.

TABLE I

<u>Year</u>	<u>Date</u>	<u>Genera</u>
1923	Nov. 7	<u>Thalassiothrix</u> , in great abundance.
1922	Sept. 8	<u>Melosira</u> , <u>Chaetoceros</u> , <u>Navicula</u> , <u>Coscinodiscus</u> .
1921	Oct. 27	<u>Rhizosolenia</u> , <u>Coscinodiscus</u> , <u>Chaetoceros</u> .
1920	Oct. 21-29	<u>Chaetoceros</u> , <u>Biddulphia</u> , <u>Pleurosigma</u> , <u>Skeletonema</u> , <u>Rhizosolenia</u> .
1919	Sept. 11	<u>Chaetoceros</u> , <u>Coscinodiscus</u> , <u>Thalassiothrix</u> .
1918	Sept. 5	Very scanty.

Table I--Showing the approximate dates of the autumn maximum of diatoms as revealed by the contents of No. 5 net.

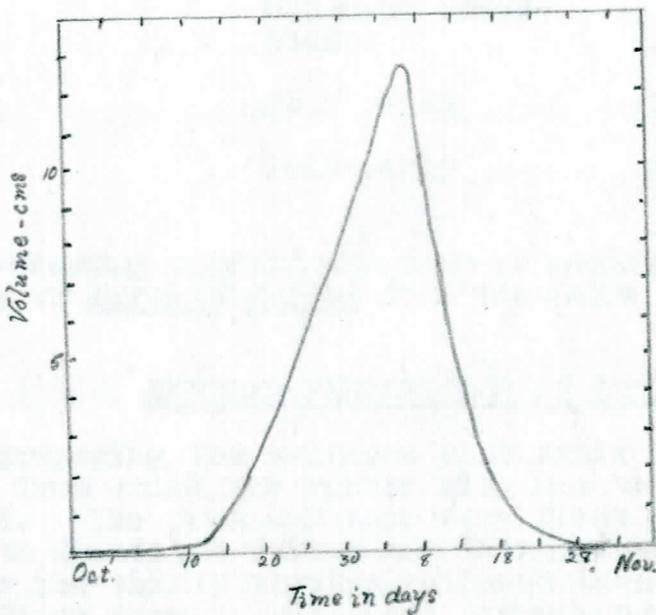


FIGURE I

Graph showing the sudden increase in diatom growth during the autumn of 1923 as revealed by contents of No. 5 net.

Examination of the tows taken with No. 0 net, at Station 6 during the summer months was then made as well as the records of those tows to determine when the jelly fish were most abundant and when they seemed to disappear. The following table gives the data as determined.

TABLE II

<u>Year</u>	<u>Maximum</u>	<u>Disappearance</u>
1923	July 20	October 1
1922	July 28-Aug.4	Sept. 15
1921	July-Aug?	Sept. 30
1920	Few jelly fish but many cteno- phores	Sept. 30
1919	July 15-30	Sept. 1
1918	July-early	Sept. 1

Table II--Showing approximate time of maximum number and disappearance of Aurelia aurita from tows with No.0 net.

GENERAL PREPARATION OF CULTURES

In preparing the cultures pint fruit jars of the perfect seal type from which the rubber ring has been removed were used throughout. The jars and tops were first carefully washed, rinsed with distilled water, and then placed in the Arnold Steam Sterilizer for thirty minutes, allowed to cool, and covered with the sterilized tops. Into each jar was poured 250 cc. of fresh sea-water taken from beyond the breakwater, the water being measured in a graduated flask. Sufficient water was taken at each time to complete a series of jars. Freshly caught jelly fish were dried on filter paper and the required quantities cut from the specimen radially and weighed in a clean beaker containing a small quantity of the 250 cc. of sea-water and then poured into the jar and the top replaced. The same jelly fish was used for the five jars in each series. Unless otherwise stated no attempt was made to reduce the jelly fish to very small pieces except what was done in cutting the required amounts. The jars were numbered 1 to 5, the fifth being the control containing only sea-water.

To provide conditions as nearly natural as possible, a series of shallow pans through which sea-water was kept running

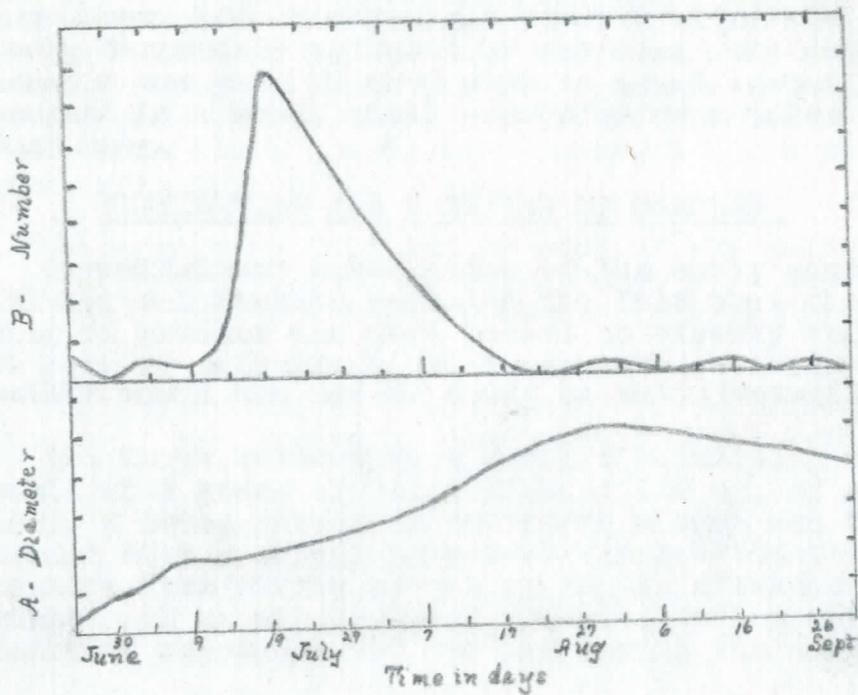


FIGURE II

Graph showing the variation in size and number of *Aurelia aurita* during the summer of 1923 as revealed by the contents and records of No. 0 net.

were placed on the roof. In each was placed a hood under which one or more series of jars could be placed in a shaded position, while other series in the same pans were exposed to full sunlight. Other cultures were placed in the north windows of the Laboratory.

Daily temperature records for the cooling pan and the Laboratory were kept. Detailed reports were made upon changes which could be observed in each jar from day to day when held up against the light. When growth as indicated by a brownish precipitate began to appear samples were taken and preserved for microscopic examination. Each sample consisted of $\frac{1}{2}$ cc. of the culture drawn off quickly in a pipette after the whole culture had been thoroughly agitated by rotating the jar in the hand. The sample was then diluted with an equal amount of 10% formalin and corked in a small vial. Samples were taken at intervals of four days.

EXAMINATION AND COUNTING OF SAMPLES

A preliminary examination of the early samples carried on at the Biological Station revealed the fact that the diatoms were growing in patches and were packed so closely together, probably about bits of jelly fish, that accurate counting would be impossible until the masses could be satisfactorily broken up.

Two large battery jars F and G containing cultures made on the basis of 2 grams of jelly fish of 250 cc. of sea-water were prepared, F being placed in the north window and G in a west window, but cooled with a stream of water from the tap to prevent the temperature from rising very high in the afternoon. Both cultures flourished and an abundance of material for experimental purposes was obtained and preserved for use during the winter.

In the first place chemical means of breaking up the growth colonies was tried. Dilute, strong, cold and hot acids were applied without satisfactory results, the difficulty being due to the silicious covering of the diatoms which were either unaffected or seemed suddenly to go into solution when boiled in the hot acid. Finally, clean quartz sand grains of fairly even size were employed as a physical means. After five to eight minutes vigorous shaking of a small quantity of sand in a 1 cc. sample, the colonies appeared to be satisfactorily broken up without injuring the diatoms to any appreciable extent. This conclusion was reached from observing the comparatively few fragments of diatoms to be found in the treated samples and from the fact that such a delicate form as Nitzschia closterium S.B.D. was found in such abundance intact.

The next essential was a suitable counting cell. First, a Levy blood counting cell was used but the quantity it contained was found to be too small. The Rafter cell of 1 cc. capacity as used by Miss Fritz presented the other extreme. An adaptation of the Rafter from a preserved sample diluted to 3 cc. The new cell had a rectangular rim 1.7 cm. x 1 cm. x 1 mm., and from careful measurement and weighing of the water it contained when covered with a slip, was found to be of .175 cc. capacity. Since each

sample diluted to 3 cc. contained 1/6 cc. of the original culture, the counting cell contained .029 cc. of the original culture. To facilitate covering the whole field of the cell, .5 mm. rulings were lightly etched on the microscope slide to which the frame was attached. More than enough to fill the cell was drawn off quickly from the sample to be counted after it had been thoroughly shaken, and the cell filled at once and the cover slip pressed down firmly.

In counting, a Spencer objective 8 mm. N. Ap. 0.50 and 10X eye piece were used which did not reveal the minute details of structure in the small forms which predominated in the cultures. In most cases, the number of diatoms in the whole field was counted. After considerable experience it was found that one half of the cell could be used with reasonable accuracy especially where the numbers were abundant since the diatoms seemed to be fairly evenly distributed over the field. In the tables these counts were recorded X x 2 where X represents the number in half of the field.

Although more detailed records of the identification as well as drawings of some doubtful forms were made, all have been grouped under three headings, namely, Nitzschia, Navicula and Other Genera.

The most abundant diatom found in the cultures was Nitzschia closterium. The Naviculae were mostly small species but very persistent. Other Genera included small chains of Fragillaria, Chaetoceros and Melosira, Surirella and Synedra Chaetoceros which was very abundant in all the tows taken at St. Andrews during the summer disappeared very quickly from the cultures which were often overgrown by N. closterium or small Naviculae. These last named Miss Fritz also found in her cultures and Prof. Bailey described as "littoral rather than pelagic or planktonic"-- (Cont. to Can. Bio. 1915-16, P. 105).

The number of diatoms recorded for each culture can at best, be only an approximation and the most general conclusions be reached from their consideration. In the first place, since some cultures developed a growth which collected in patches or on the bottom and sides of the jar more than others, the samples may not have been representative. Then, in the samples where the numbers counted were very high the period of shaking may not have reduced all the patches entirely. Only one count was made for each sample.

Since all the jars contained 250 cc. of culture medium the aggregate number of diatoms at the time the sample was taken may easily be calculated from the number counted in the cell.

CULTURES

Cultures A and B

Date:--July 10th

Notation and quantity of jelly fish: A1, 10 grams; A2, 5 grams; A3, 2.5 grams; A4, 1.25 grams; A5, control, containing only sea water. Series B was the same as Series A.

Position: The ten jars were placed in the first pan of running water on the roof, in two rows under the hood, and the position of each series changed on alternate days from the front to the back of the hood.

Light intensity: Front of hood 2.5%, side 2%, and back .05% of full sunlight.

Cultures C and D

Date:--July 10th

Notation and quantity of jelly fish: Both were made on same basis as A and B.

Position: Both were placed in same pan with A and B but in full sunlight.

For all measurements of the light intensity recorded in this paper, the writer is indebted to Prof. A.B.Klugh of Queen's University.

In all the jars except the controls, a milkiness appeared during the first two or three days but soon disappeared. The little pieces of jelly fish which had been floating in the water also disappeared except in B3 in which a small clear piece remained visible as late as August 14th. The milkiness cleared more quickly in Series C and D than in A and B. Growth as indicated by a brownish precipitate began to appear earlier in Series C and D than in A and B and was first apparent in jars No. 3 and No. 4 which had the lower concentrations of jelly fish. Later, however, jars No. 1 and No. 2 developed an even more vigorous growth which surpassed in amount the dwindling growth of jars No. 3 and No. 4. Series A and B apparently made very similar development but at no time developed as much colour as C and D. Series D developed a slightly deeper colour and greater volume of growth than series C. By Aug. 23 an alga, Enteromorpha compressa, as identified by Prof. Klugh was found to be growing in jars D1 and D2. The daily range of temperature variations as recorded for the pan of running water was from 14°-16°C.

The following tables show the number of diatoms counted in Series B and D in samples obtained on the dates recorded. It was found impossible to count all the samples and the same dates were chosen in each series for the sake of comparison.

TABLE I-- CULTURE B

Jar	Date	<u>Nitzschia</u>	<u>Navicula</u>	Other Genera	Total
B1	Jul. 29	1	43	1	45
B2	Jul. 29	0	38	15	53
B3	Jul. 29	42	85	10	137
B4	Jul. 29	9	26	8	43
B5	Jul. 29	8	41	5	54
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B1	Aug. 2	1	9	6	16x2-32
B2	Aug. 2	0	13	2	15x2-30
B3	Aug. 2	23	15	3	41x2-82
B4	Aug. 2	1	4	1	6x2-12
B5	Aug. 2	0	2	2	4x2-8
<hr/>					
B1	Aug. 6	0	21	15	36
B2	Aug. 6	1	67	3	71
B3	Aug. 6	17	54	9	80
B4	Aug. 6	4	6	10	20
B5	Aug. 6	11	33	15	59
<hr/>					
B1	Aug. 13	0	66	4	70
B2	Aug. 13	0	243	16	259
B3	Aug. 13	34	61	12	107
B4	Aug. 13	12	48	7	67
B5	Aug. 13--evaporated				
<hr/>					
B1	Aug. 22	0	19	4	23
B2	Aug. 22	0	23	2	25

TABLE II-- CULTURE D

Jar	Date	<u>Nitzschia</u>	<u>Navicula</u>	Other Genrea	Total
D1	Jul. 29	0	69	18	87
D2	Jul. 29	690	86	16	792
D3	Jul. 29	0	27	2	29
D4	Jul. 29	182	50	11	243
D5	Jul. 29	1	26	4	31
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D1	Aug. 2	0	35	0	35x2-70
D2	Aug. 2	64	57	3	124x2-248
D3	Aug. 2	0	37	16	53
D4	Aug. 2	3	10	0	13x2-26
D5	Aug. 2	0	9	3	12x2-24
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D1	Aug. 6	12	19	1	32x2-64
D2	Aug. 6	899	82	16	997x2-1994
D3	Aug. 6	0	13	9	22x2-44
D4	Aug. 6	3	37	9	49
D5	Aug. 6	0	13	4	17x2-34
<hr/>					
D1	Aug. 13	210	61	45	316x2-632
D2	Aug. 13	907	66	21	997x2-1988
D3	Aug. 13	0	11	7	18x2-36
D4	Aug. 13	3	12	0	15x2-30
D5	Aug. 13	0	10	35 (chain)	45
<hr/>					
D1	Aug. 22	0	29	3	32x2-64
D2	Aug. 22	223	24	58	305x2-610
D3	Aug. 22	1	1	2	4x2-8
D4	Aug. 22	0	10	2	12x2-24
D5	Aug. 22	0	2	0	4

From a study of the tables one would conclude that the great amount of growth obtained in the higher concentrations of jelly fish in Series D was due to N. closterium which found the light intensity more favorable than in Series B. Development in jars No. 1 and No. 2 of Series B was slower than in the same jars of Series D. It is evident samples were not taken early enough in the life of either series to get satisfactory counts for the maxima in jars B4 and D3 and D4.

Culture E

Date:--July 16th

Notation and quantity of jelly fish: E1, 10 grams; E2, 5 grams; E3, 2.5 grams; E4, 1.25 grams; E5, control.

Position:--This series was placed with the jars in a row on a support in the north window of the laboratory. The light intensity was measured as 5% of full sunlight. The temperature variations were recorded as from 16°C to 22°C.

As was observed in A,B,C,D on the roof, milkiness developed in series E in the first two or three days in all the jars but the control. Growth first appeared on the fifth day in E4 and by the eighth day was apparent in all but the control. On July 30th an alga Enteromorpha Compressa was observed to be growing to the side of the jars E1 and E4. By Aug. 22nd the whole series seemed to be dead.

In this series, E2 which corresponds to B2 and D2 in amount of jelly fish reached its first maximum by July 29th which is equivalent to five days in advance of B2 or D2 since it was set up later. The greater growth in E as compared with B was plainly due to presence of N.closterium which evidently found the double intensity of light and the higher temperature more favorable to its growth. The higher concentrations evidently afforded a better food supply and therefore, the cultures developed a greater growth.

TABLE III--CULTURE E

Jar	Date	<u>Nitzschia</u>	<u>Navicula</u>	Other Genera	Total
E2	Jul. 29	156	11	16	182
E3	Jul. 29	116	15	16	147
E4	Jul. 29	91	35	8	134
E5	Jul. 29	1	43	6	50
E2	Aug. 2	9	29	16	54
E4	Aug. 2	4	23	23	50
E2	Aug. 6	85	28	12	125
E4	Aug. 6	10	20	5	35
E1	Aug. 9	353	48	8	409
E2	Aug. 9	92	33	8	133
E3	Aug. 9	12	42	22	76
E4	Aug. 9	314	57	23	394
E5	Aug. 9	37	44	18	99
E1	Aug. 13	36	13	3	52x2-104
E2	Aug. 13	25	23	3	51
E3	Aug. 13	1	7	6	14x2-28
E4	Aug. 13	63	26	11	100
E5	Aug. 13	0	5	1	6x2-12
E2	Aug. 18	0	2	0	2x2-4
E2	Aug. 22	0	2	0	2x2-4
E4	Aug. 22	0	2	1	3x2-6

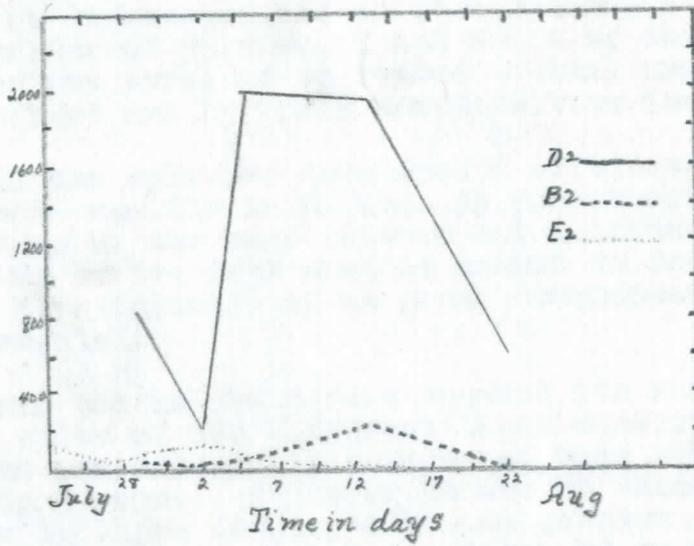


FIGURE III.

GRAPH SHOWING THE RELATIVE NUMBERS OF DIATOMS IN CULTURES B2, D2 and E2 FROM NINETEENTH TO THE THIRTY-SEVENTH DAYS OF THEIR GROWTH.

Culture H.

Date:--July 28th

Notation:--H1,H2,H3,H4,H5

Quantity of jelly fish:--Into each jar was placed 2 grams of finely divided jelly fish to 250 cc. of fresh sea-water strained through clean No. 5 netting.

Position:--H1 was placed in north window; H2 was placed one day in full sunlight in one of the pans on the roof and then removed to the north window; H3 was placed in sunlight for two days, H4 for four days and H5 for eight days and then removed to the north window.

Two grams of jelly fish was chosen as the basis of this series, as it represented an intermediate value between the concentration of jars No. 2 and No. 4 of the five previous series. The water was strained to remove animal organisms which might be contained and by their decomposition increase the food supply.

All the cultures developed a milkiness which appeared to clear more rapidly in H1 than in the others. By Aug. 5th, H4 appeared to be the best culture and continued in good condition for ten or twelve days when it seemed to fade out and die. H5 did not give evidence of as great development as the counting later revealed.

Each jar in the series reached its maximum growth by Aug. 13th in spite of the different light conditions to which the different members of the series had been subjected during the first eight days. Cultures H4 and H5 which had the longest exposure to light developed a much greater growth than the others. H3 developed twice as much as either H2 or H1 but they died off more quickly than H1 which was still retaining its maximum when the last sample was taken. The speeding up of the growth in the cultures exposed to light may have been due to a more rapid decomposition of the jelly fish.

TABLE IV--CULTURE H

Jar	Date	<u>Nitzschia</u>	<u>Navicula</u>	Other Genera	Total
H1	Aug. 6	24	40	32	96
H2	Aug. 6	3	14	9	26
H3	Aug. 6	3	28	10	41
H4	Aug. 6	10	53	30	93
H5	Aug. 6	29	35	10	74
<hr/>					
H1	Aug. 9	18	49	11	78
H2	Aug. 9	52	25	6	83
H3	Aug. 9	7	37	5	49x2-98
H4	Aug. 9	20	17	11	48
H5	Aug. 9	293	30	3	326
<hr/>					
H1	Aug. 13	162	70	17	249
H2	Aug. 13	221	30	17	268
H3	Aug. 13	475	76	9	560
H4	Aug. 13	1192	58	7	1257
H5	Aug. 13	1134	89	1	1224
<hr/>					
H1	Aug. 21	75	31	17	123x2-246
H2	Aug. 21	5	11	6	22x2-44
H3	Aug. 21	53	16	0	69x2-138
H4	Aug. 21	4	5	2	11x2-22
H5	Aug. 21	2	3	0	5x2-10

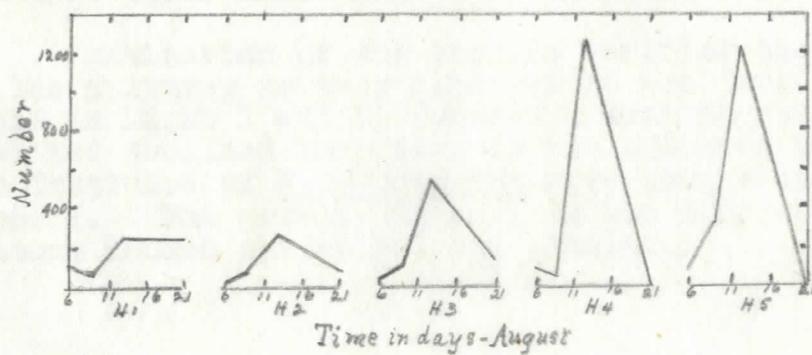


FIGURE IV.

Graph showing the comparative growth in cultures H1 to H5 as revealed by the number of diatoms counted in the samples.

Culture J

Date:--August 1

Notation:--Light 1 and 2; Shade 1 and 2; Dark 1 and 2.

Quantity of jelly fish:-- Two grams of jelly fish left as whole as possible was placed in each culture.

Position:--All the jars were placed in a pan of running sea-water; two in full sunlight, two in shade measured as 5% of full sunlight and two in covered cans.

The culture in jar Light 2 made much slower progress than Light 1, the only reason apparent being that part of the jelly fish long remained clear and floating in the sea-water. Milkiness developed in the jars in both the light and shaded positions but was less dense in the latter. The cultures in the dark exhibited no milkiness or growth and in one jar pieces of jelly fish remained clear until Aug. 24th when the last observation was made.

Examination of the samples verified the observations made on the cultures as they appeared in the jars. Growth was most rapid in Light 1 and 2, reached a much higher maximum and by Aug. 21st had declined more than in the cultures in the shaded position. The frustules of N. closterium were longer and more healthy in Shade 1. The growth, if any, in the dark was unimportant. The diatoms looked shrivelled and stunted.

TABLE V--CULTURE J

Jar	Date	<u>Nitzschia</u>	<u>Navicula</u>	Other Genera	Total
Light					
J1	Aug. 6 (missing)		59	9	248
J1	Aug. 9	180	99	6	156
J1	Aug. 13	51	12	5	17
J1	Aug. 21	0			
<hr/>					
J2	Aug. 6	0	42	10	52
J2	Aug. 9	130	24	7	161
J2	Aug. 13	2	22	3	27
J2	Aug. 21	0	15	0	15
<hr/>					
Shade					
J1	Aug. 6	3	35	6	44
J1	Aug. 9	32	18	12	62
J1	Aug. 13	54	39	30	123
J1	Aug. 21	5	29	1	35
<hr/>					
Dark					
J1	Aug. 6	1	29	5	35
J1	Aug. 13	2	17	14	33
J1	Aug. 21	0	3	3	6

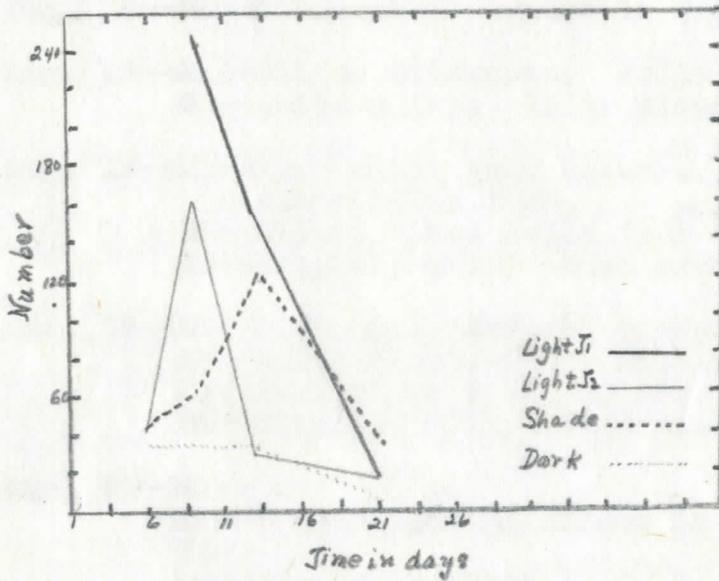


FIGURE V

Graph showing the comparative development of Culture J in full sunlight, shade and darkness.

Culture M

Date:--Aug. 14th

Notations:--M1, M2, M3, M4, M5, M6.

Quantity of jelly fish:--Two grams in 250 cc. sea-water.

Position:--A fresh culture was made each evening beginning with M1 on Aug. 14th and ending with M6 on Aug. 20th and placed on a cooling pan in full sunlight the following morning.

Purpose:-- To investigate and compare the rate of decomposition of Aurelia aurita.

Observations were made each morning about 9.30.

Aug. 16--M1--No apparent change in jelly fish.

Aug. 17--M1--Slight milkiness. Jelly fish quite clear.
M2--Quite milky. Jelly disintegrating rapidly.

Aug. 18--M1--More milky than before. Large piece of clear jelly fish.
M2--Milky. Less jelly fish than M1.
M3--Slightly milky--much unchanged jelly fish.

Aug. 19--M1--Clearing. Several pieces of clear jelly fish.
M2--Quite milky. Very little jelly fish.
M3--More milky than M2. Very little jelly fish.
M4--Slightly milky--Much unchanged jelly fish.

Aug. 20--M1--
M2--Very slight milkiness in all.
M3--
M4--More milky than 1, 2, 3.
M5--Slightly milky.

Aug. 21--M1--cleared. First traces of growth apparent.
M2--
M3--Cleared.
M4--Milky.
M5--Slightly milky.
M6--Only slightly milky. Much unchanged jelly fish.

Aug. 22--By this date growth seemed well begun in M1, M2, M3, M4. The weather during Aug. 20, 21, was cloudy and rainy and may account for M5 and M6 appearing less milky than previous cultures did in the same length of time.

Aug. 23--Growth quite marked in M1 to M4 in a descending series with M1 as maximum.

No samples of these cultures were taken. Apparently the

decomposition of Aurelia aurite went on rapidly enough in M1 in spite of the delay during the first two or three days to allow the growth to keep ahead of that which took place in the later cultures. Verification could be made only by counting.

Culture N.

Date:--Aug. 18th.

Notation:--N1, N2, N3, N4.

Quantity of jelly fish:--Two grams left as whole as possible in each culture.

Position:--All the jars were placed in a pan on the roof as follows: N1 in full sunlight; N2 covered with one sheet of paraffined writing paper which admitted 28% full sunlight, N3 covered with 4 sheets of paraffined paper admitting 8% full sunlight and N4 covered with 8 sheets admitting only 1.4% full sunlight.

The sheets were covered as evenly as possible on both sides with paraffin and then placed about the jars to meet and not overlap. Cotton batting was placed on the top of each jar to prevent entrance of light from above.

The first four days, Aug. 18th to Aug. 23rd, were cloudy and the milkiness although apparent was only slight. By Aug. 24th N1 had developed distinct signs of growth, N2 had developed distinct signs of growth, N2 had noticeably changed but N3 and N4 were unchanged. Observations recorded for Sept. 4th and Sept. 9th showed growth in all the cultures which tended to cling to the bottom of the jar. By Oct. 10th when the last observation was made the growth was quite definitely clinging to the bottom and was more brownish in N2, N3 and N4 than in N1. There was much precipitate adherent to the side of jar N4 as though some light had entered where the sheets were joined about the jar.

As shown by previous cultures full sunlight increases the rate and volume of growth over that obtained in a shaded position. The larger number of diatoms counted in the cultures on Oct. 10th may be accounted for by the way in which the final samples were taken. The precipitate was well stirred and loosened from the bottom with a small cloth attached to a stick. N4 was latest to develop and as already observed, its appearance indicated entrance of additional light, which would increase the amount of growth and explain the high maximum reached by Sept. 14th. The total count, however, in this culture is less than that obtained from N1 which had full sunlight all the time.

CULTURE N.

Jar	Date	<u>Nitzschia</u>	<u>Navicula</u>	Other Genera	Total
N1	Aug. 25	20	17	32	69
N1	Sept. 4	70	3	12	85
N1	Sept. 14	61	8	8	77
N1	Oct. 10	19	7	0	26
N2	Aug. 25	5	5	8	16
N2	Sept. 4	7	5	0	12
N2	Sept. 14	38	14	6	58
N2	Oct. 10	27	6	0	33
N3	Aug. 25	2	8	6	16
N3	Sept. 4--broken vial				
N3	Sept. 14	0	4	0	4
N3	Oct. 10	4	40	2	46
N4	Aug. 25	2	6	14	22
N4	Sept. 4	5	7	4	16
N4	Sept. 14	97	11	13	121
N4	Oct. 10	71	19	10	100

Note--All the numbers in this table except those for N4, Aug. 25 were obtained from one half the field of the counting cell.

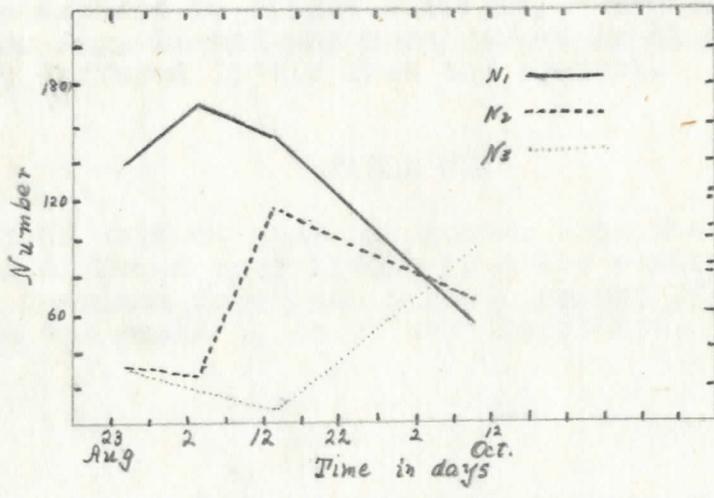


FIGURE VI.

Graphs showing the growth of Culture N in 100%, 28%, and 8% light intensity.

Culture R

Date:--Aug. 20

Notation:--R1, R2, R3, R4, R5.

Quantity of jelly fish:--R1--1.25 g. --1.
R2--.312 g. -- $\frac{1}{4}$.
R3--.09 g. -- $\frac{1}{4}$.
R4--.002 g. --1 64.
R5--control.

Positions--All were placed in the north window of the laboratory.

The purpose of this culture was to determine the rate of growth in lower concentrations of jelly fish than had been previously tried. Daily observations recorded showed that very little milkiness as compared with the higher concentrations was apparent and much less than in the cultures which had been exposed to bright sunlight. Growth was first apparent on Aug. 24 and was more marked in R1 and R2 than R3 and R4 which differed little from the control.

TABLE VII

It is evident from the counts that the growth in all the cultures differed very little from the control or that the amount of food received from such a small amount of disintegrating jelly fish was too small to be of any importance.

Jar	Date	<u>Nitzschia</u>	<u>Navicula</u>	Other Genera	Total
R1	Aug. 25	1	6	4	10x2
R2	Aug. 25	3	11	1	15x2
R3	Aug. 25	0	11	2	13x2
R4	Aug. 25	0	10	4	14
R5	Aug. 25	0	5	2	7

R1	Sept. 4	0	10	0	10x2
R2	Sept. 4	1	1	7	9
R3	Sept. 4	0	7	0	7x2
R4	Sept. 4	0	6	1	7x2
R5	Sept. 4	1	9	2	12x2

R1	Sept. 14	0	13	0	13x2
R2	Sept. 14	0	1	8	9x2
R3	Sept. 14	1	2	2	5x2
R4	Sept. 14	0	8	0	8x2
R5	Sept. 14	0	3	2	5x2

R1	Oct. 10	0	2	0	2x2
R4	Oct. 10	0	3	0	3x2
R5	Oct. 10	0	3	0	3x2

Culture S

Date:--Aug. 26

Notations:--S1, S2, S3, S4, S5.

Quantity of jelly fish:-- Two grams in each culture except S5, the control.

Position:--All the jars were left for five days in a pan on the roof in full sunlight and then removed to the north window of the laboratory. S2 was wrapped with one layer of waxed paper on the exposed side; S3 with one layer of waxed paper around the whole jar; S4 with two layers around the whole jar and S1 and S5 left uncovered.

Light intensity:--S1, 5%, S2, 1.4%, S3, 1.4%, approximately, S4, .65% of full sunlight. A yellowish deposit appeared in all but the control, and by Sept. 9th tended to adhere slightly to the bottom and sides of the jars. The last samples were taken on Oct. 10th in the manner described in Culture N.

TABLE--CULTURE S

The results from counting the samples showed less variation than in the earlier cultures. There was also a much smaller amount of growth than in Cultures H, J, and N, which were made on the basis of the same amount of jelly fish. The above mentioned cultures, however, all had higher light intensity than S2, S3 and S4. The fact that this series was latest of all and subject to several days of cloudy and cooler weather, may explain why the exposure to full sunlight for the first five days did not produce better results.

TABLE VIII CULTURE S

Jar	Date	<u>Nitzschia</u>	<u>Navicula</u>	Other Genera	Total
S1	Sept. 4	0	3	0	3x2
S3	Sept. 4	2	5	1	8x2
S4	Sept. 4	39	0	1	40x2
S5	Sept. 4	0	3	2	5x2
<hr/>					
S1	Sept. 9	1	6	0	7x2
S2	Sept. 9	0	1	0	1x2
S3	Sept. 9	1	8	3	12x2
S4	Sept. 9	4	6	0	10x2
<hr/>					
S1	Sept. 14	7	3	1	11x2
S2	Sept. 14	1	1	0	2x2
S3	Sept. 14	0	2	1	3x2
S4	Sept. 14	1	15	5	21x2
S5	Sept. 14	1	6	1	8x2
<hr/>					
S1	Oct. 10	0	5	0	5x2
S2	Oct. 10	17	4	0	21x2
S3	Oct. 10	7	6	2	15x2
S4	Oct. 10	5	6	4	15x2
S5	Oct. 10	0	3	0	3x2

Potassium Nitrate Cultures

On July 19th, three series of potassium nitrate cultures were made to be run in comparison with cultures A,B,C,D and E made with Aurelia aurita. The nutritive nitrate solution A, namely, 2 M. KNO_3 (20.2 grams in 100 cc. distilled water) recommended by Dr. E. J. Allen was used. ("On the Artificial Culture of Marine Plankton Organisms"--*Jour. Marine Bio. Assoc. Vol. VIII--P.439*). For convenience in making accurate measurements the solution was diluted one half and made in the proportion of 5.04 grams of KNO_3 in 50 cc. of distilled water and 8 cc, 4 cc., 2 cc., and 1cc. respectively placed in a litre of fresh sea water, the proportions of 2 M. KNO_3 being one half the above measurements per litre. From each litre, after being well shaken, 250 cc. was poured off and placed in a jar. Three complete series, I, II, and III, were made and numbered 1 to 5 in descending concentration. Series I and II were placed in a pan on the roof, Series I in full sunlight and Series II in 5% full sunlight. Series III was placed in the north window of the laboratory.

Growth in this series appeared very slow and of little volume. The familiar milkiness of the jelly fish cultures and the yellowish brown deposit appearing in about 5 days was absent. By Aug. 9th a slight brown colour had appeared in Series I but so little change was apparent that the whole series was closed to make room for Culture N.

TABLE IX

Only a few counts in these series were made, those which appeared most significant from the recorded observations being chosen. They showed that the addition potassium nitrate alone to natural sea-water produced results far inferior to that obtained from the addition of Aurelia aurita.

TABLE IX POTASSIUM NITRATE CULTURES

Jar	Date	<u>Nitzschia</u>	<u>Navicula</u>	Other Genera	Total
I1	July 29	0	6	1	7x2
I2	July 29	0	2	2	4x2
I1	Aug. 13	0	8	0	8x2
I2	Aug. 13	1	5	0	6x2
III-1	July 29	0	7	0	7x2
III-2	July 29	0	3	1	4x2
III-1	Aug. 6	0	6	0	6x2
III-2	Aug. 6	0	4	0	4x2
III-3	Aug. 6	1	11	2	14x2
III-4	Aug. 6	0	4	11	15x2
III-5	Aug. 6	0	2	0	2x4

SUMMARY AND CONCLUSIONS

- 1.--The addition of a small quantity of Aurelia aurita to natural sea-water stimulated the growth of diatoms present in that water provided that the quantity used was not less than about one-half per cent. by weight.
- 2.--The cultures made in the laboratory were unsuitable for the continued growth of the genera of diatoms most abundant in the water at that time but furnished favorable conditions for the development of other forms, the most common being N. closterium and small species of Navicula.
- 3.--Full sunlight produced a more rapid and greater amount of growth than light intensity of 28% or less.
- 4.--The decomposition of the jelly fish took place most rapidly in full sunlight as evidenced by the milkiness which was produced. This, Dr. Allen, in his paper "On the Culture of the Plankton Diatom Thalassiosira gravida Cleve". page 433, attributed to the growth of bacteria. Whether the growth of the diatoms was aided by the products of metabolism of those bacteria or to the presence of the organic matter of the jelly fish was not determined.
- 5.--The addition of nitrates alone to natural sea-water did not produce results at all comparable with those obtained from the addition of jelly fish.

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