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SOME ASPECTS OF THE BIOLOGY OF THE SEASNAIL, <u>LIPARIS ATLANTICUS</u> (JORDAN AND EVERMANN).

University of New Hampshire, Ph.D., 1963 Zoology

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SOME ASPECTS OF THE BIOLOGY OF THE SEASNAIL,

LIPARIS ATLANTICUS (JORDAN AND EVERMANN)

BY

ROBERT DETWYLER

B. S., State University of New York, Teachers College at New Paltz, 1954

M. S., University of New Hampshire, 1959

A THESIS

Submitted to the University of New Hampshire

In Partial Fulfillment of

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Graduate School Department of Zoology June, 1963

Marcal E Javaic George MMoore albin R. Hodydon. Philip J. Samper

1963

This thesis has been examined and approved.

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INTRODUCTION

This study of the seasnail, <u>Liparis atlanticus</u> (Jordan and Evermann), arose as a result of making intertidal investigations related to course work at the University of New Hampshire. In March 1960, three fish were captured and returned to the laboratory for identification. They proved to be specimens of the seasnail, <u>L. atlanticus</u>. This was my first introduction to this species, and they struck me as being most unusual animals. Their general appearance was that of an amphibian tadpole, with large suckers on their ventral surfaces between the bases of the pectoral fins. They were called seasnails, but they were actually fish. A literature search was undertaken to determine what had been recorded regarding this species.

I found that there were many gaps in our knowledge of the life history and habits of the seasnail. The best general account was that of Bigelow and Schroeder (1953). They reported that seasnails were inconspicuous small fish, usually found coiled up under stones or attached by their suckers to some kelp stalk or other submerged object. They have been dredged in water as deep as 50 fathoms, they have been taken in only a few feet of water, and they occur in tide pools. Young seasnails have been found living within the shells of the giant scallop (<u>Pecten magellanicus</u>). Little else appeared to be known of the life history except that it is supposed to move inshore in the winter to spawn. The spawning had not

been observed, nor had developmental stages been recorded.

It was therefore decided to initiate an investigation into the life history of the seasnail. Due to the lack of facilities, it was realized that it would be impossible to study the entire life history. Therefore, this research was limited to those portions of the life history which could be elucidated by an intertidal investigation.

Collections of seasnails were made throughout a three year period and efforts were made to determine its habitat preference in the shore area. I gathered data concerned with its food habits and growth; also, through laboratory and field observations, I investigated reproduction and the developmental stages. Lack of previous work with this species made the investigation of its parasites most productive.



Figure 1. Map of the New Hampshire and southern Maine coast, indicating the collection sites used in this study.

I.	Cape Neddick, Maine	IV.	Rye North Beach, N. H.
II.	Sea Point, Maine	· V•	Rye State Park, N. H.
III.	Fort Stark, N. H.	VI.	Rye Ledge, N. H.

MATERIALS AND METHODS

Collecting

<u>Collecting areas</u>. Although <u>L</u>. <u>atlanticus</u> is found along nearly all of the New England coast, this study was limited to the coast of New Hampshire and southern Maine. Since this expanse of our coastline is characterized by rocky shore areas, it was assumed that seasnails were distributed throughout the extent of this part of the coast. The collections for this study, however, were limited to the six areas listed below (see Figure 1):

I. Cape Neddick, York County, York, Maine,

II. Sea Point, York County, Kittery, Maine,

III. Fort Stark, Rockingham County, Newcastle, N. H.,

IV. Rye North Beach, Rockingham County, Rye, N. H.,

V. Rye State Park, Rockingham County, Rye, N. H. &

VI. Rye Ledge, Rockingham County, Rye, N. H.

These six areas proved to be well suited for the collection of seasnails. All were within reasonable distance of the University of New Hampshire and were accessible throughout the year, with the exception of Sea Point, Maine, which was inaccessible during the snowy part of the winter.

<u>Collecting methods</u>. In the areas selected as collection sites, there were many tide pools and shallow water areas which were exposed at low tide. Therefore, collections were carried out during the periods of lowest tides during each month, when weather conditions permitted. Usually the tides chosen were at least -0.5 feet or lower. Only under these conditions were the large expanses of tide pools and rocks exposed which were most productive.

The animals were exposed by turning over rocks in the tide pools and shallow areas where they were hiding. By using small dip nets, they were easily captured. Often this meant turning over nearly all the rocks in a tide pool to seek out their hiding places. It was, therefore, imperative to have collecting areas in which the rocks were small enough to move easily.

The fish were usually found in only a few inches of water, and only occasionally were they discovered under rocks that were completely above the water level. Also on rare occasions, the animals were seen while they were free-swimming and not hiding under rocks. These occurences were at night and during the periods when large numbers of small fish were in the shore areas at the beginning of the inshore migration.

As the fish were captured, they were placed in a plastic pail, and at the end of the collecting trip they were taken to the laboratory. The water was not aerated, since most of these trips were not extensive. During extremely cold periods, an insulated pail had to be used. This was necessitated by the fact that on one occasion, the water in the pail froze to an inch in thickness around its inner surface. The seasnails, which were attached to the sides of the pail by their sucking disks, were all frozen.

When low tides occurred after sunset, head lamps were used to illuminate the tide pools and to leave the hands free. Collecting at night was advantageous because the fish were not as easily disturbed and were more apt to remain attached to the rocks which made them easier to capture.

The most difficult pert of collecting was recognizing the fish once they were exposed. If they were frightened and swam off, it was difficult to follow them among the rocks. However, on many occasions they would remain attached to the rocks with their tails curled up alongside their heads. To my eyes, at least, they were well camouflaged by their color and shape. At such times, it was most difficult to recognize this blob of material attached to the rocks as a fish. With experience, however, I became accustomed to searching for these fish, and became quite skillful at their capture.

A few individuals were collected from deeper water by accompanying lobstermen as they hauled in their traps. The seasnails would be found clinging to the traps by their sucking disks. This procedure was time consuming and not very productive, so it was abandoned.

Using these collecting procedures, over 450 seasnails were captured during the three year period from March 1960 to March 1963.

Measurements and Counting of Meristic Structures

During this research it was necessary to make measurements and counts of meristic structures. As far as possible, these procedures were carried out in accordance with

methods stated in Hubbs and Lagler (1949). A few of the more pertinent measuring methods are given here.

Total length. Total length was taken as the distance from the tip of the snout to the posterior margin of the caudal fin.

<u>Standard length</u>. This measurement equals the total length minus the length of the caudal fin.

<u>Caudal fin length</u>. The length of the caudal fin was taken from the point of attachment of the caudal rays with the hypural plate to the posterior margin of the fin. To make this characteristic visible, it was necessary to remove the skin from the base of the caudal fin, place it on the glass stage of a dissecting microscope, and examine with a strong source of illumination from below.

<u>Head length</u>. The head length was measured from the tip of the snout to the posterior edge of the opercular flap. This was recorded as a figure representing the number of times the head was contained in the standard length.

<u>Disk length</u>. The length of the disk was taken from the most anterior margin of the outer flap to its posterior margin.

Eye diameter. The eye diameter was measured as the greatest distance across the cornea, that is, between the margins of the cartilaginous eye-ball.

Laboratory Confinement of Fish

The specimens held in the laboratory were confined in a recirculating salt water aquarium system in a constant tem-

perature room. This set-up consisted of a large reservoir tank with three sections. Compartment #1 is a settling tank and #2 contains a charcoal filter. Water moves from #1 to #2 and on to #3 by gravity, and is pumped from #3 to the fish tanks. It left these containers via overflows, spilled onto a large stone sea table, and drained back into the reservoir tank.

The fish tanks consisted of one large aquarium approximately 1 by 2 by $l\frac{1}{2}$ feet and smaller ones 7 by 8 by 24 inches. The smaller containers were of a type which could be partitioned by the insertion of glass plates.

The temperature of the room was regulated so that water temperatures in the aquaria containing the fish were approximately the same as the ocean temperatures $(4-10^{\circ} \text{ C}.)$. Also, at weekly intervals, distilled water was added to the reservoir to counteract evaporation and to keep the salinity of the tank water approximately that of the ocean. Salinities were checked regularly with a hydrometer.

Methods Used in Studying Developmental Stages

All eggs used in this research were naturally fertilized in the laboratory tanks. Attempts at stripping gametes from the fish, according to the methods of Costello, et al. (1957), were unsuccessful.

At first, observations of developmental stages were made from eggs still confined in masses, since individual eggs could not be separated from each other once they had water hardened. Sufficient details could not be observed, however, using this method. Attempts were made to remove the embryos

from the chorion once the blastula had formed, according to the method described by Nicholas (1927), but this could not be accomplished without injuring the embryos.

The most satisfactory method involved obtaining eggs after fertilization, but before they had water hardened. These were pressed under a glass slide in a dish of sea water until the eggs formed a single layer. Eggs treated in this manner could be observed quite easily, and it was then possible to separate individual eggs when desired.

Eggs and larvae were measured under stereoscopic dissecting and compound microscopes. Each contained an ocular micrometer and was calibrated to aid translation of the magnification of various lenses to millimeters. The eggs could only be observed for short periods before being returned to the coolness of the constant temperature room. This was necessary to prevent injury caused by increased temperatures. Usually, photomicrographs were taken at each observation and an egg sample preserved.

Fertilized eggs were kept in clean finger bowls containing sea water. The water was aerated and changed frequently during incubation. They were held at a temperature approximately equal to that of the ocean. Protozoa and fungi contaminated the eggs, but did not appear to cause any mortality, so treatment to rid the eggs of these organisms was not undertaken.

The real difficulties arose after the eggs had hatched. To date there has been very little success in efforts to rear larval fish through their critical period which extends

from the time the yolk sac is absorbed until they are able to maintain themselves on the food in their environment.

When the larvae hatched they were transferred to large plastic pans. It was impossible to place these pans in the circulating sea water system, due to the small size of the fish. Any filter, fine enough to confine them, soon clogged and the pans overflowed.

In all cases, the larvae died during their critical period, a short time after they had absorbed their yolk sacs. Various types of food in different sizes and quantities were tried. Homogenized liver, the freshly hatched larvae of <u>Artemia</u>, diatoms (<u>Thalassiosira nordenskiöldii</u>; a culture was provided by Mr. David Miller from the Biological Laboratory at Woods Hole, Mass.), and small unicellular green algae, were offered the tiny fish, but these foods were not acceptable. Even fresh plankton brought in from the sea failed to provide the proper food for the larvae. I regret to say that this part of the research was left unfinished, for no solution was found to this problem.

Method of Plankton Sampling

Besides taking plankton to feed the larvae, samples were also collected in an attempt to find the larval stages which could not be reared in the laboratory. These collections were made offshore near areas in which adults had been collected. A Turtox general purpose #20 mesh plankton net was used. Some of the samples were packed in ice and returned to the laboratory, while others were preserved immediately

in formalin. Both types of samples were poured into shallow dishes and the larvae removed with a pipette.

Methods Used in the Study of Age and Growth

All of the fish collected were measured and weighed. The total length to the nearest half millimeter and the weight to the nearest tenth of a gram were recorded. The lengthweight relationship was calculated using the method reported in Lagler (1956). When these results were plotted, it made it possible to convert length to weight and vice versa.

For the purpose of aging, attempts were made to use the otoliths since seasnails lack scales. A total of 100 pairs of otoliths were extracted. Of this number, 80 were removed from fish collected in January and February 1962, while the remaining 20 were from specimens taken at various times. It was found that the otoliths were impossible to read, due to irregularities in shape and the lack of clarity of growth rings. A number of fluids were tried in an effort to bring out the details of the zones of growth. Methyl salicylate, oil of clove, glycerine, xylene, distilled water, various concentrations of alcohol, and staining with creosote were tried. Also a series of otoliths were studied under a phase microscope, but no satisfactory results were obtained.

Of all the materials tried, methyl salicylate provided the best results. Each pair of otoliths in a watch glass with methyl salicylate were placed on a dark background. Light from a microscope lamp was directed at them from a low

angle, and the examination was made with a dissecting microscope at magnifications of 20X or 30X.

Both otoliths from each fish were examined. Opaque and translucent areas were seen in the otoliths, but they were not distinct or consistent. Otoliths from a wide size range were examined, but there was no distinct or consistent change noticeable as the fish length varied.

Vertebrae and opercular bones from several fish were also examined, but they proved to be no better than the otoliths.

Although the otoliths could not be used for aging, it was possible to measure the total length of each otolith. Measurements were made with a stereoscopic dissecting microscope with an ocular micrometer. From this data, the relationship between total otolith length and total fish length was calculated by the Lee method described in Lagler (1956).

All computations were carried out on the 1620 IBM computer at the University of New Hampshire Computation Center.

Method of Stomach Analysis

In the present work, the contents of 32 stomachs were examined, 20 from preserved fish and 12 from freshly collected individuals. The stomachs were removed and their contents placed in a watch glass. As a result of adding water and agitating gently, the food particles were separated. Each sample was examined with a dissecting microscope, in an effort to identify the fragmented parts.

Analysis of the stomach contents was carried out using the points method as described by Hynes (1950). On the basis of rough counts and visual judgement, the food items in each stomach were classified as common, frequent, etc. Care was taken to give due regard to the sizes of the organisms as well as to their abundance. Each category was then alloted a number of points and all the points gained by each food item were summed and converted to percentages. This method was essentially volumetric, but it was more rapid and required no special apparatus for measurement.

Method of Clearing and Staining

Five specimens varying in size from 40 mm to 85 mm were cleared and stained by combining and modifying methods prescribed by Hollister (1934), Evans (1948), and Russell and McCandless (1954).

The fish were first hardened in 10 per cent formalin and then transferred to two per cent KOH for varied periods of time, depending on the size of the specimen. After they had cleared sufficiently to make the skeletal parts visible through the flesh, they were transferred to the staining solution, which contained enough alizarin red S in two per cent KOH to produce a port wine color. This proportion varied according to the apparent strength of the staining reaction. After two or three days, when the skeletons were well stained, the specimens were placed in a 1:1 solution of two per cent KOH and glycerine. They remained in this solution for varied lengths of time, until the color bleached out of

the flesh. This usually necessitated several changes of the solution. Finally the specimens were stored in 100 per cent white glycerine.

Specimens treated in this manner, made possible the observation of meristic skeletal structures to confirm the counts made on preserved specimens. Larvae were also treated by this method in order to follow the development of the sucking disk, but due to the lack of different developmental stages, this study remains incomplete.

CLASSIFICATION AND DESCRIPTION

Historical Review

The most comprehensive account of these fishes is the work of Garman (1892) on the Discoboli. This was followed by a revision of the family Liparidae by Burke (1930). Some confusion existed between these two major works. However, Burke (1930), in order to facilitate a comparison of the species described, listed his conception of the species reported by Garman (1892) and other authors.

Garman (1892) combined the families Cyclopteridae, Liparopsidae, and Liparididae, into one group which he called the Discoboli, the disk-bearers. Burke (1930) placed the snail-fishes in the family Liparidae, which replaced the Liparididae. However, Berg (1940) reverted in part to the system proposed by Garman, for he once again combined the three families which Garman (1892) had called the Discoboli, into a single family. However, he gave this group the family name Cyclopteridae. The literature still contains reports using Liparidae as a distinct family for the snail-fishes. The American Fisheries Society (1960) followed the classification of Berg (1940) and did not recognize the family Liparidae.

Prior to 1898, <u>Liparis atlanticus</u> was considered to be identical with the diminutive sucker or Montagu's seasnail, <u>L. montagui</u> Donovan, found on the coast of Europe.

However, in 1898, Jordan and Evermann described this animal as a new species (Jordan and Evermann, 1898). They reported that published figures of <u>L</u>. <u>montagui</u> showed a deeper fish with a larger head and with a very low spinous dorsal fin scarcely distinct from the soft rays. Also Boulenger (in Jordan and Evermann, 1898) stated that in <u>L</u>. <u>montagui</u> the anterior dorsal looks very indistinct, has no detached portion, and none of its rays were ever produced into filaments. Jordan and Evermann (1898) placed the seasnail in the genus <u>Neoliparis</u> Steindachner, 1875, which was distinct from <u>Liparis</u>, and gave the following description of the genus <u>Neoliparis</u>:

This genus differs from <u>Liparis</u> in having a deep notch in the dorsal fin anteriorly, separating the spines from the soft rays. The species approach more nearly the Cottoid type, from which the Liparids are decended. In general the vertebrae are fewer, the fin rays fewer, the ventral disk larger, and the vertical fins better separated than in the more generate members of the family. The retention of the notch between the dorsals fully justifies the recognition of <u>Neoliparis</u> as a distinct genus.

Garman (1892) stated that the characters assigned by Steindachner to distinguish the genera <u>Neoliparis</u> and <u>Liparis</u> were not sufficient reason for separation. Burke (1930) agreed with Garman, for he believed the notch in the dorsal fin which distinguished this genus was not of specific value, since the notch is present in some specimens and absent from others. Therefore, Burke (1930) listed the seasnail as <u>L</u>. <u>atlanticus</u>, which is in agreement with the list of the American Fisheries Society (1960). After studying over 400 specimens of this species, I agree with these authors in not recognizing the genus Neoliparis.

Generic Description

The characteristics of the genus <u>Liparis</u> (Artedi) Scopoli, were summarized by Jordan and Evermann (1898) and Burke (1930).

Body rather elongate. Skin smooth; head short, flattened above; mouth horizontal, the jaws equal or the lower jaw included; teeth are trilobed and arranged in oblique rows; anterior nostrils tubular or not; ventral disk, its surface with 13 lobes; vent midway between the sucking disk and anal fin; dorsal fin continuous, undivided, its spines not differentiated; caudal well developed; dorsal fin free from the caudal or joined; pectoral fins broad, procurrent at base, emarginate and free at the tips, some of the lower rays produced; vertical fins enveloped in the lax skin; vertebrae 35 to 55.

Garman (1892) also gave a complete description of the genus <u>Liparis</u>. His description included characteristics of the internal anatomy, as well as the external features of this group.

Liparis atlanticus (Jordan and Evermann) 1898

The original description of <u>L</u>. <u>atlanticus</u> is recorded in Jordan and Evermann (1898, p. 2107), under the species <u>Neoliparis atlanticus</u> (Type - Male, No. 37215, U. S. N. M. Godbout, Quebec, 1885).

Subsequent descriptions of this species are found in Burke (1930), Bigelow and Schroeder (1953), and various brief

accounts in Breder (1929), and Perlmutter (1961).

TABLE 1. Measurements and Counts of Meristic Structures Reported for the Holotype of <u>L</u>. <u>atlanticus</u>.

	Jordan & Evermann (1898)	Burke (1930)	Lachner ⁸
Dorsal Anal Pectorals Head* Disk** Eye** Snout** Caeca Vertebrae	VI,25 23 30 4.7 1.75-2 5 3	34 27 28 4.5 1.8 5.6 30	34 27 29 4.6 1.74 6.5 39
Total Length	about 5"	109 mm	TOP mm

* - in standard length.

** - in head length.

a - Dr. Lachner provided an x-ray, photograph, and measurements of the holotype from the U. S. National Museum. This information is summarized here, as well as, the vertebrae count made from the x-ray.

Table 1 presents a summary of the measurements and counts of meristic structures reported for the holotype. There are some variations, but these are undoubtably due to the technique of the worker and changes which have taken place during preservation.

Synonyms and References

Liparis montagui, Garman, 1892, p. 47 (pl. VII, figs. 6-20, and pl. VIII, figs. 8-11).

<u>Neoliparis atlanticus</u>, Jordan and Evermann, 1898, No. 47, p. 2107.

More complete lists of synonyms and references, including those for <u>L. montagui</u>, are presented by Garman (1892), and Jordan and Evermann (1898).

Common Names

L. atlanticus, like most animals, is known by a variety of common names. Among these are New England sea-snail, dusky seaslug, snail-fish, sea-snail, and others. However, the recent publication of the American Fisheries Society (1960), A List of Common and Scientific Names of Fishes From the United States and Canada, listed seasnail as the accepted common name for <u>L. atlanticus</u>. It has been called the sea-snail in a number of previous works, but now its name is no longer hyphenated.



Figure 2. Seasnail (<u>Liparis atlanticus</u>), lateral view of an adult male of 88 mm in total length.



Figure 3. Seasnail (<u>L. atlanticus</u>), lateral view of an adult female of 87 mm in total length.

ADULT MORPHOLOGY

General

The following discussion of adult morphology is based principally on the examination of 55 preserved specimens (25 males and 30 females), and 20 fresh specimens (9 males and 11 females). Various immature individuals were examined to confirm the existence of growth differences due to age. These fish were not from a single collection, but were captured at various times and locations.

Major Adult Characters

Body (Figs. 2 and 3). The general body form is that of an amphibian tadpole. The body appears heavy anteriorly, moderately elongate, and tapering behind the abdominal cavity to the laterally compressed tail region. The skin is loose and naked.

Head (Figs. 2 and 3). The head appears blunt and the snout is truncate. The upper jaw protrudes slightly past the lower. The mouth is horizontal and terminal. The premaxillaries are protractile. The width of the head is greater than the depth. In profile, the head is slightly depressed over the eyes. The lips are distinct and interrupted by pores, which give them a scalloped appearance.

The head length in the standard length ranged from 3.6 to 4.3, with an average of 3.8 (see Table 2). From this data, it was observed that the head length changed with age, and differential growth existed. The head length varied from 3.6 to 3.8 for fish under 80 mm in total length, and 3.9 to 4.3 for those from 80 to 97 mm.

<u>Nostrils</u> (Fig. 4). There are two pairs of nostrils. The anterior nostrils are tubular and extend above the surface of the head at a tangent anterior to the eye. The posterior nostrils are more slit-like and project only slightly above the surface above the anterior part of the eye.

Eye (Figs. 2 and 3). The eyes are small. The eye diameter is reported as the number of times it is contained in the head length. The eye varied from 5.4 to 6.5, with an average of 6.2. The eyes displayed a slight differential growth, since the eyes tended to be larger in relation to the head length of the smaller fish.

<u>Pores</u> (Fig. 4). The lateral line is reduced to a series of pores on the head. The pore formula is designated as 2-6-7-2, representing the pores on one side of the head. Their description here varies only slightly from that reported by Garman (1892). There are two pores on the dorsal surface of the snout, one near the anterior edge and the other is on a diagonal between the first pore and the anterior nostril. Six pores, the maxillary series, are found in the edge of the fold just above the upper lip. Three of these pits are in the edge of the fold, forming notches. The fourth is at an angle anterior to the eye, the fifth is on the cheek posteriorly angled from the eye, and the last pore of this series is posterior to the eye. On each side of the lower jaw, the



Figure 4. Lateral view of the head of an adult seasnail, showing the characters of the head region.



Figure 5. Ventral view of an adult seasnail, showing the location and external features of the sucking disk. (Approx. 5 times natural size) seven pores of the mandibular series extend backward and curve up the sides of the head to the level of a line drawn from the eye to the gill slit. The last pore of this series is slightly posterior to the midpoint between the gill slit and the eye. The six other pores form a line along the lower margin of the side of the head, and along the lower lip producing a notched appearance. There are two suprabranchial pores above the gill slit.

Rudimentary pores were often present in the region of the lateral line or scattered in the region of the gill slit and head. Among the specimens I examined, these were always closed and resembled small white papillae.

Sucking disk (Fig. 5). One of the most unusual characters of the seasnail is the sucking disk, which is located on the ventral surface between the bases of the pectoral fins. The diameter of the disk is contained from 1.4 to 2.0 times in the head, with an average of 1.8. The proportion of the disk diameter to the head length also varied with age, being larger in younger individuals. A juvenile of 13 mm in total length had a disk 0.5 the length of the head, while one of 97 mm had a disk measurement of 1.4.

The disk has been reported to develop from the pelvic fins. An attempt was made to verify this by clearing and staining larvae of varied developmental stages, but because I was unable to rear larvae through to metamorphosis, it was impossible to complete this study. The smallest larva I was able to collect in the plankton had a total length of

24 -

8 mm and they had fully formed disks already. These individuals were observed to have present the 12 rays which support the body of the disk. In addition to the 12 rays mentioned above, there are 13 papillae or pads, one opposite each ray plus a single larger one located centrally at the anterior end of the disk. The outer margin of the disk consists of a flap, which has two notches in the anterior portion.

<u>Gill slit</u> (Fig. 4). The gill slits are reduced and restricted to the sides of the head above the bases of the pectoral fins. Nearly all of the fish examined had the gill slit above the pectoral fin; in only a few cases did it extend down in front of the first pectoral ray.

<u>Teeth</u> (Fig. 6). The teeth are trilobed and arranged in eight or nine oblique rows in each half of the jaw.



Figure 6. Teeth from the upper jaw of L. atlanticus. 60X.

Dorsal fin (Figs. 2 and 3). The dorsal fin is continuous and connected to the caudal fin. The number of rays varied from 31 to 33, with an average of 32. The dorsal rays are unbranched and the anterior five or six rays have their tips projecting above the fin membrane.

There is a further discussion of the dorsal fin under
sexual dimorphism.

Anal fin (Figs 2 and 3). The anal fin is continuous and is connected to the caudal fin. However, its area of attachment to the caudal fin was always greater than that of the dorsal fin. The anal fin rays are unbranched and varied from 24 to 26, with an average of 25.

<u>Caudal fin</u> (Figs. 2 and 3). The caudal fin is distinct and truncate. The average number of rays forming the body of the caudal fin was 10, varying from 10 to 12. After clearing and staining, many short rudimentary rays were seen on each side of the base.

<u>Pectoral fins</u> (Figs. 2 and 3). The pectoral fins are large, broad, and fan-shaped. The number of rays ranged from 28 to 30, with an average of 29. The pectoral fins are notched, with the lower lobe containing seven rays. The rays usually extend beyond the fin membrane, giving them a fringed appearance. This is usually more pronounced during the breeding season.

<u>Pyloric caeca</u>. The pyloric caeca fan out like a hand of bananas from the pylorus, but do not completely encircle the duodenum. They vary in length and are roughly in two staggered rows, with the majority on the right side. The number of pyloric caeca varied from 26 to 34, with the average of 30.

Peritoneum. The peritoneum was silvery white, with a scattering of stellate black chromatophores.

Vertebrae. The average number of vertebrae was 38. Vertebrae counts were made from 5 cleared and stained speci-

mens, and only one had 39 vertebrae.

<u>Coloration</u>. Coloration in a general way is correlated with the environment. Fish collected from among the rocks were black, gray, or dark brown. However, in the laboratory tanks they took on olive, reddish-brown, or a light tan color, which blended with the algae, stones, and bricks which were present.

The base colors of black, olive, gray, and shades of brown were usually altered by bars and blotches of various colors. The fins were frequently barred with white, blue, or pink. Often the body tended to have blotches of these colors, particularly in the area of the gill slit and top of the head.

Length. Seasnails are small fish and are generally believed to rarely reach a length of more than 100 mm. In this study, the longest specimen examined was 97 mm in total length.

TABLE 2. Synopsis of the Characters of L. atlanticus.

	A*	B*	C*
Fish examined	?	36	75
Dorsal	28-33	32-34	31-33
Anal	23-27	25-27	24-26
Pectoral s	29-30	26-28	28-30
Caudal	14-17		10-12
Head ·	4.6		3.8
Disk	1.5	1.8	1.7
Еуө	6	5.6-6.5	5.4-6.5
Snout	3		
Caeca	22	19-37	26-34
Vertebrae	38-39		38-39
* A - fr	om Garman	(1892).	
B - fr	om Burke	(1930).	
C = fr	om this s	tudy.	

DISTRIBUTION

General Range

Since <u>L. atlanticus</u> are marine fish, they are not primarily restricted in their distribution by land barriers, aside from the continents themselves. The important factor limiting their dipersal is temperature. Other major distributional influences are variations in salinity, the deep ocean waters, the effects of ocean currents, and the nature of the coastline.

The seasonal variations in water temperature are marked in the northwest Atlantic. The meeting of the cold Labrador Current with the warm Gulf Stream produces an abrupt change from frigid to subtropical conditions at varying distances off the coast of North America. The Labrador Current extends southward in-shore from the Gulf Stream, and its frigid waters are particularly evident in the Gulf of Maine.

Of the three main zoogeographical categories listed by Lagler, et al. (1962), the seasnails would be members of the shore or shelf fauna. This means that they are occupants of the continental shelf from the intertidal zone to its outer edge.

Considering the preceding factors, <u>L</u>. <u>atlanticus</u> is limited in range to the shallow waters fringing the northwest Atlantic, and occurs in greatest numbers in the Gulf of Maine. Burke (1930) believed that the southern distribution was limited by the summer isotherm of 60° F., whereas, Putnam (1874) gave the southern limit as 41° north latitude. The western boundary is the North American Continent, while on the east, the deeper waters of the Atlantic Ocean provide the barrier. Burke (1930) and Bigelow and Schroeder (1953) reported the vertical distribution from 0 - 50 fathoms. The northern limit of distribution is not so well defined, since there are scattered reports of specimens found above the coast of Newfoundland, which is believed generally to be the northern limit.

Specific Range

The range of the seasnail as reported by Bigelow and Schroeder (1953) is along the rocky shores of the North American Coast from northeastern Newfoundland, the northern part of the Gulf of St. Lawrence, and the Grand Banks to southern New England. It is rare west and south of Cape Cod. However, there are reports from Woods Hole, the coast of Connecticut, and off New Jersey.

In the Gulf of Maine, Bigelow and Schroeder (1953) reported its occurrence from Yarmouth, Nova Scotia, the Bay of Fundy, Passamaquoddy Bay, Grand Manan, Eastport, Seguin Island, off Portland, off Cape Elizabeth, at Kittery, and at various localities about Massachusetts Bay. Gordon and Backus (1957) reported three specimens from Labrador and Dunbar and Hildebrand (1952) collected one specimen from Ungava Bay. These reports indicate that the northern limit of the range will probably be extended with further invest-

igations of the northern areas.

Habitat

In the intertidel zone of the area covered by this study, the seasnails were found in tide pools along the rocky portions of the shore. Their habitat was the littoral and sublittoral pools. Littoral pools, as described by Hedgpeth, et al. (1957), are those which are regularly covered by the sea at high tide and essentially disconnected at low tide; while sublittoral pools are those which are never completely shut off from the ocean. The seasnails hide under rocks and cling to vegetation, within these pools. Specimens collected from lobster traps were from rocky areas with expanses of kelp.

It could be assumed that they would seek out a similar habitat in deeper water. However, Gordon and Backus (1957) reported that two specimens were captured in a beam trawl in 4-8 fathoms, on hard sandy mud bottom, and one specimen was seined along shore at low tide from a sand bottom. This indicates, perhaps, a different habitat preference when living in deeper water and in other parts of its range.

Seasnails were found only on the open coast, they were never encountered in brackish areas. In the laboratory they appeared capable of withstanding a considerable range of salinity. The salt content of the sea water in one of the holding tanks varied at times from 28 parts per thousand to 38 parts per thousand without appearing to cause discomfort to the seasnails.

Temperature appeared to have the greatest overall effect on the habitat selection. When the water temperature rose above 12° C., which is reached by the ocean in this area in June, large seasnails became rare in the intertidal zone. Larvae and a few small immature fish could be found occasionally. Not until the ocean temperature had lowered to 12° C. again in October, did they once more return to the shore in numbers.

Associations

Bigelow and Schroeder (1953) reported young seasnails inhabiting the shells of the scallop (<u>Pecten magellanicus</u>). During this investigation, seasnails were not observed to have any close associations with other organisms of the intertidal zone. The seasnails appeared to have a reciprocal migration with the rock gunnel, <u>Pholis gunnellus</u>, and the radiated shanny, <u>Ulvaria subbifurcata</u>. As <u>L</u>. <u>atlanticus</u> moved into the intertidal zone, these two species disappeared and were assumed to have moved out to deeper water. In the spring as the seasnails left the shore, they reappeared. This was most likely caused by differences in temperature preferences. Observations made incidentally while collecting did not indicate any other associations, except with the organisms utilized as food.

FOOD AND FEEDING

No food studies have been recorded for <u>L. atlanticus</u>. Bigelow and Schroeder (1953) presumed that seasnails feed chiefly on small crustaceans and small shellfish as does its European relatives. Garman (1892) reported that the contents of the stomachs of seasnails revealed miscellaneous lots of small marine animals (crustaceans, worms, mollusks, small fishes, etc.), mixed with quantities of seaweed.

This study was based on the examination of the stomach contents of 48 fish (35 preserved and 13 fresh specimens). The food items and their percentage of the diet are given in Table 3.

TABLE 3. Summary of the Stomach Analysis of L. atlanticus. Total lengths of fish examined 65mm - 88mm 48 Number of stomachs examined Number with food 10 83 Percentage containing food Food items No. of stomachs Percentage containing item of diet Crustaceans 10 Gammarus locusta 32 35 25 20 Jayra marina 18 Unidentifiable remains 36 10 Polychaetes Unidentifiable remains* 40 14 6 Plant material ** 5 Unidentifiable debris 8 ЛΟ *- Two worms were believed to be Harmothoë imbricata.

*- Two worms were believed to be Harmothee impricate. **- Believed to be Chondrus crispus.

Of the stomachs examined, crustaceans made up nearly 75% of the total diet, with polychaetes as the other major food item. It was impossible to identify any of the worms present, but all stomachs containing food had polychaete setae present in the debris. Only six individuals had been feeding on vegetation, this was probably <u>Chondrus crispus</u> (Irish moss), but it could not be positively identified. However, fish in laboratory tanks browsed on the <u>C. crispus</u> present.

There is some indication that seasnails feed in the early morning and evening. The stomachs of fish collected at these times were full or nearly so. The eight stomachs that were empty were from fish collected in the afternoon. However, the laboratory fish fed whenever food was offered.

THE LENGTH-WEIGHT RELATIONSHIP

Weight of fishes may be considered a function of the length. Rounsefell and Everhart (1953) stated that this relationship of the length and weight follows approximately the cube law relationship expressed by the formula, $K = W/L^3$, in which W = weight and L = length. However, a fish is continually prone to change its body proportions as it grows older so that the simple cube law does not hold throughout the life history and growth of the fish.

In the present study, the length-weight relationship was expressed by the general formula $W = aL^n$, after Lagler (1956). In this equation W = weight, L = length, <u>a</u> is a constant, and <u>n</u> an exponent. Values for <u>a</u> and <u>n</u> were determined empirically. All length and weight data were transformed to logarithms. The logarithmic form of the above formula is log W = log a + n log L. The values of log <u>a</u> and <u>n</u> were determined as follows:

 $\log a = \underline{\Sigma \log W} \cdot \underline{\Sigma (\log L)^2} - \underline{\Sigma \log L} \cdot \underline{\Sigma (\log L \cdot \log W)}$ $N \cdot \underline{\Sigma (\log L)^2} - (\underline{\Sigma \log L})^2$ $n = \underline{\Sigma \log W} - (\underline{N} \cdot \log a)$ $\underline{\Sigma \log L}$ (N = no. of individuals in sample)

By substituting the calculated values for log <u>a</u> and <u>n</u> into the formula log $W = \log a + n \log L$, calculated weights were determined by obtaining antilogs of the calculated log W for each individual. Several points, representing a variety of calculated weights, were selected (20 males, 20 females,

and 12 immatures), and used to plot the curves in Figure 7. The resulting curves provide an estimate of the probable weight of a fish of any given length and vice versa.

No single sample was collected of sufficient number to permit a length-weight analysis. It was then decided to base this relationship on the collections of an entire year, but again the size of the sample was not significant. Therefore, in the present study of the length-weight relationship, the data used was based on the fish collected from March 1960 to December 1962. Samples of each sex and the immature individuals were treated separately (118 males, 133 females, and 130 immatures).

Due to the difficulty of obtaining adequate samples throughout the year, the use of this relationship is rather limited. The length-weight relationship is probably not constant throughout the year, and is affected by such factors as availability of food, rate of feeding, maturation of the gonads, and spawning. Morrow (1951) suggested that these factors must be considered in determining the true length-weight relationship. The majority of the fish used in this study were collected during the periods of gonad maturation and spawning, therefore, its results cannot be considered as showing the typical length-weight relationship. Instead, it is the relationship that exists only during the reproductive phase of the yearly cycle.

In mature fish, ripening gonads may account for a considerable portion of the weight. But after spawning, the weight should drop without any change in length. Under these

conditions it would not be expected that the length-weight relationship would remain unchanged.

These fish were collected intertidally, and the possibility exists that their feeding habits are different durin the part of the year that they spend in deeper water. Also, their rate of feeding may be altered during the spawning period.

It seems apparent, then, that a comprehensive analysis of the length-weight relationship can be based only upon a sufficiently larger series of samples spread as evenly as possible throughout the year.



Figure 7. The average length-weight relationship for <u>L. atlanticus</u>. For solving the equations above use Naperian logarithms.

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AGE AND GROWTH

The three general methods used for determining the age of fishes are the length-frequency method, the marked fish method, and the bony structure or scale method.

The length-frequency method is based on the fact that the lengths of fish of one age tend to form a normal distribution, and that in the total length-frequency curve of the distribution of all ages of the population, the various year classes will appear as individual modes.

It was impossible to employ the length-frequency method in the present study, since the basic prerequisites for using this method, as stated by Lagler (1956), could not be met. It was not possible to collect large enough samples within a restricted period of time which were made up of an adequate representation of all of the size and age groups in the population.

The marked fish method involves the liberation and recovery of marked or tagged fish of known ages. This method was not considered, since fish of known ages were not available.

The bony structure or scale method involves the interpretation of the layers deposited in the hard parts of fish. In actual practice, the scales have proved the most useful structure in studies of age and growth, but several bones and the otoliths have been used successfully, too. However, seasnails are scaleless, so otoliths were used in this study. The otolith used was the saccular otolith or sagitta, which is the largest of the three otoliths present on each side of the head in the sacculus of the inner ear. The otolith is laid down in concentric layers, a process which is probably going on at all times. Unknown factors, possibly connected with food or season, cause variations in structure which produce definite laminations which appear as opaque and translucent areas when viewed after proper treatment and under diffuse light.

In determining age and growth from otoliths, it must be established that the rings observed are annual, or represent some other definite period of time. Also, all or most of the fish in the population should form a ring at the same time. A relationship should exist between the length of the fish and the length of the otolith.

In seasnails, the otoliths were impossible to read, due to irregularities in shape and the lack of clarity of growth rings. Opaque and translucent areas were seen, but they were not distinct or consistent. Otoliths from a wide size range and samples from various times of the year were examined, but there were no distinct or consistent changes noticeable as fish length and time of capture varied. It was not possible to determine, therefore, the annual nature of the growth zones or whether the rings were formed at the same time in all individuals.

The relationship between otolith length and fish length was determined by using the Lee method, as described

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in Lagler (1956). The general formula is L = a + cS, where L = fish length, S = otolith length, <u>a</u> and <u>c</u> are constants. In this method, a modification of a direct proportion is assumed to exist between otolith and body growth. The resulting regression formed a straight line, which is shown in Figure 8. The coefficient of correlation was 0.92, which indicated that the otolith does grow at a proportional rate with the body. It is unfortunate that easily seen concentric rings are not produced within the otolith as it increases in size.

This graph is only an approximation of the relationship between fish length and otolith length since the regression line did not pass through the 0, 0 intercept. There are several factors which could be responsible for this variation. The samples on which this study was based were not all collected at the same time and were not of sufficient variation in fish length. There were three times as many fish above the length of 50 mm as there were smaller individuals, which would provide a "top heavy" sample. This would alter the result, since Lagler (1956) stated that the values for a increase successively with age. Thus age and length distribution of the sample must influence estimation of the constant. Whitney and Carlander (1956) stated that temperature, parasitism, and perhaps other environmental factors may modify the body-scale relationship, so it is possible the same is true in the case of otoliths.

Since no satisfactory method of aging was found, it

was impossible to determine age classes and to further investigate the growth of the seasnail.

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Figure 8. The regression line for the body length to otolith length for L. atlanticus.

REPRODUCTION

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General

Our information concerning the breeding and spawning habits of <u>L. atlanticus</u> is very incomplete. To the best of my knowledge, no studies have been made directly on this subject, and the little information that is available is contained in the works of Putnam (1874), Burke (1930), and Bigelow and Schroeder (1953). This material is concerned with the fact that seasnails move inshore during the winter to spawn, and varied reports of the spawning time.

Information regarding the reproductive phase of this study was gathered incidentally while collecting and while observing individuals maintained in laboratory tanks. The material presented here was compiled from the collection and study of individuals over a three year period, accompanied by laboratory observations through two spawning periods.

Sexual Dimorphism

Among the male seasnails, the anterior five or six dorsal rays are elongated and fleshy during the breeding season (see Fig. 2). The body is also covered with "thumb-tack" prickles which are nuptial tubercles such as are found in several other species of fish. The prickles consist of minute styliform spines with round flat heads which are embedded in the skin (see Fig. 9). The prickles are absent from the ventral surface of the body, but are distributed over the dorsal and lateral surfaces of the head, body, and on to the fins. Burke (1930) stated that the only other species of <u>Liparis</u> showing this character is <u>L. rutteri</u> of the Pacific coast of North America.



Figure 9. Skin of male, showing the breeding tubercles. From Garman (1892). 20X

Burke (1930) stated that in 1904 Jordan and Snyder proposed a new genus based on the presence of "thumb-tack" prickles. This was good evidence that these structures had escaped serious consideration previous to this time. They had been noted and figured, however, by Lütken (1886) and Garman (1892). Jordan and Evermann (1898) described the seasnail as having small, scattered, light to bluish dots over the body. These were probably the bases of the prickles which they failed to notice.

The females lack the prickles. Some do show, however, an elongation of the anterior dorsal fin rays, but to a lesser degree than the males. During the breeding season, the females are more pot-bellied than the males, particularly when they are ripe or near-ripe for spawning. Because of the relatively large quantities of eggs which the ripe female con-

tains, the distended body wall in the gonadal region appears pink or light orange due to the pigmentation of the eggs showing through.

Sexual Maturity

Because no satisfactory method of aging was found, the age at which individuals reached sexual maturity can only be determined by inference. However, there is evidence which indicates that sexual maturity is reached when a total length between 60 to 70 mm is attained and the fish are entering their second year.

Of all the fish collected, none with a total length of less than 60 mm showed any indication of sexual dimorphism, and the gonads did not reveal any outward signs of maturation. The males and females among the smaller individuals could not be differentiated externally.

Individuals which ranged from 60 to 70 mm in total length showed definite maturation of the gonads and sexual dimorphism became apparent. At this point in the study, females were used to follow gonad maturation since the changes in the ovaries could be determined more readily than those progressing within the testes of the males. As the spawning period approached, the females in this length range had ovaries which became more granular in appearance and started taking on a light pink coloration. All the eggs in the ovaries, except the apical area of the general egg stock, matured at the same rate as shown by an increase in size and pigmentation. The eggs were of two sizes, the maturing eggs were 1 mm in diameter, while all others were 0.1 mm. No intermediate sizes were present. This homogeneity of egg size in these smaller ovaries was interpreted to be the characteristic appearance at the time of the first spawning. Although these individuals were observed during the spawning season, they produced eggs only once. Ovarian examinations of larger individuals showed no sharp separation between the general egg stock and the maturing eggs. Several stages of egg maturation were present in the ovaries. Eggs were of several sizes, the diameters were 0.1, 0.2, 0.5, 0.7, and 1.1 mm. These larger specimens were also observed to spawn several times in one spawning period.

Considering gonad maturation and the lengths of individuals collected throughout the year over a three year period, fish in the 60 to 70 mm total length range appeared to be entering their second year. However, this could not be substantiated through actual aging. Tosh (1894) believed that females of <u>L</u>. <u>montagui</u> with an age of one year plus 10 months were bearing eggs for the first time.

Spawning Season

The spawning of <u>L</u>. <u>atlanticus</u> has never been observed in its normal habitat. Bigelow and Schroeder (1953) stated that spawning probably takes place from March until midsummer in the Gulf of Maine. Perlmutter (1961) believed it to spawn in the winter, while Putnam (1874) recorded that the spawning season was in March.

In this study, the plotting of fish length against

the numbers collected indicated that the seasnails have a definite spawning period (see Fig. 10). These graphs, starting in June, show that small fish were collected along with the last few large spawning fish still inshore. In subsequent months throughout the year, the peaks of the curves shifted slightly to the right during the late fall, winter and spring. I feel that this shift to the larger size ranges indicated a definite spawning period. If the curves showed no tendency to shift to the right, it would indicate a situation similar to that found by Turner (1938) in which the curves for Brachyrhaphis episcopi, a poeciliid fish, showed no tendency to move to the right during any season and it was assumed that it maintained its position by a continual new supply of young. Therefore, there was no definite spawning period but continual reproduction throughout the year. However, this is not the case with the seasnails; there is a definite egg laying period.

The results of this investigation indicated that the spawning season extends from midwinter to midsummer. Females swollen with eggs were collected during late January and early February, and others ready to reproduce continued to inhabit the intertidal zone throughout the spring until June. Other indications of an extensive spawning period are the sizes of larvae and immature forms which have been observed. Huntsman, as reported by Bigelow and Schroeder (1953), found larvae in Passamaquoddy Bay as early as April, while they towed one little seasnail only 7 mm long on German Bank as



Figure 10. Graphs showing the relative numbers of fish collected of each length group for each month over a three year period.

late as September 2. It is possible, however, that this specimen could have been <u>Liparis liparis</u>, the striped seasnail. I have collected only two individuals of this species, but both of these were collected offshore. Juveniles of 10.5, 13 and 14 mm in total length were collected in July and August, which indicates that they were spawned in March or early April. However, during this same period many larvae were collected in plankton samples. These ranged from 4 to 8 mm, and most likely were spawned in the middle and late spring.

Individuals maintained in laboratory tanks first spawned in February and continued until May. However, since these fish were not kept under natural conditions, this cannot be used as a definite indication of the spawning period. But this data does correspond to other reports, and I feel it is indicative of the extent of the spawning period.

Another indication of a protracted spawning period is the fact that the only egg masses collected in tide pools were found in May, and they were in an early stage of development. Also a male still bearing prickles was collected in August.

Spawning Migration

Throughout its range, the seasnail is considered a resident species, undertaking no considerable migration north or south along the coast. However, its abundance varies in a definite cycle throughout the year, apparently as a result of a more or less definite onshore and offshore

movement (Bigelow and Schroeder, 1953; Burke, 1930).

The number of seasnails collected intertidally declined after June, and only a few small immature individuals were found during the summer months and early fall. During October there was a marked increase in the number of seasnails frequenting the intertidal area. The majority of these early arrivals ranged in total length from 40 - 60 mm. This group consisted of immatures and a few small males. The arrival of these smaller fish marked the beginning of the inshore or spawning migration. This is most likely a short migration since seasnails are not strong swimmers and it would seem improbable that they would be capable of traveling any great distance. Starting in October, they are continually found in considerable numbers in the intertidal zone, except during periods of extreme cold or strong surf, until the last spawners disappear in June.

This inshore migration has a considerable effect on the numbers and sizes of fish captured in the intertidal zone. Figure 10 shows graphically how the numbers of these inhabitants of the tide pools vary throughout the year.

However, these graphs cannot be interpreted as a true picture of the intertidal phase of the migration. They are only indicative of the data available from this study. Collections made in the shore area are dependent upon the tides and weather conditions. Therefore, the amount of collecting is varied throughout these periods, and this has a direct effect upon the numbers of fish captured. As previously stated, the seasnails move out into deeper water during

periods of extreme cold and rough surf. This was especially true during the months from December to March. The interpretation of Figure 10 must be carried out with these facts in mind.

The greatest number of fish was collected during October and November. However, the conditions for collecting were usually favorable at this time. The peak in the total length range for this period was from 50 - 59.9 mm. In December, the size range shifted to the 60 - 69.9 mm group, and there was an increase in the number of maturing females. Near the end of December, there was nearly an equal number of males and females in each collection. Also, there was a decrease in the number of fish collected. In January and February, there was an increase in the number of larger fish, and the smaller females were swollen with eggs. Through the spring the number of fish captured was reduced, but there was a shift to the larger size ranges and there were increased numbers of females.

Therefore, the overall results of the migration caused an increase in the number of immature fish frequenting the inshore spawning areas early in the fall. They were soon displaced by the smaller adults which were spawning for the first time. As time passed, this group spawned and was replaced by reduced numbers of larger fish. The study of the spawning habits of the larger and older individuals in the laboratory has shown that as size increases they spawn more than once. This helps explain the fact that the larger females stay in the spawning areas for a longer time. This is shown in Fig. 10 by the fact that after March there was very

little change in the size range until the spawning period was over and the adults left the spawning areas.

Spawning Behavior

Unfortunately spawning was not observed in nature, but it was seen among fish confined in laboratory tanks. In February 1961, fish were placed in laboratory tanks to determine if they would spawn under artificial conditions. These individuals ranged in size from 60 - 70 mm in total length. Most of the group was separated into pairs (one female and one male) and placed in partitioned aquaria, but no spawning occurred. However, on March 2, two small egg masses were found on the bottom of a large tank containing two small females and one male. These individuals had total lengths of 60 - 64 mm. These eggs proved to be fertile, and they hatched after an incubation period of 27 days at 7.5° C.

Since the larger fish which had been separated into pairs failed to spawn after a period of six weeks had passed, attempts were made to strip the eggs from the females and fertilize them artificially. However, this method was unsuccessful.

During May of this same year, four females bulging with eggs and ranging in size from 70 - 83 mm were collected. These were placed in a tank with a single male from a previous collection. In a period of three days all four of the females spawned. The eggs of the four masses also proved to be fertile and hatched after 22 days at 9° C. On none of these occasions was actual spawning observed.

In February 1962, laboratory attempts were made once again to observe spawning. After the experiences of the previous year, no attempt was made to segregate pairs, and all fish were placed in large tanks with a sex ratio of about three females to each male. Stones on which Irish moss, <u>Chondrus crispus</u>, was growing were also placed in the tanks.

On February 16, one female was observed to be quite active while all of the remaining fish in the tank were curled up and attached to the stones and sides of the tank which was their usual habit. Frequent observations over the next three hours showed the same female continuing her activity. She would swim rapidly to the surface with all her fins expanded and continue along the surface with her snout breaking the surface of the water. Following this she would dive vertically to the bottom of the tank at which time she would brush along the sides of one particular male and on several occasions bit at his tail or fins. The male soon expanded his fins, especially the elongated rays of the anterior portion of the dorsal fin.

Four hours after the female started her initial advances, the male became quite vicious and drove all of the other fish from one of the stones by biting their fins and butting them with his snout. After this was accomplished he began to nibble at and butt his snout against a holdfast of the Irish moss attached to this particular stone. This cleaning procedure was accompanied by much fluttering of the fins and was interrupted several times to make short swimming

excursions with the female which involved biting and fin flapping. This continued for approximately one-half hour.

Finally the male ceased his activity, and took up a position next to the spot he had prepared. At this point all of his fins were expanded to their fullest and he began to quiver. The female by now had a prominent bulge in the area of the vent. Shortly after the male began to quiver, she swam over the area prepared by the male and deposited a small mass of eggs. The male wriggled violently around the eggs and it was assumed he was fertilizing them although no milt was visible. He then nuzzled the egg mass against the holdfast and tamped them down with his snout. Four more spawning passes were made by the female over a period of about an hour. In this manner the egg mass was built up. After this the female became inactive.

After the egg mass was deposited, the male took a position along side of it and fanned it with his fins. For the next several days he guarded the eggs by violently attacking any intruders. This protection of the eggs by the male is similar to that reported for the lumpfish, <u>Cyclopterus lumpus</u>, by McIntosh and Masterman (1897). However, his attention to the egg mass lasted for only a few days. In nature it may continue longer since the laboratory conditions were quite unnatural. There was a limited area for spawning and this led to considerable fighting among males for selection of spawning sites.

Spawning was observed on several occasions following this initial episode. The same general pattern of activity

was followed each time. The spawning activity was initiated by the female. She would select a particular male and through her activity stimulated his preparation of a spawning site. After readying the spawning site, the male would take up a position next to it and start his quivering. This seemed to signal the female to start her spawning passes. This number varied from three passes by smaller females to as many as seven by the larger ones. There were often some spectacular gyrations by the male when the egg mass was deposited by the female some distance from the spawning site. The male would butt them with his snout, fan them with his fins, and in one case balanced them on the top of his head to return them to the desired position. On several occasions, eggs that had fallen off the spawning rock were abandoned on the bottom of the tank after the male made several unsuccessful attempts at retrieving them. In one such case, they were retrieved by a male guarding another egg mass and attached to his cluster.

My observations in the laboratory indicate that a single egg mass may represent the spawning accomplishments of a single pair, or it can result from the activity of one male and several females. A single male was observed while he accumulated an egg mass from the efforts of several females. On the examination of this type of egg mass it was found that they often contained eggs of different coloration and at several stages of development.

At no time during the spawning observations did the fish attempt to eat any of the eggs. Although on several

occasions females disturbed during spawning scattered eggs over the bottom of the tank, no attempts were made to eat them. Several fish were sacrificed and their stomach contents examined, but no eggs were ever found.

Although most of the animals used in this spawning study eventually died, I do not believe that this is necessarily the fate of reproducing individuals in nature. Most of the fish which died were badly injured. Since most of the egg masses were left where they were deposited, and I had not provided many spawning rocks, this led to a great deal of fighting among the males which were guarding the eggs (see Fig. 11). Many of the fish had most of their caudal fin missing and their other fins torn. It was assumed that they died as a result of these injuries.



Figure 11. Attached egg masses (arrows) of the seasnail which were spawned in laboratory tanks. The Irish moss has been cut away to expose the small clusters. (approx. .75X)

The Eggs

The eggs of <u>L</u>. <u>atlanticus</u> are demersal, that is they have a specific gravity slightly higher than water and sink to the bottom, where they are attached to the substratum or submerged objects (see Fig. 11). When the eggs are first deposited, they are quite sticky, although they usually needed to be forced against some object such as was done by the males during spawning. Single eggs and small masses which fell to the bottom of the tanks usually did not become attached since their impact with the substratum did not supply enough force to initiate attachment.

However, once the eggs became attached and water hardened, they were extremely difficult to remove. All attempts at separating the eggs of a mass once they had water hardened were futile. The chorion became very horny, but remained transparent. The color of the eggs varied from a pale yellow to salmon pink depending on the pigmentation of the yolk. The clumps of eggs were very difficult to observe since they were usually placed near the holdfasts of the vegetation or in between the rocks which were among the vegetation. In addition, colors of these relatively small egg masses blended quite well with their surroundings.

The ovaries of 12 females were examined in an attempt to determine fecundity. The mature eggs were easily separated from the smaller immature eggs and the number of eggs reaching maturity was determined by actual count. The results indicated that from 475 to 700 eggs reached maturity at one

time. Therefore, the number of eggs deposited at each spawning is relatively small. Since a female may spawn from three to four times in one season, a single female may produce from 1400 to 3000 eggs.

In all, spawning was observed on six occasions during a period of approximately three and a half months, and thirty egg masses were deposited. The water temperature during this time varied from 7° C. to 10° C. No attempt was made to control the amount of light to which the fish were exposed. The room in which the tanks were located was usually lighted during the day, but the lights were turned off at night. However, most of the spawning activity took place during the periods when the lights were turned on. On several occasions when spawning activities were in progress, the lights were turned off and the fish became inactive.

Many of the eggs were left at the site of deposition until they were eyed and then they were removed to smaller containers until they hatched. This was necessary because the larvae were so small that they would pass through the filters at the tank outlets and into the circulating water system.

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DEVELOPMENTAL STAGES

Embryonic Development

The embryonic development of <u>L</u>. <u>atlanticus</u> has never been described. Bigelow and Schroeder (1953) briefly reported some of the development of the European seasnail, <u>Liparis</u> <u>montagui</u>, from the works of Ehrenbaum (1904) and McIntosh and Masterman (1897). Erhenbaum (1904) confined his study to the European seasnails and devoted a major portion to the larval development, with only brief and scattered descriptions of the embryology. This was also the case with McIntosh and Masterman (1897); they discussed the eggs and the later larval stages of the European species.

Hence, there has been relatively little information recorded concerning the development of the seasnails. This part of my study, therfore, was an attempt to follow the development of <u>L</u>. <u>atlanticus</u>, and to present a somewhat continuous sequence of events from fertilization to hatching to supplement the scattered information already printed. Only the descriptive aspects of development are included and no attempt was made to reveal the mechanisms of the developmental processes, which would include the interaction of genetic and environmental forces which determine characteristics of the individual. Such extensive studies as the latter have been carried out in recent years, but for the purpose of this research, it was decided that a descriptive study was ample. It was guided by such works as the development of

Contraction of the

<u>Fundulus heteroclitis</u> by Oppenheimer (1937); a study of the development of six California fishes by Budd (1940); <u>Abudefduf</u> <u>sexatalis</u>, the sergeant major, by Shaw (1955); <u>Gobiesox</u> <u>strumosus</u>, the cling fish, by Runyan (1961); and the studies by Hildebrand and Cable (1930, 1938).

Oppenheimer (1937, p. 1) in her study of the normal development of Fundulus heteroclitis stated:

The very nature of the embryonic processes renders it impossible to choose criteria absolutely valid for subdividing the total course of development. The chronological age of a teleostean embryo, expressed in hours or days, does not represent its actual age, which varies according to conditions of temperature, oxygen supply, etc. The precise state of development of older embryos cannot be expressed in somite numbers, since this varies, when compared with the differentiation of other organ systems, in embryos maintained under different conditions, and probably in embryos differing in genetic constitution.

Therefore, in this study no attempt was made to label stages as to time or to seek histological characteristics. Development was broken down into stages by criteria which were easily visible and of sufficient duration so that minor differences between various embryos of the same stage would be minimized.

The development of <u>L. atlanticus</u>, from before fertilization to the prolarva, was arbitrarily divided into twentyfive stages, which are figured in the following plates and are pictured in the appendix.

<u>Stage 1. Unfertilized egg</u> (Figure 12). Normally <u>L. atlanticus</u> deposits its eggs in a small clump. In this mass each individual egg becomes flattened at points of contact where they press against each other or the substrate. Therefore, the measurement of egg diameter was difficult due to the irregular shape of the eggs. However, some eggs were taken immediately after spawning, placed in sea water and separated before they water hardened. Eggs so treated had a range from 0.8 mm to 1.4 mm, with an average diameter of 1.1 mm.

The transparent chorion of newly deposited eggs had an irregularly sculptured surface and was soft and pliable, but very tough even though quite fragile in appearance. Due to the irregularity of the chorion, no micropyle was observed. The lightly pigmented yolk contained many granules or yolk platelets which gave it an opaque appearance. Throughout the yolk several large oil globules and numerous widely scattered smaller ones were dispersed.

<u>Stage 2.</u> Fertilized egg (Figure 13). At fertilization, the chorion lost its irregular appearance and as it water hardened became very horny. The perivitelline space formed as the chorion separated from the plasma membrane. The perivitelline space was small. This is probably due to the fact that the eggs are demersal and are not exposed to the hazards that pelagic eggs experience. In the fertilized egg, the yolk platelets seemed to dissolve and gave the yolk a clear homogeneous appearance. Protoplasm invested in the yolk slowly began to stream toward, and accumulate at, the animal pole. Thus the raised blastodisk was formed. It was continuous at its periphery with the invisible plasma membrane that surrounded the entire yolk.

Stage 3. Two-cell stage (Figure 14). As cleavage
began, many small oil globules concentrated at the pole opposite the blastodisk. The first division was preceded by a slight lengthening of the blastodisk in the axis perpendicular to the first cleavage plane. The first cleavage plane, which was meridional, divided the blastodisk into two cells or blastomeres of equal size.

<u>Stage 4.</u> Four-cell stage (Figure 15). The second cleavage plane, also meridional, was perpendicular to the first and divided the blastodisk or blastoderm into four smaller cells equal in size.

<u>Stage 5. Eight-cell stage</u> (Figure 16). The third cleavage plane, which was double, was parallel to the first and resulted in an eight-celled blastoderm made up of two rows of four cells each. The resulting blastoderm was elongated in the axis of the second cleavage plane.

<u>Stage 6.</u> Sixteen-cell stage (Figure 17). The fourth cleavage plane, double and parallel to the second, divided the blastoderm into four rows containing four cells each. Usually the blastoderm became oval in outline and the rows of cells were indistinguishable.

<u>Stage 7. Thirty-two-cell stage</u> (Figure 18). As cleavage continued, the actual divisions were difficult to follow. However, after the fifth cleavage the blastoderm consisted of thirty-two cells, and formed a cap of cells resting on the yolk. Since all of the divisions were not occuring simultaneously, the symmetrical pattern was interrupted and the cells were irregularly arranged.

Stage 8. Late cleavage stage (Figure 19). Continued

PLATE I.

Figures 12 - 17. Early developmental stages of the eggs of the seasnail, <u>L</u>.

atlanticus. 50X.

Figure	12.	-	Stage	1.	Unfertilized egg.
Figure	13.	-	Stage	2.	Fertilized egg.
Figure	14.	-	Stage	3.	Two-cell stage.
Figure	15.	-	Stage	4.	Four-cell stage.
Figure	16.	-	Stage	5.	Eight-cell stage.
Figure	17.	-	Stage	6.	Sixteen-cell stage



Figure 16.

PLATE II.

Figures 18 - 23. Developmental stages of the eggs of the seasnail, <u>L. atlanticus</u>. 50X.

Figure	18.	~	Stage	7.	Thirty-two-cell stage.
Figure	19.		Stage	8.	Late cleavage stage.
Figure	20.	-	Stage	9.	Early blastula_stage.
Figure	21.	-	Stage	10.	High blastula stage.
Figure	22.	-	Stage	11.	Flat blastula stage.
Figure	23.	-	Stage	12.	Expanding blastula stage



cleavage resulted in cells which were smaller; however, the blastoderm still retained its previous volume.

Stage 9. Early blastula stage (Figure 20). With the formation of the blastula, the cells piled up due to both horizontal and vertical cleavages which resulted in a high distinct cap of cells on the yolk. At this stage it was quite evident that the preceding divisions had not cut entirely through the blastoderm, but had left a layer of protoplasm between the blastoderm and the yolk called the central periblast.

Stage 10. High blastula stage (Figure 21). At the high blastula stage, the blastoderm had not changed its shape, but was composed of many more still smaller cells than before.

Stage 11. Flat blastula stage (Figure 22). The blastula having reached its maximum height in the previous stage now began to flatten and cover more of the yolk. As the flattening continued, the first indication of the blastocoel or segmentation cavity became visible between the outer blastoderm and the central periblast that covered the yolk beneath the blastoderm.

Stage 12. Expanding blastula stage (Figure 23). The blastoderm continued to flatten, subsequently expanding to cover a greater portion of the yolk. The segmentation cavity was now distinctly visible beneath the blastoderm. This cavity in the blastodermal cap increased in size as the blastoderm lifted away from the periblast.

Stage 13. Early gastrula stage (Figure 24). As the

blastodermal cap continued to expand over the yolk, its cells piled up at the periphery at the expense of the center. The thinning central portion of the blastoderm was the extraembryonic membrane which formed yolk sac epithelium. The thickened peripheral area of the blastoderm was the germ wing. Once the germ ring formed, there was a thickening which was greater at one portion and formed the embryonic shield. This was the region of embryo formation which at first was a small extension of cells protruding from the germ ring into the segmentation cavity and it is here that gastrulation began.

<u>Stage 14.</u> Late gastrula stage (Figure 25). As gastrulation continued, the embryonic shield increased in size by the addition of cells anteriorly, laterally, and posteriorly. Shield formation was indicated by a localized accumulation of cells which was the keel of the central nervous system. By the time the yolk was one half covered, the general body outline of the embryo was visible with its ventral surface pressed into the yolk and the thickened cephalic region raised into the perivitelline space.

<u>Stage 15. Yolk plug stage</u> (Figure 26). At this stage, the germ ring had advanced to a position where it enclosed almost the entire yolk within the extra-embryonic membrane. The confining action of the ring caused the exposed portion of the yolk to bulge outward forming the yolk plug. This will gradually be forced inside as the blastopore closes. There was a great thickening of the entire embryo. Three or four mesodermal somites were visible in the midbody

PLATE III.

Figures 24 - 29. Developmental stages of the embryos of the seasnail, <u>L. atlanticus</u>. 50X.

Figure 24. - Stage 13. Early gastrula stage.
Figure 25. - Stage 14. Late gastrula stage.
Figure 26. - Stage 15. Yolk plug stage.
Figure 27. - Stage 16. Closure of the blastopore.
Figure 28. - Stage 17. Formation of the optic vesicles.
Figure 29. - Stage 18. Formation of the auditory vesicles.



Figure 28.

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Figure 29.

region; a tail thickening was present and the eyes were vaguely distinguishable.

Stage 16. Closure of the blastopore (Figure 27). The blastopore closed at a point immediately posterior to the tail end of the embryo. The lips of the germ ring met and fused, leaving no trace of the blastopore. The head of the embryo by this time was quite well formed. Mesodermal segmentation continued (10-12 somites), while the brain was outpocketing to form the optic vesicles. The embryo had extended slightly more than one half the circumference of the yolk.

<u>Stage 17.</u> Formation of the optic vesicles (Figure 28). The optic vesicles were now quite evident and the three primary regions of the brain became apparent at this stage. The tail was beginning to lift free of the yolk. There was a vague indication of differentiation in the area in which the auditory placode developed later. Previous to this stage there were several large oil globules, but now they had fused to form one prominent globule which persisted throughout the existence of the yolk.

<u>Stage 18.</u> Formation of the auditory vesicles (Figure 29). The ear had become vesicular, mesodermal segmentation continued, and the embryo nearly surrounded the yolk. The caudal portion was free of the yolk and a fin fold was evident running from the ventral point of attachment, around the tail and along the dorsal surface. The lens of the eye was distinct and the choroid fissure was present. Cells had gathered to form a faint bud in the region at which the

pectoral fins later formed.

Stage 19. Motility, circulation and pigmented eyes. (Figure 30). The embryo had increased considerably in size, and more than circled the yolk. The eyes were darkly pigmented, and the lens of the eye was distinctly visible. The embryo was quite active and was able to rotate inside the chorion, prior to this there were only feeble twitchings of the tail. The heart was beating, whereas previous to this only a pulsating vessel had been visible. Circulation over the yolk was also visible. The pectoral fin buds were apparent.

Stage 20. Otoliths in the auditory capsules (Figure 31). This stage showed some increase in activity, a further darkening of the eyes, and continued increase in size. The most prominent development was the appearance of calcareous concretions in the auditory capsules; formerly small and dispersed, they had aggregated to form the otoliths.

<u>Stage 21. Prominent fin buds</u> (Figure 32). A gathering of cells had been visible in the region of the pectoral fins since stage 18, but at this time the fin buds were greatly enlarged and quite distinct. The notochord could be seen to extend from below the posterior portion of the eyes to the tip of the tail which now bore a distinct transparent fin fold.

Stage 22. Rounded fins (Figure 33). The pectoral fins had now changed in shape from buds to flattened round structures. The embryo circled the yolk nearly l_2^1 times. Movements were more vigorous.

PLATE IV.

Figures 30 - 34. Developmental stages of the embryos of the seasnail, <u>L. atlanticus</u>. 50X.

Figure 30. - Stage 19. Motility, circulation, and pigmented eyes. Figure 31. - Stage 20. Otoliths in the auditory capsules.

Figure 32. - Stage 21. Prominent fin buds. Figure 33. - Stage 22. Rounded fins.

Figure 34. - Stage 23. Pigmentation of the fins.









Figure 34.

Stage 23. Pigmentation of the fins (Figure 34). At this stage, chromatophores appeared on the pectoral fins. The pigment was dark and in a stellate pattern. The embryo had enlarged to such an extent that it filled the cavity of the chorion completely. The tail was greatly elongated and was wrapped tightly around the yolk and across the head. Pigmentation was also visible along the base of the ventral portion of the fin fold.

<u>Stage 24. Hatching</u>. The chorion at this stage had become quite soft and pliable. Just prior to hatching the embryo became quite active and changed its position frecuently. Due to lack of space within the chorion, these movements seemed to require great effort and manipulation. During one of these periods of activity, the chorion split and the tail of the embryo slipped out first. With a few short periods of rest and activity, the larve twisted the remainder of its body free of the chorion. Since the head emerged last, frequently it became trapped momentarily inside the crumpled chorion. Usually it took several days for all of the developing eggs in a single mass to hatch.

<u>Stage 25. Prolarva</u> (Figures 35a and 35b). The prolarva, as stated by Hubbs (1943), is a larva still bearing yolk, and should be known as such until the yolk is exhausted. The prolarva described here is an early one shortly after hatching. The newly hatched animals ranged in total length from 3.2 mm to 3.9 mm, with the average length of 3.7 mm. They still bore large pale pink to orange yolk sacs with the one persistent oil globule in the anterior portion.

The most distinct feature was the large black eyes, which composed nearly half of the head length and were flecked with irridescent green and blue pigment. The mouth was well formed and open, while the nostrils were present, they were very difficult to observe due to the transparency of the head. The otocysts and otoliths were very distinct.

The pectoral fins were large and flecked with dark brown pigment, while the fin fold was transparent except for a row of chromatophores along the base of the ventral portion.

The notochord was clearly visible, curving upward above the eyes and extending posteriorly to the tail. Circulation was extensive, many blood vessels extended over the yolk and through the head. The dorsal aorta could be seen running ventral to the notochord. The viscera of the body was an indistinct mass except for the greenish-yellow gall bladder.

The prolarvae were quite active and had very little difficulty in maneuvering. They fluttered their tails very rapidly, while fanning the large pectoral fins to keep their bodies balanced. They congregated near the surface of the water and toward the side of the container nearest a light source. The larvae were quite gregarious, for when they were dispersed mechanically, they soon congregated once more.

The incubation period varied from 22 to 30 days. This was undoubtably due to the fact that the conditions were not kept constant and the egg masses were not all handled in the same manner. About 75 to 80 per cent of the eggs in each mass





Figure 35. a. <u>Liparis atlanticus</u> prolarva just hatched. Lateral view. 3.7 mm. b. <u>L. atlanticus</u> prolarva. Dorsal view. 3.7 mm.

hatched, which, considering conditions, was rather high and greater than expected. However, about 4 to 6 per cent of those which hatched had some type of malformation.

Although the larvae were kept alive for varying periods after hatching, some as long as two weeks, very little change was noted, except for a decrease in yolk sac size. The body lengthened slightly, coloration became a bit darker, but no radical changes occured.

Transitional or Larval Development

Since it was impossible to raise the larvae beyond the prolarval stage, the postlarval period, which includes the transformation to the juvenile, was not observed.

The next phase of this study was carried out through the collection of plankton samples in an attempt to observe some of the postlarval stages from natural spawning areas. However, it was extremely difficult to distinguish between the early postlarvae of <u>L</u>. <u>atlanticus</u> and other small fish present in the samples. Some larvae which appeared to be postlarval stages of <u>L</u>. <u>atlanticus</u>, on closer examination resembled sculpin larvae.

Late postlarval stages with the sucking disk developed were collected, and were assumed to be seasnails, since spawning adults had been found in these areas. The only other fish spawning in the shore area which would have a disk would be <u>Cyclopterus lumpus</u>. The chances that these were seasnail larvae was good, since these larvae did not resemble any of the larval stages of C. lumpus figured by Garman (1892). There was a possibility that these were stages of <u>Liparis liparis</u>, the striped seasnail, but this species had never been collected in the areas where these samples were captured. The only specimens of <u>L. liparis</u> collected were taken from deeper waters, 5 fathoms or more, and were never found intertidally.

The postlarval stages that were collected, resembled very closely those figured by Erhenbaum (1904, plate IV, fig. 25 and plate V, fig. 37) of <u>L</u>. <u>montagui</u>. The sucking disk was well developed even in those small individuals which were only 7 to 8 mm long. Examination of specimens cleared and stained with alizarin red S showed that the disk was supported by 12 rays. Staining also demonstrated that the vertebrae were considerably cossified as were many of the bones in the head region.

It was impossible to make figures of these stages for they were greatly distorted by preservation. I failed in all my attempts to return them to the laboratory alive. At the time when this collecting was in progress, there were many small medusae present in the area, and when samples were concentrated in the collecting procedure, most of the larvae were preyed upon by them. Only a few specimens were in recognizable condition, and these were cleared and stained in an attempt to follow the development of the sucking disk.

The best method for following postlarval development will require a further refinement of technique so that the prolarvae can be induced to feed. Then they can be reared

through the juvenile stage. This method will establish larvae of known origin and approximate age, thereby eliminating the confusion with other species.

Juvenile

The smallest transformed seasnail observed was 10.5 mm in total length. This individual was collected on July 7, 1961 in about three fathoms of water where it was clinging to the stipe of Laminaria. There was no difficulty in identifying this as a seasnail, since it had nearly all of the adult characteristics. The only differences were that the head was quite round and had not become depressed to any degree. The body was rounded, nearly transparent, and possessed a light scattering of stellate chromatophores covering it.

Other small individuals were also found in the course of tide pool collecting. One of 13 mm in total length (Figure 36) was found on June 2, 1962, and one 14 mm long was collected on August 17, 1962. The only differences between these individuals and the preceding one was greater pigmentation and a slimmer body form.



Figure 36. Juvenile. 13 mm. Showing the establishment of the adult body form.

PARASITES

General

No studies of the parasites of <u>L</u>. <u>atlanticus</u> have been recorded. In the examination of the gonads, analysis of stomach contents, and counting pyloric caeca, a number of parasites were encountered. A thorough investigation of the macroscopic parasites was undertaken with thirty fish captured in two collections during November and December 1962. All of the parasites found were new reports for the species, and one trematode is believed to be a new species.

Cestoda

The commonest parasite of the seasnails was a tapeworm, <u>Spathebothrium simplex</u> Linton, 1922. Wardle and Mcleod (1952) stated that <u>Spathebothrium</u> is the type and only known genus, with one species, <u>simplex</u>, of the family Spathebothriidae. This worm was first reported from the striped seasnail, <u>Liparis</u> <u>liparis</u>, at Woods Hole, Massachusetts. Linton (1922) described the genus <u>Spathebothrium</u> as having no distinct scolex; strobile taenaeiform, bluntly rounded at the extremities, proglottides not distinct, reproductive apertures on the median line and irregularly alternate. Its life cycle is unknown.

<u>S. simplex</u> were found mainly in the anterior portion of the intestine, and frequently with their anterior ends protruding into the pyloric caeca. One fish with a total length of 78 mm, contained 36 of these tapeworms. The examination of 30 fish produced a total of 273 tapeworms. I do not believe a single intestine was examined which did not contain this animal.

The first specimens encountered were identified by Dr. Wilbur L. Bullock of the Zoology Department at the University of New Hampshire.

Trematoda

The second most frequently encountered parasite was a trematode which is believed to be a new species. Five specimens were forwarded to Dr. Thomas C. Cheng of the Biology Department at Lafayette College. Dr. Cheng (personal commuication) stated the following:

All the specimens which you sent are of the same species. These are members of the genus <u>Pycnadenoides</u> Yamaguti, 1938 of the family Allocreadiidae. As far as I know and can find out, only two species of this genus are known: <u>P. pagrosomi</u> Yamaguti, 1938, the type species, found in the marine fish <u>Pagrosomus unicolor</u> in the Inland Sea of Japan. The other species is <u>P. salami</u> Manter, 1947 from <u>Calamus</u> <u>bajonado</u>, another marine fish, off the coast of Florida. Your specimens belong to neither of these species and I suspect that it is a new one. When one considers that the type species was found in a different species of fish in Japan, and the second species in still another species of fish in Florida, it is not surprising that yours, which is from <u>Liparis atlanticus</u> from the New England coast, is new. Think of the ecological barriers and differences.

The maximum infection encountered was 23 trematodes from a fish 71 mm in total length. Examination of 30 fish produced 245 specimens. The trematodes inhabited the upper intestine and the pyloric caeca. Where these parasites were found in large quantities, the numbers of <u>S</u>. <u>simplex</u> was reduced, and vice versa.

Acanthocephala

A rather rare parasite was the acanthocephalan, <u>Echinorhynchus gadi</u>. This spiny-headed worm was found in the intestine of only a few fish. Five specimens were collected, three of these were from a series of thirty individuals. The identification of this species was confirmed by Arthur J. West, a graduate student in parasitology, in the Zoology Department at the University of New Hampshire.

<u>E. gadi</u> has been reported by Meyer (1932) from <u>Prionotus evolans, Lophius piscatorius, Gadus callarias,</u> <u>Melanogrammus aeglefinus, Pseudopleuronectes americanus,</u> <u>Paralichthys dentatus, Roccus saxatilus, Limanda ferruginea,</u> <u>Myoxocephalus aeneus, and Stenotomus chrysops</u>. Several of these species are found within the range of <u>L. atlanticus</u>. The grubby sculpin, <u>M. aeneus</u>, was frequently found in the tide pools with the seasnails.

Nybelin (1923) reported <u>Gammarus locusta</u>, <u>Amphithoë</u> <u>rubricata</u>, <u>Calliopius rathkei</u>, and <u>Dexamine spinosa</u>, all amphipods, as the intermediate hosts of <u>E. gadi</u>. Since the seasnails feed mainly upon crustaceans, especially <u>Gammarus</u> <u>locusta</u>, it is not unusual that they are parasitized by this organism.

Arthropoda

One specimen of the parasitic copepod <u>Lernaeocera</u> <u>branchialis</u> was found. This specimen was identified by Dr. Alan G. Lewis of the Zoology Department at the University of New Hampshire. This was a male of the mature copepodid stage. Cameron (1958) reported that this species is a common parasite of the Gadidae. The males are supposed to remain on flatfish, while females seek out the Gadidae and attach to the gills. Since only one specimen was found, its appearance here may have been only accidental.

Nematoda

About one in seven of the specimens examined, contained large nematodes. These parasites were found under the loose skin, free in the coelom, and in the ovaries of the females. These organisms were not identified.

Protozoa

One fish which died in the laboratory tank was examined by Dr. Wilbur L. Bullock. He found protozoa of the genus <u>Trichodina</u> living on the gills. Hymen (1940) reported that members of this genus are ectoparasites which inhabit the surface of a variety of animals, such as hydras, sponges, planarians, tadpoles, and fishes. I do not know if the specimen was parasitized by this protozoan when I collected it, or whether the infection developed in the aquarium.

SUMMARY

Intertidal collections of the seasnail, <u>Liparis</u> <u>atlanticus</u>, were carried out over a three year period in an effort to fill the gaps in our knowledge of the life history of this species.

A review of the classification of the seasnail determined that the most correct name for this species is <u>Liperis</u> <u>atlanticus</u>. This is suggested by the American Fisheries Society, therefore, the genus <u>Neoliparis</u> under which it was originally described was not recognized. The latest taxonomic designation places <u>L. atlanticus</u> in the family Cyclopteridae and gives seasnail as the common name.

Examination has been made of various body parts. Some differential growth was found to exist in the head, eye, and disk. The comparison of the adult characteristics and of various measurements made on the specimens collected during this research varied only slightly from earlier reports, although a greater number of fish were examined than in any previous study.

The seasnail has a general range from Newfoundland to southern New England, but recent reports indicate that the northern limit of the range may be extended by further investigation. The habitat preference in the intertidal zone is the rocky areas of the open coast. No close associations with other organisms of the intertidal area were observed.

The food of the seasnail was found to be chiefly

crustaceans and some polychaetes.

The length-weight relationship was determined, but was considered to be most indicative of the condition that exists during the inshore or reproductive phase of the life history, since this relationship is believed to vary throughout the year.

It was not possible to determine the age of the seasnails, since no structure was found that demonstrated growth rings that could be satisfactorily interpreted.

Reproduction was investigated through field and laboratory observations. Males demonstrated sexual dimorphism by fleshy development of the elongated anterior dorsal fin rays and the appearance of breeding tubercles on the upper portions of the body, head, and fins. Gonad maturation was traced and it was shown that sexual maturity is gained between the length of 60-70 mm. At this time the fish are entering their second year. The spawning season extended from March until midsummer. Seasnails begin their inshore migration in October. The first fish to arrive are small immature individuals, which are later joined by those which are spawning for the first time. Through the winter and spring fish size increased and the females became more abundant. The larger and older females were found to spawn several times, which caused them to be in the spawning areas for a longer period. The last spawners left the shore in June. During the summer only a few larvae and juveniles were captured.

Spawning was observed among laboratory confined fish. Reproductive behavior was initiated by the female, who induced

the male to select and prepare a spawning site. The females deposited several small masses of eggs which the males fertilized and gathered into a single attached egg mass. The eggs were about 1.1 mm in diameter and their color ranged from pale yellow to salmon pink, depending on the pigmentation of the yolk. The clumps of eggs were well hidden among the stones and algae in the laboratory tanks.

The stages of embryonic development were observed and described from the laboratory deposited eggs. The transitional or larval development was not recorded, because the larvae could not be reared past the absorption of the yolk sac. Collection attempts for these stages from the plankton were unsuccessful, too.

The smallest fish collected was 10.5 mm in total length. The body form of this juvenile resembled very closely that of the adult.

The seasnails were examined for parasites, and all of the specimens observed were new reports for the species. The commonest parasite was the tapeworm <u>Spathebothrium simplex</u>. A trematode, which is probably a new species, was very abundant in the intestine and pyloric caeca. The acanthocephalan <u>Echinorhynchus gadi</u> was found in only a few cases, and one specimen of the parasitic copepod <u>Lernaeocera branchialis</u> was collected. Unidentified nematodes and protozoa of the genus Trichodina were also present.

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APPENDIX

Photomicrographs of the embryonic development of L. atlanticus. (A - Z approx. 50X)

A. - Unfertilized egg. Small yolk platelets are visible throughout the yolk.

B. - Fertilized egg. Lateral view of the fertilized egg, showing the building up of the blastodisk in preparation for the first cleavage. Many small oil globules are concentrated at the negative pole.

C. - First cleavage. At about $6-6\frac{1}{2}$ hours the eggs had begun their meroblastic cleavage.

D. - Two-cell stage. The first cleavage plane, which was meridional, divided the blastodisk into two cells or blastomeres of equal size.

E. - Four-cell stage. The second cleavage plane, also meridional, was perpendicular to the first, and divided the blastoderm into a rosette of four cells equal in size.

F. - Eight-cell stage. The third cleavage plane, was double and parallel to the first, and resulted in an eight celled blastoderm made up of two rows of four cells each.





c.













F.

G. - Sixteen-cell stage. The fourth cleavage plane, double and parallel to the second, divided the blastoderm into four rows of four cells each, which often appeared as an oval as shown here.

H. - Thirty-two-cell stage. At these later cleavage stages, the major changes were in the number and size of the cells.

I. - Late cleavage stage. Although the cells were smaller, the blastoderm still retained its previous volume.

J. - Early blastula stage. The blastoderm at this stage was similar to the blastoderm of the late cleavage stages, but was composed of smaller cells.

K. - Early blastula stage. A polar view of the early blastula.

L. - High blastula stage. The blastoderm changed very little in shape, but was composed of still smaller cells than before.









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M. - High blastula stage. A polar view of the high blastula. Note the persistent oil globules.

N. - Flat blastula stage. The blastoderm has flattened to form a flat lenticular structure capping the yolk. The first indication of the blastocoel or segmentation cavity was observed at this stage.

0. - Expanding blastula stage. The blastoderm continued to flatten, simultaneously expanding to cover a greater portion of the yolk. At this stage the segmentation cavity was distinctly visible.

P. - Early gastrula stage. As the blastoderm expanded over the yolk, its cells piled up at the periphery at the expense of the center. The central thin area of the blastoderm was the extra-embryonic membrane which will form yolk sac epithelium. The thickened rim was the germ ring. As shown here, the yolk is nearly one-third covered by the yolk sac epithelium, and the embryo has formed as an extension of cells protruding into the segmentation cavity.

Q. - Late gastrula stage. The germ ring was almost equatorial in position, while the slightly concave tip marked the anterior end of the embryo. The general body outline was visible and the cephalic region was beginning to thicken.

R. - Yolk plug stage. The yolk was over two-thirds covered by the yolk sac epithelium. Its pinching action caused the uncovered portion of the yolk to bulge outward to form the yolk plug. Three or four somites were visible in the mid-body region; a tail thickening was present and the eyes were vaguely apparent.

98







Q.







R.

S. - Closure of the blastopore. The blastopore closed at a point immediately posterior to the tail end of the embryo. The head was quite well formed and the brain was outpocketing to form the optic vesicles. Mesodermal segmentation was apparent.

T. - Formation of the optic vesicles. The optic vesicles were quite evident and the three primary regions of the brain were apparent at this stage. The tail was beginning to lift free of the yolk.

U. - Formation of the auditory vesicles. The ear was now vesicular, mesodermal segmentation continued and the embryo nearly surrounded the yolk. The tail was now free of the yolk and possessed the primary fin fold.

V. - Motility, circulation, and pigmentation of the eyes. The embryo had increased considerably and more than circled the yolk. The eyes were darkly pigmented and the lens was visible. The embryo at this stage was quite active and was able to spin around inside the chorion. The heart was beating and circulation was visible over the surface of the yolk.

W. - Otoliths in the auditory capsules. Calcareous concretions in the auditory capsules, formerly small and scattered, have aggregated to form the otoliths.

X. - Prominent fin buds. A gathering of cells had been visible in the region of the pectoral fins, but now the fin buds were greatly enlarged and quite distinct. The body now bore a continuous transparent fin fold.







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Y. - Rounded fins. The pectoral fins had changed in shape from buds to flattened round structures.

Z. - Pigmentation of the fins. At this stage, chromatophores appeared on the pectoral fins. The embryo had enlarged to such an extent that it filled the cavity of the chorion completely.

A-l - Hatching. In this picture, the tail is projecting through a slit in the chorion, while the rest of the body is still enclosed in the crumpled chorion. (approx. 40X)

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A-1.