PERCENTAGE OF STARVING NORTHERN ANCHOVY, *ENGRAULIS MORDAX*, LARVAE IN THE SEA AS ESTIMATED BY HISTOLOGICAL METHODS

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ABSTRACT

The proportion of starving larvae of northern anchovy, *Engraulis mordax*, was estimated for the Southern California Bight in March 1977 from histological examination of larvae for 64 1 m net tow samples. The number of larvae in the tows varied from 0 to about 400. Approximately 6 per tow were sectioned and examined with the light microscope. Twenty-six specimens were identified as emaciated from anomalies of the trunk musculature and digestive tract. Some of the emaciated larvae occurred as isolated cases at widely scattered locations, but most were from a few nearshore tows, indicating "patches" of starving larvae. Temperature and plankton volume data indicate that the patches were associated with fluctuating environmental conditions. The samples indicate that about 8% of northern anchovy larvae in the Southern California Bight were starving.

One of the goals in investigations of pelagic fish stocks is that of predicting how large year classes will be at the time they are recruited to the fishery. One of the primary approaches to this problem has been the estimation of larval mortality rates based on abundance estimates from egg and larval surveys. While such surveys will probably continue to be the most reliable source of information on abundance at early ages, the high costs and time delays in processing samples are reasons for seeking alternative approaches (Hunter 1976b). Recently, Lasker (1975, in press) has developed a promising index based on availability of food in concentrations suitable for survival of early feeding stages of the northern anchovy, *Engraulis mordax*.

This study reports another approach that could provide an independent prediction of year class strength for the northern anchovy; namely, estimation by histological methods of the proportion of larvae in the sea showing symptoms of starvation. Since level of mortality in a population is likely to be some function of the proportion of larvae observed to be starving, the proportion, if based on adequate sampling, could be an indicator of ultimate year class success.

Condition factor (Blaxter 1971), chemical indices (Ehrlich 1974), morphometric analyses (Shelbourne 1957; Nakai et al. 1969; Ehrlich et al. 1976; Theilacker 1978), and histological analyses (Umeda and Ochiai 1975; O'Connell 1976; Theilacker 1978) have all been used with some success to characterize the starving condition in larvae of various marine species, in most cases under controlled laboratory conditions. The histological approach differs from the others in that the criteria of starvation are based on qualitative changes in the character of cells and tissues, not on quantitative measurements. Histological criteria developed earlier for northern anchovy larvae starved in the laboratory (O'Connell 1976) were the principal guidelines for evaluating the condition of ocean-caught larvae in this study.

METHODS

In March 1977, 64 net tows were taken over a 12-d period from the NOAA ship *David Starr Jordan* to obtain northern anchovy larvae for histological study. Almost half of the tows were taken between 2 and 10 mi (3.7-18.5 km) from the coast, most were near Newport Beach, Calif., where northern anchovy eggs and larvae were abundant, but some were much farther offshore. A surface temperature was taken by bucket thermometer at each net tow station.

Net tows were taken with a 1 m plankton net on which the cod end was a cylindrical Plexiglas[2]

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bucket (length 30.5 cm, diameter 10 cm) that could be removed quickly. Tows were of short duration to minimize the time larvae would be under stress during capture. The net was let out at a fast steady rate to a depth of 20 m, and then retrieved at a moderate rate. The mean tow time was 3 min (SD = 40 s).

The net drained rapidly as it was hauled out of the water, leaving the cod end cylinder filled with water containing a light to moderate amount of plankton. The contents of the cylinder were immediately poured into a small sieve made of 0.505 Nitex mesh netting, and the sieve was then suspended in Bouin's fluid to fix the concentrated plankton. The total elapsed time from start of tow, when the plankton net first entered the sea surface, to submergence in Bouin's averaged 5 min, 8 s (SD = 1 min, 9 s). After the initial fixation, about 10 or 15 min, the sample was transferred from the sieve to a jar of fresh Bouin's, and this was replaced by 70% ethyl alcohol 2 or 3 d later.

In carrying out the above procedure the inside of the plankton net was not washed down after retrieval until the cod end containing the sample had been removed. After the cod end was removed, the inside of the net was hosed down thoroughly in preparation for the next tow.

Subsequent to the cruise, all northern anchovy and other fish larvae were sorted out of the samples and counted. From those tows containing only a few northern anchovy larvae, all were set aside for sectioning. From those tows containing many larvae, about half a dozen were chosen for sectioning. The number was approximately doubled for a few samples of special interest, e.g., offshore banks. Specimens were picked at random by putting all northern anchovy larvae from a given tow in a shallow, wide-mouth container and repeatedly dipping with a vial as the contents were swirling slowly. During this procedure small specimens with obvious yolk sacs were rejected because they represented nonfeeding larvae not yet vulnerable to starvation.

The total number of larvae selected for sectioning was 318. Standard length was measured with an ocular micrometer, then each specimen was imbedded in paraffin, sectioned serially as close to the sagittal plane as feasible, and stained in Harris' hematoxylin and eosin-phloxine B. Prior to microscope examination the mounted specimens were put in random order with their identities concealed.

Histological criteria similar to those diagnostic for laboratory starved larvae were readily established for ocean-caught larvae by preliminary examination of a few dozen (unidentified) specimens, after which all ocean-caught larvae were classified as to condition. Under Results, the histological indications of condition are described first, and then the classification of larvae is examined in relation to other variables, i.e., standard length, geographical distribution, temperature, and plankton volume.

RESULTS

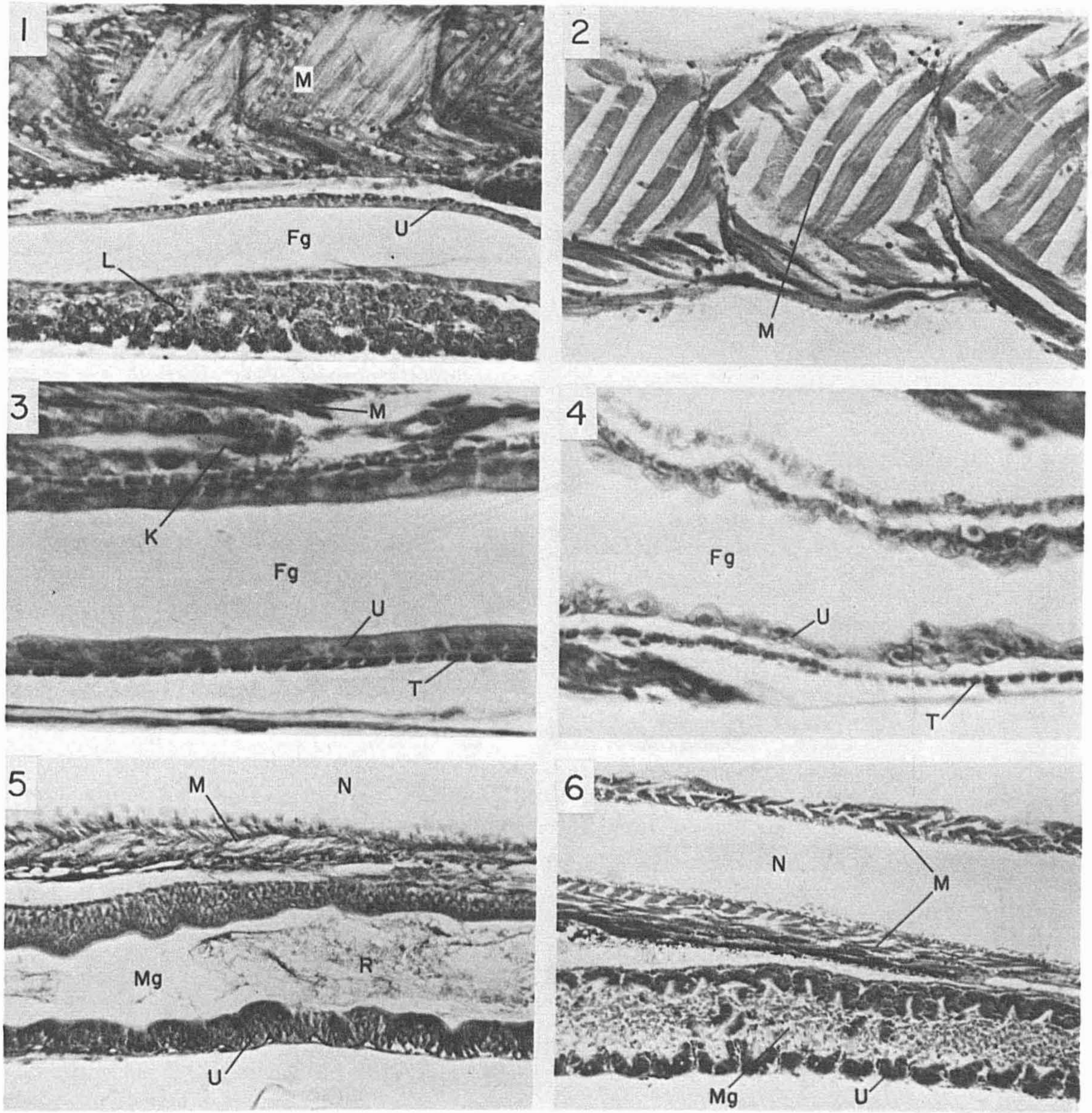
Histological Characteristics of Condition

O'Connell (1976) found the most noticeable effects of artificial starvation of northern anchovy larvae just beyond yolk absorption (3-5 mm) to be cellular dissociation with loss of zymogen in the pancreas, separation and hyalinization of trunk muscle fibers, and shrinkage of the notochord. In the ocean-caught material examined in the present study, specimens ranging from 2.5 to 10 mm showed anomalies in the trunk musculature and notochord, and occasionally also in the pancreas, that closely resembled the effects of starvation in the laboratory material. These larvae almost always showed, in addition, certain irregularities in the histology of the foregut and the midgut that were more striking than effects seen in the digestive tracts of artificially starved larvae.

Trunk Musculature

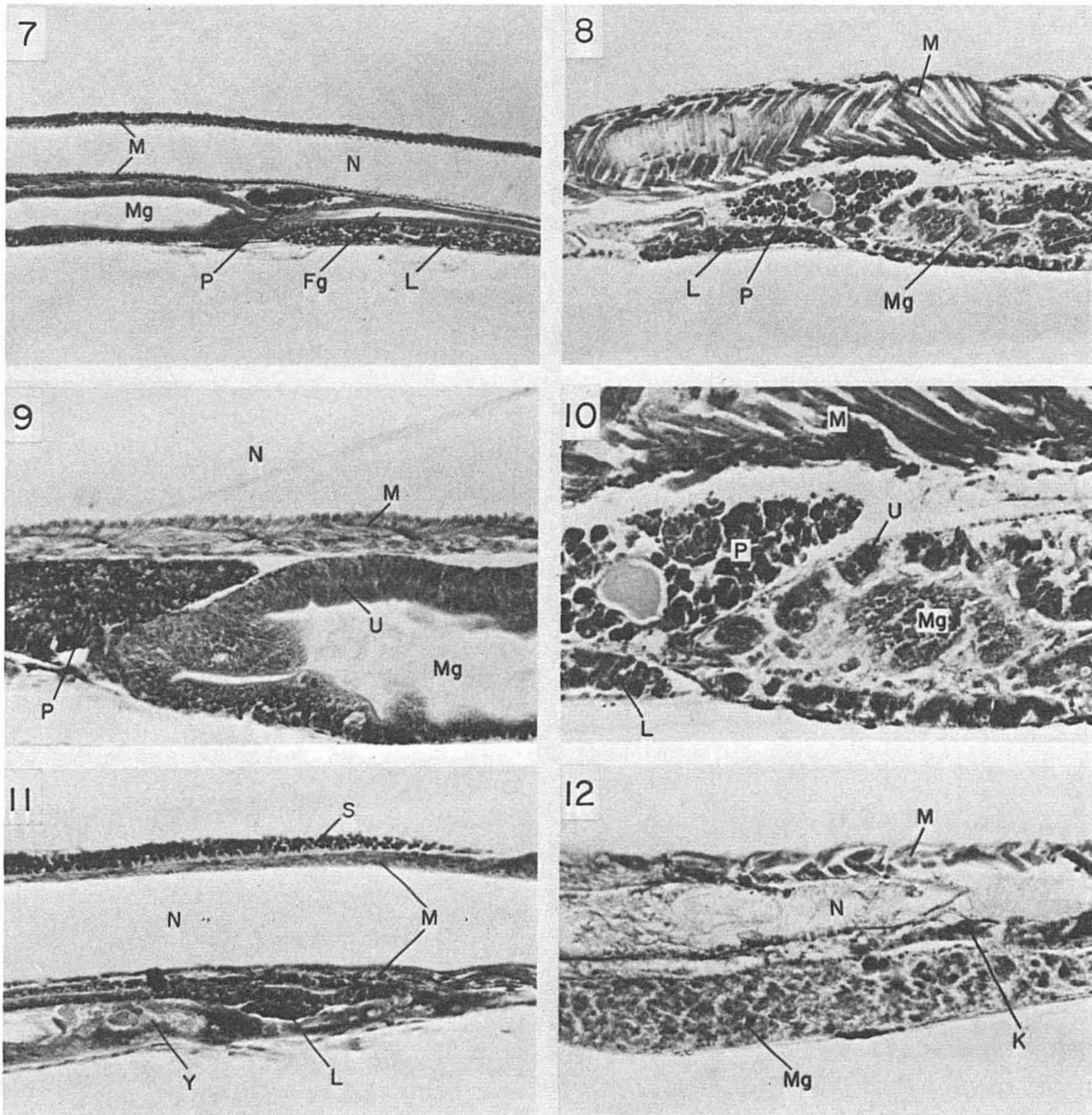
The trunk musculature in the majority of larvae showed good integrity and texture, forming a compact, solid sheet over the lateral surfaces of the notochord, with evident intermuscular matrix tissue and only occasional small separations (Figure 1). The notochord in such larvae generally had a smooth profile and was rarely separated from the musculature. In some specimens, however, the muscle fibers were noticeably separated from each other throughout, indicating an anomalous condition. In the more extreme cases (Figure 2) the fibers were widely separated with degraded fibril clarity, and matrix tissue was greatly reduced. In such specimens the notochord was also irregular in profile, imparting a "lumpy" shape to the trunk of the animal as a whole.

Degraded musculature, of course, might be the result of some process other than starvation. One possibility is capture myopathy, which has been



FIGURES 1-6.—Histological comparisons of healthy (left) and emaciated (right) northern anchovy larvae. All sections are approximately sagittal. 1) The trunk musculature, cut tangential to the body surface, forms a solid sheet. The foregut mucosa is composed of uniform cuboidal cells. 6.1 mm SL; 250×. 2) The trunk musculature, cut tangentially, shows widely separated fibers. 8.5 mm SL; 250×. 3)The foregut mucosa is composed of uniform thick cuboidal cells. 7.0 mm SL; 630×. 4) The foregut is irregular and the cells of the mucosa are diminished in size. 6.1 mm SL; 630×. 5) The mucosa of the midgut shows good integrity and organization, and the lumen contains moderate food residue. 6.3 mm SL; 250×. 6) The midgut is filled with disassociated cellular debris and the mucosa is fragmentary. Separated trunk muscle fibers are also visible. 8.5 mm SL; 250×. Symbols: Fg, foregut; K, kidney; L, liver; M, trunk muscle; Mg, midgut; N, notochord; R, food residue; T, transverse muscle coat of digestive tract; U, mucosa.

reported for a number of ungulates, primates, and birds (Harthoorn 1977) and in the swordfish, *Xiphias gladius* (Tibbo et al. 1961). The outstanding symptom is degeneration of skeletal muscle, presumably from acidemia from overexertion during intense flight and struggle in capture. Though the possibility cannot be entirely dismissed, capture myopathy seems an unlikely cause of the



FIGURES 7-12.—Histological comparisons of healthy (left) and emaciated (right) northern anchovy larvae.—Continued. 7) The trunk muscles cut in cross section above and below the notochord are compact. The foregut and midgut have mucosa of good thickness and integrity. 5.5 mm SL; 100×. 8) Trunk muscles are separated and uneven, the midgut is filled with disassociated cellular material, and the pancreas is severely disassociated. 5.5 mm SL; 100×. 9) Portions of the midgut, the pancreas, and the trunk musculature below the notochord all show good integrity. 5.0 mm SL; 250×. 10) Enlargement of Figure 8 for comparison of pancreas to that in Figure 9. 250×. 11) The foregut is regular and the trunk muscles are compact. The foregut is not yet expanded, and the midgut is not visible in the section. The remnant of the yolk sac, primarily periblast tissue, is prominent. 2.5 mm SL; 250×. 12) The midgut is a solid mass of necrotic debris. The notochord and trunk muscles are collapsed and irregular. The kidney is necrotic. 2.2 mm SL; 250×. Symbols: Fg, foregut; K, kidney; L, liver; M, trunk muscle; Mg, midgut; N, notochord; P, pancreas; S, spinal cord; U, mucosa; Y, yolk.

muscle deterioration observed in the northern anchovy larva. It is essentially a phenomenon of protracted struggle in large animals. In the case of the northern anchovy larvae, time and physical manipulation were minimized in capture.

Depletion of tissues arising from starvation seems a more likely cause of muscle deterioration. Love (1974), in summarizing depletion in (adult) fishes, states that "... when fish are starved the lipid reserves ... decrease to a certain point beyond which the muscle protein is mobilized. As the protein decreases, the water increases, and this change is mainly brought about by shrinkage of the cells and a corresponding increase in the fluid between them." In conjunction with these findings it had been shown that extracellular spaces appeared in the musculature of Atlantic cod, *Gadus morhua* (Love et al. 1968), and American plaice, *Hippoglossoides platessoides* (Templeman and Andrews 1956). Deterioration of the musculature was a prominent effect following artificial starvation of early postyolk-sac northern anchovy larvae (O'Connell 1976) and of jack mackerel, *Trachurus symmetricus*, larvae (Theilacker 1978). Deterioration of musculature during starvation has also been observed in larval herring, *Clupea harengus*, and plaice, *Pleuronectes platessa* (Ehrlich et al. 1976).

Digestive Tract

In most larvae the long foregut was straight with a uniform lumen and a smooth surfaced mucosa of cuboidal or thick squamous cells (Figure 3; see also Figures 1 and 7). The foregut was considered anomalous if the profile was noticeably irregular in sagittal view. The mucosa usually also showed some variation in cell thickness and shape. In the more extreme cases the mucosa cells tended to be reduced to little more than the nucleus (Figure 4), and the lumen sometimes contained a few sloughed cells.

The midgut varied greatly in appearance, depending on the degree of longitudinal folding and transverse ridge development and on the plane of a particular section. Nevertheless, in most specimens a substantial lumen, sometimes containing food, and a simple mucosa of low columnar cells could be traced (Figures 5, 7, 9). Although the infranuclear portions of the cells were often narrowed and slightly separated, the supranuclear portions were always well joined.

The midgut was judged anomalous when little or no lumen could be traced, or when the traceable lumen contained a considerable bulk of loose nuclei and necrotic cellular debris, and the mucosa proper was fragmenting (Figures 6, 8, 10). In the worst case the midgut was a homogeneous mass of necrotic debris enclosed in only a basement membrane, with no trace of either lumen or mucosa (Figure 12). This specimen, which happened to be the smallest examined, also showed a severely collapsed notochord and degenerate musculature. The degree of organ development, including fully pigmented eyes, indicated that it had shrunken. Healthy specimens of comparable size, which still had unpigmented eyes, showed good notochord and musculature, and often a sizable remnant of the yolk sac (Figure 11). Shrinkage has been shown in laboratory starved larvae of both the Atlantic herring (Blaxter and Hempel 1963) and the northern anchovy (O'Connell 1976; O'Connell and Raymond 1970).

Whereas the muscle and foregut anomalies are at least logically acceptable as consequences of inadequate nourishment, interpretation of the midgut anomaly is problematical. In artificially starved northern anchovy larvae the midgut showed thinning and increased separation of cells, and the loss of some cells (O'Connell 1976), but not strong contraction followed by general fragmentation and necrosis of the mucosa as seen in some of the ocean-caught specimens. It may be, of course, that the symptoms were different because the laboratory and ocean situations were different. Laboratory animals were starved without ever having an opportunity to feed, whereas the oceanic larvae showing symptoms undoubtedly did have the opportunity to feed. Most in fact had processed food through the digestive tract, as indicated by the presence of supranuclear inclusion bodies in the mucosa cells of the hindgut, though the hindgut and the inclusion bodies were sometimes in a state of disintegration. Such inclusion bodies were never found in laboratory specimens deprived of food from time of hatching (O'Connell 1976).

Contraction and congestion resembling that seen in the midguts of the emaciated northern anchovy larvae from the ocean have been described for some other fishes, but they can be symptomatic of disease as well as of starvation. *Clupea harengus* larvae of 9-13 mm, for example, are vulnerable to a nematode that grows in the body cavity and deforms the gut, often resulting in occlusion of the lumen that blocks food intake and/or defecation (Margolis 1970). There are other nematodes whose larvae attack the gut walls of certain fishes, causing inflammatory and degenerative changes, including localized necrosis and infiltration of abundant lymphocytes (Margolis 1970). There are also protozoans, such as

Nosema anomala, which invade the intestinal wall of the young of the threespine stickleback, *Gasterosteus aculeatus*, and generate hypertrophied cells filled with its vegetative and reproductive stages (Lom 1970). Certain viral diseases produce degenerative changes that include necrosis and sloughing of intestinal epithelium (Yasutake 1975). On the other hand, starvation of immature salmon cause, among other things, marked atrophy of the stomach with degeneration of the epithelium, which ". . . could presumably be used for nourishment" (Love 1974). Starvation of

TABLE 1.—Numbers of northern anchovy larvae and other data by net tow for March 1977, off southern California. Number emaciated and standard length pertain only to sectioned larvae. See Figures 13 and 14 for tow locations.

Tow	Date	Hour	Temperature (°C)	Plankton volume (ml)	Number of Larvae		Mean standard length	Number emaciated		
					In tow	Sectioned		Severe	Moderate	Incipient
1	17	1530	13.2	1	0					
2		1750	13.8	5	9	6	4.6			
3		2210	13.5	4	33	7	4.0	1		
4	18	0825	14.2	2	9	5	5.0			
5		1035	14.5	2	9	5	4.9			
6		1440	14.7	3	14	6	6.4			
7		1910	14.7	3	68	6	5.4			
8		2340	14.8	2	47	6	4.7			
9	19	0350	13.7	3	54	6	4.5	3	1	
10		1010	15.2	4	7	7	4.9	3		1
11		1215	15.6	3	2	2	3.9		1	
12		1550	15.5	1	32	6	5.1			
[1]13		1600		8	[2]200	6	5.2			2
14		2215	14.6	11	43	6	4.7			
15	20	0005	13.4	4	24	10	6.7	4	1	2
16		0735	13.6	4	59	6	4.7			
17		1155	14.0	10	20	7	5.5			
[1]18		1205		3	15	6	4.2			
19		2045	15.0	7	70	6	5.5			
20	21	0005	14.9	4	65	7	6.5	4	2	1
21		2045	14.2	21	[2]400	6	5.0			1
22	22	1920	14.2	7	16	6	5.7			
23		2255	13.6	32	[2]350	7	6.0			
24	23	0945	14.6	6	40	6	4.2			
[1]25		0955		10	54	6	4.4			
26		1110	14.7	3	5	5	4.4			
27		2045	14.1	12	180	6	4.9			
28		2315	13.4	7	56	5	5.4		1	
29	24	1020	14.7	14	62	6	5.3			
[1]30		1030		8	67	7	5.3			
31		1440	13.7	7	8	6	5.4			1
32		1915	14.8	11	[2]200	6	4.7			1
33	25	0120	13.8	35	[2]300	6	4.9			
34		1550	13.1	14	97	7	4.8			
[1]35		1555		5	22	6	5.7			
36		2310	13.4	6	122	7	6.9			
37	26	0140	13.1	38	[2]250	6	7.1			
38		0345	12.7	10	19	6	6.6			
39		0550	12.3	33	4	4	12.5			
40		0730	12.8	17	0					
[1]41		0740		29	0					
42		0927	12.7	106	1	1	8.0			
43		1300	14.4	4	1	1	5.9			
[1]44		1325		21	1	1	12.2			
45		1620	14.1	1	0					
[1]46		1630		12	1	1	8.5			
47		1745	14.5	7	0					
48		2030	14.2	13	2	2	10.6			
49		2205	14.2	21	2	2	9.3			
50		2345	14.3	15	3	3	7.2		1	
51	27	0120	13.8	19	2	2	8.2			
52		0240	13.2	29	16	16	12.3			
53		0410	13.2	42	35	15	11.0		1	
54		0645	14.2	7	0					
55		1340	14.5	10	0					
56		1450	14.6	20	0					
57		1600	14.5	7	0					
[1]58		1610		30	0					
59		1839	14.1	27	5	4	9.8	1		2
60		2115	14.0	17	3	3	12.8		1	
61		2330	13.8	12	1	1	11.7			
62	28	0145	14.4	15	11	7	9.4			
63		0415	15.2	5	42	14	7.1			
64		0640	15.2	2	22	15	6.9	1		

[1]Taken at same location as preceding tow at almost twice the depth.
[2]Number estimated from count of a substantial fraction.

mummichog, *Fundulus heteroclitus*, up to 8 days resulted in a decrease in the quantity of lipid droplets in cells of the digestive tract and contraction of the intestine such that the lumen was small, sometimes scarcely traceable (Ciullo 1975). While these considerations suggest that disease could be the cause of the midgut anomaly present in certain of the ocean-caught northern anchovy larvae, starvation is the more tenable explanation because there was no evidence of parasites or pathogens in the hematoxylin- and eosin-stained specimens, and other anomalies in these specimens were consistent with demonstrated effects of starvation.

Other Organs

Deterioration was sometimes evident in other organs, particularly in the specimens with the most severe anomalies in the musculature and digestive tract. The pancreas and liver, for example, showed good integrity in most larvae (Figures 1, 9), but some showed an unusual degree of dissociation in these organs (Figures 8, 10), and in a few, both organs had undergone considerable lysis. The kidney ducts were intact in all specimens, but in a few the cells of the ducts were unusually thin, or necrotic (Figure 12). In several the mantle layer of the brain showed poor integrity, perhaps from a reduction of neuroglia.

Classification of Larvae

During the course of microscope examination, each larva was designated healthy, incipient emaciation, moderate emaciation, or severe emaciation. The three classes pertaining to larvae with anomalies are not rigorous, but they imply the following: incipient, slight looseness of the trunk muscles; moderate, obvious separation of the trunk muscle fibers, some irregularity of the notochord and possibly the foregut, strong contraction of the midgut, and sometimes a high incidence of hypertrophic cells in the midgut mucosa; and severe, obvious separation and hyalinization, and sometimes disarray, of the muscle fibers, notable irregularity in the profile of the notochord and foregut, depletion of foregut muscosa cells, and fragmentation of midgut mucosa, with a central core of dissociated and necrotic cellular debris. Of the 318 larvae sectioned, 26 were classified as severely or moderately emaciated, and another 11 were classified as incipient. These are listed by net tow in Table 1 along with the raw data for all tows. The larvae classified as incipient are included with the healthy rather than with the emaciated or "starving" group in the sections that follow.

Relation of Emaciated Larvae to Other Variables

Standard Length

Emaciated larvae were all <10 mm SL and were distributed almost proportionately over the range 2-10 mm SL (Table 2). Larvae classified incipient were similarly distributed. In the lowest size category, 2.1-4.0 mm SL, only half of the 46 larvae examined had exhausted their yolk and become vulnerable to starvation. The emaciated individuals were part of this contingent. The absence of emaciated larvae in the categories above 10 mm SL may be a chance result of the relatively fewer numbers of larger larvae present in the tows, but there may also be some actual reduction in starvation effects at this size because of increasing lipid reserves with growth (Love 1974).

TABLE 2.—Standard length distribution of the northern anchovy larvae sectioned and examined and of those classified as emaciated or incipient.

Standard length	Number examined	Number emaciated	Number incipient
2.1- 4.0	46	6	3
4.1- 6.0	137	8	3
6.1- 8.0	60	8	3
8.1-10.0	30	4	1
10.1-12.0	26		1
12.1-14.0	14		
14.1-16.0	3		
16.1-20.0	3		

Geographical Distribution

More than half of the tows were spread over a large offshore area where abundance of northern anchovy larvae was generally low (Figure 13, Table 1) and where samples from six tows each contained a single emaciated larva. The remainder of the tows occurred in an area of a few hundred square miles off Newport Beach where larval abundance was high (Figure 14, Table 1) and where samples from four tows each contained several emaciated larvae. The fact that these four samples showed a high proportion of emaciated larvae, while others from nearby tows showed only healthy larvae, indicates that there was a contagious or patchy distribution of such larvae off Newport Beach.

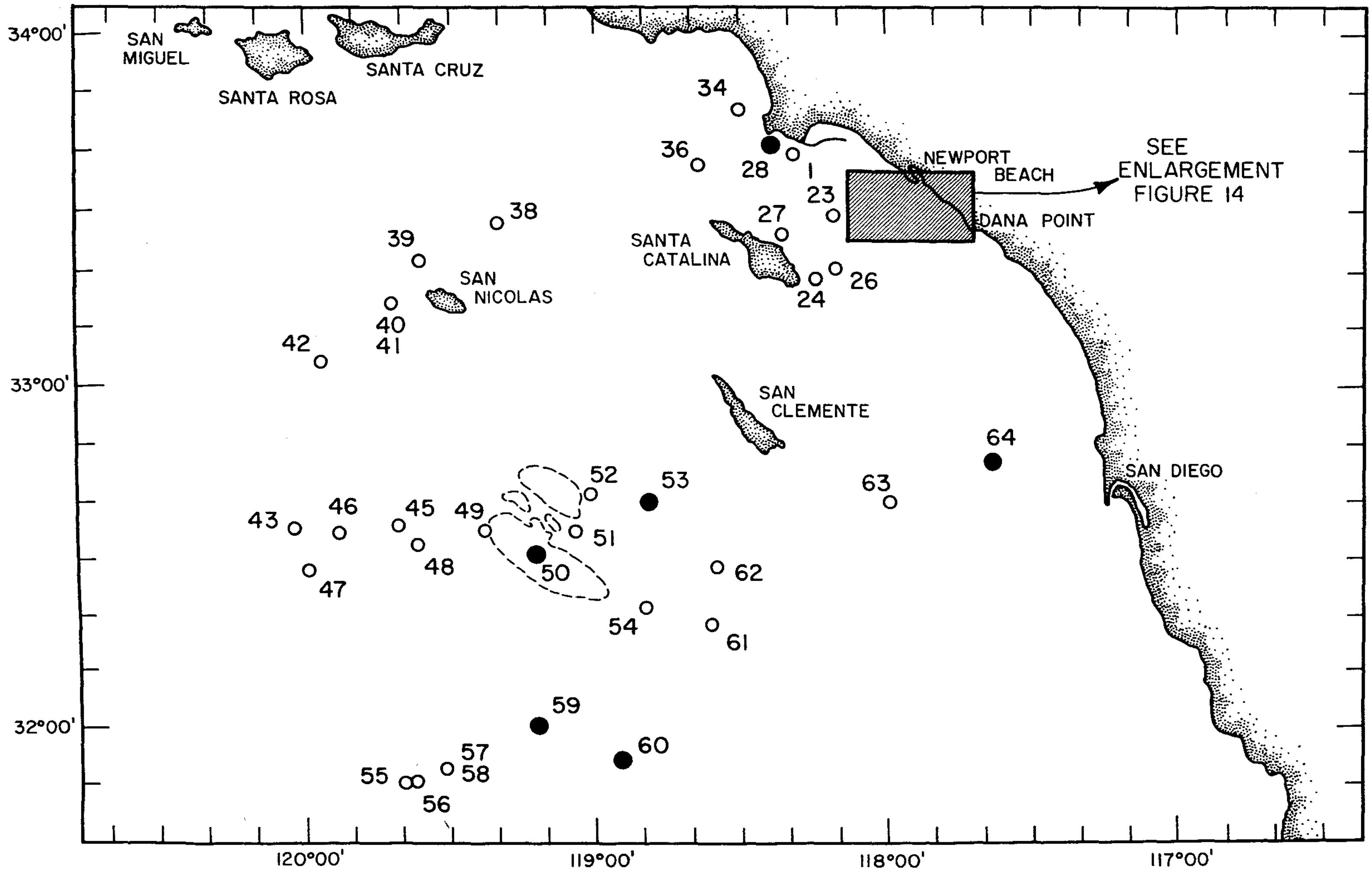


FIGURE 13.—The locations and sequence numbers of tows taken in March 1977 over the Southern California Bight, exclusive of the Newport Beach area. Open circles indicate tows from which all sectioned larvae were healthy, and dots indicate tows in which one of the sectioned larvae was emaciated. See Table 1 for data on tows.

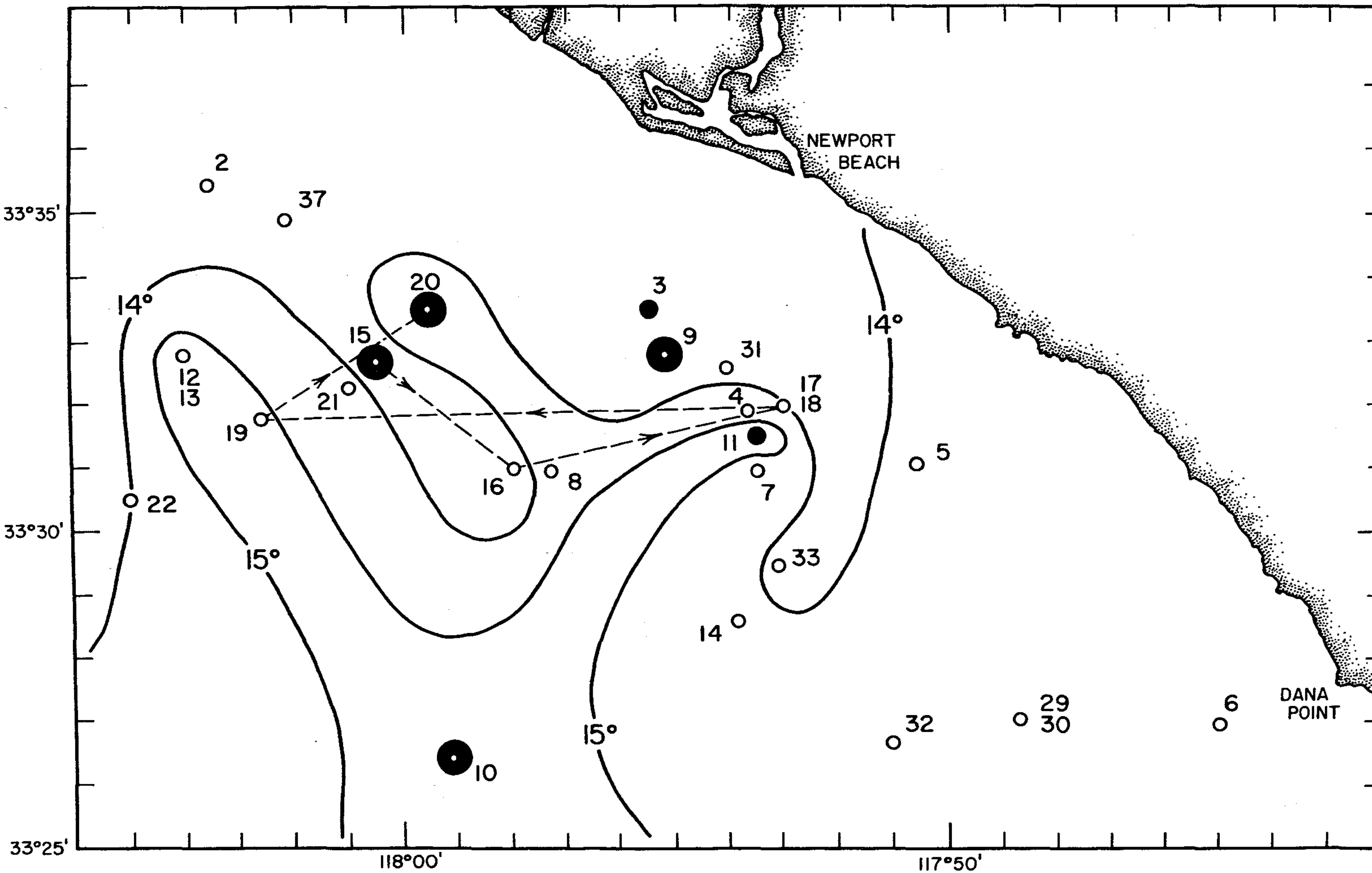


FIGURE 14.—The locations and sequence numbers of tows taken in the Newport Beach area, southern California. Open circles indicate tows from which all sectioned larvae were healthy; dots indicate tows from which one of the sectioned larvae was emaciated; larger dark circles with open centers indicate tows from which most of the sectioned larvae were emaciated; 1° C surface isotherms show that there was considerable temperature variation in the area; and dashed line is the cruise track for one 24-h period, and it indicates that the presence of emaciated larvae (tows 15 and 20) was persistent over at least this time span. See Table 1 for data on tows.

The impression of "patches" of larvae in poor condition indicated by the histological samples from the Newport Beach area was further strengthened by subsequent examination of the unsectioned larvae remaining from all net tows. Figure 15 shows a random portion of tow 23, which contained over 300 larvae and produced only healthy larvae in the histological sampling. The specimens are full-bodied with good symmetry and are straight, or at worst gently curved. Figure 16 shows a random portion of tow 9, which produced a high proportion of emaciated larvae in the histological sampling. Several of these larvae have angular body bends, trunks and digestive tracts that are lumpy and sinuous, and heads often misshapen with loose or missing eyes. They also appeared to be less intensely colored by the fixing solution than the others. When viewed in toto, this and the other three tow collections of larvae that produced histologically poor samples were readily distinguishable from all others.

The emaciated larvae consititute a percentage of the number of larvae examined, but the magnitude of this percentage depends on the portion of the total samples that are considered (Table 3). The four tows with a high incidence of emaciation, considered by themselves, indicate 60% emaciated larvae within local patches. This drops sharply to 12% when coverage is expanded to a few dozen tows in approximately 200 mi^2 off Newport Beach, and to 10% when an additional 10 tows, rather widely scattered over the San Pedro Channel area are included (inshore set). By comparison, the pooled offshore samples (offshore set) which represent perhaps 6,000 mi^2, indicate 5% emaciated, and samples pooled for the entire cruise show an intermediate value of 8% emaciated.

Day and night subsets of the inshore and offshore sets show differences in both the available population and the percentage of emaciated larvae. The lower daytime catches imply that the population was less available during this period, probably because much of it migrated below the 20 m depth of the tows during the day and probably also because larvae have some success in visually avoiding the net during the day. As a simple binomial function, the 15% emaciated larvae for the inshore night group is significantly higher

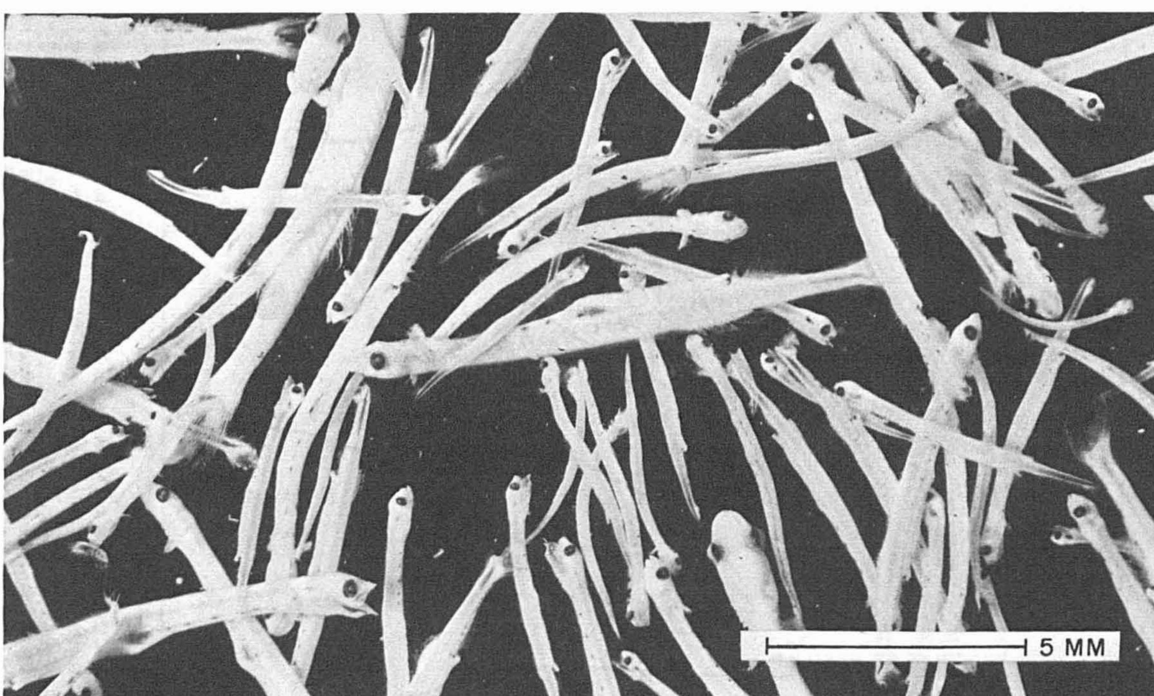


FIGURE 15.—A random portion of the northern anchovy larvae from tow 23, in which the larvae show generally good body form.

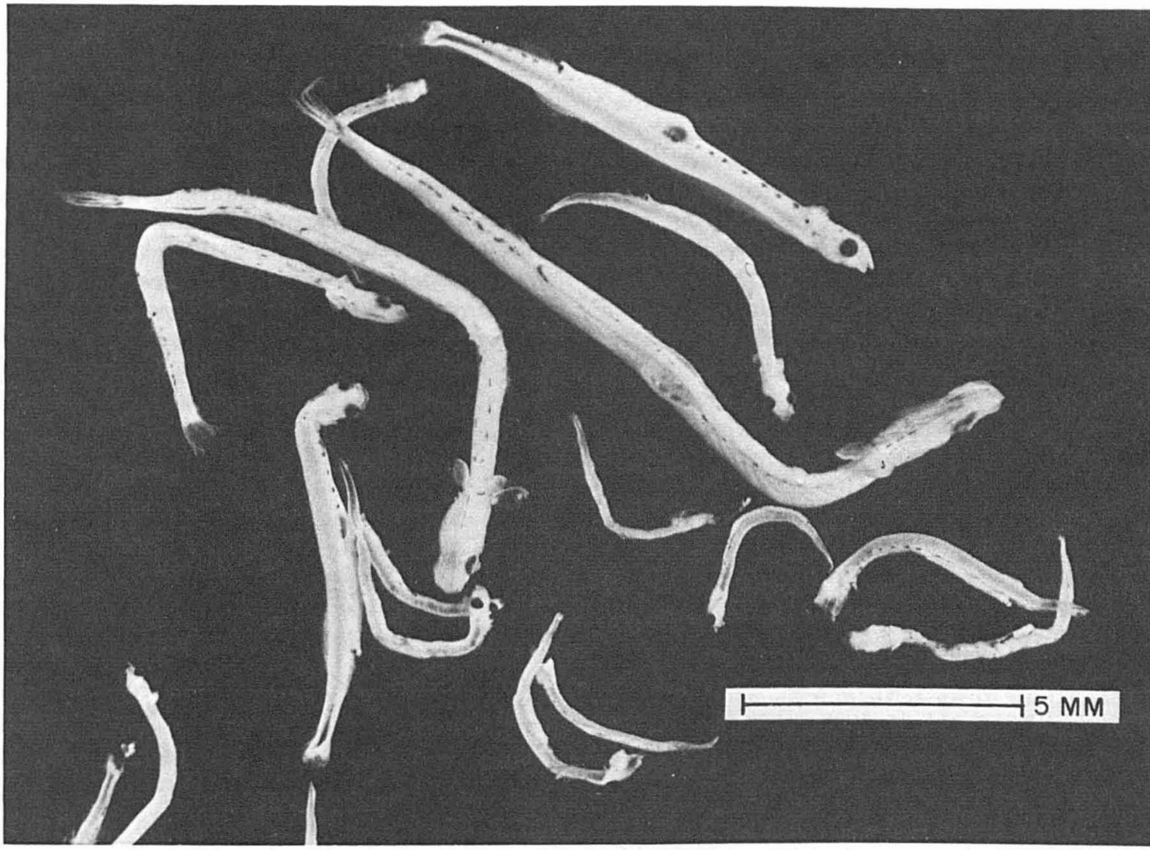


FIGURE 16.—A random portion of the northern anchovy larvae from tow 9, in which the larvae show generally irregular body form.

TABLE 3.—Percentage of emaciated northern anchovy larvae for different sample arrays.

Sample arrays	Number of tows	Larvae/ tow	Number of larvae examined	Number of larvae emaciated	Percent emaciated	95% confidence limits
Samples with high incidence of emaciation	4	37.5	30	18	60.0	±17.5
200 mi² coastal area includes above	27	77.1	165	20	12.1	± 5.0
Inshore set (tows 1-37):						
Day (0600-1800 h)	20	36.6	111	4	3.6	± 3.5
Night (1800-0600 h)	17	134.0	109	17	15.6	± 6.8
Pooled	37	81.3	220	21	9.6	± 3.9
Offshore set (tows 38-64):						
Day (0600-1800 h)	14	1.9	19	1	5.3	±10.0
Night (1800-0600 h)	13	11.2	79	4	5.1	± 4.8
Pooled	27	6.3	98	5	5.1	± 4.4
Total cruise pooled	64	50.0	318	26	8.2	± 3.0

than the other three subset percentages, but the obvious contagious nature of the distribution of these larvae casts doubt on the value of such tests. It is probably safer here to choose a lower percentage, such as the 8% for the entire cruise pooled, as representative, and assume that the observed subset differences are nothing more than sampling variation.

Surface Temperature

The tows with a high incidence of emaciated larvae were not associated with a given level of temperature, but it appears that they were located in an area of variable temperature (Figure 14). The 13° and 15° C tongues of water may have been basically persistent water masses, as Methot and

Kramer (1979) suggested, but the irregularity of the isotherms defining these tongues off Newport Beach implies local shifting of temperatures. Comparison of time as well as temperature differences along and adjacent to the cruise track for 20 March indicates, in fact, that notable temperature differences between closely located tows were probably more a matter of change over time than of static gradients between locations. Tows 15 and 20 are of particular interest because they both had a high incidence of emaciated larvae and were close together, but differed in temperature by 1.5° C. However, tow 20 was taken 24 h later than tow 15, and the thermograph record showed that the higher temperature applied to tow 15, as well as to the tow 19 and tow 20 positions, at the later time. Such short-term temperature shifts indicate that there was water mass instability or movement in the area.

Plankton Volume

Plankton volume averaged appreciably lower for the four tows with a high proportion of emaciated larvae than for either the inshore or the offshore set of tows (Table 4). The average number of larvae was also relatively low in these four tows, being about half the average for all tows off Newport Beach, or in the San Pedro Channel area (inshore set). Number of larvae and plankton volume, in fact, tend to be associated for the inshore set (Figure 17), and while the four tows with high incidence of emaciated larvae do not show the lowest values, they are among the tows with low values.

Plankton volume did not relate to temperature in the inshore area, but there were some interesting changes with time (Figure 18). From 17 March to midnight of 20 March all volumes were 11 ml or less. This includes the four tows with a high incidence of emaciated larvae, one of which (tow 20) was the last taken in this time period. Twenty-four hours later tow 21, the first tow with a markedly higher volume (21 ml), was taken at a nearby position (Figure 14). Three more tows of progressively higher volume were taken during night hours over the next few days, but there were also several tows with lower volumes (3-14 ml) taken during this period, some at night. This pattern indicates that there was a striking change in the plankton regime off Newport Beach starting on 21 March, with volume tending to be higher, especially at night, than it had been during the preced-

TABLE 4.—Mean plankton displacement volume (milliliters) for different sample arrays in the study of northern anchovy larvae off southern California.

Item	No. of tows	Larvae/ tow	Plankton Volume/ tow	Plankton SD
Samples with high incidence of emaciation	4	37.5	3.8	0.5
200 mi² coastal area includes above	27	77.1	8.3	9.3
Inshore set includes above	37	81.3	8.7	9.0
Offshore set	27	6.3	19.7	20.1
Total cruise	64	50.0	13.3	15.6

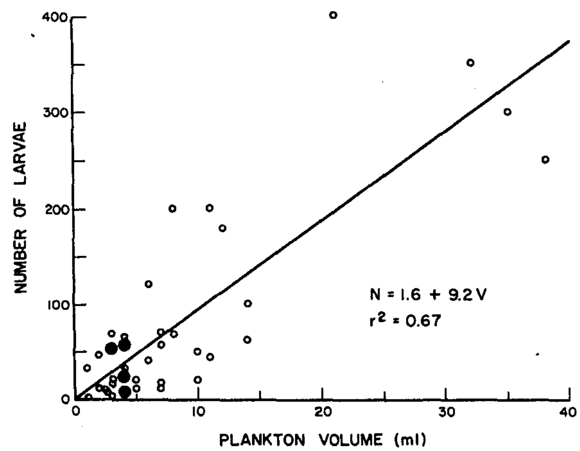


FIGURE 17.—The regression of number of anchovy larvae on displacement volume of plankton (larvae excluded) for the inshore tows, 1-37. The solid circles indicate the four tows with a high incidence of emaciated larvae.

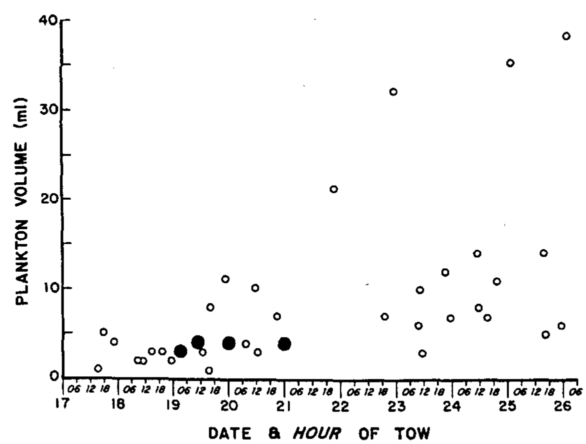


FIGURE 18.—Displacement volume of plankton on date (March 1977) and hour of tow. Dots indicate the four tows with a high incidence of emaciated larvae. Dates are in bold type and located at midnight points on the hour scale.

ing few days, when the tows with a high incidence of emaciated larvae were taken.

DISCUSSION

The most significant result of this study is that northern anchovy larvae showing symptoms indicative of starvation were indeed found in the ocean and were thus identified on the basis of their individual appearance. The emaciated condition of these larvae is very similar to that induced in laboratory animals by total food deprivation (O'Connell 1976), which implies that circumstances of insufficient food were responsible for their occurrence in the ocean, especially where samples contained many such larvae, as off Newport Beach. Other causes are possible, and one of the most obvious is discharge from sewage outfalls in the area. However, the discharges are now diffused rapidly and in general appear to be harmless and perhaps even beneficial to nearby young and adult fish (Southern California Coastal Water Research Project[3]).

Zones of insufficient food might well have existed in the Newport Beach area at the time of the survey. The variations in both temperature and plankton volume, which were clearly dynamic in nature, indicate that there was appreciable water mass movement or instability, and such conditions can alter plankton abundance. Lasker (1975, in press) found, for example, that phytoplankton blooms believed to be advantageous for first feeding anchovy larvae were variously dissipated and suppressed as the water column became unstable in turbulent weather and sea conditions. In the present study the time sequence of plankton volume change off Newport Beach shows the possibility that water with relatively low plankton levels and "patches" of emaciated larvae was being replaced by water with greater plankton abundance, in which larvae were also more abundant and generally healthy.

Although the occurrence of zones of insufficient food is a reasonable hypothesis in regard to the "patches" of emaciated larvae off Newport Beach, it is not a plausible explanation for some of the other samples, where occasional emaciated individuals occurred among abundant healthy larvae. It can only be surmised that in any location, and despite generally good conditions, there will be some instances of starvation through detrimental combinations of genetic constitution, accident, and chance failure to capture food.

If circumstances of poor food availability are produced by water mass activity, as suggested above for the Newport Beach area, they are likely to be more or less transient phenomena, which raises the question of short-term susceptibility of northern anchovy larvae. Immediately after yolk absorption, northern anchovy larvae will survive only a day or two without food (Lasker et al. 1970), and they will show visible effects before dying (O'Connell 1976). Protein components are quickly affected because early postyolk-sac fish larvae have negligible lipid reserves (Ehrlich 1974), though such reserves obviously increase with growth and become a buffer against insufficient food (Love 1974). Even so, northern anchovy larvae of relatively large size, 35 mm SL, survived only 2 wk, on the average, after feeding was stopped, and during this period the average lipid content of living larvae was declining while mortality in the population was rising, with smaller individuals dying sooner than larger ones (Hunter 1976a). Since larvae examined in the present survey were appreciably smaller than the above, thus having lower lipid reserves, the signs of emaciation could have resulted from relatively few days of insufficient food.

Most of the larvae showing histological signs of emaciation in this study also showed signs of previous feeding, not by the presence of food residue, but rather by remnants of eosinophilic inclusion bodies in the hindgut mucosa cells. The starvation and previous feeding indications are not incompatible. Laboratory feeding studies have shown that growth, lipid content, and survival all decline quickly under limited or discontinued feeding (O'Connell and Raymond 1970; Hunter 1976a), and it is probable that histological signs of deterioration would also appear quickly, especially in early larval stages.

If, as proposed by Hjort (1914, 1926), the level of mortality suffered by the early feeding stages of fish populations is a decisive component of the prerecruitment mortality, some measure of that mortality should be a useful indicator of ultimate year class success. The proportion of larvae observed to be starving may be one such useful indicator: It is directly visible, and it may reflect a substantial part of total daily mortality. Zweifel and Smith (in press) have estimated average daily

[3]Southern California Coastal Water Research Project. 1978. The effects of the ocean disposal of municipal waste. Summary Report of the Commission of the Coastal Water Research Project, June 1978, 27 p. Filed at 1500 East Imperial Highway, El Segundo, CA 90245.

population abundance of larval anchovies by length and region for the 1967 through 1975 California Cooperative Oceanic Fisheries Investigations net tow data, and length dependent mortality rates were calculated from the abundance estimates. For larvae of 7.5 mm SL, which approximates the median length of those showing symptoms of starvation in the present study, the estimated average daily mortality rate in the San Pedro Channel area was 21%. If it is assumed that all larvae showing symptoms will die directly or indirectly from starvation, the observed 8% with symptoms in the March 1977 survey could indicate a net daily mortality from starvation of 8%, which is 40% of the average total daily mortality for this length group. If starvation tends to contribute this substantially to total mortality, variations in the proportion of larvae observed to be starving may relate reasonably well to the magnitude of ongoing total mortality and consequently to recruitment from the year class.

How well the proportion of starving larvae from a given sampling in 1 yr will predict the eventual recruitment of that year class will only be evident from correlation of the two variables for at least a few years. As for 1977, with a northern anchovy "starvation ratio" of 8%, there were indications that recruitment would be good. The winter and early spring were relatively mild, a condition conducive to development of high density patches of larval food organisms, particularly from dinoflagellate blooms (Lasker in press). Growth rate of northern anchovy larvae was also shown to be above average in the San Pedro Channel area for March 1977 (Methot and Kramer 1979). The above average growth rate may have been valid for much of the population developing in the region without applying to the patches of emaciated larvae, which were taken at different locations than the growth samples. Estimates from recent catch data indicate that the 1977 year class is of moderate size, as compared with the large 1976 and 1978 year classes and the small 1974 and 1975 year classes (J. S. Sunada[4]). Thus, to the extent that 8% starving larvae is a reliable estimate of that parameter for 1977 and to the extent that the parameter is associated with recruitment, both higher and lower occurrences of starving larvae are likely possibilities from future surveys.

[4]J. S. Sunada, Assistant Biologist, Marine Resources Region, California Department of Fish and Game, 350 Golden Shore, Long Beach, CA 90802, pers. commun. May 1979.

Reliability of the estimate of starving larvae is probably reasonably good for 1977 in that tows were most concentrated in a region of high abundance, but reliability in future sampling efforts could probably be improved by more diligent stratification in respect to population distribution. In addition to sampling at more than one point in time, an effective strategy might be to expand sampling in several areas that show high abundance, particularly of larvae under 10 mm SL, along a preplanned survey track. Expanding sampling in this way would likely result in a quantity of samples that would be formidable if analysis is entirely dependent on histological or physiological parameters. The distinctive appearance of the aggregated larvae from those few tows of the present survey that contained predominantly emaciated larvae, however, suggests that histological analysis can be greatly reduced.

Assuming that starvation of any consequence will occur in patches, a stereomicroscope scan of the total aggregation of larvae from each tow should suffice for the identification and enumeration of such patches, with histological processing reserved for selected verification. Undoubtedly some of the starving larvae that occur as scattered single cases would be missed under such a procedure, but this should have little effect on the overall estimate.

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