Staff 1946

F. A. Brown, Jr., Northwestern University (Annelida), In charge of course
T. H. Bullock, University of Missouri (Coeleenterata)
W. D. Burbank, Drury College (Protozoa, Porifera)
O. G. Goodchild, Southwest Missouri State Teachers College (Helminthes)
J. H. Lochhead, University of Vermont (Arthropoda)
Madeleine E. Pierce, Vassar College (Mollusca)
W. M. Reid, Monmouth College (Echinodermata)
Mary D. Rogick, College of New Rochelle (Molluscoidea, Protochordata)

Laboratory Assistant

Antoinette Baca, Vassar College
Marine Biological Laboratory, Course in Invertebrate Zoology

Memorandum to students on what to bring to Woods Hole;

Equipment. Compound and dissecting binocular microscopes are essential, and you should make every effort to secure them from your own institution. As stated in the catalog, a limited number are available for rental at the Laboratory. Pack the microscopes securely. They may be insured at a low rate during transit and while at the Laboratory by advance arrangement through Mr. MacNaught's office. You must take the initiative in this. If microscopes are shipped, rather than carried as personal baggage, they ought to be dispatched early, as delays in transportation may otherwise leave you without an instrument for several days. Ship by express, not freight. Other essential equipment are dissecting kit and 6-8X hand lens. Drew's "Laboratory Manual of Invertebrate Zoology", 5th edition will be used in the course. These may be brought by the students, purchased at the Laboratory Supply Dept. after arrival or rented from the course supply for a nominal fee. Haecker's "Invertebrate Zoology" or Borradaile and Pott's "The Invertebrate" will be useful to you, in addition to the strongly advisable identification manual (Pratt) mentioned in the catalog. A microscope lamp and a spotlight for the binocular will be very useful.

Clothes. Woods Hole is very informal, and slacks and sport shirts are the rule. You may, however, wish to bring some "presentable" clothes for evening wear or for Saturday night dances at the M. B. L. Club (not formal evening dress). Raincoat and hat are essential, as is at least one outfit of warm clothes (heavy sweater, etc.). For collecting trips you will need an old pair of sneakers, bathing suit, old dungarees or overalls, old shirt and old sweater that can stand salt water.

Suggested miscellaneous and options. Sun glasses; sunburn cream; beach towel or robe; tennis racquet and balls; sketchbook or water colors, camera and film, etc. A few phonograph records to lend for the dances or for the Monday night symphony concerts will be appreciated.

Remember that you are expected to arrive and check in with the laboratory assistant, Miss Baca, before 5 P.M. on Monday, July 22nd. At 7:30 some introductory remarks about Woods Hole, the Laboratory and the Course will be made. These last will be followed by a lecture on the sea by Professor Redfield of the Woods Hole Oceanographic Institution.
August 30, 1946

Dr. Charles Packard, Director
Marine Biological Laboratory
Woods Hole, Massachusetts

Dear Dr. Packard,

May I take this opportunity to make the following report with regard to the Invertebrate Course for the summer of 1946.

We were very fortunate in being able to keep the same excellent staff as of 1945. The course received 107 applications (58 men and 69 women). Fifty-five were selected (26 men and 29 women) representing a total of 58 institutions (see student list attached). 64% of the class were either graduate students or post-doctorates; 27% held senior standing in their home institutions; only 7% were college juniors and these were all students with very good recommendations. At the last minute, filling a vacancy, one college sophomore was admitted on the basis of his presence at Woods Hole, and a glowing recommendation from Dr. Tashiro whom he had assisted in a medical school course in Physiology. Twenty-five students (45%) had already taken at least one course in invertebrate zoology. Thirteen students (24%) were veterans of World War II. The tuition of 17 students (31%) was paid by their institutions. Student ages ranged from 19 to 36 with a mean of 24.2 years. All students with the exception of one were voted and awarded certificates of satisfactory performance in the course.

In addition to the routine program of the course (see attached 1946 program) there were eight special lectures. Six of these by members of the M.B.I. staff constituted a series on general aspects of Oceanography which the staff of the Invertebrate Course felt was basic to an understanding of marine biology. A seventh introduced the students to the plant life of the region. The eighth was a general lecture upon some broad phylogenetic problems. All these lectures I felt were excellent. The following is a list of the titles, dates, and speakers:

Lectures on Some General Aspects of Oceanography

1. "The sea as an Environment" Monday, July 22 8:00 p.m.
   Dr. A. G. Redfield

2. "Ocean Currents" Thursday, July 25 4:00 p.m.
   Mr. O'D. Ieelin

3. "The Tide" Monday, July 29 7:30 p.m.
   Dr. A. G. Redfield

4. "Geographical Distribution of Marine Animals" Thursday, Aug. 1 8:00 p.m.
   Dr. L. Huntins

5. "Nutrition in the Sea" Monday, August 5 7:30 p.m.
   Dr. B. L. Ketcham

6. "Food Resources of the Sea" Thursday, August 8 8:00 p.m.
   Dr. C. L. Clarke

This section redacted due to personal information.
Other Lectures

7. "Algae of the Woods-Hole Region" Thursday, August 15 7:30 p. m.
   Dr. Hannah Crossdale

8. "Some problems of Invertebrate Morphology" Thursday, August 29 9:00 a. m.
   Dr. Libbie Hyman

The possibility of election of special research projects to be carried on concurrently with the regular course subject matter has been described in the Laboratory Bulletin for several years but has not been encouraged. This year it was called to the attention of the student body on the first day of the course and eleven (20%) of the class selected special introductory research projects with staff members. Four elected to work with Dr. Barberocks upon Protozoa (3) and Porifera (1). Two worked with Dr. Bullock in problems in Ccelenterates (1) and Osteophora (1). One worked with Dr. Reid on Cestodes of marine fishes; one with Dr. Goodchild on digenetic trematodes; one with Dr. Pierce on early embryology of some common marine invertebrates and two worked with me on endocrines in Crustacea. These research projects, I might emphasize again, were carried on in addition to the minimum lecture and laboratory work scheduled for the course.

The laboratory directions have been revised slightly over last year, including additions to the lists of references to the original literature. We have supplemented the mimeographed material with references to Drew's "Invertebrate Zoology" as has been customary for many years. With this latter volume now out of print, and with only enough copies remaining to take care of one more year the staff is making preparations to extend the mimeographed manual for 1948 to make it completely independent of Drew.

Proposed Plans and Needs for 1947

It is now my hope that the current excellent staff of the course can be held together for another year. No member of the staff has been associated with the course for more than four years. With Dr. Bullock's move to California, however, there is a strong possibility that he will ask to be replaced some time before next summer. The staff is 100% with major professional interests in the invertebrates and nearly 100% are active research workers in their respective fields. It would be difficult in my opinion to assemble a more enthusiastic and cooperative staff.

The following are our needs for the summer of 1947 as far as we can foresee them at this time:

**Laboratory of Invertebrate Zoology:**
- Blackboards resurfaced.
- Goose neck lamps for every student.
- Water-table piping checked and repaired.
Space Requirements:
Other O.K. Rooms
For the Whole Season: #21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, and 45 or 46.
From July 23 to Sept. 1 #82, 56.

We would like to have small sinks installed in O.K. rooms 27, 24 or 25, 30, and 22 or 23. If at least two of these four are installed we would be able to get along in a moderately satisfactory fashion without #46 or 45 as requested earlier. It is my understanding that several small soapstone sinks are being replaced in the Brick Building. These latter would probably serve adequately our needs.

Budget for Instruction
See attached sheet.

Consultant in Invertebrate Zoology
We would like to have the name, Dr. Libbie H. Hyman added to the list.

Assistants in Invertebrate Zoology
The following people have been appointed to serve as assistants. The last two will serve as assistants without stipend.
Marie Wilson, Western Maryland College
Virginia Fogerson, Vassar College
Ames L. Hopkins, Jr., Harvard University.

Sincerely yours,

Frank L. Brown, Jr.
Instructor in Charge
Department of Invertebrate Zoology

FARNES
14-Waters
15-Enders
16-Hopp
17-Hend
18-Elberull
19-Gae
20-Case
21-Markhoto
22-Endes
23-Peters
24-Vivian
25-Edward
26-Osborn
27-Anderson
28-Church
Invertebrate Class 1946

1- Morris
2- Sanderson
3- Robinson
4- Rice
5- Candelas
6- VanHoesen
7- Banner
8- Warner
9- Williams
10- Saslow
11- Berreau
12- Hopkins
13- Ehrentheil
14- Warters
15- Enders
16- Hopp
17- Hand
18- Liberti
19- Gese
20- Gese
21- Meinkoth
22- Mendes
23- Peters
24- Vivian
25- Edwards
26- Crocker
27- Pierce
28- Baca
29- Burbanck
30- Rogick
31- Lochhead
32- Brown
33- Reid
34- Bullock
35- Goodchild
36- Gehr
37- Netmore
38- Liu
39- Amberson
40- Bergquist
41- Ferguson
42- Foley
43- Pollens
44- Kramer
45- Moulton
46- Emerson
47- Foreman
48- Feld
49- Bingham
50- Chadwick
51- Thompson
52- Chivers
53- Sullivan
54- Seitner
55- Swanson
56- Jakowska
57- Rafferty
58- Fullerton
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60- Humphrey
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62- Smith
63- Cattell
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### Time of Low Tide

**Woods Hole Oceanographic Institution Wharf**

**1946**

**Eastern Daylight Saving Time**

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INVERTEBRATE ZOOLOGY CLASS PROGRAM
1946

(Subject to change according to the weather)

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<tr>
<td>Mon</td>
<td>July 22</td>
<td>7:30 P.M. Introductory remarks and lecture on the sea</td>
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<tr>
<td>Tue</td>
<td>23</td>
<td>Protozoa (Burbanck and Rogick)</td>
<td>STONY BEACH (8:45--10:28-11:30) Lab Rev 1:45-4:00</td>
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<tr>
<td>Wed</td>
<td>24</td>
<td>Protozoa (Burbanck and Rogick)</td>
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<tr>
<td>Thur</td>
<td>25</td>
<td>Porifera (Burbanck and Rogick)</td>
<td>NOBSKA (1:00-1:56-3:30) Lab Rev 4:30-5:30</td>
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<tr>
<td>Fri</td>
<td>26</td>
<td>Porifera (Burbanck and Rogick)</td>
<td>COELENTERATA (Bullock and Pierce) 1:45-4:15</td>
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<tr>
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<td>27</td>
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<td>28</td>
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<td>Mon</td>
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<tr>
<td>Tue</td>
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<td>Wed</td>
<td>31</td>
<td>Platynhemminthes (Goodchild and Reid)</td>
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<td>Sat</td>
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<td>Brazil (Rogick and Brown)</td>
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<td>Tues</td>
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<tr>
<td>Wed</td>
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<td>Thur</td>
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Unless otherwise specified, class hours are from 9:00-12:00 and 1:45-4:15
## INVERTEBRATE ZOOLOGY CLASS
### 1946
#### Seating Plan

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**Lecture Table**

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### Teams and Instructors for Field Trips

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<th>Team</th>
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<th>WDB</th>
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<td>Capt. 3</td>
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<td>Lagoon Pond</td>
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<td>9:00-1:42-3:00</td>
<td>2</td>
<td>3</td>
<td>X</td>
<td>6</td>
<td>Capt. 4</td>
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<td>Hadley Harbor</td>
<td>Wed. Aug. 14</td>
<td>1:30-3:25-4:30</td>
<td>6</td>
<td>Capt. 4</td>
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<td>Cuttyhunk</td>
<td>Mon. August 26</td>
<td>9:30-1:55-3:30</td>
<td>6</td>
<td>Capt. 4</td>
<td>3</td>
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Before the start of each field trip all members of each team are to provide themselves with the proper collecting apparatus (according to the "Team Organization" list) and report to the proper instructor (according to the above chart) in the laboratory. Students are not to proceed to the boat until given specific permission by the Team's Instructor. N. B. that each team member is entirely responsible for the care and return in clean condition of his or her assigned piece of equipment.

Immediately upon return from a trip, the team members must help the Ark Angel remove all specimens to cool, fresh salt-water in appropriately sized finger bowls, watch glasses, etc. with numbered labels (pencil on paper) corresponding to the Ark bottle numbers. This must be done at once, before leaving for meals or to change clothes.

Teams will meet in the laboratory with their respective instructors, usually at 7:30, to review the names and characteristics of the forms collected, to preserve specimens where desirable, to identify small and doubtful forms, and to prepare field trip reports when and as directed. Angel must check and hand in team collecting record.
Directions to Students on Conduct of Field Trips and Use of Assigned Apparatus

This should be read in its entirety by each student at the beginning of the course, because each will eventually use all pieces of equipment, and in the meantime can help his classmates to remember the operation of their respective equipment.

Recording Angel ("Angel").

This is perhaps the most important single job. Carry a pencil, Angel Book Check list (which is mainly for reference on spelling of names, etc.) and a piece of board on which are pinned several sheets of paper to use in recording the results of the collecting. Follow the instructor as he supervises the collecting, and record (1) the tentative identification of each specimen found; (2) the number of the bottle into which it is put by the Ark Angel; (3) any notes about behavior, breeding condition, exact location, parasites, ecological or faunal relations, etc. etc. (4) Any identification characters which will be helpful in identifying the animal again in the field - e.g., "Shimmy Worm" for Nephthys, "rough shell, sharp spire" for Littorina littorea, "smooth shell, sharp spire" for Littorina rudis and "smooth shell, blunt spire" for Littorina palliata. (Each team member may then enter these field characteristics in his second checklist (not the one used as a trip record) and use this as a short of field key to help in identifications. This, however, is not required, and is simply recommended as being very helpful.) Use a separate sheet for each area collected, heading the sheet with the number of the area (according to the map), a brief description of the area (as suggested above in the directions for the Field Trip Report), and any other ecological peculiarity. Be sure that the forms you record are typical inhabitants of the area, rather than incidental or accidentally. For example, Bryozoa are practically never found in mud, but may occur on rocks projecting above the surface of the mud, and should thus be described. Likewise, Spirorbis is indeed found on rocky beaches, but not on the rocks (on seaweed).

After returning to the laboratory, supply the labels for the specimens when they are transferred from the numbered Ark bottles to fresh seawater in preparation for the Lab. Review.

During the Lab. Review, correct and amplify the Field Notes. After identifications are completed, rule off, on the Angel Book checklist, one column for each area collected. Put in the appropriate columns, opposite the proper name of the animal, the estimates of abundance which were recorded on the Field Record sheets. The special notes on various species should not go in the Angel Book Checklist, unless the instructor so directs, as this list serves only as a record of the total number of forms found by your team on a given trip. The special notes, however, should be recorded by each student individually either in his Field Key Checklist, or in his Field Trip Report.

Finally, after each team member has finished with the complete team record, give the Angel Book Checklist to the Laboratory Assistant for storage as a permanent record.

Ark Angel ("Ark")

The Ark has the important task of gathering the specimens collected and seeing that they arrive safely at the laboratory. Before starting on the trip check over the Ark and see that all bottles are present and clean. When collecting starts, follow the instructor and put each
specimen, as collected, into a bottle of appropriate size. Make cer­
tain that the Recording Angel puts down the number of the bottle in
which the specimen is put. Do not crowd the bottles - worms and
criustaeas, in particular, need plenty of space. Leave some air space
in the bottles. If "doubling up" becomes necessary, be very careful
not to mix up the records by putting two animals of different species
from different areas together, and be careful not to mix animals which
will prey on one another. Be sure the bottles are securely corked
while the Ark is in motion, but whenever a lengthy pause occurs (par­
ticularly at lunch time or on the trip home) take the opportunity to
uncork the bottles to permit aeration, replace turbid water, and splash
the bottles in cool water. Don't dip the whole Ark, as the bottles
will float away. It is your responsibility to see that the animals
reach home in good shape. You may either carry the whole Ark with you
(watch out for slippery rocks) continuously, or set it on shore and
just carry out a knapsack full of bottles, returning for a new load
when necessary. In any case, be sure the Ark, when not being carried,
is on a flat solid support, out of reach of the tide, and in the shade,
if possible; and be sure to dash water over it frequently to keep the
bottles cool.

Immediately upon return from a trip, and before changing clothes or
leaving for supper, transfer all specimens from the bottles to fresh
sea water in watch glasses or finger bowls. Be sure each is accompani­
ed by a label (in pencil) bearing its proper bottle number, and be
certain that the bottles are really empty - small forms often stick
around the shoulder of the bottle or on the cork. Gastropods and
some crustaceas will often climb out, so should be loosely covered,
Don't mix forms which will prey on one another. Rinse out Ark bottles
return in proper order, and replace the Ark in the Storeroom.

Sea Bucket
Do not use the See Bucket for carrying anything, and do not let others
put anything in the bucket, as the window breaks easily and is diffi­
cult to replace. Be careful not to sink it or let it float away from
you. Never try to fill any buck when the boat is in motion. In using
the See Bucket be sure the window is clean. Visibility is improved by
wetting the upper surface of the glass and by putting the face close
to the glass.

Scrape Net
Be careful not to catch the netting between the steel edge and the
piling.

Sieve
Hold by bottom, rather than by hand holes, as shovel may bruise your
fingers. Shake gently, particularly when gravel is present, as other­
wise delicate animals will be macerated. With thick mud, water should
be splashed in from above.

White Pan, Finger Bowl, Watch Glass, Forceps, etc. These are for
examining more closely the animals caught in net or sieve. Stick
close to the instructor and be ready to take charge of the animal and
transfer it to the Ark Angel when the identification is made.
Habitat Symbols to be used in angell sheets and check lists.

1. In sand or mud:
   - CS = Clean sand
   - MS = Muddy sand
   - M = Pure Mud
   - PS = Peaty sand
   - G = among grass roots.

   Use figures to indicate depth in inches, e.g., MS 6 = 6" down in muddy sand; CS O = at surface of clean sand.

2. Attached to or crawling on rocks in still water:
   - Ex = On exposed surface
   - U = On underside

3. Attached to or crawling on rocks exposed to waves or to strong currents:
   - Ex = Exposed surface
   - U = On underside

4. On or among algae in still water:
   - U = Ulva
   - A = Acetabularia
   - F = Fucus
   - E = Enteromorpha
   - R = Red Alga

   Use same key letters as in category 4.

5. On or among algae exposed to waves or strong currents:
   - NR = Not recognized

6. Free Swimming or floating:
   - TP = In tide pools
   - S = Over sand or mud
   - O = In open water (Tow net collections)
   - SP = Spray pools

7. On pilings:
   - T = In tidal zone
   - B = Below tidal zone
   - U = Unattached

8. Dredged in deep water:
   - S = On sand bottom
   - G = On gravel bottom
   - A = On algae
### Invertebrate Zoology Course 1946

<table>
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<tr>
<th>Team 1</th>
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<th>Team III</th>
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<td>1. Fullerton</td>
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<td>7. Cattell</td>
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Table indicating who is responsible for each piece of field equipment for each of the filed trips. The numbers of the individuals of each team are given above.

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<th>Al</th>
<th>Had</th>
<th>H</th>
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Abbreviations: P - white pan; SB - see bucket; WG - watch glass; FB - finger bowl; Pi - pipette; F-forceps; CD - crystalizing dish.
CHECK LIST OF INVERTEBRATE ANIMALS
Commonly or occasionally found by the Invertebrate Zoology Classes
at the
Marine Biological Laboratory
Woods Hole, Mass.

Forms marked (*) are most commonly found or are conspicuous for other reasons. Where a name used in Pratt's "Manual of the Common Invertebrate Animals" (revised edition) differs from the name used in the check list, the name used in Pratt follows the check list name, and is enclosed in parentheses.

Woods Hole, Mass.

*******
PHYLM: PROTOZOA

(not listed)

PHYLM: PORIFERA

Class Calcarea
*Leucosolenia botryoides
*Sycon ciliatum

Class Demospongia
*Chalina arbuscula
*Cliona celata
*Halicondria panicea
*Microciona proliferata
Suberites compacta
Tethya gravida
Class Hydrozoa

Abietinaria abietina
*Bougainvillia
  B. caroliensis
  B. supercilialis
*Campanularia
  C. calceolifera
  C. rilexuosa
Clava leptostyla
Clytia
  C. edwardsi
  C. johnstoni
  (j. bicophora)
Cordylophora lacustris
Corynitis agassizi
  (Gemmaria genmosa)
Ectopleura ochracea
*Eudendrium
  E. album
  E. carneum
  E. ramosum
  E. tumuc
Eutima mira
Gonionemus nurbachi
Halecium halecinum
*Hydractinia echinata
Nemopsis bachei
*Obelia
  O. commissuralis
  O. geniculata
  O. bisuspidata
*Pennaria tiarella
Physalia pelagica
Podocoryne
  P. carneas
  P. fulgurans
*Schizotricha tenella
*Sertularia pumila
Stylactis hooperi
Thuiaria argentea
*Tubularia
  T. crocea
  T. larynx
Turritopsis nutricula

Class Scyphozoa

Aurelia aurita
Cyanea capillata
Dactylometra quinquecirrha

Class Anthozoa

Aleyonim carneum
*Astrangia danae
*Edwardsia
  E. elegans
  E. leidyi
Eloactis producta
Haicampa farinacea
*Metridium dianthus
*Sagartia
  S. leucolena
  S. luciae
  S. modesta

PHYLUM CTENOPHORA

*Neonlropsis leidyi
PHYLUM PLATYHELMINTHES
Class Turbellaria
Sub-class Acoela
*Polychoerus caudatus
Order Tricladida
*Bdelloura candida
Procerodes wheatlandi
Syncoelidium pellucidum
Order Polycladida
Eustylochus ellipticus
Planocera
P. inquilina
P. nebulosa
Stylochus zebra

PHYLUM NEMERTEA
Order Paleonemertea
Cephalothrix spiralis
Order Heteronemertea
*Cerebratulus lacteus
*Lineus
L. bicolor
L. ruber
L. socialis
*Micrura leidyi
Order Haplonemertea
Amphiporus ochraceus
*Tetrastrama
T. candidum
T. vermiculum

PHYLUM NEMATHELMINTHES
(not listed)

PHYLUM TROCHELMINTHES
(not listed)
PHYLUM MOLLUSCOIDEA

Class Entoprocta
Barentsia
*Pedicellina

Class Ectoprocta
*Acanthodesia tenuis
Actea anguina
Aevernilla setigera
Alcyonidium
*Bowerbankia gracilis var.
*Bugula
*B. flabellata
*B. turrita
Callopora aurita
*Crisia eburnea
*Cryptosula pallasiana
Electra
E. hastingsae
*E. pilosa
Flustrella hispida
Hippodiplosia
H. americana
*H. pertusa
Hippoporina contracta
Hippothoa hyalina
Membranipora
M. crustulenta *(M. lacrullae)
M. tuberculata
Microporella ciliata
Schizmopora avicularis
Schizomavella auriculata
Schizoporella unicornis
*Smittina trispinosa var. nitida
*Stephanosella biaperta
Tegella unicornis
PHYLUM ANDELEIDA
Class Chaetopoda

Sub-class Polychaeta

Ampharete setosa

*Amphitrite
  A. brunnea
  A. ornata

Anaitides ctenula

*Arabella iricolor

*Arenicola
  A. cristata
  A. marina

Aricia ornata

*Autolytus sp.

Chaetopterus variopedatus

*Cirratulus grandis

*Clinedaphes Gouldi

*Clymenella torquata

*Diopatra cuprea

Dodecaceria coralii

*Enchytreus albidus

*Enoplobranchus sanguineus

Eteone sp.

*Eulalia annulata

Eumidia sanguinea

Flabelligera affinis

*Glycera
  G. americana
  G. dibranchiata

Harmothoe imbricata

*Hydroides dianthus

Laonice viridis

*Lepidodaphes squamatus

Lumbrineris
  L. hebes
  L. tenuis

Maldane urceolata

Marphysa sanguinea

*Neanthes
  N. succinea
  N. viridens

Nepthys
  N. bucera
  N. incisa

*Nereis pelagica

Nicolea zostericola

Ninoe nigripes

Notomastus
  N. filiformis
  N. luridus

Parasabella microphthalmia

Pholoe minuta

Pista palmata

Platynereis dumerillii
Class Chaetopoda
Sub-class Polychaeta
Podarke obscura
*Polycirrus eximius
Polydora sp.
Pseudopotamilla oculifera
Sabellaria vulgaris
*Scoloplos
  S. fragilis
  S. robustus
Spio setosa
*Spiorbis spiorbis
Stauronereis sp.
*Sthenelais leidyi
Syllis sp.
Terebella rubra
Terebellides stroemi
Thelepus cincinnatus
Travisia forbesi
Sub-class Oligochaeta
Enchytraeus albidus
Lumbricillus agilis
Class Gephyrea
*Phascolosoma gouldi
PHYLUM MOLLUSCA
Class Amphineura
* Chaetopleura apiculata
Class Pelecypoda
* Anomia simplex
* Arca
  A. campechiensis pexata
  A. transversa
Astarte castanea
Barnea truncata
* Brachidontes demissus plicatilis
Cardium pinnatulum
Corbula contracta
* Cumingia tellinoides
* ENSIS directus
Gemma gemma
Gouldia mactracea
Laevicardium mortoni
Lyonsia hyalina
Maoma
  M. balthica
  M. tenta
Mactra solidissima
* Modiolus modiolus
Mulinia lateralis
* Mya arenaria
* Mytilus edulis
Noetia ponderosa
Nucula sp.
Ostrea virginica
Pandora gouldiana
* Pecten irradians
Periploma leanum
Petricola pholadiformis
* Solemya velum
Tagelus
  T. divisus
  T. gibbus
* Tellina tenera
* Teredo navalis
* Venus mercenaria
Yoldia limatula
Class Gastropoda
Acanthodoris sp.
* Acmaea testudinalis
Aeolidia sp.
Alexia myosotis
Anachis avara
* Bittium alternatum
Busycon
  B. canaliculatum
  B. carica
Class Gastropoda (Continued)

Caecum pulchellum

*Cerithiopsis
  *C. adamsii
  C. greeni
  C. subulata

*Coryphella* sp.

*Crepidula
  *C. fornicata
  C. glauca
  C. plana

Elysia sp.

Epitonium sp.

Eupleura caudatum

Facelina bostoniensis

Haminoea solitaria

*Lacuna vincta

Littorina
  *L. littorea
  L. obtusata
  L. saxitilis

*Melampus lineatus

Melanella intermedia

*Mitrella lunata

*Nassa obsoleta
  N. trivittata
  N. vibex

*Natica clausa

*Odostomia* sp.

Polinices
  *P. duplicata
  P. heros

Rissoa sp.

Thais lapillus

Turbonilla sp.

Turritella sp.

*Urosalpinx cinereus

Vermicularia spirata

Class Cephalopoda

Loligo pealei
PHYLUM ARTHROPODA
Sub-phyllum Crustacea
(Sub-order Cladocera and Classes Ostracoda, Copepoda, and Branchiura not listed.)

Class Cirripedia
Order Thoracica
*Balanus
  B. balanoides
  B. eburneus
*Chthamalus fragilis
Lepas
  L. anatifera
  L. fascicularis

Order Rhizocephala
Peltogaster paguri

Class Malacostraca
Order Mysidacea
*Heteromysis formosa
  Michtheimysis stenolepis

Order Cumacea
Diastylis sp.

Order Tanaidacea
Leptochelia savignyi
Tanais cavolinii

Order Isopoda
Chiridotea
  C. caeca
  C. tuftsii
Cirolana concharum
Cyathura carinata
Edotea triloba
Erichsonella filiformis
*Idothea
  I. baltica
  I. metallica
  I. phosphorea
*Jaeera marina
Limnoria lignorum
Philoscia muscorum
Probopyrus pandalicola
Sphaeroma quadridentatum

Order Amphipoda
Aeginella longicornis
Allorchestes littoralis
Ampelisca macrocephala
Amphithoe
  A. longimana
  A. rubricata
*Caprella acutifrons
Carinogammarus mucronatus
Chelura terebrans
*Corophium cylindricum
Elasmopus levis
*Gammarus
  G. Annulatus
  G. locusta
  G. marinus
<table>
<thead>
<tr>
<th><strong>Class Malacostraca</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Order Amphipoda</strong></td>
</tr>
<tr>
<td>Grubia compta</td>
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<tr>
<td>Haustorius arenarius</td>
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<tr>
<td>Jassa marmorata</td>
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<tr>
<td>Lembos smithi</td>
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<tr>
<td>Leptocheirus pinguis</td>
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<tr>
<td>Lysianopeis alba</td>
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<tr>
<td>Melita nitida</td>
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<tr>
<td>Microdeutopus gryllotalpa</td>
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<tr>
<td><em>Orchestia platensis</em></td>
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<tr>
<td>Stenothoe</td>
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<tr>
<td>S. cypris</td>
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<tr>
<td>S. minuta</td>
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<tr>
<td><em>Talorchestia longicornis</em></td>
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<tr>
<td><em>Ucniola irrorata</em></td>
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<tr>
<td><strong>Order Decapoda</strong></td>
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<tr>
<td>Callianassa stimpsoni</td>
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<tr>
<td><em>Callinectes sapidus</em></td>
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<tr>
<td><em>Cancer</em></td>
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<tr>
<td>C. borealis</td>
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<tr>
<td>C. irroratus</td>
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<tr>
<td><em>Carcinides maenas</em></td>
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<tr>
<td><em>Crago septemspinosus</em></td>
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<tr>
<td>Emerita talpoida</td>
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<tr>
<td>Eurypanopeus depressus</td>
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<tr>
<td>Heterocrypta granulata</td>
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<tr>
<td>Homarus americanus</td>
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<tr>
<td><em>Libinia</em></td>
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<tr>
<td>L. dubia</td>
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<tr>
<td>L. emarginata</td>
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<tr>
<td><em>Neopanope texana</em></td>
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<tr>
<td><em>Ovalipes ocellatus</em></td>
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<tr>
<td><em>Pagurus</em></td>
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<tr>
<td>P. acadianus</td>
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<tr>
<td>P. longicarpus</td>
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<tr>
<td>P. pollicaris</td>
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<tr>
<td>P. pubescens</td>
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<tr>
<td><em>Palaemonetes vulgaris</em></td>
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<tr>
<td>Panopeus herbsti</td>
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<tr>
<td>Pelia mutica</td>
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<td>Pinnixa</td>
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<tr>
<td>P. chaetopterana</td>
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<tr>
<td>P. cylindrica</td>
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<tr>
<td>P. sayana</td>
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<tr>
<td><em>Pinnotheres maculatus</em></td>
</tr>
<tr>
<td><em><strong>Uca</strong></em></td>
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<tr>
<td>U. minax</td>
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<tr>
<td>U. pugilator</td>
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<tr>
<td>U. pugnax</td>
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<tr>
<td>Upogebia affinis</td>
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<tr>
<td><em>Virbius zostericola</em></td>
</tr>
<tr>
<td><strong>Order Stomatopoda</strong></td>
</tr>
<tr>
<td><em>Chloridella empusa</em></td>
</tr>
</tbody>
</table>
PHYLUM ARTHROPODA (Cont'd)

Sub-phylum Insecta
Class Apterygota
Order Collembola
  *Anurida maritima

Sub-phylum Arachnida
Class Xiphosura
  *Limulus polyphemus
Class Acarina
  Family Halacaridae
  Various species
Class Pseudoscorpionidea
  One species (Chelonethida)
Class Pycnogonida
  Anoplodactylus lentus
  Pallene empusa
  Tanystylum orbiculare
Incertae sedis
Class Tardigrada
  Various species
PHYLUM ECHINODERMATA

Class Asteroidea
  *Asterias
    A. forbesi
    A. vulgaris
  *Henricia sanguinolenta

Class Ophiuroidea
  *Amphipholis squamata
  *Ophioderma brevispinum
  Ophiopolis aculeata
  Ophiura sp.

Class Echinoidea
  *Arbacia punctulata
  *Echinarchinus parma
  Strongylocentrotus drobachiensis

Class Holothuroidea
  *Leptosynapta
    L. inhaerens
    L. roseola
  *Thyone briareus
PHYLUM CHORDATA
Subphylum Enteropneusta (Hemichordata)
  *Saccoglossus kowalevskyi
Subphylum Tunicata (Urochordata)
  Amaroucium
    *A. constellatum
    A. pellucidum
    A. stellatum
  Appendicularia
  *Botryllus schlosseri
  Ciona intestinalis
  Didemnum
    *D. candidum latarium
    D. albidum
  Molgula
    M. arenata
    *M. manhattensis
  *Perophora viridis
  Salpa
  *Styela partita
NOBSKA POINT

Inset 200 ft. west of main map

Freshwater Pond

Private Road

Nobska Beach

1/2 mile

1. Sandy Beach

Nobska Light

2 Sandy Bottom

Rocks with Seaweed

3

Radio Tower

4 Small Rocks

3a Large rocks with seaweed

21 Rocks with Seaweed
INVERTEBRATE ZOOLOGY COURSE - 1945 - Student Field Trip Report

Name __________________________ Team ___ Trip ________ Date _______

Time of Low Tide ______ Collected from ___m. to ___m. Air Temp. ___

Water temp. ______ (indicate local variations from this below under habitat description) Weather: Wind _____________ Sky _______________

Precipitation __________________________________________________________________

I. Describe each of the habitats studied. Refer to appended map. Include such factors as: nature of substratum (be more specific than "mud" or "rocks"); degree of exposure to or protection from wave action (estimate importance of scouring and impact); strong currents; temperature fluctuations, direct sunlight, salinity fluctuations, silt, pollution; tidal levels involved; important plants.

II. List the most abundant species, in the order of abundance, in each habitat. If more than one natural grouping or definable association of abundant species occurs in a habitat, distinguish them. Distinguish the few (5 or 4) most important species which characterize or define each association.

Species Ecologic notes
### III. Probable or possible ecological correlations or questions:

Think about the area and the animals studied, their physical and biotic environment, their needs, adaptations and problems. Using your imagination and common sense, but without "loose" thinking, suggest pertinent questions about this area and attempt explanations of the presence and absence of particular species. Some examples of general questions: Are predators, herbivores, scavengers, or plankton feeders markedly more important here, one than the other? Why? What general groups of animals are absent or poorly represented? Do they have a common denominator which might explain this? What are the most important problems of existence of each habitat which all its fauna must overcome? What differences are discernible between the exact situation and habit of life of species belonging to genera or families of which we have more than one member in this vicinity? Point out correlations between structure or habit of life and physical conditions imposed by the environment (e.g. byssus threads in mussels which live exposed to currents, permanent tube-dwellers more common, wandering burrowers less common in clay-mud where gas exchange below surface is probably poor and cohesion of particles may impede burrowing). Use back of sheet or extra pages if necessary.

<table>
<thead>
<tr>
<th>Species</th>
<th>Ecologic notes</th>
</tr>
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<tbody>
<tr>
<td>1</td>
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<td>9</td>
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<td>10</td>
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</tr>
</tbody>
</table>

### IV. Notes about rare, striking or otherwise interesting forms not covered elsewhere.
<table>
<thead>
<tr>
<th>Trip No.</th>
<th>Locality</th>
<th>Map area</th>
</tr>
</thead>
<tbody>
<tr>
<td>Date</td>
<td>Weather</td>
<td></td>
</tr>
<tr>
<td>Temp. (a) Air. °C</td>
<td>Water. °C</td>
<td>Low tide</td>
</tr>
<tr>
<td>Exposed to waves?</td>
<td>Stagnant currents?</td>
<td>Silt?</td>
</tr>
<tr>
<td>Any fresh water?</td>
<td>Pollution</td>
<td></td>
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<tr>
<td>General description of area</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Field identification</th>
<th>Bottle</th>
<th>Habitat (use key)</th>
<th>Abundance</th>
<th>Ecological notes etc.</th>
</tr>
</thead>
</table>

(Continued over)
The display of Protozoa in the Woods Hole region is no less spectacular than the larger invertebrates, but because of their small size, we do not intentionally collect them on our field trips. Two outstanding reasons for studying these interesting animals are as follows: it is generally agreed that the Protozoa are the ancestors of the Metazoa; and, the Protozoa play a very important role in the complicated food cycles of the littoral and pelagic regions of the ocean.

Any living material is relatively difficult to study and the size and rapid movements of Protozoa aggravate this situation. Since we cannot observe individual protozoans with the unaided eye, we have insisted (See Annual Announcement of Marine Biological Laboratory) that you equip yourself with good optical instruments. In 1943, Buck described a method for quieting free-moving Protozoa which we shall use and thus put the study of these animals on a par with their larger descendants.

You may think of your work for the next few days as an ecology field trip brought indoors. An opportunity will be afforded for you to study succession, behavior, social organization (commensalism, parasitism, and symbiosis), and adaptation to environment. For the more skilled, experienced, or ardent student, we have arranged for additional observations and experiments to be carried out.

We have obviously served you with a larger "helping" than most of you will be able to "consume" in the time allotted. Conduct yourself as you would at your favorite Smorgasbord and sample as many things as possible. Not only make a wide choice, but also select the larger "helpings" (larger protozoans). These will be easier to observe and identify with accuracy and in most cases, they are just as typical of a particular habitat as the smaller forms.

The following is a suggested procedure:

NOTE: In all your work, share exceptionally good preparations with others at your table.

A. ECOLOGICAL STUDY

1. After you have lightly coated your optical instruments with vaseline, it might be well for you to start your study by obtaining from the water-table both slides and Syracuse dishes that have been submerged for a week and two weeks in live casts in the Eel pond and the dishes have been submerged for three days and a week in running sea water. This material can be used for the study of succession since the slides and dishes have been submerged for different lengths of time. IT IS BEST TO BEGIN BY STUDYING THE ATTACHED FORMS --KEEP SLIDES AND DISHES FOR FURTHER STUDY ON FREE-MOVING FORMS.

On the glass slides from the Eel Pond other attached forms
can be found by placing a clean syracuse dish in a finger bowl and placing the slide in the finger bowl on top of the syracuse dish and quickly filling the finger bowl until the slide is just covered with sea water. The finger bowl containing the dish and slide may then be placed on the stage of your dissecting scope. The general arrangement of the colonial forms may be worked out in this way, and when higher magnification is necessary for detail, the bottom of the slide may be wiped clean and a cover-slip put on the preparation. On these slides you will probably find such forms as Actinophrys, Heterophrys, Zoothamnium, Cothurnia, and Vorticella, etc. DON'T BE CONFUSED BY YOUNG BRYOZOA OF PROTOCHORDATES.

II. To study the effect of other substrates on distribution of additional sessile forms, you may make slide preparations of small "sprigs" of algae and hydroids (coelenterates) which have been scraped from wharf pilings and boats. In such material, you will probably find such members of the Class Suctoria as Ephelota, Podophry Acineta, etc. Paracineta limbata may often be found not only in the above material, but also growing attached to bryozoa.

III. Using the method described by Buck (1943) you may now wish to study the free-moving animals. With a dissecting needle, put a small amount of yeast, to which Congo Red has been added, into a drop of sea water containing the motile forms. If you ring your cover-slip with a small amount of vaseline, the preparation will last longer and in 10 minutes the animals will be slowed down enough so that you may readily study them. In all probability, you will see red food vacuoles form which will later turn blue.

Your slides and syracuse dishes used earlier will still prove to be a good source of motile forms, but the hydroid and algae buckets will probably have more types such as Urostyla, Uronychia, Holosticha, Anisonema, Pelomyxa, Lacrymaria, Euplotes, Philaster, Dysteria, Aspidisca, Diophyra, Gymia, Lombus, Raphidiophrys, Massula, etc.

If, for some reason, the forms are not too numerous, place some of the algae or hydroids in a finger bowl of sea water and keep until the next day. Also, if material is removed from the bottoms of the buckets on the water-table with a long pipette, a large number of animals may be found.

IV. Protozoa from brackish ponds (fresh water ponds made somewhat salty when sea water is added during storms or by seepage) are quite interesting, numerous, and rather different from those found in a strictly marine habitat. (The famous Lillie's ditch is the source of some of this material -- see labels on jars.) You may expect to find such forms as Frontonia, Condylactoma, Paramecium, Childonella, Loxophyllum, Dilileptus, Loxodes, Prorodon, Holophyra, Coleps, etc.

(USE THE YEAST - CONGO RED SUSPENSION ON THESE TOO)

V. Open water is quite different in its protozoan types from more enclosed sea water or brackish water. Typical of this open sea environment is the ciliate, Tintinnopsis. You will recognize it by its "house" (lorica) made of sand crystals (Calkins) and the adoral zone enclosed by tentacle-like membranelles. A SMALL JAR CONTAINING TOW MATERIAL WILL BE FOUND ON THE BACK TABLE.
It is not likely that you found many living Foraminifera or Radiolaria so prepared slides of these animals are provided for you. Dinoflagellates are tremendously important in the economy of the ocean, and if you have not found any living ones, there are also prepared slide of Noctiluca. THESE SLIDES ARE ALSO ON THE BACK TABLE.

B. SOCIAL ORGANIZATION

NOTE: Most easily found forms are marked with an asterisk (*). Use Kudo instead of Pratt for identification.

1. Commensals

**Protozoa**

**HOST**

*Boweria teredinidi*  
*Teredo navalis* (Shipworm)

Tease the gill lamellae found on the end of the animal near the valves. Mount teased material on a slide in sea water. Look for the commensals attached to the lamellae. If you do not find them the first time, try another *Teredo*. (If you are not familiar with the anatomy of the shipworm, see figure in Borradaile and Potts, P. 587)

*Lichnophora macfarlandi*  
*Crepidula plana* (Boat shell-
mollusk)

Lichnophora is found on the egg cases of *Crepidula*. The egg cases may be found by separating the host from the substrate to which it is attached. If eggs are present, they will appear as yellowish or greyish mass adhering to the substrate. Remove some of the eggs to a slide, cover, and examine the egg cases with low power--other ciliates may be present.

*Ancistrum isseli*  
*Modiolus modiolus* (bearded mussel)

Cut the adductor muscles by inserting a scalpel between the valves. Pry them apart but do not separate. Wash the surface of the mantle cavity and foot into a syracuse dish by means of a pipette. The ciliate remains quiescent and can be easily studied, or may be transferred to a slide. CAUTION: use just a little water for washing.

*Ancistrum mytili*  
*Mytilus edulis* (Edible mussel)

*Conochrophthirius mytili*  
*Modiolus modiolus* (bearded mussel)

Open in the manner described above for *Modiolus*. The smaller more numerous ciliate is *Ancistrum*. It looks much like the form found in *Modiolus*. *Conochrophthirius* is much larger and less abundant. It sticks to the bottom of the dish or the surface film.

*Chilodonella* sp.  
*Orchestia* sp. & *Talorchestia longicornis*

*Allosphaerium* sp  
*Orchestia* sp & *Talorchestia (Sand fleas) longicornis*

Kill host with a dissecting needle and then place the sand-flea in a syracuse dish of sea water. The commensals may be found by use of
the dissecting scope using transmitted light. Both ciliates are found on the exoskeleton of the host. Chilodonella has a conspicuous oral basket and is smaller on the average than Allosphaerium.

*Folliculina sp*  
Bdelloura egg cases (Flatworm)  
on  
Limulus (Horseshoe crab)

Bdelloura egg cases are small, elliptical yellowish brown bodies on the gill books of Limulus. If you remove these and put them in a syracuse dish of sea water, you can find Folliculina by using your dissecting scope and then, if necessary, transfer the protozoan to a slide.

**Trichophrya salparum**  
Molgula manhattanesis (Protochordate)

Fill finger bowl with enough sea water to cover the host. Look for suctorian with dissecting scope and remove it to a slide.

**11. Parasites**

*Trichodina sp*  
Thyone briareus (Sea cucumber-Echinoderm)

Place Thyone in a weak amonia solution (2%) and, when it softens slightly, return it to the sea water. The animal will immediately eviscerate the alimentary tract. Cut off portions, open and wash contents on to a slide. The peritrich is barrel-shaped and may move fairly rapidly.

*Schizocystis sipunculi*  
Phascolosoma gouldi (Annelid)

Remove the intestine of the host, open it and wash the contents into a syracuse dish. Transfer the parasite to a slide. It is pointed at both ends, curved, and wriggles slowly.

Haplozoan clymenella  
Clymenella torquata (Bamboo worm--annelid)

**Monocystis clymenella**

Procedure like that for Phascolosoma above.

*Paravorticella clymenella*  
Clymenella torquata

Procedure the same as above, but be sure to get the extreme posterior end of the intestine, the colon. **NOTE:** Posterior end is marked by a caudal funnel.

Selenidium echinatum  
Dodecaceria (Blackish annelid)

Procedure the same as above, The parasite is located in the intestine.

Anoplophrya orchestii  
Orchestia agilis (sand-flea)

Crush the sand-flea on a slide and add sea water. The parasitic ciliate is small but abundant in the infected host. Examine a leg. In an infected host, the lacunae will be packed with the parasites.
You may have to try several hosts, but one infected one may supply several students.

*Protophrya ovicola*  
*Littorina saxitilis* (Small snail)  
(Same as *L. rudis*)

This parasite infests the females of *L. saxitilis*. The ciliate is located in the uterus. Break the shell of the snail and place the animal in a syracuse dish of sea water. Tense apart the visceral hump between the red ovary and the head. When the host is well infected, enough parasites will probably be present for several preparations. You may have to try several snails before finding an infected one.

**DEMONSTRATIONS OF PARASITIC FORMS IF OBTAINABLE**

**Porospora gigantea**  
*Homarus americanus* (Lobster)  
Intestine

**Nematopsis sp**  
*Primary host--Mud crab*  
*Panopeus herbsti or Eurypanopeus depressus*  
*Secondary host--Ostrea virginica* (Oyster)  
Spores in muscles, gills and mantle

**Meroregarina amarouci**

**A gregarine**  
*Balanus eburneus* (Barnacle)  
Intestine

**Trichodina sp**  
*Puffer fish*  
Gills and gill chamber

**III. Symbionts**

In the Animal Kingdom, there is no better example of symbiosis than that between the termite and the flagellates that inhabit its intestinal tract, therefore, since you are studying the Protozoa, we include this material.

**Trichonympha sp.**  
Termites of this region

**Dinemynpha sp**  
"  
**Pyronympha sp**  
"  
**Spirotrichonympha sp.**  
"  
Grasp the head of the termite in one pair of forceps and the tip of the abdomen in the other. Pull the latter gently. By this means, the intestine can be pulled out of the body. Tease it gently and add a drop or two of 4% saline solution. The intestinal flagellates are abundant. The genus *Trichonympha* is the largest and has the longest flagella. *Dinemynpha* is next largest and is flask-shaped with the anterior end pointed. *Pyronympha* is smallest and moves in a corkscrew manner. *Spirotrichonympha* is compact and has a spiral structure of the pellicle.
REAGENTS AVAILABLE ON THE TABLE IN THE REAR OF THE LABORATORY


   How it is prepared: One-four cake of yeast and 30-40 mgs. of Congo Red boiled gently for 5-10 minutes. Keep in refrigerator.

2. Carmine suspended in sea water for "feeding" Protozoa. Carmine will form red vacuoles. Mix a very small amount of carmine suspension with the Protozoa.

3. Two nuclear stains are saturated methyl green in 1% acetic acid and aceto-carmine (supersaturated carmine in 45% glacial acetic). A drop of either of these stains run under the cover-slip will kill the organism and stain the nucleus. Caution: Either dispose of slide and cover-slip after using these stains or wash both carefully before using again.

4. Methylene blue and Neutral Red are both vital stains for staining living Protozoa.

5. For evisceration of Thryene, 2% sea water solution of ammonia.

6. For termite flagellates, 0.4% saline solution.

C. EXPERIMENTAL WORK

The work outlines here will not have to be completed in the time allotted for the study of protozoa. It is not expected that most or even the majority of you will decide to do additional work with this group of animals. In all fairness to the Protozoa, it should be pointed out that significant work can be done in a relative short time because the organisms multiply so rapidly.

The following are suggested problems and if any others occur to you, they will, of course, be acceptable if they will not take too much time from your study of the other Phyla or your participation in the activities of your collecting them:

1. Methods Of Sterilization:

   a. Use of the Migration-dilution apparatus developed by Mr. C. Lloyd Claff.* The apparatus to be used has graciously been lent to the Invertebrate Zoology course by Mr. Claff, consequently, we can only show our appreciation by using it with the utmost care. This method is used for sterilizing many Protozoa at once.

   *Research Fellow in Surgery, Harvard Medical School

   b. Use of Parpart's method. This method is used for sterilizing single or very few Protozoa at one time.

2. Mating types.

Dr. Ralph Wichterman of Temple University has graciously supplied the course with enough Paramecium bursaria for the whole class to follow through the process of conjugation.
3. Permanent staining of Protozoa by use of standard methods of staining on the cover-slip with the use of the small Columbia staining dishes.

PLEASE NOTE

1. DRAW as many of the forms suggested in Sections A and B as time will allow. Divide your time evenly between the two sections. Work in Section C. may be extended past the time allotted for the Protozoa.

2. IDENTIFY the Protozoa as closely as possible. Try to identify the animal WHILE YOU ARE STUDYING IT, DON'T TRY TO IDENTIFY FROM A DRAWING. You all have at your disposal the revised edition of Pratt. There are also several copies of Kudo "floating" around although, they should be returned to the front table immediately after using. In addition, the excellent treatise by Kahl is extremely valuable. It is in German, but the drawings are all in English.

3. Include with your drawings notes on your observations such as approximate size, behavior, substrate, vacuole formation (food and contractile), and source of material. Anything else of interest that you observe please add.

THESE RECORDS ARE TO BE TURNED IN JUST BEFORE THE LECTURE ON PORIFERA.
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PORIFERA

The laboratory outline is prepared for a full day of work on the Porifera. Therefore, organize your work so that at the end of the day you will have accomplished those parts of the laboratory study which are most interesting and beneficial to you.

The regeneration experiments on Microciona will be started first in order that the material may be observed during the day.

Regeneration of Microciona. Each student will need a finger bowl, a Syracuse watch-glass and a slide. The group at each table will need an additional finger bowl to be used in common. Fill the finger bowls about two-thirds full of sea-water. Place the watch-glass on the bottom and the slide on top of the watch glass. Into the common bowl, cells and broken fragments from Microciona will be pressed through fine meshed bolting cloth. Pipette a small quantity of this material into the finger bowls containing the slides. As soon as the cells have had time to settle on the slide, carefully lift it out and examine it. Return it carefully to the finger bowl. Study several times during the day. Have the cells shown a tendency to aggregate?

The structure of the reunion mass and the cells involved are discussed in papers by Wilson and Galtsoff. Many of the cell types, the syncytium which forms the surface layer and all of the skeleton are retained by the bolting cloth. Wilson considers three types as represented in the free cells, one, the choanocytes which remain specific and produce a new gastric epithelium, two, cells with nucleolate nuclei and possessing granules to a variable degree in their cytosomes and, three, non-nucleolate cells possessing in their cytosomes fine granules of uniform size which gives to the cells a grayish color. He regards the nucleolate type as a primordial cell which produces in the reunion mass, as in the adult, skeletal and reproductive cells. The non-nucleolate cell produces the syncytium which covers the body and lines those canals not occupied by choanocytes. Some indication of cell types can be learned from fresh preparations but fixed and stained material has been found more satisfactory. Do not spend too much time trying to identify the different types of cells.

Ascon type. (Leucosolenia). Note relation of individuals to each other and prevalence of buds. Make a habit sketch.

Select an individual which is relatively free from debris. With a razor or sharp scalpel cut it lengthwise and place the halves on a slide so that both inner and outer surfaces may be viewed under the same cover glass. Note the different types of spicules and especially their general arrangement. Note, also, the lack of cementing material (spongin) to hold them together such as will be found in some monaxonid sponges, e. g. Halichondria and Chalina. Focus on the gastric surface and observe the beating of the choanocyte flagella (The details of the collar cells may be studied better in Sycon). Note the numerous porocytes with their intracellular openings. Draw a porocyte and some adjacent choanocytes.

A spicules in this form and also in Sycon, Chalina, Halichondria and Microciona can be studied best by examining the residue which remains
after sponges have been boiled in potassium hydroxide. Material has been treated in this way for you and is available on the preparation table. Draw acetic acid under the cover glasses of your several preparations. Note the results. Illustrations and names of some typical spicule shapes are found in Pratt's Manual and Hyman's Invertebrates, p. 298. Compare the descriptions given in the text for species represented in Woods Hole with your observations. Draw each type of spicule found in each of the sponges available. For the non-calcarea refer to Vosmaer's The sponge of the Bay of Naples. Write the scientific term under each spicule. See pp. 95-105 in Delage and Herouard. Emphasis has been placed on the form of the spicules because the spicule is frequently the most constant morphological structure available in the classification of some sponges.

**Sycon type.** (Sycon called Grantia in older texts and Scypha by de Laubenfels.) Suggestions for study of this sponge are given in Draw (pp. 37-40, 5th edit.). The remarks here are supplementary to the text. The relative sizes of incumbent and radial canals and their relation to each other may be seen clearly in a tangential section of a dried skeleton made as follows: Two cuts are made parallel to the long axis; the plane of the first passes tangentially to the middle of the body wall, the plane of the second, parallel to the first, is made sufficiently near the long axis so as to include part of the inner surface.

Study of living material. Observation on the direction of current flow can be carried out either in Sycon according to directions in Drew or in Halichondria which shows a more vigorous flow of current if good material is available.

It is suggested that, instead of tangential sections mentioned in Drew for living material that you try to make cross-sections sufficiently thin to allow study with a ×4 mm. objective and yet, in making the sections, to retain typical relations of the animal's structures. Sections a quarter to a fifth of a millimeter are about right. The choanocytes lining the radial canal will be visible and with proper adjustment of condenser and illumination one may observe flagella, collars, and the prosopyles. Draw a choanocyte if you did not observe one in the fragments of Microciona.

Observe also in this preparation germ cells and developing embryos which may be present. Compare those amphiblastulae with the larvae of Microciona. These latter may be obtained by dissecting pieces of the adult sponge with needles. When freed from the parent the larvae swim out into the surrounding water. Draw whatever embryonic stages, either in sycon or microciona that may be available in your material.

**Loucon type.** 1. (Chalina). Observe the general organization of the colony. Identify the oscula. Only a rough idea can be gained of the anatomy of the sponge from thin cross-sections but such a preparation will reveal the manner in which short monaxonid spicules are effectively held together by spongin. Use magnification of about 400 x. Make a drawing to illustrate this.

2. (Halichondria). Place a piece of a colony in a Syracuse watch glass and look for an osculum. Note the vigorous flow of water from it. A thin membrane which forms the osculum is a syncytium. It covers the surface of the sponge forming its dermal
layer. The spicules beneath support it like so many tent poles.
Numerous ostia are present in the membrane which lead into the
extensive subdermal space below. From this lead the incumbent canals
directing the water toward the flagellated chambers.

Other sponges. Examine such other sponges as may be available in the
laboratory, probably Tethya and Cliona. Cliona is the sulphur sponge—
smell it. It is able to bore into shells, but apparently not by the
use of an acid.

All records of your work on the Porifera are due not later than to-
morrow morning at 9:00 a. m.

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when treated with narcotics or attacked by aquarium
Classification of the Phylum Porifera

Class I. Calcarea of Calcispongia, the calcareous sponges.
Skeleton composed of separate calcareous spicules, one-, three-, four-rayed, not divisible into megascleres and microscleres.
Order 1. Homosclera, the ascoc sponges. Structure ascocoid.
Order 2. Heterosclera. Structure ascyconoid or leuconoid.

Class II. Hexactinellida or Triaxonida or Hyalospongiae, the glass sponges. Skeleton composed of triaxon (six-rayed) siliceous spicules or some modification of the triaxon form, separate or united into networks; choanocyte limited to finger-shaped chambers arranged in a simple or folded layer; without surface epithelium.
Order 2. Amphidiscophora. With amphidisks, no hexasters.

Class III. Demospongiae. Skeleton of siliceous spicules or horny fibers or both; siliceous spicules not triaxon, generally differentiated into megascleres and microscleres; flagellated chambers mostly small, round, of the leuconoid type.
Subclass I. Tetractinellida. With tetraaxon spicules; no sponging; spicules sometimes wanting.
Order 1. Myxospongia. Without spicules; structure simple.
Order 2. Carnosa or Homosclerophora or microsclerophora. Megascleres and microscleres not sharply differentiated; mostly without triaenes; asters may be present.
Suborder 1. Astrophora. Microscleres include asters.
Suborder 2. Sigmatophora. Microscleres when present are sigmas.
Subclass II. Monaxonida. Megascleres monaxonal; with or without spongia.
Order 4. Hadromerina or Astromonaxonellida. Megascleres mostly tylostyles; microscleres when present some form of aster; without spongia. (Cliona, Suberites, Tethys).
Order 5. Halichondrina. Megascleres mostly of two or more kinds; microscleres wanting or are raphias, with little spongia. (Halichondria)
Order 6. Poecilosclerina. Megascleres often of two or more sorts, localized in distribution; reticulate, united by more or less spongia; often with echinating spicules; microscleres include sigmas, chelae, and toxas. (Microciona)
Order 7. Haplosclerina. Megascleres of one kind, diactinal, without special localization; with or without microscleres; spongia generally present. (Chelina)
Subclass III. Keratos, the horn sponges. Skeleton composed of spongic fibers, without siliceous spicules.

Taken from "The Invertebrates" by L. H. Hyman.
Wood Hole offers especially favorable opportunities for the study of certain aspects of coelenterates and ctenophores. Some of these are listed below in the order in which suggestions for study are presented on the following pages. In order to allow for differences in interest and preparation more subjects are listed than there is time for any one student to undertake. Since the class will not be working together it will be necessary to choose the topics you wish to study and to organize your time according to the topics chosen, your preparation and the materials available. It is recommended that everyone who has not already a good background in the general morphology of these groups do I and II. Those who have such a background should at least examine the living material offered. Unless special interests dictate otherwise one or more of the topics listed in each of III and IV may then be studied.

I. Gross morphology of Coelenterate
   A. The medusoid form - structure of various hydrozoan and scyphozoan medusae
   B. The planula stage - examples from several genera
   C. The polypoid form - structure of a few types

II. Gross morphology of Ctenophore - chiefly of one large species common in this area

III. Taxonomy and life cycles
   A. Classification of hydroids and the use of keys
   B. Life histories and reproduction in representative genera of hydroids

IV. Biology, behavior and responses - physiology of the receptors, effectors and nervous system.
   A. Behavior of hydroids.
   B. Spawning in Hydactinia
   C. Feeding behavior of anemones
   D. Feeding behavior of medusae
   E. Behavior, responses and luminescence in a ctenophore
   F. Regeneration in a hydroid
   G. Regeneration and grafting in a ctenophore
   H. Structure and types of nematocysts
   I. Excitation of nematocysts
   J. Structure of mesoglea in different groups
   K. The nerve net: experiments on anemones
   L. The nerve net: experiments on medusae
I. GROSS MORPHOLOGY OF COELENTERATA

A. THE MELUSOID FORM

Class Hydroscoa

1. Order Hydroida

   a. Anthomedusae (Suborder Gymnoblastae), Padocaryne, Bougainvillae.

The medusae available are very young ones, recently produced, asexually, by the hydroid polyp. Place in depression slide. Examine under binocular by reflected light, with black surface below slide, and by transmitted light. Observe general form, swimming movements, contractions of tentacles, etc. Examine under compound microscope. If necessary, quiet with a drop or two of magnesium sulphate. Make an outline drawing of the general structure of this or one of the other hydromedusae.

   Note: Exumbrella, the outer, convex, boral side of the dome-like bell. Subumbrella, the inner, concave, oral side. The surface of both is ectodermal. Velum, the muscular diaphragm partly closing in the subumbrellar space, perforated by a large central opening. It is characteristic of hydromedusae, sometimes called craspedote medusae. Manubrium, the stalk pendant from center of subumbrella. At its tip is the mouth. If the medusa were mature gonads would be formed in the walls of the manubrium (cf. Leptomedusae). Gastrovascular system: the mouth opens into a gut or manubrial cavity at the base of which is a sac, the stomach. Four chalices, the radial canals, lead from the stomach to the edge of the bell. These canals define the periradial, adradial and interradial axes. A circular canal at the edge of the bell connects the radial canals. These connected spaces are lined with ciliated endoderm and serve to prepare and distribute ingested food as well as to carry oxygen and to dispose of wastes for the inner tissues. Mesoglea is the noncellular jelly between ectoderm and endoderm. The tentacles are muscular filaments with an endodermal core. Their structure and arrangements differ characteristicly in the various kinds of meduses and they may be at the tip of the manubrium as well as at the margin of the bell. Sense organs: ocelli are present in some but not all kinds of meduses; they are pigmented light-sensitive spots on the tentacle-bulbs at the edge of the bell. These and the statocysts of leptomedusae are the first sense organ (distinguished from sense organelles of protozoans) in the animal kingdom. Nematocysts occur in concentrations especially on tentacles and lips, but are better studied in polyps such as Pannaria.
b. Leptomedusae (Suborder calyptratea), Obelia

Examine as for Anthomedusae, noting differences. This disc-shaped medusa sometimes lies "inside out" with its sub-umbreller surface more convex than the exumbreller one. Its velum is poorly developed, and it does not employ the water-jet method of swimming. Its circular canal is obscure. It lacks ocelli but has special sense organs of balance, the statocysts (not found in Anthomedusae), which are transparent cups containing concretions. Immature gonads may be seen if the medusae are not too young, as four small swellings on the radial canals near the base of the manubrium. This position in the radial canals is characteristic for the suborder.

2. Order Trachyline, Gonionemus

This relatively large form is very favorable for study and has been much used in experiments on behavior. It was once abundant in the Eel-Pond but disappeared some years ago after the eel grass died out. Examine preserved demonstration material. Consult Drew, pp. 48-51.

3. Order Siphonophora, Physalia

Some hydromedusae in addition to sexual reproduction also exhibit budding of new medusae from the bell margin or manubrium. In siphonophores this process is carried to an extreme and the budded individuals remain adherent to the original one. In the colony thus formed, various individuals are specialized for various functions, polymorphism is exhibited. Physalia is a highly integrated colony which appears and behaves as an individual but the gas-filled float, tactile tentacles, mouth-bearing manubrium-like feeding zooids and gonad-bearing stalks, all may be regarded as highly modified interconnected medusoid and polypoid zooids. Examine demonstration material and consult texts on demonstration table to identify structures. If living Physalia is available observe killing of Fundulus by it.

Class Scyphozoa

1. Order Semaecostomeae, Cyanea and Aurelia

Observe swimming movements in a large bowl of fresh sea water. Compare with the locomotion of hydromedusae. What anatomical bases for the differences can you find? Dissect a specimen in a finger bowl. Make sketches, for example an oval view of one quadrant or more and a vertical section through a selected axis.
Note the absence of a velum. In the center of the sub-umbrella four curtain-like oval arms hang from the corners of the mouth. These arms are ciliated and armed with nematocysts. Food entangled in mucus is borne to the mouth along the arms which also serve for incubation of the eggs. The mouth opens by a short pullet into a large flattened stomach. The mouth and oval arms are supported by four heavy radial ribs of jelly, spanning the stomach. Between these ribs the subumbrella is a thin walled, folded membrane on the aboral side of which, in the floor of the stomach, lie the brown gonads. In the sub-umbrellar ectoderm of *Cyanea* at about the level of the peripheral margin of the stomach is a band of circular muscle. Radiating outward from the circular muscle are sixteen strips of radial muscle, each anchored to the jelly of the disc by a radial septum in its midline. The muscular system in *Cyanea* is in contrast to that of *Aurelia* in that (1) the muscle is optically differentiated and visible in the living animal in *Cyanea*, transparent and invisible in *Aurelia*. (2) the musculature is essentially uniformly distributed over the subumbrella in *Aurelia*, not concentrated in bands, and (3) consists virtually entirely of circular muscle.

The distribution of the nerve net is related to that of the muscles. It can be worked out by simple experiments. These are outlined under IV, below.

On the subumbrellas of *Cyanea* a peripheral to the circular muscle originate eight adradial groups of hollow, heavily armed, contractile tentacles. The edge of the disc is cut into sixteen lappets. Set in sensory niches between the lappets in the perradii and interradii are the eight rhopalia or tentaculocysts. These are club-shaped bodies bearing sense organs and special nervous structures including organs of equilibrium containing concretions of calcium sulphate, and centers which maintain the rhythmic pulsations of the bell. Cut off part of the edge of the bell and examine the rhopalia in detail.

In the floor of the stomach just central to the gonads is a zone of gastric filaments which secrete digestive enzymes. Beyond the level of the gonads the septa anchoring the radial muscles divide the gastrovascular cavity into sixteen gastric pouches, which near the edge of the bell give off numerous short branches into the lappets. There is no circular canal.

Consult Draw for directions on *Aurelia*. 
B. THE PLANULA STAGE

Class Scyphozoa

The eggs or sperm of *Cyanea* medusae are shed into the stomach and passed out of the mouth. The eggs are fertilized in the stomach of the mother by sperm in the seawater and become adherent in clusters to the oral arms, where they develop into planulae. The planulae are released as minute, yellow, pear-shaped larvae with ciliated ectoderm and thick endoderm. Nematocysts are present. The larvae swim rapidly for a time then settle down hind end up. In our *Cyanea* from Lagoon Pond the settled planula flattens and encysts instead of becoming a polyp at once. In autumn the cyst hatches into a small polyp, the scyphistoma.

Examine planulae of *Cyanea* with compound scope, under a coverslip. Set planulae in a watch glass aside in a cool place for 4-6 hours and observe encystment. Examine photograph of hatching cysts.

Class Hydrozoa

The eggs or sperm of mature hydrozoans, *Bougainvillea*, *Podocoryn*, *Obelia*, etc., are shed directly into the sea. The fertilized eggs develop in a few hours or days into a planula which for a time swims free or creeps over the substrate. Examine a watch-glass containing planulae of the Hydrozoan *Hydractina*, settled in several stages and developing into polyps. Detach a well-developed polyp, place it under a coverslip and examine it briefly under the compound microscope.

C. THE POLYPOID FORM

Class Hydrozoa

The planula of Coelenterata develops into a polyp which usually but not always reproduces sexually by budding off other similar individuals. A complex colony of polyps may be formed if the budded individuals do not become separated from each other. When fully developed, the colony may bud medusae which detach themselves and swim away. In many Hydrozoa, however, the medusa-buds remain attached to the colony until their sexual products are ripe; and in most Hydrozoa, the medusa-buds are mere vestiges, and the genera appear to be borne by the polyp itself.

Place a branch of a colony in a watch-glass. Note the regular arrangement of the parts. The individuals or hydrenths are connected by common stems of coenosarc covered with a horny sheath, the perisarc or periderm. The hydrenths bear tentacles of two types, threadlike and knobbled, differently placed. The mouth is terminal, at the end of the proboscis. Just above the circle of threadlike tentacles are medusa-buds. These ripen their gonads while still attached to the polyp. Their tentacles are rudimentary, and they are not equipped for a free life.

If examined after 8 o'clock at night, the ripe medusae buds will be seen to be expanded. They pulsate, and discharge the eggs and sperm from the gonads in their membranous wall, shortly before or after which the medusa becomes detached and later dies.

Make accurate sketches of the polyps and developing medusa-buds of Penneria, dissecting out parts if necessary. Observe the ectoderm and endoderm. Are they differently colored? Can you find cell-wells? The gastrovascular cavity, lined with ciliated endoderm, continues from the cavity of the hydranth into the stems and other parts of the colony. Is the food current by one hydranth available to another which has caught nothing?

Note the differences in the nematocysts of the different kinds of tentacles. Run a small drop of methyl green under the coverslip, drawing it in by application of absorbent paper to the far side. Watch the large nematocysts discharge their thread as the dye reaches them. Draw discharged nematocysts. Can you observe the thread inside undischarged nematocysts? Can you find the sensory "trigger" (cnidocil) of the cell (cnidoblast) which secretes and encloses the nematocyst?

2. Structure of a Calkytoplasm Hydroid.

In the Calkytoplasm, the perisarc continues over the hydranth as a protective cup. The gonad-producing medusoids (conophores) are borne only by special modified hydrants (gonozoids). These gonozoids (which may be reduced to a mere stalk or blastostyle) plus the conophores, are encased in a sheath of perisarc called the gonotheca. The whole is termed a gonangium. Examine Obelia, or Campanularia, following Drew (pp. 45-48).

Class Scyphozoz

Living polyps may be available from Cyanene or Aurelia. If not, examine preserved demonstration material of scyphopolyps and ephyre. Read Drew (pp. 57-9 on the development of the polyp (scyphistem) and the strobilization of the young medusae (ephyre) from it. Draw a scyphistoma or strobile and an ephyre.
Class Anthozoa

Subclass Zoantharia

1. Order Actinaria, Metridium and Nematostella, if available

Make a thorough study of Metridium, following Drew, pp. 59-61.

In connection with Drew, section 3, mount a living tentacle together with the scutia, and run methyl green under the coverslip as directed for staining nematocysts of Hydrozoa. Note the difference between nematocysts of tentacle (some of which are adhesive rather than penetrant) and of scutia. Crushing, by pressure on the coverslip may bring out other nematocyst types not discharged by methyl green.

Study the structure of a living Nematostella, a simple anemone. This animal has been found in the Mill Pond at Woods Hole for the past few years. Otherwise it is known only from the Isle of Wight. Its transparency makes it an excellent laboratory animal. The water where it is found is very slightly diluted and may be considerably so following heavy rains. It lies in very soft mud and debris with the mouth and tentacles extended at the surface.

Allow the animal to expand in a watch glass. Generally the expansion is more rapid and complete in darkness.

Under a dissecting microscope the following feature may be observed:

External parts: Tentacles (How many?) Mouth. Column, differentiated into a thin-walled upper region (capitulum); a thick-walled lower region (scapus) and an expanded basal bulb (physa).

Internal parts: Mesenteries (How many?) How arranged with reference to the tentacle arrangement?). Are all mesenteries primary, i.e., joined to the pharynx? Pharynx. Look for small spheroid bodies lying in the angles where the mesenteries are inserted at the body wall. These are clusters of cells containing nematocysts. They may be dislodged and will swim about in the coelenteron. Nothing is known as to their origin or function. Observe the extent of the mesenterial filaments on the edges of the mesenteries. There are no scutia. Study the activities of Nematostella. How does its locomotion differ from that of Metridium? What foods will be accepted? What is the effect of feeding on peristaltic movements concerned in burrowing?

2. Order Madreporaria, Astrangia

Study and draw under the dissecting microscope the structure of the skeleton of a colony of the star-coral, Astrangia danae. This is the only stony coral the range of which extends
Mnemiopsis is the only common ctenophore in this area. It is a large form and favorable for many kinds of study, including regeneration, polarity and dominance, nervous control of cilia and luminescence. Belonging to the Order Hobata it is considerably modified from the form of the commonly described cydippid types. Consult the account of a cydippid in Drew, and lobate modifications in Hyman pp. 688-9, 683.

Visualize the form of Mnemiopsis as that of a solid sphere modified in the following ways: (1) elongate in the oral-aboral axis. (2) compressed in one side to side axis (called the tentacular axis). (3) expanded at each end of the other side-to-side axis (called the stomodeal axis), into great oral lobes, so that an oral view or cross section might suggest the side view of a

spool or dumb-bell. At the end of the animal where all the lobes hang free is the slit-like mouth, elongate in the stomodeal axis (or stomodeal or "sagittal" plane). At each side of the mouth, defining an axis at right angles to the stomodeal axis is a single tentacle in a small pocket. (These tentacles are much smaller than in most ctenophores and have lost the usual tentacle sheath). Hanging free just to each side of each tentacle is a tongue-like auricle, four in all. These are fringed with well developed cilia.

The most conspicuous and diagnostic feature of ctenophores is the system of ctenophoral or comb-plate rows of ciliated swimming plates. Eight such rows radiate from the aboral pole and extend
meridionally towards the oral end. One pair extends down each oral lobe, these are the adesophageal, subsegittal or terminal plate rows. A shorter row runs into the base of each auricle. These four rows are the adtentacular, subtentacular or lateral rows. Notice that the comb plate rows may be considered adradial. At the aboral pole is a sense organ, including a statocyst, presumably an organ of equilibrium, and possibly other types of receptors. The comb plate rows converge and their synchronism is effected here also.

The internal anatomy is simple. The mouth leads into a long, flattened ectodermal stomodeum. Near the aboral pole this opens into a small endodermal stomach or Infundibulum. Extending aborally from the stomach is a funnel tube which opens to the outside by a tiny eccentric pore in one of its four interradial corners. The stomach gives off eight canals: one in each interradius and four in the per-radial of the tentacular plane, two paragastric or pharyngeal canals which hug the sides of the stomodeum and two to the bases of the tentacles. The inter-radial canals dichotomize very soon into adradial canals which lie under the eight comb plate rows, extending orally beyond their ends and anastomosing in great loops, the two adesophageal of each oral lobe with each other and the adentacular with each other and with the pharyngeal canals. The hermaphroditic gonads are in the walls of the meridional canals. These canals are also the source of the brilliant green luminescence produced on stimulation after dark adaptation.

Ctenophores are micro-predators feeding on mollusce larvae and other plankton forms. Ciliary currents on the surface of the body carry the food into the auricular grooves where it is caught by adhesive cells or colloblasts of the numerous small tentacles of the lobial ridge and transferred to the mouth. There are no nematocysts. Study the action of the comb plates, determine the direction of effective beat and of the metachronal wave passing along the row. This relation is peculiar in ctenophores.

In order to study details of the apical and oral regions, the canals and the action of comb plates cut a specimen into suitable fragments and manipulate these to obtain favorable views. Make general and detailed sketches, semidiagrammatic when necessary. Emphasize and be sure you understand the biradial type of symmetry.
III TAXONOMY AND LIFE CYCLES

A. CLASSIFICATION OF HYDROIDS AND THE USE OF KEYS

Begin by running the species studied under IG through the keys to suborders, genera and species, as though you did not already know their names. Be sure you understand the construction and use of a key.

Obtain unknown forms and run them through the key. Write your identification on your notesheet, then check it by reference to the back of the numbered card accompanying the supply bowl. Note on your sheet the reason for any error you may have made.

As you examine each form, before going on to the next, make both general and detailed sketches, labelling special features and comparing with forms previously studied. In addition to information in the key, employ the outlines below and consult the texts in order to make an adequate study. Make drawings and notes on each form on a separate sheet.

Outline descriptions of common hydroids


Cenepenaria. For study of other parts than gonosome, follow directions for Obelia. For gonosome, see Drew, pp. 47-48.

Clove. A very simple form, the zooids of which are slender naked hydrenths arising singly from a mesh-work of perisepo-covered hydorhiza. Gonophores are sporosacs borne below the proximal tentacles in compact clusters.

Eudendrium. The gonosomes of Eudendrium are complicated relative to those of most Gymnoblastae. The sexes of the colonies are separate. The male gonophores appear to form linear series of two or more sporosacs, several of which series are attached to the hydrenths just proximal to the tentacles. Note the spadix running through each series of sporosacs as a central column. The female gonophores are sporosacs clustered around the base of the hydrenth or tip of the pedicle. Note the distally bifurcated spadix partly encircling the eggs. In some species of Eudendrium the hydrenth bearing the gonophores is highly aborted and becomes a mere blastostyle.

Obelia. 1. Drew, pp. 45-7. For study of cell layers, follow Drew, pp. 43-44, sections 4 and 5 of Hydra. For study of nematocysts follow Drew, p. 44, sections 6 and 7 of Hydra, using a drop of methyl green instead of safranin as reagent. Compare with nematocysts of Pennaria and Hydractinia. Note that the blastostyle of the gonangium represents the gonozoid.
Compare medusae of Obelia with other forms that are available. Look for differences that may be of taxonomic significance. See the note at the end of the key about hydromedusae whose polypoid form is not known.

Pennaria.  1. Proceed as for Tubularia, Drew, p. 51, through section 3.

2. Note the difference between basal and distal tentacles. The knob at end of the capitata tentacles is formed by batteries of stinging cells. Are the tentacles hollow or solid?

3. Observe the relation of the gonophores to the hydranth body. The gonophores of Pennaria are medusae which are usually set free when ripe. Observe the rudimentary tentacles, volvm, radial canals, manubrium. Try to find a series of stages of development of the medusa. The colonies of Pennaria are of separate sex. Examine both a male colony, in which the medusae-buds have whitish spermaries in the manubrium, and a female colony in which the manubrium of the gonophore bears several opaque or pinkish eggs. The medusae are usually liberated, and eggs and sperm shed into the water from them, between 7 and 9 p.m.

Sertularia.  For gonosome, see Drew, p. 48.

Notice that the hydrothecae are sessile, not supported on pedicels as in Obelia or Cerebranularia. The hydrotheca is not open at the tip when the hydranth is retracted, but is closed by a delicate hinged operculum.

Schizotricha.  The perisarc of the stem and of the hydrocladia (main side-branches) is jointed. The divisions between these joints are internodes. The branches arise from the shorter internodes of the main stem.

Regularly arranged on the branches are large and small hydrothecae. The latter is a nemostome containing a mouthless zooid called the sercoestyle, which is armed with batteries of nematocysts and with adhesive organs for food gathering and defensive use.

The gonosomes are large and curved. They arise from the bases of hydrothecae.


B. LIFE HISTORIES AND REPRODUCTION

Begin by briefly reviewing the systematic divisions of the Hydrozoa and their distinctions, especially with respect to reproduction, the sexual bodies and embryology. Run down two or three species through the key so that its use is understood and the kind of species differences that occur is realized.
Then, according to material available and inclination investigate (1) methods of reproduction, (2) structures involved and (3) differences between species and groups. Examine yourself as much live material as possible, taking full advantage of whatever material is available by careful study as opposed to casual observation. Make numerous simple but accurate sketches, general views and details of structures and stages involved in reproduction, with special emphasis on comparison of the same process in different forms. Make use of the texts, the species key and the outline descriptions under IIIA. Consider the significance of the differences between species as respects (1) phylogeny and our interpretations of primitive and derived conditions in coelenterates as a whole and (2) the life and habits of the respective species - limitations imposed and advantages of each condition for particular habitats.

Note the series presented by the order Hydroidea. See representatives of as many steps in the series as possible.

(1) Species with free living medusae produced by a poorly developed polypoid stage, (Conionemus). The polyp is so reduced and inconspicuous as rarely to be found. In some related forms there is no polypoid phase at all.

(2) Species with free living medusae produced by well developed polypoid phase, (Bougainvillea, Podocoryne, Obelia). When released the medusa is not sexually mature and the animal, which possesses all essential organs, swims for some time, feeding and growing.

(3) Species which liberate reduced medusae, unfit for a long free life and with gonads already ripe which are soon discharged. (Stylophora, Penneria)

(4) Species with reduced gonophores, readily recognizable as medusoid, not liberated but producing sexual products while still attached to the polyp. (Tubularia)

(5) Species with greatly reduced gonophores. In some cases the gonophores are sparsely not easily recognized as medusoid, so that the polyp phase appears itself to bear the gonads. (Clava, Hydridinia, Zonaria, Campenularia, Sertularia, Schizotheca)

Note that this extreme reduction of the medusoid is scattered among gonochorys and lower olychorys (compare Podocoryne with the closely related Hydridinia and Obelia or Clytia with Campenularia), whereas it is of universal occurrence among higher olychorys.

Examine as many stages of larval development as possible. Look carefully in bowls in which hydroids have stood for a day without running water, for planulae of Campenularia, embryos and planulae of Penneria.
Actiniae of Tubulæria. Actiniae are essentially planulae which have advanced to the polyp stage before attachment. How does the method of locomotion in actiniae differ from that in planulae. The larvae can with care and luck be maintained for several days in finger bowls until they attach and develop into primary colonies. Compare with planulae and polyps of scyphomeduse.

Note the phenomenon of polymorphism. Most hydroids are dimorphic in the sense that they produce individuals of special form (medusoids) which bear the gonads. Many hydroids also produce special zooids which produce the sexual individuals; and some have still other types of non-nutritive zooids; in other words, they are polymorphic.

(1) Eudendrium renseum: Calve. Are there any regular differences between different zooids of the colony (as between those with and those without gonophores?)

(2) Eudendrium cornutum: Stylectis. Sterile (nutritive) zooids larger and with larger or more numerous tentacles than reproductive ones.

(3) Podocoryne. Nutritive zooids larger and with more numerous tentacles than reproductive ones, although the letter can feed. Sterile, non-nutritive zooids lacking tentacles, of two types, occur.

(4) Hydractinia. Like Podocoryne but reproductive zooids differ much more from the nutritive ones in lacking tentacles and in not feeding.

(5) Schizotrichia. Specialized reproductive zooids (gonozooi); and also specialized sterile non-nutritive zooids. Are there simple dimorphic forms like Calve and Eudendrium renseum among the Calyptoblast hydroids which you have examined?

(6) Class Hydrozoa, Order Siphonophora. Highly polymorphic, free-swimming or floating colonial Hydrozoa, composed of several types of individuals attached to a stem or disc supported by floats. Examine the demonstration specimen.

Study the phenomenon of asexual reproduction, highly developed in coelenterates. The colonies of hydroids such as we have examined usually arise from a single primary zooid formed from the planula. This primary polyp gives rise by asexual reproduction to the rest of the colony, and sometimes to new colonies also.

(1). Solitary hydroid. A manner of growth found in a few Siphonophora, as Caryophyllia (in which asexual reproduction usually does not occur). Sometimes found in Tubulæria corynæa, in which buds are produced which may pinch off from the parent so that no colony is produced, although usually remaining loosely associated with the parent zooid.
(2) **Hydrorhizal colonies.** The zooids bud out singly and unbranched from the creeping hydrorhiza, and consequently there is no hydroc8ulus. In mature colonies of *Clava* and *Hydrectina* the hydrorhiza forms a thick, tangled mat. In such Celyptohlests as *Clytie johnstoni* the hydrorhiza branches to some extent but on the whole runs in clear lines across the substratum.

(3) **Hydroc8uline colonies.** The usual plant-like erect forms. New hydroc8uli may arise at intervals from the common hydrorhizal network, just as do the single polyps of hydrorhizal colonies; thus hydroc8uline colonies may be compound, i.e., include many of the erect groups, all joined by a common coenosarc.

(3e) **With oldest hydranths at tips of colony.** Found in *gymnoblasts*. The terminal hydranth of each branch is the oldest one on that branch. The terminal hydranth of the main stem is theoretically the oldest of the colony and should represent the original hydrorhiza.

The stem and branches elongate indefinitely by means of a growth zone just below each hydranth. Below each growth zone new hydranths are budded.

(3b) (i) **With youngest hydranths at tips of colony.** Stems and branches tipped by hydranths; a method of growth found in the lower families of Celypto-blaster hydroids (e.g., *Compenulariidae*). The hydranths are at the base of the stems or branches. Young hydranths are budded off at tips of each stem, so that the terminal hydranth is constantly losing this position to the next youngest one.

(ii) **With stems and branches tipped by permanent growing points.** In this method of growth, a modification of (2) found in the higher Celypto-blaster (Sertulariidae and Plumulariidae), the tips of the stem and branches continue to grow, and the hydranths arise just proximal to this growing tip. The most distal hydranths are the youngest, as in the other Celypto-blaster, but instead of the growing zone arising proximal to the terminal hydranth as in the Compenulariidae, the terminal hydranth of the Sertulariidae and Plumulariidae arises proximal to the growing zone in which the stem ends.

(3c) **In many Compenulariidae and other hydroids, under adverse conditions the coenosarc is withdrawn from the hydranths, and long, free, hydrorhiza-like stolons are given off from pedicels or branches. These stolons may break off, float away, and on adhering to a suitable substratum produce a new colony. Observe stolons in material set aside yesterday.**
IV. PROBLEMS IN BIOLOGY AND BEHAVIOR

A. Behavior of hydroids

Hydroids are of particular interest because they are the simplest animals to display a nervous system and a muscular system (as opposed to isolated contractile cells, general irritability and cell organelles of lower forms). They are a strikingly successful group of aquatic predators and the state of the receptors, effectors and nervous system illustrates well the relation between anatomy, physiology and habit of life.

The largest and most favorable hydroids for experiments on behavior are not common in this region, but a great deal can be learned by estate observation of simple stimulus-response experiments even if controlled cutting is impossible. In making these observations do not confine your attention to startling phenomena. These animals do not often stand on their heads or throw convulsions. Consider everything the animal or each part of the animal is doing - or not doing - and contrast that with what it might have done, both at "rest" and as a result of planned stimulation. Then consider what this must mean and what it might mean as respects the organization of the sensory and neuromuscular systems. Read the classical account of experiments on Corymorpha by G. H. Parker in "The Elementary Nervous System."

We are especially poor at Woods Hole in large hydrozoan medusae. Students who are interested in this subject are, however, urged to read the classical accounts of Romance in "Jellyfish, Starfish and Sea-urchins" and Loeb in "The Comparative Physiology of the Brain."

Hydrectinia colonies regularly grow on small shells occupied by hermit crabs. The relation is not obligatory for they occur on rocks, pilings, Limulus, etc., and in arctic regions the shells of live snails are utilized, but in these circumstances the colony characteristically lacks one form of dactylozooid present on hermit crab shells. Hold a snail shell with a Hydrectinia colony under water in a finger bowl, by means of forceps placed so their one point, in the mouth of the shell, discourages the crab from emerging. Using very small pieces of mussel meat or other animal food, gently squirt food from a pipette or place with forceps upon the colony. Note which zooids react and how they do so. Follow the process for some time. Try breaking the shell and using fragments with Hydrectinia. Arrange conditions so that the action of nematocysts can be observed. Notice which nematocysts or zooids do not discharge. A piece of cellulose rubbed in iced juice can be used so that observation under the compound scope is possible.

Using an intact shell and holding as before, note the fringe of bent or coiled spiral zooids along the spire-side of the opening of the shell. Scrape the shell with a needle and watch these zooids. What do other zooids do? Note that the same synchronous response of the spiral
zooiids is also given for other stimuli - allow the crab to move in and out, stimulate the crab, jar the dish, determine what stimuli are ineffective. Carefully stimulate one zooid alone, moderately and severely (pinch with fine forceps) to determine the effect on the same and other zooiids. Very little is known about the general problem of the organization of the nervous system in colonial animals; for interested students a bibliography is available.


Test for luminescence in a number of genera of hydroids. Some of them are reported to exhibit it.

Investigate the behavior and responses of Tubularia. Consider both macro- and micro-behavior, i.e., movements of tentacles, activity of cilia and nematocysts. What is the significance of its relative passivity and lack of spontaneous activity?

If well developed hydromedusa are available experiments may be devised to demonstrate various features of the nervous system, depending on the material. Compare rate of pulsation in large and small specimens; in warmer and cooler water; in the presence of excess K ion and low K ion (ask for solutions). Compare the effect of a drug like strychnine on healthy medusae and on small Nereis, Palamometes or other free swimming higher forms. Test for responses to light. Analyze the mechanism of swimming and compare it in different hydromedusae. From your observations on the fact of and the character of the pulsations define and specify problems and experiments which you think would be appropriate on medusae of size sufficient to permit operative procedures.

Examine closely and analyze the "resting" behavior of Physalia and its responses to mechanical stimuli, clam juice or other liquid food, small pieces of solid food and live fish (Fundulus). Ordinarily this organism is too rare here to be able to carry out experimental operations, but when it can be done experiments on the great undifferentiated neuro-muscular sheet of the float and on the various kinds of tentacles and zooiids are very revealing.

From what you have learned of the nervous and muscular systems of these organisms and of their habit of life and food supply evaluate the importance of nematocysts in the coelenterate economy. Suggestions for study of nematocysts are given under IV-H and I.

Write down all your observations and interpretations. This step is as important as any other; the facts you have learned will be virtually useless to you unless they are written down, promptly and completely. Use simple drawings and animated-cartoon-type sketches liberally.
B. The control of spawning in Hydractinia

Hydroid colonies of those species whose gonophores are not liberated as free swimming medusae include a large number of gonad bearing individuals with their sex products in various stages of maturity. These may be shed over a period of two months or more in the year. Each colony is typically of one sex only. From these facts it can be seen that it would be a great advantage to the species if the eggs and sperm were shed periodically in great concentration instead of continuously in small concentration, and synchronously in all gonocytes of a colony and in the colonies of the two sexes. Just this has been described for a number of forms. The mechanism for Hydractinia has been worked out by Ballard (1942, Biol. Bull., 82). A certain sequence of light and dark is apparently necessary and induces spawning quite predictably.

Choose a number of hermit crab shells with Hydractinia colonies. These should in most cases be sexually mature in June and July at Woods Hole. Place them in large dishes of sea water avoiding crowding. Arrange the dishes so that they are not likely to be heated, by direct sunlight or otherwise. If necessary the dishes should be cooled by running water around them. Provide a means of darkening the dishes, such as cardboard boxes that can be inverted over them, and means of illuminating the colonies, such as a desk lamp. Keep some colonies where the natural illumination of day and night can reach them and no artificial light at night can. Look for evidence of spawning every few hours during the day, especially in the early morning, beginning with the first daylight. Keep other colonies under conditions of continuous dark and still others under continuous illumination for more than 24 hours, then look for eggs that have been shed. Two further groups should be subjected to alternating light and dark periods of about two hours each, beginning in the one case with colonies that have been twelve hours or more in the dark, in the other with colonies that have been twelve hours or more in the light. Observe them at intervals of fifteen minutes or less while in the light. The results of these experiments should "rehearse" the phenomenon and further tests to determine more precisely the adequate stimulus for shedding should suggest themselves. Set up what you regard as the appropriate tests, on paper, carefully defining in each case what the question is that you are trying to answer and then carry out any of them that are feasible with the time and facilities available.

Write up your experiments, observations and interpretations. This need not take much time but should be as carefully and thoughtfully done as possible. Preferably after you have finished your work, consult Ballard's paper.
C. Feeding behavior in Anthozoa

Primitive and sessile forms like anemones exhibit little overt behavior. But the study of the organization of the nervous system requires the observation of responses to stimuli. The responses to food include a large share of the reactions of which animals like these are capable and when analyzed often turn out to be surprisingly complex. The experiments suggested here on feeding are closely related to those outlined under IV-I, K and M. The student is urged to consult appropriate sections of Parker's "The Elementary Nervous System" and Panton and Panton, 1943, J. Exp. Biol., 20.

A number of large specimens of *Metridium* attached and expanded in large dishes of sea water should be secured. Arrange them so that running sea water may be turned on when the specimens are left for long periods to relax or to recover from overfeeding and so that the running water may be turned off and observations made without moving the dishes. The dishes should be just deep enough so that the upper surface of the anemone when expanded is an inch or two under water.

Elicit the feeding reaction in a normal way by dropping pieces of animal food (fish, clam or crab meat) onto the oral disk of an expanded anemone that has not been fed for twelve hours or more. Turn off the running water when making experiments. Observe the entire course of the response closely; write down each successive part of the sequence, note lengths of time for each, consider what muscles or other effectors must be involved in each and possible hypotheses of the activation of the respective effectors. (For example can the effector be an "independent effector", directly stimulated by the food, is there a local reflex involving sensory and nervous structures, is there a non-local reflex and what could be its path, is a nerve net involved and what properties of the conducting or coordinating system are displayed.) Repeat the observation of the normal reaction several times, in the same and different specimens to distinguish the constant features from the variable. Discover by plying one individual with food as fast as it will be accepted what the alterations in the reaction as a result of overfeeding are. What do you think is the cause of the change? Test your hypothesis, for example repeat the whole experiment with a fresh animal but removing the piece of food each time just before it is taken in. Invent other experiments which will give information on the mechanism of the normal response and the change with overfeeding. For example, ply an animal with food until it is clearly overfed but always allowing the food to touch only one side of the oral disk, then test the other side. Repeat without allowing any actual ingestion.

Squirt plain sea water with a pipette onto the mouth of a fresh animal, is there any response? Repeat with weak acid (M/20 HCl). Is the response essentially the same as that to food? Now stimulate in
the same ways the outer tentacles without allowing any of the food or
acid to reach the mouth, observe the response of the mouth. Can you
draw any conclusions about the conducting system. Variations of this
experiment and combinations of it with cuts in the oral disk will
suggest themselves.

What do you think is the adequate stimulus for the feeding
reaction? Set up various hypotheses and test them. For example, test
the response to mechanical stimulation of various kinds, stimulate one
tentacle alone, more than one - close together and far apart, many
tentacles, other parts of the body. Even if there is not a feeding
reaction carefully note what response is brought about, significant
conclusions about the plan of the nervous system can be drawn from
these tests. Compare the result of the same mechanical stimulus in
individuals that have not long since fed, but are not surfeited, and
ones starved for 24 hours or more. Do you think there is a real quali-
tative difference in the response to inert objects and food or are
there all grades between the response of a starved animal to an inert
object and that of a surfeited animal to food? Compare the response
to an inert object (piece of cotton, sponge or filter paper) in sea
water and after stirring into the dish a little clam juice. Compare
the response to the same inert object soaked in sea water and soaked
in food juice.

Test various chemicals or mixtures in liquid form using the
same kind of inert object to carry the liquid. Thus, try soaking
cotton in clam mucus, in clam or other blood free of contamination with
mucus, in mucus from anemones of the same species; try tentacles cut
off from anemones of the same species, the same after boiling; test
human saliva in various dilutions, bread, pure protein. If time permits
other substances may be tested and fairly definite conclusions as to
the class of chemicals that are responsible for normal stimulation may
be drawn; see Pratin and Pratin, 1943. Interested students may attempt
to duplicate these responses with electrical stimulation in order to
explain the difference between mechanical and chemical stimuli and to
harmoise the mechanism of the feeding reaction with the general
physiology of the anthozoan nervous system as worked out in IV-K and M.
A stimulator which can deliver pulses brief enough to act as single
stimuli, setting up single impulses in the nervous system at a time,
and repeated at low frequencies (down to one in ten seconds) is essential.

Write down every observation you make, as you make it. This is
particularly important in experiments like these where so much individual
variation occurs. Include, but do not rely solely on, your general
impression of the typical or most characteristic result of any given
stimulation; the detailed, objective record of each individual experiment
often reveals unexpected significance when reviewed in the light of later
experiments. Draw conclusions or state what possibilities are ruled
cut or what alternatives seem to be left and define the problems that
remain or are created by findings so far; devise experiments that seem
to you possible and appropriate to answer some of these questions, even
though you cannot do them.
D. Feeding behavior of medusae

The best forms for the experimental study of reactions to food in medusae are not regularly found at Woods Hole, but a general impression of the mechanism of obtaining food in these forms based on observations of Cyanea and Aurelia is very worthwhile. If Physalia is available it should be studied as exemplifying a quite different type of mechanism with, however many fundamental similarities. See IV-A.

Attempt to obtain a complete feeding reaction in Cyanea and/or Aurelia by placing pieces of animal food in contact with the oral arms. If no response is obtained in medusae that have not been fed for 24 hours or more, try paralyzing a medusa in order to stop the swimming movements and make possible more prolonged contact of the food with any one spot. Cut out each of the eight marginal bodies and place the preparation oral side up on the bottom of a shallow dish. Again attempt to elicit a feeding reaction by placing food on the oral arms. Look closely for micro-responses, i.e., ciliary, nematocyst and local muscular responses. If a fairly reproducible reaction is obtained (look out for sensory adaptation of "fatigue") try placing the food in contact with other parts of the animal. Try mechanical stimulation with inert objects and chemical stimulation with liquid food. Test for cooperation of the oral arms and the nervous system of the bell by running a knife around close to the base of the arms and through the subumbrellar neuro-muscular sheet; is the food reaction altered?

Is the response essentially different when the oral arms are cut away from the body or a piece of one arm isolated? What is the significance of neuro-muscular autonomy? Test isolated mouth arms of Aurelia or small fragments of Cyanea mouth arms with mussel or clam juice, egg white, mucus from various sources, bread, sugar solutions and other substances as time or facilities permit. Grade the responses on some rough quantitative basis, as one plus, two plus and three plus and compare the effectiveness of these chemical stimuli, straight and in various dilutions, using a number of specimens and as comparable conditions as possible. Consult Hensel, 1935, Wiss. Meeresuntersuch., Abt. Kiel, N.F. 27.

Record your observations as carefully as you can and make as many deductions as they permit. Consider the general significance of the properties of the nerve-muscle system exhibited in this form of behavior. The interested student is urged to read appropriate sections of Romanes "Jellyfish, Starfish and Sea-urchins".
Behavior of a ctenophore

Besides possessing a number of interesting features peculiar to itself, the ctenophore nervous system has been given extra significance by phylogenetic speculation such as the orthogonal theory of Henstridge. Two forms of response are here especially suitable for study. These are ciliary activity and luminescence.

Observe carefully the activity of the comb plates in an intact specimen of Mnemiopsis. As you watch it, write down a list of the characteristics of this activity (times, directions, coordination, variability, etc.) and of the questions these raise in your mind. Devise your own experiments to answer as many of these questions as you can. The following suggestions are only offered to supplement your own ingenuity.

Do the successive waves that pass down the plate rows from the apical body depend on a spontaneous rhythmic center there for their initiation? Cut out the apical body or isolate an oral lobe with its comb plate rows to see whether spontaneous waves will be initiated in them. Isolate as small a piece of plate row as you can and look for spontaneous activity. What do you think is the normal pacemaker and how are the waves coordinated? Cut across a row, leaving it in place. What happens to the activity on the two sides of the cut, in the first few seconds, after some minutes, after several days and after many hours? Try this on several specimens, at different levels and in isolated oral lobes or smaller pieces.

Do you think the wave may be propagated by the mechanical stimulus each plate contributes to the next when it beats? In an isolated row or oral lobe arranged for close observation stimulate locally by touching a plate row and notice the sinking in of the row beneath the surface as though for protection. Notice whether the plates thus covered over still beat and whether the wave is propagated beyond this region. By very careful manipulation it is possible to hold one or more plates still with fine needles and then to look for propagation of the metachronal wave across the immobilized region; if this is not easily accomplished try stropping a few plates by applying a few strands of cotton, holding them across a row. Is the continuity of the jelly or of any nerves that may be in it necessary for the propagation of the metachronal wave? Cut through as much of the jelly as possible in an isolated oral lobe, leaving as little continuity as you can between the two ends of the row; this must be done carefully so as not to strain the row. How fast does the metachronal wave travel? Compare this with the speeds of other kinds of conducted waves of excitation.

Look for general reflex responses of the whole plate row system. In other species ciliary arrest and reversal of beat are common and conspicuous features of this system, but in Mnemiopsis reversal is very
difficult to elicit. Squirt a small quantity of clam juice or the like onto the suricles of an intact specimen while watching the plates closely; look for a prompt but very transient response of the whole plate system. Notice what the animal does when it bumps into an obstacle. Does it ever change the direction of progression when swimming free in a large aquarium?

Attempt to demonstrate a difference between intact animals and preparations with the apical body removed as respects orientation to gravity.

At night or after some hours in a dark room look for luminescence in Mnemiopsis. Does it occur spontaneously? What is its character, location and pattern of response to stimuli? To what kinds of stimuli in what parts of the body will this response occur? How local or general is the response in each case? What kinds of cuts through the ectoderm alone and through the jelly will circumscribe the response, i.e., what is the simplest operation that will confine the response? What kinds of cuts can the response circumvent or have no effect on it? What happens to the ability to luminesce when the animal is exposed to light? Try various intensities or times of exposure and test for luminescence after varying periods in the dark. Try light of different colors obtained by means of filters of known transmission, compensating for lowered intensity. Speculate on the function of this elaborate mechanism in the general economy of the organism.

Consult the appropriate sections in Parker. A bibliography is available for those interested. Record your observations and interpretations as carefully and fully as possible.

F. Regeneration in a hydroid

Many hydroid polyps offer very favorable material for study of regeneration, especially such topics as polarity, dominance and factors controlling differentiation.

Cut stems from a "colony" of Tubularia, sort in a finger bowl selecting healthy looking smaller stems, about 10 mm. long. With sharp scissors cut off the hydroids a short ways below the base. Handling the stems with a medicine dropper transfer to watch glasses closed inside large dishes with running sea water. Use a number of stems for each experiment.

Prepare pieces of stem from various levels and of various lengths. Compare the speed as well as the character of regeneration. In some pieces tie a ligature around the stem at a chosen level so that the continuity of the coenosarc is interrupted. Look for signs of regeneration after 30 hours or so.
Organize your study into definite experiments designed to answer definite questions, plan controls for each group. Record your results, telling what each experiment was designed to test, and your interpretation of the results. Use simple sketches wherever appropriate. Consult Berth, 1940, Biol. Rev., 15 and other papers referred to there.

G. Regeneration and grafting in Mnemiopsis

Regeneration in ctenophores - and medusae - is especially suitable for the study of differentiation of more complex structures and repair of the nervous system, as opposed to Tubularia. These animals also lend themselves to experiments on grafting, pieces from different individuals may be held together in various unnatural positions and after a few hours will "take" and eventually achieve true organic continuity.

Cut across plate rows in intact individuals and in isolated rows or oral lobes end at various levels; look for recovery of synchronism of beat of the plates on the two sides of the cut. Cut out a piece of a row including several plates; watch for the formation of new plates and for evidence of conduction across the gap. Repeat with larger gaps. Cut out the apical body or cut off the entire aboral end and describe the steps by which the organism reorganizes. Cut off the oral end; divide animals in two in each of the meridional planes; cut off both oral and aboral ends. Other kinds of operations will occur to the interested student and should be tried. Keep the animals or pieces in uncrushed large dishes or in slowly running sea water, in the former case change the water at least daily.

Cut two individuals meridionally but just to one side of the midline so that two pieces are obtained which have apical body and midline structures and two which do not. Graft each of these pairs together by arranging a few strands of cotton across the pieces so that they are immobilized and held together for a few hours, until they tend to stick together; look for evidences of the two pieces becoming effectively one organism, and compare the performance of the two differently constituted pairs. Try grafting an extra apical body into normal animal in any convenient position; look for competition of the new and the old apical bodies. Graft pieces cut equatorially - two aboral ends, two central pieces with neither aboral nor oral ends. Try other combinations that occur to you.

Describe in detail each experiment, its results and your interpretations, distinguishing between the more limited, carefully stated necessary conclusions ("this must mean...") and the more general, tentative, possible hypotheses ("this may mean..."). Consult Conklin, 1937, Biol. Bull., 72, 72, 73, Proc. Nat. Acad. Sci., 23, and 1936, Biol. Bull., 71.
H. Structure and types of nematocysts

These intracellular organelles are diagnostic of coelenterates. They are remarkable for the high degree of morphologic specialization in such very small structures. Numerous distinct types occur, with characteristic distribution among species and within the individual. They are of great taxonomic significance, of great functional significance to the coelenterate, and present a number of interesting problems to the biologist: the cytologic development, mechanism of discharge, stimulus to discharge and possible nervous control, the evolution and function of such a variety of types, the chemistry and pharmacology of the potent toxins they carry.

The study of the structure and types of nematocysts will require (1) a good microscope with oil immersion lens, good light and a real understanding of how to use the microscope, (2) ingenuity in obtaining and preparing nematocysts for study. Detailed directions cannot be given here, but some suggestions may be made. Explore as wide a range of species and groups and as many parts of each animal as possible. Prepare small pieces for the observation of undischarged nematocysts. Try various ways of discharging them - not all types are discharged by the same treatment. Some agents to use are dilute acid or base, dye solutions, fresh water, mechanical stimulation, pressure food. Dehydrate and clear where possible, studying in balsam or etc. under the highest power. Consult Hymen for general description, classification of types and references.

I. The excitation of nematocysts

Nematocysts are apparently independent effectors, containing within themselves very specific receptors. The analysis of the adequate stimulus for discharge under normal conditions has proven very difficult, many workers contributing observations which did not seem to fit into a consistent picture. Pure mechanical stimulation does not induce any considerable discharge, many chemicals are powerful exciters of nematocysts but natural food juices do not cause spontaneous discharge and many very effective solid foods are water-insoluble. Only recently has the problem been illuminated by the excellent work of Pentin (1942, J. Exp. Biol., 19) which the student is encouraged to read.

Using the tentacles and acontia of Actidium observe the discharged nematocysts of two types by touching a cut off tentacle or acontial thread to a cover slip on which saliva has been spread and dried. Stain some such slips with methylene blue and others with acid fuchsine.
Arrange an excised tentacle under the microscope so that its edge is seen in profile. Use low power. Touch the tentacle gently with a human hair. A good discharge of nematocysts should be seen, entangling the base of the hair so that the tentacle tends to cling to the hair as it is moved. Touch in the same way with a chemically clean glass rod, drawn out to a conveniently fine diameter, (Even handling the glass will provide enough chemical stimulus to spoil the results.) Avoiding mechanical stimulation of any kind, even rubbing against the glass slide or dish, allow pure food juices to come in contact with the tentacle (clam mucus, meat juice free of particles, or etc.). If carefully done neither of these pure forms of stimulation will excite any considerable discharge.

Using very small wisps of cotton, test the effectiveness of the following three types of stimulation in causing nematocysts to entangle and adhere to the cotton: (1) cotton wet with sea water, (2) wet with a food juice and (3) wet with a 1% solution of bile salt in sea water. The cotton is not "held" by the first because no nematocysts are discharged nor by the last because they are all discharged by the powerful chemical exciter before the cotton is close enough to be entangled. This experiment underlines the importance in the normal mechanism of the timing of the discharge; it is useless if set off by diffusible substances that reach the cells before the solid object is close enough to be shot by the tiny threads.

Set up two tentacles from the same anemone under the same conditions but in two separate depression slides. Provide one with a medium of sea water the other with a medium of saline: sea water, 1:20, or with particle-free clam juice. Stimulate with the chemically clean glass rod and observe the great difference in the result. The natural chemical stimulus does not discharge the nematocysts but it greatly lowers the threshold to mechanical stimulation. Correct timing is thus automatically ensured.

Time will probably not permit extensive testing of a number of chemicals or the demonstration of the properties of the actual substances in food that sensitize nematocysts to mechanical stimuli. In general the chemicals that have any effect on these organelles may be grouped into (1) those that produce violent discharge, not inhibited by Mg (the excitability of nematocysts like that of other sensory structures is depressed by excess Mg in the sea water), (2) substances that cause extrusion of undischarged nematocysts, (3) substances producing discharge, inhibited by Mg, and (4) substances that cause no visible change but greatly increase the sensitiveness to mechanical stimuli. The first three groups are all chemicals that are not normally encountered by the coelenterate, the last group includes some unnatural substances and the unknown natural chemical. Pentin’s work makes it likely that the active substance in natural food is not pure protein or carbohydrate but something behaving like a surface active lipid which is strongly adsorbed onto protein (it
will not pass a collodion membrane and is non-extractible with ether, but extractible with alcohol. It is thus not the same as the chemical probably responsible for activating the feeding reaction of the tentacles and oral disc (protein) but is very closely associated with that substance. Note that it is essentially non-diffusible and does not reach the nematocyst through the water. Dry skin, insoluble foods, well washed keratin, etc. very effectively discharge nematocysts and many foods do not even sensitize nematocysts in the neighborhood of those actually in contact with it. It seems likely that the surface active agents are directly conveyed by contact and act on the cell membrane of the nematocyst. There is evidently considerable specificity in the effective agent for nematocysts commonly discharged by many but not by certain of the normally encountered organisms in the coelenterate's environment.

Attempt to obtain discharge at a distance from the point of stimulation. Using the human hair as an effective local stimulus, look for response a short distance away. Test the state of the nematocysts in a saturated anemone: ply an anemone with food, always giving it to one side of the oral disc and avoiding any stimulation of tentacles on the other, till food is not accepted any more. Then test the responsiveness of nematocysts on excised tentacles from the two sides to normal stimuli. What is the significance of your observations?

Write up your experiments, results and interpretations as carefully as you can.

J. Structure of mesogloea in different groups

Most of the structural differentiations in the mesogloea of coelenterates and the mesenchymatous jelly of ctenophores have not yet with certainty been identified. The microscopic appearance is very different in different groups. It is worthwhile to examine and describe the visible elements in the jelly even though very little can be definitely labelled as yet. Using fresh, living fragments examine under a variety of lighting conditions, with and without such vital dyes as are available and determine its appearance and formed inclusions in various regions and its relation to ectodermal and endodermal structures covering it. Compare Aurelia, Clyene, Nematocidae and hydrooids, including hydromedusae. Make simple sketches and describe as fully as possible what you can see.
K. The nerve net: experiments on sea anemones

The properties of the nerve net and its plan of organization may well be studied in two typical cases, in the anthozoan Metridium and the scyphozoan Aurelia. The experiments of Parker, reviewed in "The elementary Nervous System" and those of Pantin, 1935, J. Exp. Biol., 12: 139-155 form a good basis for demonstrating the plan of the nervous system in Metridium. Consult these two sources.

Obtain several large specimens of Metridium, attached in separate dishes. Determine by trial the normal response of a fully expanded, not recently stimulated anemone to stimulation of various parts of the column, oral and pedal discs by simple contact with a glass rod and by series of single electric shocks repeated from once in five seconds to five times a second. Compare the sensitivity of different regions, notice that the same muscles can be called into action from many points, learn what the range of different responses of which the anemone is capable and how to produce them. Already at this point several significant generalizations about the nervous system can be made.

Choose a well attached anemone and with a sharp knife cut into the pedal disc in such a way as to produce a spiral strip about 5 mm. wide extending half way or more around the base, free at one end and still attached to the anemone at the other. Allow the anemone to rest and expand, then stimulate at the free end of the strip or tongue. Note that you are forcing the impulse to take a certain route and by making such cuts in various positions something can be said about the pathways that must exist. Make cuts in the body wall so as to free tongue shaped strips in both horizontal and vertical directions and test similarly. Is conduction in each direction possible, is there any difference?

The question whether nerve fibers cross the mesoglea from ectoderm to endoderm has never been satisfactorily answered anatomically, i.e., by microscopic examination, but physiologically a clear answer is possible. Cut through the ectoderm and wall into the body wall in a horizontal circle, right around the column of an anemone, interrupting all possible paths from the ectoderm of the pedal disc over the oral disc and into the endoderm through the mouth. Stimulate the edge of the pedal disc and look for contraction of the longitudinal mesenteries.

Cut anemones almost into two by vertical incisions that leave only a bridge of tissue in one place or another - oral disc, pedal disc, column. Test for conduction of excitation from one half to the other. In a preparation with only the pedal disc attaching two halves, note the reaction time as carefully as possible without special means and then cut through all the mesenteries on one side, forcing the impulse to travel in the ectoderm of the column. Test for conduction and compare the reaction time with the previous. By this last means, somewhat refined, Pantin has worked out the specialized conduction tracts in the anemone.
Write up your experiments, their results and your conclusions from them. This is a very important part of the values to be had and should be done carefully even though briefly and not elaborately.

L. The nerve net: experiments on medusae

Use Aurelia if available, Cyanea if necessary. Compare the properties of the intact medusa, the preparation with oral arms removed and the preparation with oral arms and marginal bodies cut out. For stimulation use single electric shocks or series of single shocks repeated from one in ten seconds to once per second (single induction shocks, make or break shocks, or condenser discharge shocks may be used). Cut the center out of the bell and remove the oral arms of a medusa, thus making a doughnut preparation, then cut through any radius making a strip preparation. Remove all the marginal bodies but one at one end of the strip. Make about four radial cuts from the inside 2/3 or the width of the strip, and four more from the outer edge 2/3 of the way to the inner edge and half way between the first four so that the cuts interdigitate. Is the excitation wave able to make its way around all these cuts? Use either the electric shock or the spontaneously initiated impulse of the marginal body. Increase the complexity of the path by further cuts; these may be through the subumbrellar neuromuscular sheet only in order to leave the jelly to support the preparation. Remove the last marginal body and stimulate at various points along the strip to demonstrate that the impulse can travel in any direction through the nerve net. Start with a fresh doughnut preparation and cut spirally about three times around the bell to make an orange peel preparation. Can the excitation wave circumvent this severe degree of interruption of conduction paths?

If Cyanea is the only form available the following experiment should be done first. Make a doughnut preparation, cutting just inside the circular muscles. Convert this into a strip preparation. Make interdigitating cuts across the strip but placing them carefully with relation to the radii, not any place as in Aurelia. In one preparation place the cuts which extend from the inside towards the margin in the radii of the radial muscles or in every other such radius, and the cuts which extend from the outer margin towards the inner in any radius between the first cuts. In another preparation make the first set of cuts in the radii between radial muscle bands and test the conductivity of the strip before making a second set of cuts. What can you conclude about the distribution of the nerve net in this species? The other experiments, except the spirally cut preparation, can be done on Cyanea if this distribution is kept in mind.

Make a doughnut preparation and lengthen the conduction path around the circle by a series of interdigitating cuts part way across its width. The preparation should have no marginal bodies and no spontaneous contractions. Stimulate at one point and observe the two exci-
Tetanic waves start from that point, travel in opposite directions around
the circle and meet at the other side, extinguishing each other because
each finds the region of nerve net in front of it refractory. Repeat
with tetanic stimulation maintained for a quarter of a second or so,
very the strength from just threshold to many times threshold. Watch
the result of each stimulus. Usually sooner or later, under conditions
that are not understood, the two waves will be asymmetric and one will
not be blocked by the other. There is nothing to prevent this wave from
completing the circle and starting around again. An entrapped circuit
wave is thus initiated and will continue to pass around the circle many
times. What is the significance of this behavior? What can stop the
wave? Count revolutions per minute and determine whether the rate of
conduction changes significantly or systematically (temperature readings
are necessary to rule out changes due to this factor).

Cut very small pieces out of the bell of a medusa. Test for
their ability to respond to stimuli. If stimuli near threshold strength
are used it is probable that any responses are true reflexes, mediated
through the nervous system. What is the significance of your observa-
tion, from a specific anatomical point of view and from a general biologi-
cal point of view?

The interested student is urged to read portions of Hommes' "Jellyfish, Star-fish and Sea-urchins" and Perker's "The Elementary
Nervous System". Write up your experiments, results and interpretations;
use sketches and diagrams wherever possible.
KEY TO THE MORE COMMON HYDROIDS OF THE
WOODS HOLE AREA.

Key to SUBORDERS OF HYDROIDA

I. Hydranth without a protective cup (theca) into which it can be
retracted (although perisarc continues over hydranth to base of
tentacles in Bougainvillea.)

GIMNOBLASTEA

II. Hydranth with a protective theca into which it can be retracted
(although the cup is too reduced to cover the hydranth in
Halecium.)

Calyptoblastea

Key to FAMILIES and GENERA of GIMNOBLASTEA

I. Hydranths with scattered tentacles only, not arranged in a
whorl.

IA. Tentacles thread-like (filiform, scattered on distal
part of hydranth. (Cleavidae)---------------------

IA1. Colony branched.

1. Cordylophora

IA2. Colony unbranched, zooids arising directly
from the creeping hydrorhiza.

2. Clava

IB. Tentacles knobbed at tip (capitate), scattered
over whole hydranth. (Corynidae)--------------------

3. Corynitis

II. Hydranths with some or all tentacles in a whorl around
base of hypostome.

IIA. Hydranths with distal tentacles in addition to
the basal whorl.

IIA1. Hydranths with scattered capitate tentacles
distal to the basal filiform whorl. (Pennaridae)---

4. Pennaria

IIA2. Hydranths with distal and basal whorls of
filiform tentacles.

IIA2a. Zooids usually colonial; perisarc
well developed; medusa buds not
liberated. (Tubularidae)---------------------

5. Tubularia
IIA2b. Zooids solitary; perisarc weak or absent; free medusae liberated.

(Corymorphidae)------------------

6. CORYMORPHA

IIB. Hydranths with basal whorl of tentacles only.

IIB1. Colony erect and branching, bush-like.

(Bougainvillidae)------------------

IIB1a. Perisarc continued over hydranth to base of tentacle; hypostome conical; free medusae liberated.

7. BOUGAINVILLEA

IIB1b. Perisarc ends below hydranth; hypostome trumpet-shaped; gonophores are sporosacs. (Eudendridae)------------------

8. EUDENDRIUM

IIB2. Colony encrusting, zooids arising directly from the hydorhiza.

IIB2a. Hydrorhizal spines rough. Tentacles of gonozooids reduced to rudiments; gonophores are sporosacs. (Hydractinidae)

9. HYDRACTINIA

IIB2b. Hydrorhizal spines smooth. Tentacles of gonozooids well developed; free medusae liberated. (Podocorynidae)------------------

IIB2b (1). No spiral zooids; medusa degenerate, and with ripe gonads when liberated.

10. STYLACTIS

IIB2b (2). Spiral zooids occur; medusa with well developed tentacles, and unripe when liberated.

11. PODOCORYNE

Key to FAMILIES and GENERA of CAMPYTOBLASTEA

I. Hydrotheca supported on a pedicel.

IA. Hydrotheca large enough to enclose hydranth.

IAl. Hydrotheca bell shaped; open at distal end.
IIAla. Colony unbranched or not regularly branched; hydrothecal rim toothed; gonothecae ringed.

12. CLYTIA

IIAlb. Colony regularly branched (in the terminal twigs at least); hydrothecal rim untoothed (except Obelia bicuspida); gonothecae not ringed.

IIAlb (1). Gonophore a sporosac.

13. CAMPANULARIA

IIAlb (2). Gonangia liberating free medusae.

14. OBELIA

15. CALYCELLA

IA2. Hydrotheca tubular; distal end closes into a cone over retracted hydranths by means of long hinged tooth-like flaps. (Campanulariidae) -----------

16. HALECIUM

II. Hydrothecae sessile, without pedicel.

IIB. Hydrothecae placed on both sides of stems.

(Sertulariidae) ---------------

IIB1. Hydrothecae arranged in opposite pairs.

17. SERTULARIIDA

IIB2. Hydrothecae of the two sides of the stem alternate.

IIB2a. Hydrotheca bottle-necked, its rim smooth.

18. ABIETINIRIA

IIB2b. Hydrotheca not bottle-necked, its rim with two opposite teeth.

19. THUARIIDA

IIB. Hydrothecae placed on one side of stem only.

(Fluminulariidae) ---------------

20. SCHIZOPTRICHA
Alphabetic List of GENERA, with Key to or

Description of SPECIES of each.

(Number following name of genus gives its position in generic key).

Abietinaria (18). A. ABETINA, in deep water. Branching of colony poundly feather-like (pinnate); gonothecae as small as hydrothecae, placed in rows on upper sides of branches, with acrocyst; gonophore sessile. Horn-colored, height to 12 inches.

Bougainvillea (7). B. CAROLINENSIS, on rocks, algae and pilings. Perisarc, dense, horny; gonophores are free medusae, budded singly from branches and pedicels. Perisarc greenish, hydranth reddish; height 2 to 8 inches.

Calycella (15). C. SYRINGA, Creeping over other hydroids, bryozoa, algae. Unbranched; hydrothiza linear; gonosome with acrocyst; gonophore sessile. Perisarc dark horn-colored; height 1/8 inch.

Clava (2). C. LEPTOSTYLL, on algae and under stones. Gonophores are sporosacs clustered below tentacles of hydranth. Color reddish, height 1/4 inch.

Campanularia (13). Four common species. Difficult to distinguish from Obelia and Clytia except by gonophore, which is sessile.

I. Main stem bearing many branches, which themselves branch

C. AMPHORA, in shallow water. Gonothecae on the hydrocaulus, oval, slender, with very small apertures at tip. Height, 6 inches.

II. Stem bearing pedicels only, or a few irregular branches.

IIA. Stem stout, its internodes (between successive pedicels) short, only four to five times as long as thick.

C. FLEXUOSA, on algae, rocks, pilings. Gonothecae on the hydrocaulus, large tubular, with truncate tip. Stem pale horn-color; height 1½ inches.

IIB. Main stem slender, its internodes long, seven to eight times as long as thick.


C. ANGULAT., on eelgrass; height 3/4 inch.

IIB2. Stem only slightly zig-zag (flexuose). Gonangia on the hydrocaulus, with the tip peculiarly folded over.

C. CLICOLIFER, on Mytilus and pilings; height 1 inch.
Olyria (12). Two common species. Medusae with only 4 to 8 tentacles when liberated.

I. Colony unbranched or sparsely branched; rim of hydrotheca with about 16 rounded teeth; gonangia deeply and regularly ringed (annulate).

C. JOHNSTONI, on rocks and algae. White; height 1/2 inch.

II. Colony branched profusely but without well defined main stems; rim of hydrotheca with about 12 to 14 sharp teeth; gonangia irregularly annulate.

C. EDWARDSI, on piles. White; height 1 inch.

Cordylophora (1). C. LACUSTRIS, in brackish-water ponds. Gonophores are sporosacs borne singly on branches and bases of pedicels. Height 1 inch.

Corynitis (3). C. AGASSIZII, on shells and pilings, associated with encrusting Bryozoa. Gonophores are free medusae (Gemmaria remmusa) budded in a cluster near base of hydranth. Pink; height 1/3 inch.

Corymorpha (6). C. PENDULA, in deep water. Gonophores are free medusae (Hydrocoodon pendula) budded in bunches (racemes) above proximal tentacular whorl of hydranth. Pink; height 1 to 4 inches.

Eudendrium (8). Four common species. Male gonophores are strings of sporosacs radiating from gonozoids, which may be ordinary hydranths or mere blastostyles. Female gonophores are spherical sporosacs budded from gonozoid or pedicel.

I. Main stem compound (fascicled); colony regularly branched.

IA. Gonozoids not completely reduced to blastostyles; male gonophores, 2 or 3 chambered.

E. RAMOSUM, on rocks and piles, and in deeper water. Hydranths and male gonophore red, female gonophore orange; height 4 to 6 inches.

IB. Gonozoids aborted to blastostyles; male gonophores 4 or 5 chambered.

E. CARNEUM, on piles and algae. Red; height 2 to 5 inches.

II. Main stem not compound; colony irregularly and sparsely branched.

IIA. Gonozoids not completely reduced to blastostyles; male gonophores 2 to 3 chambered.

E. ALBUM, on piles, rocks, algae. White, male gonophores yellow; height 1 inch.

IIB. Male gonozoids completely aborted to blastostyles, the gonophores 4 or 5 chambered; female gonophores scattered on pedicels and caulus as well as hydranth body.

E. TENUE, on piles. Bright pink; height 1/2 to 1 inch.

Macleocium (16). E. TALENTINUM, on piles and shells and stones in shallow water. Colony pinnately branched, fan-like;
stem fascicled; gonangia in rows on upper side of branches, the female ones shouldered, the male slender; gonophores are sessile. Height 6 to 10 inches.

Hydractinia (9). Echinata, usually on shells of the snail Littorina inhabited by hermit crabs, widespread. Special defensive zooids, without tentacles; gonophores are sporangia on reduced zooids. Pale pink, female gonophores red; height 1/4 inch or more.

Obelia (14). Three common species. Medusae with 12 or more tentacles when liberated.

I. Main stem compound (fascicled); hydrothecae deep, ribbed, the rim with 14 to 20 double-pointed teeth; gonangia not bottle-necked.
   0. EIOUSPIDATA, on seagrass and piles and in deeper water. Gonangium smaller than hydrotheca, tip broad and truncate. Height to 33 inches.

II. Main stem simple; hydrothecae shallow, smooth, toothless; gonangia bottle-necked.
   III. Main stem usually unbranched, giving off regularly alternating pedicels; internodes of stem short and thick, about three times as thick as pedicels.
   0. GENICULATA, on algae and piles. Gonangia very large, about five times as long as hydrotheca. Height 1 inch or less.

IIIB. Main stem with many compound branches; internodes or terminal branches moderately long and slender, twice or less as thick as pedicels.
   0. COMMISURATA, on shells, stones, seagrass, rocks, piles. Gonangia of moderate size, about three times as long as hydrotheca. Height 6 to 8 inches.

Pennaria (4). F. TIARELLA, on piles, rocks, seagrass. Branching of colony feather-like. Gonophores are reduced but free medusae budded singly above proximal tentacles of hydranth. Pericarce dark, hydranths pink or red; height to 6 inches.

Podocoryne (11). F. GINN, usually on shells of the snail Nassa trivittata inhabited by hermit crabs, on sand or seagrass. Special defensive zooids, without tentacles. Gonophores are free medusae budded below in a cluster tentacles of gonozoids. White or pale pink, gonophore reddish; height ½ inch or more.

Schizotricha (20). S. TENELLA, on piles. Colony dichotomously branched, plume-like, delicate; small special zooids present, in trumpet-shaped thecae; gonothecae large curved corncupias; the gonophores are sessile. White; 1 to 3 inches high.

Sertularia (17). S. FUMILLA, on rocks, algeas, piles, seagrass. Colony branched or not. Members of the pairs of hydrothecae not in contact. Gonotheca bottle-necked, not annulated, with acrocyst. Dark horn color; height ½ to 1 ½ inches.

Stylactis (10) S. ROOPERI, on shells of living Nassa obsoleta, on seagrass. Hydranths very slender. Gonophores are free but rudimentary medusae budded below tentacles of gonozoids. Pink (?); height 3/4 inch.
Thalia (19). T. ARGENTEA, in deep water; empty perisarc only found in summer. Branching of colony forked (dichotomous), forming a cluster of long bushy plumes; gonothecae larger than hydrothecae, with two spines at tip, and acrocyst. Stems dark, branches silvery; height 12 inches.

Tubularia (5). Three species. Gonophores are rudimentary medusae, not set free, borne in bunches above proximal tentacles of hydanth.

I. Colony branched; stems extensively annulated.
   T. LARYNGE, on piles and algae. Stem yellow, hydrothecae and gonophores bright pink; height 1 to 1 1/2 inch.

II. Colony branched sparingly or not at all; stems annulated only at intervals.

IIIA. Colony formed of unbranched clusters of 5 to 10 individuals; 30 to 40 tentacles in proximal whorl.
   T. COUTHOUYI, sandy and stony bottoms; dead in summer in shallow water. Stem and gonophores scarlet; height 5 to 7 inches, spread of tentacles 1 inch.

IIIB. Colony a dense, sparingly branched tuft; 20 to 25 tentacles in proximal whorl.
   T. CRASSA, pilings just below low tide, sometimes in brackish water. Stems pale, hydrothecae and gonophores rose; height 3 to 5 inches.

NOTE: Several of the species of hydromedusae common in the plankton at Woods Hole have hydroid stages which are uncommon or unknown. Of these the Gymnoblasts (Anthomedusae) are Bougainvillea superciliaris; Ectopleura ochracea (hydroid solitary but otherwise much like Tubularia); Nemopsis bachei (hydroid a Bougainvillea); Podocoryne fulgurans (which reproduces medusae asexually and may produce a hydroid stage infrequently or not at all). The Calyptoblastae (Lepto-

...
PLATYHELMINTHES - LABORATORY DIRECTIONS

First Laboratory Period - one-half day

Class: Turbellaria

A. Bdelloura candida: Observe this worm in its natural environment on the ventral surface of the horseshoe crab, Limulus. Note its general distribution on the host. Is the worm parasitic or commensal? What does it eat? Find the cocoons of the worm on the lamellae of the gill book; why are they deposited there?

1. The pharyngeal-probascis mechanism: Transfer a worm to a drop of sea water on a slide; add several drops of 7% ethyl alcohol and watch for pharynx protrusion. This protrusion usually occurs within a few minutes. Now add a cover-slip and examine the musculature of the pharynx under high power.

2. External ciliation (a taxonomic character): Add a drop of sea-water carmine suspension to a slide with Bdelloura and observe the water currents set up by the epithelial cilia. Observe an edge of the worm under high power to determine the manner of cillum movement.

3. Morphology: Place a medium-sized Bdelloura between two glass slides with vaselined edges and gradually compress until the worm is quite flat and quiet. Follow directions in Drow (p. 68-70). If necessary, for clearness, make separate diagrams of the different organ systems. These systems may be included in a single composite drawing. To determine details of the entire morphology may require a study of several specimens. Certain details of these systems are best seen in small, immature worms.

B. Optional: For students who have studied Bdelloura previously and for those who want to make additional comparative and embryological studies:

1. Make a comparative morphological and taxonomic study of several species of Turbellaria in the laboratory.

2. Study the eggs and developing larvae of Euplana gracilis. This polyclad is often found in abundance on ulva in the Bel Pond. Use Kükenthal on the front desk for reference.

Second Laboratory Period - one-half day.

Class: Trematoda

1. Adult Digenetic Trematode.


Obtain several specimens from the assistant or locate them for yourself from pieces of gull small intestine and mount on a slide.
If you locate other trematodes identify and save for observation under the next section. Vaseline the edges of the cover-slip and apply slight pressure until the worms are fairly well flattened. Watch a few minutes for egg discharge. As the worm flattens, body details may be observed more clearly. Beginning anteriorly, look for the following features: oral sucker, surrounding the mouth; short prepharynx; bulbous pharynx; narrow esophagus, which branches into two long intestinal crura (or ceca) that extend laterally to almost the posterior end; acetabulum (or ventral sucker), at about mid-body length, followed by the genital pore (the last two structures are slightly different from the condition in other trematodes; they are both invaginated from the surface and open into a common chamber—hence the name Cryptocotyle, which means "hidden sucker"); uterus filled with yellow eggs; dextral lobate ovary; median vitelline or yolk reservoir; dextral ovate seminal receptacle; two obliquely situated testes; median excretory bladder, extending as a slender tube to the posterior tip, emptying posterior at the excretory pore; the vitellaria or yolk follicles are located in the lateral fields, they extend from the posterior end to a point about midway between the acetabulum and the pharynx. Certain details of these organ systems are more easily seen after treatment of the living worm with dilute vital dyes. Try: neutral red, methylene blue, Bismarck brown, etc. Observe, also, stained permanent specimens for additional morphological data.

b. Optional:

1. The adult of Parorchis acanthus (= Parorchis avitus (Lin-ton) from the cloacas of piscivorous birds including the herring gull, Larus argentatus, the common gull, Larus canus, the common tern, Sterna hirundo and the roseate tern, Sterna dougalli is, in many ways, better for class room study than Cryptocotyle linlina. If P. acanthus is available it may be used in place of, or in addition to, C. linita.

Obtain a specimen of this worm and mount it on a slide in a small quantity of avian salt solution. Place a cover glass on the preparation and while looking through the microscope carefully withdraw the excess solution from the edge of the cover glass with a piece of absorbent paper; continue the withdrawal until the worm is flattened, quiet and relatively transparent. Now carefully vaseline the edges of the cover glass. The following parts should be identified and studied. At the anterior end is the oral sucker surrounded by a flared collar. Estimate, or count, the number of spines in the collar. The mouth which is in the center of the oral sucker leads back into a short prepharynx which, in turn, joins the muscular pharynx. The narrow esophagus runs posteriorly and shortly branches into the two, thin walled, narrow intestinal ceca (caecum). The large, cup-shaped acetabulum (ventral sucker) is between the anterior heavily cuticular spined region and the posterior more weakly cuticular spined area. The genital pore opens to the ventral surface immediately anterior to this sucker. From the pore the uterus and vas deferens pass posterior and finally connect to the median ovary and the testes. The testes lie side by side (Gr. par, beside; orchis, testis) and are irregular in outline. Contiguous to the ovary posteriorly is a small
Land which is also connected, by vitelline ducts, to the laterally lying vitellaria. The excretory system consists of a posterio-dorsally emptying, irregularly shaped, conspicuous excretory bladder which receives, antero-laterally, two much branched excretory trunks. The ultimate unit of the excretory system is the flame cell (solenocyte, terminal flame bulb) visible only under higher power. Note the eggs and free miracidia in the terminal part of the uterus. Each miracidium contains a single, fully formed redia and the life cycle is, therefore, without a sporocyst generation. Study the miracidium and understand its full importance.

2. Study and identify, if possible, any other living trematodes that you may find in the mouth, stomach, intestine, cloaca, or excretory system, etc., of the other vertebrate hosts available for parasitological examination.

Third Laboratory period - one-half day

Larval Forms of a Digenetic Trematode.

1. Redia. In a finger bowl at your desk are specimens of Littorina littorea infected with the larval stages of C. lingua. The largest mass of these larvae is usually to be found in the "liver" of the snail. Notice the texture and color changes between the uninfected snail liver and the infected one. Locate an undamaged redia and mount on a slide. Note the pharynx, short intestine, antero-lateral birth-pore, and the developing cercariae, ranging from undifferentiated germ-balls at the posterior end to mature cercariae near the pore may be observed. Draw a mature redia containing cercariae in various stages of development.

2. Cercaria.

b. Obtain a drop of sea water containing mature cercariae of C. lingua, add a drop of 1:10,000 solution of neutral red, and cover with a cover-slip. As the water evaporates the pressure of the cover-slip flattens and quiets the cercariae; the body details then become more distinguishable. Observe the general resemblance of the body of the cercaria to that of the adult fluke. Find oral sucker, pharynx, intestinal crura, penetration glands (how many and of what use?) with ducts leading forward to empty near the anterior tip, cystogenous glands (of what function?) scattered over the whole body surface, germinal mass, excretory bladder, eye spots, and tail. Note the distribution of the fin on the tail, and spines on the body. See part b. for physiological experimentation suggestions for use with this cercaria. Determine the usefulness to the cercaria of any adaptations you find.

b. Optional: Make a comparative study of the various cercariae in the laboratory after examining the cercaria of C. lingua. See part 3b. Determine the probable type of life-cycle of each form from their cercarial specializations. Note: tail size and motility, eye spots, reaction to light and shadows, swimming motions, etc.

3. Encystment of the Cercaria to form the meta cercaria:
a. The metacercariae of *C. lingua* are found in fishes (especially the Cunner). Add *C. lingua* cercariae to a fingerbowl containing a small Fundulus. Locate metacercariae in the fins a few hours later. Add a small piece of Cunner fin to a watch glass containing cercariae; observe and make a series of sketches showing the activities of the cercariae during penetration and encystment.

b. Optional: Other cercariae and hosts will be available for additional experimentation. The following table lists the trematodes from the Woods Hole area whose life-cycles are known. Expose the proper metacercarial hosts to the cercariae available and determine the methods of infestation. If time and hosts are available feed the infected metacercarial hosts to the proper adult host and reclaim the young encysted adult worms.

<table>
<thead>
<tr>
<th>Adult worm</th>
<th>Cercaria in *</th>
<th>Metacercaria in Adult In</th>
<th>See</th>
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</thead>
<tbody>
<tr>
<td><em>Zoogonus rubellus</em></td>
<td>Nassa obsoleta</td>
<td>Nercis</td>
<td><em>Fishes</em></td>
</tr>
<tr>
<td><em>Himasthla quissotensis</em></td>
<td>Nassa obsoleta</td>
<td><em>Nercis and bivalve molluscs</em></td>
<td></td>
</tr>
<tr>
<td><em>Lepeocreadium setiferoides</em></td>
<td>Nassa obsoleta</td>
<td>Turbellaria and Flat annelids</td>
<td><em>Fishes</em></td>
</tr>
<tr>
<td><em>Stephanostomum tenue</em></td>
<td>Nassa obsoleta</td>
<td>Menidia (liver)</td>
<td><em>Puffer &amp; B.B.</em></td>
</tr>
<tr>
<td><em>Gynacotyla hassicola</em></td>
<td>Nassa obsoleta</td>
<td>Talorchestia</td>
<td><em>Sandpipers</em></td>
</tr>
<tr>
<td><em>Cryptocotyle lingua</em></td>
<td><em>Littorina</em></td>
<td>Skin of fishes</td>
<td><em>Piscivorous birds</em>, '30 J. Morp*</td>
</tr>
<tr>
<td><em>Podocotyle atomon</em></td>
<td><em>Littorea</em></td>
<td>Crustaceans</td>
<td><em>Fishes</em></td>
</tr>
<tr>
<td><em>Opecoeloides Manteri</em></td>
<td><em>Columbella</em></td>
<td>Amphipods</td>
<td><em>Fishes</em></td>
</tr>
<tr>
<td><em>Zoogonoides laevis</em></td>
<td><em>Columbella</em></td>
<td><em>Nercis</em></td>
<td><em>Tautog</em></td>
</tr>
<tr>
<td><em>Parorchis acanthus</em></td>
<td><em>Urosalpinx cinereus &amp; Thais tapillus</em></td>
<td>Open</td>
<td><em>Gulls</em></td>
</tr>
</tbody>
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Stunkard ’38, B. B.
Stunkard ’38, B. B.
Stunkard ’38
Martin ’38
Stunkard ’40
Martin ’38
B. B.
Martin ’38
Hunninen & Cable ’43
Tr Am. Micr. Soc.
Hunninen & Cable ’41
B. B.
Stunkard ’42 J Paras.
Stunkard & Cable ’32
B. B.
| Speletrema nicolli | Bittium alternatum | Blue-crabs | Gulls | Stunkard & Cable '38 B. B. |
| Siphodera vinal edwardsee | Bittium alternatum | Flounders | Toadfish | Cable and Hunninen, '42 J. Parasit. |
| Deropristis inflata | Bittium alternatum | Nereis | Eels | Cable and Hunninen, '42 B. B. |
| Lecithaster confusus | Odostomia | Copepods | Sticklebacks and other fish | Hunninen & Cable '41 J. Parasit. |
| Monorchideidae cumingiae | Cumingia tellinoides Tenera | Siphons of cercarial hosts | Flounders and eels | Martin 1940 B. B. |
| Maritrema arenaria | Undiscovered Barnacles | Ruddy Turnstone Castle, '40 (shore bird) | |

* The species name used here is that given in the paper cited.

4. **Metacercaria.**

a. Study the metacercaria of *C. lingua* as it appears encysted in a Cunner fin. Carefully remove, with well-sharpened needles, a metacercaria from a cyst and study it. Note that except for maturity it resembles the adult worm; can you find any points of difference? What changes occur after the metacercaria becomes established in the final host?

b. Optional: The following experiments may be substituted for the metacercaria of *C. lingua*.

1) Remove and study other metacercariae obtained as a result of your experimentation under part 3 b.

2) Remove and study the metacercaria of *Maritrema arenaria* from the barnacle, *Balanus balanoides*. This worm matures in the Ruddy Turnstone (*Arenaria interpres*), a shore bird. These cysts occur in large numbers throughout the tissue of the barnacle and may be teased out easily with needles under the dissecting microscope. Place the cysts on a slide in a drop of water. Slight pressure of the cover-slip usually encysts the worms.

**Fourth Laboratory Period - one-half day**

**Class: Cestoda.**

1. **Scolex:** Examine living scolices of *Rhynchobothrium* and *Calliobothrium* (both from the spiral valve of the smooth dogfish), and if available, *Crossobothrium* from the sand shark. Look for bothria, hooks, suckers, proboscides with their sheaths and
contractile bulbs, the unsegmented neck region, excretory tubes with adjacent flame-cells, and the nerve trunks.

2. Mature proglottid: Compress a mature proglottid between a slide and cover-slip and study. Follow Drew (p. 74), as far as possible.

3. Plerocercous (Cysticercoid): The hexacanth embryo of the tapeworm, Otobothrium, from the Hammerhead shark, enter the body of the butterfish, their immediate host to form the cysticercoid larvae which are small white dots in the dorsal body muscles. Tease a cysticercoid out of its sheath of host connective tissue and carefully tease it apart with needles. Cover and study in flattened position. As the larva unfolds, notice the fine proboscides like those in Rhynchobothrium.

Class: Nemertea. Optional:

1. Amphiporus: Follow the directions for Tetrastemma in Drew (p. 77-78). The chief difference in gross morphology between the two forms consists of the larger number of eye-spots in Amphiporus.

2. Make a comparative study of nemertines in the laboratory.

Phylum Nematohelminthes - Class Nematoda: Optional:

Follow directions on mimeographed sheets distributed with this outline to work out the detailed morphology of Metoncholaimus pristiurus.
**Oncorcholaimus pristiurus**

This form is a free-living Nematode found in the mud in shallow salt water. It belongs to the large marine group, the Oncokolaiminae (type genus, Oncorcholaimus, "tooth in the throat").

**Preparation for Examination:**

Study several specimens in a syracuse dish with a binocular dissecting microscope to observe the continual coiling and uncoiling characteristic of many nematodes and to distinguish the blunt anterior form the more pointed posterior end. Note that some large specimens contain several large bead-like structures at about the middle of the body. These are eggs and indicate the animal is an adult female.

Place such a specimen in a drop of fresh water for one to two minutes until quiet and then mount at once in clear sea water. Flatten the animal slightly by removing water from under the cover-glass. Under these conditions the worm should be quiet except for slow movements of the digestive tract which will help observation.

**The Digestive System**

Note that the posterior end tapers very rapidly and is slightly curved. The anterior end tapers gradually. Along the sides of both ends are numerous sensory setae. At the truncated extremity of the anterior end is seen the mouth opening. Behind it is a short pharynx in which there are three sharply-pointed teeth, the onchia. The thick-walled tube running backward from the pharynx is the oesophagus. At its posterior end is a spincter valve (the cardia) marking the beginning of the intestine, which is a yellowish-brown tube running throughout nearly the entire length of the body. Careful focussing on the anterior part of the intestine will show that its wall is composed of typical columnar epithelium. The inner ends of most of the epithelial cells are filled with granules which give the color to the intestinal wall. About halfway along the tapering tail (ventrally) is seen the anus and running forward from it at an angle is the rectum.

**Tail and Spinneret:**

The tail is first conoid and then cylindroid in the posterior fourth where it ends in a somewhat blunt, almost imperceptibly swollen rounded spinneret showing internally the three slightly swollen ampullae of the three caudal glands. The cement-like secretion of these glands is poured out of the minute pore at the extreme tip of the tail to be used in temporarily cementing the work by the tail to the substratum. A spinneret valve (hemispherical posteriorly and tapering anteriorly) is fastened by a contractile fiber to the ampullae and the contraction of this fiber withdraws the valve to allow outflow of the secretion. The caudal glands are ellipsoidal, arranged in tandem fashion from a point about five body diameters anterior to the anus to a point about 10 body diameters anterior to the anus. Each caudal gland is connected with an ampulla by a long, slender caudal gland duct.

**Excretory System:**

This system consists of a single "renette" cell which is a fusiform, ventral cell located ventrally about four body-widths behind the neck. This cell is connected by the renette duct to a single ventral excretory pore located about one-fourth the distance from the anterior end of the body to the nerve ring. The renette duct enlarges near the excretory pore to form a minute excretory visicle.
The Nervous System:
The chief concentration of the nervous system is the thick, semi-translucent nerve-ring which encircles the esophagus about midway of its length. Before and behind the nerve-ring are numerous distinctly nucleated ganglion cells. Other ganglion cells may be seen along the ventral nerve and in connection with sensory setae of the tail may be seen in demonstration specimens stained with methylene blue. The longitudinal nerve cords are not well developed in Metoncholaimus.

The Female Reproductive System:
A short distance proximal to the large thick shelled eggs, which are in the uterus, may be seen a row of cuboidal cells nearly as large as the diameter of the body. The most proximal of these cells, the oocytes, marks the posterior end of the ovary. From this point, anteriorly, the ovary continues forward showing progressively more advanced stages in egg development. The broad reflexed ovary is continuous with the much narrower oviduct which turns posteriorly to connect with the uterus near the posterior end of the ovary. Posteriorly, the uterus connects by a short, transverse vagina to the slightly elevated vulva, the ventrally located external opening of the female system.

The Demanian System:
This system is found only in the female. It consists of the following structures: A short distance anterior to the rectum are two large, clear, cross-striped tubes, the moniliform glands, which open posteriorly by separate pores. Anteriorly these tubes unite near the conspicuous, rosette-like uvette. From the uvette a tube runs to the intestine, and another tube, the efferent uterus which joins the uterus in the vicinity of the vulva. The demanian vessels elaborate a copious, elastic, sticky, non-water-soluble secretion possibly utilized during copulation and also presumably to protect and preserve eggs after deposition.

Make a large drawing of a female Metoncholaimus to show as many of the above features as you have been able to identify.

The Male Metoncholaimus:
In the manner already described mount a male specimen and examine the demanian system is absent although possibly represented by obscure homologous structures. The tail of the male diminishes suddenly in size at the arms and is armed with about ten small "supplementary organs" ventrally located, which give the tail a serrated appearance, giving rise to the specific name, pristiurus (saw-tailed). There are also about thirty short ventral sensory setae. Supplementary organs and setae are alike sensory in function. The opening of the male genital system is just anterior to the anus. Extending forward from it are two slender, rod-like spicula, about seven times as long as the anal body diameter. There are two testes, the anterior testes and the posterior, extending in opposite directions along the middle third of the body. The two testes join the long vas deferens which connects with the ejaculatory duct which in turn opens posteriorly through the genital pore. In each testes there is a progression of stages in sperm development from the blind end of the testis toward the junction with the vas deferens. Draw to show the male genital organs and the extreme posterior end of the male worm.
PLATYHELMINTHES AND NEMATHELMINTHES
SELECTED BIBLIOGRAPHY

(Good reference lists, original research and investigations based on the Woods Hole fauna have been the criteria used in compiling this bibliography. It is not an exhaustive review of the literature of these groups, but is one designed for quick orientation in the several groups.)

GENERAL

Kukenthal, W. und T. Krumbach. 1928. Handbuch der Zoologie. Vol. 2, first half. An excellent general discussion of the morphology, physiology, and taxonomy of all groups included under the Platyhelminthes and Nemathelminthes. Following each group discussion is a good selected bibliography up to 1930. Most of the other references in this list are supplementary to those in Kükenthal.

PLATYHELMINTHES:
Class: Turbellaria


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Class: Cestoda


**Class: Nemertea**


**Phylum Nemathelminthes**

**Class: Nematoda**


**Ecology and Physiology of Parasites**


KEY TO THE MORE COMMON NEMERTEANS OF THE WOODS HOLE AREA

**NEMERTEA:** Soft, very contractile, often brightly colored, mostly free swimming; body elongate, tapetalike or filiform; proboscis often protruded on stimulation.

<table>
<thead>
<tr>
<th>Step</th>
<th>Description</th>
<th>Code</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (20)</td>
<td>Eyes present</td>
<td>2</td>
</tr>
<tr>
<td>2 (15)</td>
<td>Eyes few in number, less than 20</td>
<td>3</td>
</tr>
<tr>
<td>3 (10)</td>
<td>Four eyes, forming a rectangle</td>
<td>4</td>
</tr>
<tr>
<td>4 (5)</td>
<td>Body stout</td>
<td>Tetrastemma vittatum&lt;br&gt;(Green or yellowish, with 1 or 2 dorsal stripes; 5cm x 4mm; on muddy bottom at low tide)</td>
</tr>
<tr>
<td>5 (4)</td>
<td>Body slender</td>
<td>6</td>
</tr>
<tr>
<td>6 (7)</td>
<td>Body tapering from middle both ways</td>
<td>Tetrastemma elegans&lt;br&gt;(2 cm x 1mm; median dorsal yellow and two lateral brown stripes; among weeds and stones)</td>
</tr>
<tr>
<td>7 (6)</td>
<td>Body wider in front, tapering posteriorly</td>
<td>8</td>
</tr>
<tr>
<td>8 (9)</td>
<td>Body pale yellow or reddish, spotted; found on muddy bottom</td>
<td>Tetrastemma vermiculum</td>
</tr>
<tr>
<td>9 (8)</td>
<td>Body white, light green, or yellowish; not spotted; among algae</td>
<td>Tetrastemma candidum</td>
</tr>
<tr>
<td>10 (3)</td>
<td>Eyes 4 - 14, lateral, never forming a rectangle</td>
<td>11</td>
</tr>
<tr>
<td>11 (12)</td>
<td>Body small, somewhat flattened, dark green with mid-dorsal yellowish stripe</td>
<td>Lineus bicolor&lt;br&gt;(45mmx 1.5 mm; single row of 8-14 eyes on each side; among algae and hydrooids in shallow water)</td>
</tr>
<tr>
<td>12 (11)</td>
<td>Body long and slender, not flattened</td>
<td>13</td>
</tr>
<tr>
<td>13 (14)</td>
<td>Body coiled in tight spiral when contracted</td>
<td>Lineus socialis&lt;br&gt;(15 cm x 1.5 mm; green or brown with a few narrow encircling light rings; single row of 4-6 very small eyes on each side of head; under stones between tides)</td>
</tr>
<tr>
<td>14 (13)</td>
<td>Body not coiled in tight spiral when contracted</td>
<td>Lineus ruber&lt;br&gt;(Green, brown, or reddish; single row of 4-8 eyes on each side of head; 20 cm x 6mm; under stones between tides)</td>
</tr>
<tr>
<td>15 (2)</td>
<td>Eyes numerous, more than 20</td>
<td>16</td>
</tr>
<tr>
<td>16 (17)</td>
<td>Body slender, 4 cm long, light green; eyes in 2 or 3 parallel rows along each side of body; between tides</td>
<td>Zygonemertes virascens</td>
</tr>
<tr>
<td>17 (16)</td>
<td>Body relatively short and thick; eyes never in parallel rows</td>
<td>18</td>
</tr>
<tr>
<td>18 (19)</td>
<td>Eyes converging backwards; yellowish; 7cm x 3mm; under stones between tides</td>
<td>Amphiporus ochraceus</td>
</tr>
<tr>
<td>19 (18)</td>
<td>Eyes arranged in 2 frontal clusters on the white marginal area, and in 2 dorsal groups; reddish or brown; head wider, set off from body, white in front with a white spot on each side and an H-shaped figure in the middle; under stones between tides</td>
<td>Amphiporus angulatus</td>
</tr>
<tr>
<td>20 (1)</td>
<td>Eyes absent</td>
<td>21</td>
</tr>
<tr>
<td>21 (28)</td>
<td>Caudal cirrus present</td>
<td>22</td>
</tr>
<tr>
<td>22 (23)</td>
<td>Body very long, 2m or less x 25mm; broad, flat; head lancet-shaped with lateral groove; flesh-colored; in sand</td>
<td>Cerebratulus lacteus</td>
</tr>
</tbody>
</table>
23 (22) Body long and slender; head without lateral groove...
24 (25) Head long and pointed, pure white; body whitish; in sand between tides... Zygeropola rubens
25 (24) Head short and triangular; body flat... Zygopollia saecula
26 (27) Body dark brown or yellow; in sand...
27 (26) Body red or purple dorsally with light median lines; 15mm x 4mm; in sand between tides... Micura leidyi
28 (21) Caudal cirrus absent...
29 (30) Body short, flat, thick, with large sucker at posterior end; in branchial cavity of Mya, Venus, etc...
30 (29) Body slender, cylindrical, no sucker, free-swimming...
31 (34) Body cylindrical in front, flattened behind...
32 (33) Color orange; 25 cm x 10mm; at low tide mark...
33 (32) Buff in color; head white, flattened, rounded in front; 12cm x 3 mm...
34 (31) Body cylindrical and filiform, not flattened...
35 (36) Body very small, 25mm x 0.5mm; head large and distinctly set off; whitish; among annelid tubes at low tide mark... Tubulanus pellucidus
36 (35) Body long, slender, tapering to pointed anterior end; 15 cm x 1 mm; worm coils body in spiral; flesh-colored; under stones and in sand between tides... Cephalothrix spiralis
**KEY TO THE MORE COMMON POLYCLAD TURBELLARIANS OF THE WOODS HOLE AREA**

POLYCLADIDA: Fairly large turbellarians, with thin, leaf-like body; intestine with very numerous branches which ramify throughout the body; eyes usually numerous.

1. (6) Tentacles absent
   2. (3) Marginal eyes present
   3. (2) Marginal eyes absent
   4. (5) Four conspicuous eye clusters near anterior end
   5. (4) Eyes, few, about six on each side, not in conspicuous clusters
   6. (1) Tentacles present
   7. (6) Tentacles formed by upfolded anterior margin
   8. (7) Tentacles formed just posterior to anterior body margin
   9. (12) Marginal eyes present, surrounding or nearly surrounding body edge
   10. (11) Color pattern of alternating yellowish or white and brown cross bars of which the most anterior and posterior ones are V-shaped; usually found in hermit crab shells (Pagurus pollicaris); often on wharf piles
   11. (10) Color usually cream or yellow; body with undulated margins; under stones in shallow water and tide pools
   12. (9) Marginal eyes absent
   13. (16) Oval form; tentacles without eyes scattered through them
   14. (15) Living in mantle cavity of Busycon; white in color
   15. (14) Living on Sargassum weed; flesh-colored
   16. (13) Elongate forms; tentacles with eyes scattered through them
   17. (18) Color green with median dorsal light stripe, 29mm x 10mm; on algae
   18. (17) Color yellowish, with brownish spots, 6-8 mm long; on algae

- **Coronadona mutabilis**
- **Notoplana atomata**
- **Eurylepta maculosa**
- **Euplana gracilis**
- **Stylochus zebra**
- **Stylochus ellipticus**
- **Hepolapla inquilina**
- **Hoploplana grubei**
- **Planocera nebulosa**
- **Gnesioceros floridana**
PART I.—TERMINOLOGY


PART II.—COMPILED BIBLIOGRAPHIES.


PART III.—PHYSIOLOGICAL, MORPHOLOGICAL, EMBRYOLOGICAL, etc. PAPERS.


PART IV.—TAXONOMIC SECTION


REvised CHECK LIST of Bryozoa most commonly encountered around Woods Hole, --- their present-day names and the names previously used.

MODERN TERMINOLOGY

Phylum Molluscoidea
Class Entoprocta

Parentesia
Podiocellina

Class Ectoprocta

Acanthodosia tenuis ------------ Membranipora tenuis
Aeta anguina
Aevmallia sestigera ------------ Bussia or Hippurarria
Alcyonidium
Bowerbankia gracilis
Bugula flabellata
Bugula turrita
Callopora aurita --------------- Membranipora aurita
Crisia eburnea
Cryptosula pallasiana ----------- Lepralia pallasiana
Electra hastingsae -------------- Membranipora monostachys
Electra pilosa ---------------- Membranipora pilosa
Flustrrella hispida
Hippodiplosia americana--------- Lepralia americana
Hippodiplosia portusa----------- Lepralia portusa
Hippoporina contracta---------- Lepralia serrata
Hippothoa hyalina
Membranipora crustulenta?-------- Membranipora lacroixii
Membranipora tuberculata--------- Membranipora tehuelcha
Microporella ciliata
Schizmopora avicularis---------- Calllepora americana
Schizomavella auriculata ------- Schizoporella auriculata
Schizoporella unicornis
Smittina trispinosa var. nitida
Stephanosella biaperta----------- Schizoporella biaperta
Tegella unicorns---------------- Membranipora unicorns

Roughly about 60 more spp. occur around the Woods Hole region but the above are the more common forms encountered.
INVERT.ZOOL. ♦ Bryozoa

CLASS ENTOPROCTA Nitsch 1869.
Family PEDICELLINIDAE Hincks 1880

PEDICELLINA Sars 1885 or BARENTSIA Hincks 1880.

A. General directions:
1. Obtain some slides which have been immersed in sea water for 2 weeks or so and study them for Bryozoan colonies. Some of these slides will undoubtedly contain lovely colonies of Barentsia, Pedicellina, Cryptosula, Bugula and a variety of other forms and phyla.
2. These slides should be placed in Petri dishes containing some sea water so that the free behavior of the specimens can be studied.
3. Study the Petri dishes first under a binocular dissecting scope to get a general view of the slide. Identify the forms found on it, then transfer the Petri dish and its contents to the stage of the compound microscope, first using the 50x then later the 100x magnification.
4. After you have completed your study of the slides return them to the water table because later in the period or tomorrow you may need them again.
5. In case the slides are not satisfactory or do not contain the desired form return them and obtain some material from the central tank table.

B. Specific directions
1. Answer the questions asked henceforth, thru observation.
2. Select a Pedicellina or Barentsia colony for study.
3. The colony of Pedicellina is stolonoate, that is, the individuals or zoids are stalked and the stalks are connected by a delicate tube or stolon. The same condition obtains in Barentsia.
4. Is the stolon perfectly straight or may it bend & twist about?
5. Is the stolon entirely adherent, everywhere touching the substratum?
6. Are there any septa or partitions along the stolon? Are there any septa in the immediate vicinity of the stalk or peduncle?
7. Are there any spines or hairy processes along the stolon, peduncle or calyx?
8. Does the stolon continue as a single line or does it branch, sending side lines outward?
9. Are the peduncles of uniform diameter from stolon to calyx?
   If you have a Barentsia specimen there will be a marked enlargement at the base of the peduncle. Also there might be "rings" or a ringed appearance around the enlargement.
10. Are the peduncles of the same diameter as the stolons?
11. Are the peduncles hollow?
12. Is the stolon hollow?
13. Do any peduncles branch?
14. Is the head or calyx slightly flatter on one side than on the other?
15. Is the head like a ball (contracted), with tentacles folded inward, or is it like a flower, with tentacles unrolled outward, like petals?
16. Are there any peduncles without heads?
17. Where are younger peduncles placed on the stolon? Are they at one end or do they seem to occur haphazardly?
How can you tell a young peduncle from an old one?

19. Take a fine dissecting needle or a hair and touch a tentacle of an expanded zoid or animal. Repeat several times, after a period of rest, to see if you get the same response.

20. If 1 tentacle is touched do the others on the same head retract or fold in?

21. What is the nature of the reaction—i.e., does the tentacle shorten or does it merely fold in?

22. If 1 tentacle is touched does the response involve other heads on the same stolon?

23. Count the number of tentacles on several individuals. Is there a constant number for the entire stolon or is there variation?

24. Compare the tentacle number with that obtained by your neighbor.

25. Does size of calyx seem to have anything to do with tentacle number?

26. Are tentacles ciliated all around or just on one side?

27. Is there a definite pattern of ciliary action?

28. Do the tentacles show any ability for independent action or do all do the same thing upon stimulation—in the same manner and at the same time?

29. The tentacles are borne on a ridge of skin or body wall known as the lophophore. Is the lophophore circular or horse-shoe-shaped in these forms?

30. Observe the plane or the slant of the lophophore. Is it at right angles to the main axis of the stalk and calyx? If not obliquely it is not Pedicellina but another genus.

31. If the specimen is so oriented that the flat and gibbose sides of the calyx are both evident then either the right or the left side of the specimen will be facing you. The esophagus runs along the flat side, the intestines along the gibbose side. Figure out whether you are looking at the right or the left side of the animal.

32. The tentacles fold or curl into the atrium or vestibule, a space within the cirrlet of tentacles at the top of the calyx. There are several openings into this atrium from the inside. See diagram at right.

33. A brood room is present around the anal opening. In some calyces it may be small, in others large.

34. The tentacle ring is interrupted in the region of the anal opening. This is evident only in the top view of the lophophore.

35. Is the mouth round or crescent shaped?

36. A scarcely evident fold of skin, the epistome partly guards the mouth from the tentacular side.

37. If the animal can not be conveniently studied in the Petri dish you might gently scrape off an area under the binocular dissecting scope, place the scraped material on a slide with a cover slip and some sea water. Then, study for details of anatomy, using 100x, then 215x and finally 430x.

38. The digestive tract is simple, consisting of mouth, esophagus, stomach, intestine, rectum and anus. The mouth, as well as a good deal of the rest of the tract is ciliated.
39. The tubular esophagus goes downward into the calyx from the mouth, following the latter side of the calyx.

40. It enters the greatly expanded stomach which occupies most of the "bowl" part of the calyx. The stomach wall is considerably thickened, in spots.

41. From the stomach leads the narrower, somewhat ovoid intestine, which in turn is followed by still another ovoid extension, the rectum. The intestine is still somewhat horizontal in position in the gibbose part of the calyx while the rectum turns upward vertically along the gibbose wall up toward the tentacle base region. A sphincter exists between the intestine and rectum and at the anus.

42. Above the stomach and intestine are the gonads, brood chamber, ganglion and the nephridial tubes. There seems to be conflicting opinion regarding the reproductive organs of Pedicellina. In P. cernua there occur male individuals, female individuals and hermaphrodites, (Marcus 1939, pp.275-276). The testes of P. cernua are much bigger than the ovaries in the unisexual individuals. The following description fits P. cernua (after Marcus).

43. The 2 testes open directly into a large seminal vesicle. The ejaculatory duct is long, ciliated and ends close below the anus in position but anterior to it. (The seminal vesicle apparently seems to take the place of the brood pouch of the hermaphrodite??) The testes may be extensive, partially surrounding the esophagus, the upper part of the stomach and intestine.

44. The ovaries are bag-shaped and contain ova of various sizes. A female zoid may have some tissue on which embryos attach and grow. Ripe eggs measure 40 x 50 x 60 micra. Eggs are fertilized inside the animal.

45. The hermaphroditos of P. cernua resemble the above described female zoid except for the presence of 2 small testes and other minor details.

46. The small testes open each by a duct into a common gonad duct which also receives the 2 ducts from the ovaries. A shell gland is present in the vicinity of the junction of the gonad ducts.

47. The nephridial organ is a Y-shaped tube. It is located between the mouth and ganglion. The ganglion is a small elongate ovoid body just dorsal to the stomach and posterior to the esophagus.

48. Nerves, invisible to our present study methods, lead from the ganglion to the tentacles and other parts of the body.

49. Muscle fibers lead vertically from the base of the calyx along each side of the stomach to the atrial region being responsible for retraction of the atrium.

50. There are circularly arranged muscle fibers about the esophageal wall which are constrictors.

51. Extensors of the esophagus are 2 slender fan-shaped muscles extending from the body wall anteriorly and horizontally toward the esophageal wall. There are other muscle also but these are most conspicuous.

52. There is a circular sphincter in the flap of skin around the edge of the lophophore, at the outer base of the tentacles. This constricts when the tentacles are folded in.

53. The area between the body wall and the digestive tract normally occupied by the coelom is filled with a parenchyma.

54. Suggested work on Pedicellina or Barentsia:
   a. Answer such questions as you can from observation.
   b. Suggested drawings:—make such as you have time for.
54. **Fig. 1**—Habit sketch showing stolon, peduncles, calyces of several individuals of a colony either of *Pedicellina* or *Barentsia* or both.

**Fig. 2**—Draw a patch of stolon on both sides of a peduncle & also a peduncle., enlarged.

**Fig. 3**—Sketch a top view of a calyx showing tentacles expanded, the vestibule and the various openings.

**Fig. 4**—Side view of calyx, enlarged, showing the digestive tract, musculature and such other organs as you can find.

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**CLASS ENTOPROCTA** Nitsche 1869

Order STENOSTOCHATA, Formerly CYCLOSTOMATA

Family Crisiidae Johnston 1847

Genus Crisia Linnouaux 1312

1. It is better to study young Crisia colonies from submerged slides but if these are unsatisfactory then colonies from sea weed will be suitable for study.

2. The colonies are delicate, white, lacy, branching masses. On what kind of sea weed are they growing?

3. How tall are the colonies?

4. Observe the masses first with a dissecting 'scope, then study under the compound microscope, with the light shining down on the specimen if you find transmitted light unsatisfactory. Use 100x magnification.

5. Note the mode of attachment to the sea weed or to the slide. Is there a small circular disc—the PRIMARY DISC—to which the colony attaches?

6. Does there seem to be a constricted JOINT between the disc and the first or PRIMARY ZOUD? This BASAL TUBULUS is always present in some spp. of Crisiidae (Crisiella producta) but sometimes absent, sometimes present in Crisia oburnea.

7. How many individuals sprout out immediately from the primary disc?
6. Which is the oldest and which the youngest part of the colony? Use terms "distal" and "proximal" with respect to the primary disc.

9. Do you find any tiny holes, PSEUDOPORSES, in the white calcareous part of the colony (in the tubes & disc)?

10. Are the pseudopores bigger & more abundant in the younger parts of the colony which are thinner and more transparent than in the older basal, more calcified parts of the colony?

11. What is the shape of the pseudopores? Is it the same on the inside as on the outside? Breaking off a chip or crushing another piece of colony on another slide, then spreading the pieces might show this point.

12. Observe the PRIARY ZOID which is attached to the primary disc or substratum. Is the zoid of uniform diameter throughout?

13. Is there a bud coming distally from it? If there is, this bud is the 2nd pod. Is there a 3rd individual on the other side? At times in C. cbornea 2 lateral branches are developed from the same primary zoid.

14. Is the distal, open end of the primary zoid bent in a definite direction because of the bud sprouting from it? Is this mode of bending & direction of bending carried out in succeeding zoids?

15. Do all the zoids face in the same direction?

16. Do you find any brown pinched-in places or rings along the colony? These are the NODES. The white tubes and branches between the nodes are the INTERNODES.

17. Each tube in the internode is a zoid or ZOOECIUM. The number of zoecia in the internodes is diagnostic for the various spp. of Crisiaidae. Count the # of zoecia in each internode of your colony. Compare with your neighbor's results.

18. Compare the number of zoids in the basal internode with successive internodes. Does the number differ consistently?

19. Are the nodes the same color at the proximal as at the distal end of the colony?

20. The female reproductive cells are developed in Crisia in individu­als called GONOZOIDS or BROOD CHAMBERS, which are located in place of some ordinary zoids in the internodes. They are pear-shaped or club-shaped, much more swollen than ordinary zoids which are tubular. They occur with a definite regularity, at a definite place on the internode. Can you find a gonozoid in your colonies?

21. Is there more than 1 gonozoid per colony? A colony does not reach its full development until it has gonozoids. Internodes containing gonozoids are fertile internodes, those lacking gonozoids are called sterile internodes. The latter internodes have slightly different looking individuals from the XIXIXIX XXXXXX fertile internodes. Do you notice any difference in your specimens of both kinds? This is an example of dimorphism.

22. Is there the same number of zoids in sterile internodes as in fertile internodes?

23. Each gonozoid has the following parts:
   a. PROXIMAL PART—this is narrow, wedged in between neighboring zoids & sometimes partly hidden by them.
   b. MIDDLE PART— strongly dilated, continuous with the narrowed proximal part. Here embryos are found.
   c. DISTAL PART—the constricted, tubo-like exit from the middle part, thru which the larvac leave the gonozoid.
24. Are the pseudopores visible in the gonozoid? Are they more densely "peppered" over the surface than those of the autozoids (ordinary zooids)?

25. What is the shape of the OOECIOSTOME—the tube or channel by which the larvae leave the swollen part of the gonozoid?

26. What is its location with respect to the neighboring zooid openings?

27. What is its rim like? Does it curve, flare or appear perfectly straight & of uniform diameter?

28. Is its opening, the OOEOCIOPORE, of the same diameter as that of the neighboring zoocia? The ooeciostome is quite important in the identification of Crisia spp.

29. If it is possible to study some living colonies of Crisia undisturbed, the zooids will be found to have & tentacles.

30. Tear apart a zooid, & a gonozoid. Are there any fleshy parts & larvae in these structures?

Suggested activity for this exercise:

a. Answer all questions is you have time.

b. Suggested figures for drawing:

Fig. 5. Make a "mathematical formula" or diagram showing the exact arrangement and mode of branching of autozoids, gonozoids, branches, etc. using the following symbols:

\[ PA \text{ --- parent zooid, if any} \quad AZ \text{ --- autozoid} \]

\[ GZ \text{ --- gonozoid} \quad BR \text{ --- branch} \]

Joint

Rough example: \[ PA & BR & 3AZ & GZ \ldots \quad 6AZ & GZ & BR \ldots \]

Fig. 6. Make a habit sketch of a colony.

Fig. 7. Make a detailed view of a gonozoid & its ooeciostome.

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j. Smit,F.A. 1863. Bidrag till kändedomen om Hafs-Bryozoornas utveckling. ÖfSALA UIV., ARSSKRIPT (brood chamber)


l. Smit,F.A. 1865. Om Hafs-Bryozoornas utveckling och fett-kroppar. Ibid.

m. Smit,F.A. 1867. Krit. Forteckning, etc. II. Ibid, Arg. 23.
1. Obtain colonies of Bowerbankia from the side table. They will form a soft grayish fume on vegetation, on Bugula, hydroids, rocks, etc. Study first in a watch glass with some sea water, under the dissecting scope.

2. The zoids are closely clustered together, giving the impression of tall transparent columns or sky-scrapers. Their bases are so close together that it is difficult to study them. The zoids are irregularly arranged along a slender STOLON, occurring sometimes on both sides and other times along one side of the stolon. Sometimes the zoids are bilaterally placed, other times irregularly placed. Are there any zoids on the top of a stolon?

3. Is there a caudal, pointed process on the basal side of a zoid?

4. Do all zoids in a colony have these basal processes?

5. Is there more than 1 caudal process per zoid?

6. Are the zoids which have caudal processes older or younger than those which do not have them?

7. What is the shape of each zoid?

8. Carefully observe the free edge of the zoid. Is there anything peculiar or different about its shape? Is its shape different from that of the rest of the zoid?

9. Are all zoids of the same thickness and length? How can you explain this?

10. Watch for protrusion of tentacles. If any are protruded count them. Compare the number with that obtained by your neighbor.

11. Does it take long for the zoids to protrude their tentacles once the colonies have been allowed to remain undisturbed?

12. After individuals protrude their tentacles touch a tentacle lightly and note results. What happens to the individual which was touched? Are all the neighboring individuals affected?

13. Place a small amount of carmine in the water, setting the preparation aside for a few minutes until the zoids have had a chance to ingest some of the material. While you are waiting for that to happen you might observe the ciliary currents around the tentacles. Is there any rejection of the colonizing material?

14. Carefully dissect away or tear out 2 or 3 zoids from the colony. Use dissecting needles or fine forceps. Place these zoids on a slide with some sea water and a coverslip.

15. If your microscope is calibrated, measure the: zoocelial length, width, stolon width, etc. How do your measurements agree with the following:

<table>
<thead>
<tr>
<th>Measurements</th>
<th>B. gracilis</th>
<th>B. caudata</th>
<th>B. gracilis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zoocelial length</td>
<td>0.42-0.6 mm</td>
<td>1.0-1.5 mm</td>
<td>0.35-1.2 mm</td>
</tr>
<tr>
<td>Zoocelial width</td>
<td>0.17-0.18 mm</td>
<td>0.16-0.24 mm</td>
<td>0.16-0.2 mm</td>
</tr>
<tr>
<td>Stolon width</td>
<td>0.09-0.11 mm</td>
<td>0.07-0.1 mm</td>
<td>0.1 mm</td>
</tr>
<tr>
<td>Stomach diameter</td>
<td>0.1 mm</td>
<td>0.09-0.12 mm</td>
<td></td>
</tr>
<tr>
<td>Gizzard width</td>
<td>0.1 mm</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

If you do not know how to calibrate your scope or how to make measurements under the scope & wish to learn, ask the instructors & you will be shown.

Dr. Marcus considers B. gracilis & B. caudata 2 separate spp. while Dr. Osburn considers them 1.
16. Notice that this species has a very definite body cavity, unlike Pediocellina.

17. If the zoid is retracted, can you identify the tentacles?

18. Are the tentacles pulled in, in a straight line or are they folded up or twisted about? Are they all parallel to each other?

19. In the retracted zoids you may see a channel leading from the external opening into the zoid down to the tip of the tentacles. This is the VESTIBULE. Toward its lower end are several groups of muscles leading from the body wall to its wall. These are the PARIETO-VAGINAL muscles. How many of them are there?

20. Are there other muscles arranged hoop-like, horizontally, along the body wall proper, in the mid region? These are the PARIETAL MUSCLES. How many bands of them are there? Is the number the same on all the zooecia?

21. Within the circle of tentacles at their base, is located the mouth. If the zoid is retracted you will not be able to see the mouth. However, beneath the tentacles is the narrow tubular esophagus. The next digestive tract organ after the esophagus is the rounded, bulbous, hard gizzard or PROVENTRICULUS, regarded by Marcus as the cardiac part of the stomach. It looks like a ball. Is it colored differently from the rest of the tract?

22. The proventriculus opens into the enlarged somewhat baggy J-shaped stomach, which extends downward to the base of the zooecium then turns upward to parallel the esophagus. A slight constriction separates the stomach from the intestine which is parallel with the tentacles, generally, for a distance, if the polypide or zoid is retracted. Note the point of termination of the intestine. This is the anus. Why is this species a member of the ECTOprocta? What is the main difference between an ECTOproct and an ENDOproct?

23. Do you find any muscles extending from the lophophore or general lower tentacular & upper esophageal regions down to the body wall, in the same general direction as the downward half of the digestive tract? These are the RETRACTORS. The pull in the polypide.

24. If your slide does not have any sizable length of stolon on it tear off a piece from a fresh colony & place on your slide. Are there any knobs along the stolon anywhere? Are these of the same size or are some bigger than others? What might these be?

25. Is there anything inside these buds? Can you recognize tentacles or digestive tract at this early stage?

26. Are there any partitions or SEPTA along the stolon? Where are they located with respect to the zooecia?

27. Does the stolon branch?

28. Is it entirely adherent?

29. Are the zooecia arranged perpendicular to the stolon or are they set in at a slant or angle?

30. Are there any empty-looking large zooecia along the stolon? How do you account for their size and yet their emptiness?

31. Do any of the zooecia proper give rise to other zooecia?---i.e. have branches on their side?

32. Have any of the zooecia ingested the red carmine?

33. Suggested work for this form:-
   a. Answer the questions from observation, if time permits.
   b. Fig. 8. A habit sketch of the colony.
   c. Fig. 9. An enlarged view of 1 or 2 zoids attached to the stolon.
Bibliography on Bowrorbancida.

a. Calvet, 1900. pp. 227-230, 232-234; Pl. VI, Fig. 13, Pl. VII, Fig. 4.

Bryozoairea Ectoproctea Marins. MONTPELLIER.


A.R.Q., do P.C.S. PARA-BAINSE. I (1): 7-36, p. 29, fig. 1 also.

d. Marcus, 1937. pp. 127-139, TAF. XXVIII, fig. 74.

e. Marcus, 1935. pp. 55-67, TAF. XIII, Fig. 33


g. Osburn, 1940. p. 341 & Fig.

Class ECTOPROCTA Niitsche 1869
Order CHEILOSTOMATA Busk 1852
Family ESCHARELLIDAE Levinson 1909
Cryptosula pallasiana (Moll 1803)

1. Obtain slides which have been immersed in EEL Pond for a couple of weeks. Place the slide in a Petri dish containing enough water/sea to cover the specimens so colonies can expand their tentacles.

2. On these slides will be some other forms in addition to the Cryptosula. See Pratt, p. 301 for small sketch of this species.

3. Cryptosula colonies are flatly encrusting, calcareous. In this young stage they will be translucent in spite of their calcification.

4. Each colony is flat and becomes circular in time. It consists of a number of compartments or zooecia arranged in juxtaposition (side by side) & spreading out radially from the antennal zoid, the ancestrula.

5. How many zooids or zooecia are there in the biggest colony on the slide?

6. Can you identify the ancestrula from which the colony sprang?

7. Are all the zooecia of approximately the same size?

8. How many zooids touch the ancestrula directly?

9. Are the zooids at the very edge of the colony as complete as the ancestrula so far as walls are concerned?

10. The biggest opening or hole in each zooid is the zoosocial aperture. The shape of the aperture is one of the most important key characters in Bryozoan classification. Each species has a characteristic aperture. What is its shape in this species? Is the aperture longer than wide?

11. There is a delicate membrane, the operculum, closing the aperture. It has a reinforced chitinous rim. It is open or closed, depending upon the location of the tentacles. If the tentacles are protruded the operculum is open. If the tentacles are withdrawn, the operculum is closed. How does the operculum open?—sideward, downward, upward, inward or outward? Do you see any muscle attachments to the operculum—like the chains of a draw-bridge?

12. The lateral margin of the aperture has a small tooth, denticle or cardella (synonyms here) on each side, toward the posterior border. The operculum pivots on these cardelles. The cardelles occur at the angles formed by the wider outslung lower border and the flatter upper narrower half of the aperture. The narrower upper part of the aperture, thru which the tentacles are extruded is the
Cryptosula pallasiana.

12. porta. The wider lower half of the aperture is the vanna. The
vanna is the opening into the compensation sac or compensatrix.
the anter is the part of the operculum closing the porta. The
poster is the part of the operculum closing the vanna.

13. The compensatrix becomes filled during the extrusion of the poly-
pide. The compensation sac is crescent shaped from the front and
located around and beneath the widest part of the aperture, around
the vanna. Can you identify it in some of the most transparent
individuals? In the Ascothorea, to which Cryptosula belongs, the
polyptide can emerge from the zooecium only if an equal volume of
water compensates this extrusion. Canu & Bassler, 1920, p. 58,
state that when the polypide withdraws again into the zooecium
the water is emptied again from the compensatrix.

14. Borg, 1930, gives these measurements for the aperture or Cryptosula
pallasiana: Length 0.23 mm., Width 0.15 mm. at widest part. Check
these measurements if your microscope is calibrated.

15. The calcareous front wall of the zooecium is the tremocyst. It is
perforated by a number of large pores, the tremopores. How many
tremopores are there? Is the number constant, i.e. identical for
all zooecia in the colony? Check with your neighbors.

16. The 2 patches of muscle attached to the opercular anter are the
occlusor muscles. Each is fan-shaped, with the narrow end attached
to the operculum?

17. How many tentacles does your specimen have? Calvet says 16-17 but
Canu & Bassler think that there might be more. Compare with your
neighbor.

18. In the most translucent specimens can you identify the tract? If
there is a colony on the reverse side of the slide study that for
a moment. If not, turn the slide over carefully & study the
back of the colony which you have been observing up to now. Are the
internal organs more visible in this view? Are there any pores
visible on the basal or back wall?

19. Are pores visible on the distal zooecial wall—the one above the
apertura? On the proximal wall? We will come back to this point
later when studying mature calcined colonies.

20. Find a very young colony on your slide where the calcification has
not progressed far. Are the tremopores much larger & differently
shaped than xxx in older specimens?

21. Obtain a dried mature colony from the supply table. This very pro-
bably will be attached firmly to a rock or shell. If convenient,
place the rock or shell under your binocular. If the specimen is
too large for that you should make a calcined mount of it. Direc-
tions for calcining are given on page 12-17 "Directions for perma-
nent preparations of calcareous Bryozoa.

22. Do you find any globose bodies, oavicells, distal to the apertura?
Canu & Bassler say oavicells & avicularia are present but Osburn
says that they are wanting. If you find one be sure to call the
instructor’s attention to it, as it will settle a debated point.

23. Take a small fragment of the dry or calcined colony & either crush
it gently or pick off the frontal wall so that the lateral, distal &
proximal walls of the zooecium show. The calcined colony is pre-
ferrable to the natural untreated colony. You may have to turn the
slide about or hold it at an angle so that you can see the various
walls plainly. Wall perforations called septulae are present. If they
consist of a single pore or hole they are uniporous. If they
CRYPTOSULA PALLASIANA

23. consist of a series of pores in a plate they are multiporous or rosette plates. Which do you find in your specimen along the lateral walls? Which types occur along the distal wall? In the proximal wall? If possible, break off a lateral, distal & proximal wall to study under 100x and 430x.

24. suggested work on Cryptosula:
   a. Answer all questions.
   b. Possible drawings:
      1. Fig. 10: A habit sketch of the colony.
      2. Fig. 11: A zoid from a very young colony showing as much of the internal anatomy as possible.
      3. Fig. 12: Enlarged view of septulae.
      3. Fig. 13: Several contiguous zoociae of the calcined or mature well calcified specimen.

25. References on Cryptosula pallasiana:
   c. Canu & Bassler 1929, pp. 325-326
   d. Davenport 1891, pp. 47-49, 53, 54, Pl.VIII, Fig. 71, Pl.IX, Fig. 72, 73, 81, Pl. X, Figs. 84, 86, 91.

Class ECTOBRIOCTA Mitsche 1869
Order CHILOSTOMATA Bask 1852
Family Bugulidae Gray 1843
Genus Bugula Oken 1815

1. This second member of the Cheilostomata is chosen because it is exceedingly common here.

2. Two spp. of Bugula are available: B. flabellata (J.W. Thompson) 1847 or 1848 & B. turrita (Desor) 1843. They can be distinguished by their mode of growth. Place a sprig of each in a fingerbowl of sea water to see their growth habit better. B. turrita has a delicate spiral growth habit while B. flabellata is stubby & fan-like.

3. Place a very small sprig of each in a watch glass containing enough sea water to cover them. Place this on your dissecting scope, allowing sufficient time for polypides to protrude their tentacles. When they do protrude, count them & record for each spp.

4. Spread a small sprig of each species on a slide, arranging it in such a manner that there are some zoociae "face" up and others "face" down. Cover with a coverslip. Try to answer these questions from observation.

5. What is the color of each spp.?

6. Bugula is a colonial Bryozoan. It consists of a number of box-like or roughly tubular filled chambers called zoids. The entire colony called the zoarium.

7. Are the zoids visible to the naked eye?

8. How many rows of zoids are there in each branch? The number is important, being used as a species criterion in this group,—along with some other characters. The number is different in the 2 spp.

9. Do the zoids on each branch arise alternately with respect to each other or do they arise abreast of each other?
ZOOL.—BRYOZOA

BUGULA

10. The walls of the zooids or the chamberlike shells housing each living individual are called **zooecia**. They are transparent but yellowish in color & are formed of the hardened cuticula.

11. Are there any zooecia which are empty of soft inner parts? The poly-pide consists of digestive tract and tentacles. Sometimes these soft poly-pides within the zooecia die, leaving empty cuticulae which because of their hardness or firmness remain thus for quite a long time.

12. Observe the free distal edge of the zooecia. Is it straight or provided with spines, hooks or flap-like processes?

13. If provided with flaps or spines how many per zoid?

14. Are there any on the inner distal border?

15. Compare the 2 spires, the "face" up one and the "face" down one. Is the proximal border or point of junction with the extypicid zoid below (on the "face" down specimen) perfectly straight?

16. The different individuals of the colony arise by budding. The basal individual, by which the colony is attached to the substratum developed from a sexually produced larva. This basal individual, or ancestrula is the parent from which the other members sprang, directly or indirectly by budding.

17. Do you find any small or incomplete individuals at the tips of the branches, — i.e. some without spines, without a fully developed poly-pide? These are very probably young buds. Do the zooecia near the point of origin of the colony differ in appearance from those higher up along the stalk? In what way?

18. On the "face" up zooids note the large thinner, or more transparent area, oval at the base, occupying the upper 1/3 or 2/3rds of the wall. This is the apertura. It is covered over by a thin membrane which is a part of the body wall of the animal. Is there a semicircular flap, the operculum, at the top of the apertura, between the flaps or spines? The tentacles come out thru this region when the operculum is opened.

19. Observe carefully the inner and the outer edges of the zooecia. Are there any peculiar, bird-head-shaped structures, the avicularia? They look like the head of an eagle or hawk and are stalked. They are distinct, specialized individuals. Their jaw opens & closes periodically. The whole "head" sways also. This activity can best be seen on unflattened individuals, i.e. individuals not under a coverslip. Observe the individuals which are already under the dissecting scope. Stimulate them with a hair. The avicularia should "snap" at it and if the jaws clamp together you may be able to lift the whole colony out of the water by the hair. The jaw or mandible is moved by a huge fan-shaped muscle which is present in the head.

20. The avicularia are stalked. Are they arranged upside down, right side up, out in front or at the side of the zooecium?

21. Is there any avicularium present on every zoid? Or, are the avicularia restricted to a single branch? Or, are avicularia scattered apparently indiscriminately over the colony?
Vert. Zool.—Bryozoa

SUGULA

18. Are the ovicells empty or full? Those which are full have developing larvae in them. Those which are empty have either served as incubators or are about to serve as such. What color are the full ones?

19. Do those which have ovicells have the same size avicularia as the non-ovicelled ones?

20. Do those zoids which have ovicells also have the same number & type of spines or flaps as those which are not ovicelled?

21. The tentacles are borne on a circular ridge of flesh, the lophophore which surrounds the mouth. The tentacles when withdrawn into the zoecium are in a fold of skin called the introvert or tentacular sheath. It is closely applied to them. When the tentacles are everted or pushed outside, this introvert turns inside out, bringing the tentacles out. The tentacles & introvert are pulled in or pushed out by the retractor muscles. The retractor is a longitudinal muscle stretching from the lophophore downward to the body wall. It is fibrous in appearance. Can you find it?

22. The digestive tract is U or V-shaped. It consists of a pharynx (just below the tentacles), a slight constriction, then a short narrow tube, the esophagus, a stomach which is the bottom of the V (called caecum at the very bottom), then the intestine which terminates at the anus, located near the mouth but outside the circle of tentacles. Can you find all these structures?

23. The bottom of the stomach, the caecum, is attached to the body wall by a strand of tissue, the funiculus. The funiculus bears a small fluffy growth or ball of tissue, the testis, which consists of germinal tissue & sperms. The testes may not always be sufficiently developed to be noticeable.

24. The anus opens thru the introvert. Can you find where the intestine is attached to the introvert in a retracted polypide?

25. Bugula is monocious, i.e. has both male & female reproductive organs in the same animal. The testes have already been located. The ovary is a lump or mass of tissue attached to the body wall, more distal than the testes & near the middle part of the body cavity. The animal must be properly oriented for you to be able to see these organs. Do you find the ovary? The egg which is produced is fertilized supposedly by the sperms from the same zoid, then passes out into the ovicell where it developed further.

26. A small knob, the ganglion lies between the upper part of the pharynx & intestine. This is the only easily visible part of the nervous system.

27. Examine the youngest zoids. Are their tentacles of the same length as those of the rest of the zoecia? Is their digestive tract recognizable?

28. Suggested work on Bugula—
   a. Answer questions & make what drawings you can.
   b. Drawings suggested:
      Fig. 14. Habit sketch showing mode of growth of colony
      Fig. 15. Enlarged view of the front surface of the zoid
      Fig. 16. Enlarged view of the back surface of the zoid.
      Fig. 17. An ovicell, enlarged.
      Fig. 18. An avicularium, enlarged.

29. References on Bugula.
   a. Forbes, A. 1933. Conditions affecting the response of the avicularia of Bugula. BIOL. BULL. LXV (3): 469-479. (see its references
DIRECTIONS for PERMANENT PREPARATIONS of
CALCAREOUS BRYOZOA.

Supplies needed:
1. a blowpipe, either of metal or glass, the former preferable.
2. an alcohol lamp or gas burner (Bunsen burner) & matches.
3. An old spoon or crucible for holding tiny specimens
4. binocular dissecting scope, or compound microscope of low power
5. forceps, dissecting needles or small brush, glass slides, labels.
6. very thick balsam or mucilage
7. dry specimens of calcareous bryozoa.

Purpose of exercise:
1. a. To prepare permanent slides of the skeletons of calcareous bryozoa.
2. b. To burn the organic matter which obscures the diagnostic features
3. of the colony.
4. c. To lift the firmly attached colonies from the substratum without
doing too much damage to the colony.

Method of procedure:
1. Have all supplies on hand before starting.
2. Select a good-sized dry rock with a vigorous colony on it.
3. The idea is to blow thru the blowpipe in such a manner that the
fire of the lamp or burner will be bent toward the colony and
focused directly upon it. The colony will turn black at first,
then may become red hot, then turn white. By that time it should
be lifting off or cracking off the rock substratum. Be careful
that rock fragments do not hit you in the eye.
4. Do not burn the colony until it becomes a crumbly snowy white mass
because by that time the calcining will have gone too far and
your colony will be useless. Continue the calcining only as long
as necessary, when some white begins to show and as long as the
colony seems to retain its fine diagnostic pattern without any
crumbling or flakiness. Placing the colony under the microscope
will aid you in determining when to stop.
5. If the colony comes off in chips save these. Gently transfer
them to a slide on which you have placed a thick drop of balsam.
Set this balsam and allow to harden in time. It might
be an excellent idea to mount a natural uncalcined fragment near
the calcined one on the same slide, for comparison. Make slides
of a number of different bryozoan spp. Leave slides at your
4
5
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7
2
3
1
5
6
7
1--blowpipe
2--balsam bottle
3--slide
4--spoon
5--lamp or burner
6--flame
7--bryozoan colony
on rock

1--brow pipe
2--balsam bottle
3--slide
4--spoon
5--lamp or burner
6--flame
7--bryozoan colony
on rock
GLOSSARY

1. ANCESTRULA----The primary zoid resulting from the metamorphosis of a larva. It is the ancestor of the entire colony.

2. APERTURA----An opening into the zoid which is entirely closed by an operculum.

3. AREOLES-----Pores bordering the edge of a calcified frontal wall.

4. AUTOZOID----A normal individual in a bryozoan colony.

5. AVICULARIUM--Avicularia are incomplete, variously shaped individuals. Avicularia may have various shapes: bird's head, oval, elliptical, spatulate, etc. They may occur in various locations: on the front, at the side, and between zooecia; around the apertura in various locations; or on the sides of ovicells, etc. They may be imbedded in the calcareous wall or may swing freely from the zoid.

6. BROOD CHAMBER--Spaces or compartments in which Bryozoan female germ cells and larvae develop. Is a more inclusive or more general term than ooeicum, ooeicum or gonozoid.

7. BROWN BODY----A mass of tissue representing a degenerated or degenerating polypide.

8. FRONTAL SIDE--The uppermost surface of an attached encrusting colony. It is the side on which the apertura is located.

9. FUNDICULUS----A large strand of tissue connecting the aboral end of the alimentary canal with the aboral wall of the zooecium.

10. GONOZOID-----A zoid or individual adopted or transformed for the production of embryos. Another name for ooeicum or ooeicum.

11. HETEROZOID--Any individual which differs distinctly in some way or otherwise from the normal zoids but which because of various reasons like mode of origin, must be considered as homologous with the normal zoids. Heterozoids would include gonozoids, kenozoids, etc.

12. INTERNODE ---The part between joints or nodes of the Cyclostomata. It contains the zooscltal tubes.

13. LOPHOPHORE---The stage or disc surrounding the mouth and bearing the tentacles.

14. LYRULA------A tooth or plate of various shapes or dimensions located at the posterior, inner part of the apertura, in the midline. It protects the opening of the compensatrix. Found in Smittina.

15. MUCRO-------A skeletal bump or cone-shaped growth present on the frontal surface of an zoid, near or before the apertura.

16. NODE--------The joint in a Crisiidae colony.

17. OOECIOPORE--The actual external orifice of the gonozoid.

18. OOECEOSTOME--The tube leading from the ooeiopore into the ooeicum, if the ooeicum happens to be of such a type.

19. OOECEIUM----The brood chamber or ooeicum of the Cheilostomata.

20. OOELEUCUM----A delicate membraneous valve closing the apertura. May be calcified in some spp.

21. OPESIUM-----The uncalcified or more transparent frontal area of a zoid. Part of it is closed by an operculum. When its opening is completely closed or filled by the operculum it becomes the apertura.

22. OVICELL------Brood chamber for developing larvae. Often used synonymously with brood chamber, ooeicum, gonozoid, but is a very inappropriate term for some Bryozoa.

23. PLEUROCYST--A calcareous deposit, often granular, on the zoecia. Often forms the interareolar costules. Rarely porous. Part of the calcified wall of the zoecium. In Smittina & other forms.