# ROLE OF THE OIL GLOBULE IN SURVIVAL AND GROWTH OF STRIPED BASS (Morone saxatilis) LARVAE

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## Introduction

After years of studying the striped bass (Morone saxatilis) population in the Sacramento-San Joaquin Estuary it was recently concluded that the number of bass entering the fishery is largely a function of survival during the early life stages (Stevens, 1977). Although they found survival was positively correlated with river flows, the direct causative factors which affect egg and larval mortality have not been determined. At the National Marine Fisheries Service, Tiburon Laboratory, we have been working with the assistance of the California Department of Fish and Game on experiments to determine how different inherent and environmental variables affect survival, growth and development in striped bass eggs and larvae. In the course of these experiments we noticed the vital role the oil globule played in allowing feeding larvae to adapt to stresses. In this paper we present results which show how striped bass larvae depend on the energy rich oil globule to meet metabolic requirements under various food densities.

## Methods and Materials

Eggs artificially fertilized by a single striped bass male were obtained from the California Dept. of Fish and Game Central Valleys Hatchery at Elk Grove, California during the 1976 and 1977 spawning season. After water hardening, eggs were transported in styrofoam containers to the Tiburon Laboratory and incubated at 18° C in glass McDonald jars. Larvae hatched approximately 48 hours after fertilization and were allowed to collect in a 50-gallon fiberglass tank. The fourth day after fertilization larvae were transferred to green acrylic plastic containers shaped like 1/2 spheres. Containers used for subsampling of larvae were stocked with 150 larvae per pan. Containers used for measuring survival were stocked with 25 larvae per pan. Two replicates for subsampling and two for survival were filled for each food concentration. Larvae were fed Artemia salina in 3 densities of 6.2, 4.0 and 2.2 per ml, hereafter referred to as high, medium and low concentrations. These fed larvae were compared to unfed larvae in subsequent measurements. Temperature, pH, and dissolved oxygen were measured daily (Table 1). Salinity was maintained at 3.0 ppt from hatching

until day 9 at which time it was increased to 10.5 ppt. Water and food were changed on alternate days. Survival was determined daily by making 3 replicate visual counts in each pan. Preliminary point-of-no-return experiments were run in which 2 pans with 25 larvae each were starved for one week and fed. Two more pans with 25 larvae each were starved for 2 weeks, then fed. Samples of 5 to 10 larvae were taken daily for measurements of growth and development, the specimens being anesthetized first with ms 222 and preserved. The oil globule in each egg and larvae was measured for volume (ellipsoid volume = $(\pi LH^2)/6$) with an ocular micrometer in a dissecting microscope. One half of each day's specimens of each treatment was dissected to remove the oil globule and food from the gut. We removed only food from the guts of remaining larvae and left the oil globule intact. Both sets of specimens were dried at 66°C for 24 hours and weighed on a Cahn electrobalance to the nearest microgram. Dry weights of the oil globules were obtained by subtracting the average dry weight values of the dissected larvae from those of the intact larvae. Tissue weights were represented by larvae with the oil and food removed.

Caloric contents of oil, tissue and whole eggs were determined with a Parr adiabatic calorimeter. Oil was extracted and concentrated by means of a freon solvent extraction. Tissue samples were taken from larvae which had completed total oil consumption.

Percent dry weight composition of the lipid fraction in freshly spawned eggs was calculated by a 2:1 chloroform/methanol extraction in a micro-Sohxlet extractor. Results were checked by using heptane as a solvent and less than 1% variation was obtained. The chemical content of the extract was analyzed by NMR (Nuclear Magnetic Resonance) analysis at the California State University, San Francisco.

Results - (35 mm colored and black and white photomicrographs accompany this section illustrating growth and development of the embryo and larva and the progressive consumption of the oil globule.)

Description -

Maturation of the ovum prior to fertilization is characterized by a coalescence of many oil globules into a single large globule (Bonn et al, 1976; Mansueti, 1958). The dominant features of the relatively large, (average diameter of 3.4 mm), pelagic egg are the chorion, large perivitelline space, yolk and oil globule. Eggs averaged 280 micrograms (µg) in total weight. Estimates of the percent dry weight composition of the oil globule differed according to the solvent used for extraction. Freon extraction estimates of two females from 1976 were 56.8 and 38.0% (2 replicates/female). Chloroform / methanol (2:1) extraction of a third female from 1977 showed 63.4% (4 replicates).

The varying amounts of oil in eggs from different females resulted in different caloric contents per unit weight of eggs, 8323 and 7816 cals/gm. Eggs averaged 2.21 calories per egg, higher than most other fish species. This original egg energy consisted of 1.38 oil calories and 0.83 yolk calories. Oil was found to be a high energy source with 9291 cals/gm.

In recent (1977 season) tests of the chemical content of the oil it was found by NMR analysis to consist primarily of tryglycerides. This agrees with descriptions of oil globules in other fish eggs (Smith, 1957).

Utilization of the Oil Globule -

Prior to feeding the embryo and yolksac larva rely solely on endogenous energy sources--those contained in the yolk and oil globule. We found the yolk was consumed first. Total yolk consumption coincided with the onset of feeding, 7 days after fertilization, 5 days after hatching. The oil globule, however, remained approximately the same in size and weight until this time.

The rate of oil globule consumption was markedly different between fed and starved larvae (Figures 1, 2 and 3). Starved larvae conserved their oil globule as shown in the volume dry weight determinations and did not grow or develop at all until their deaths. In contrast fed larvae consumed their oil in nearly identical patterns. As larvae fed, they grew in direct relation to the food available (Figure 2). The only effect of food concentration on oil consumption in larvae was at the low concentration. These larvae contained oil for a longer period (up to day 27). The high density larvae had no oil after day 18. Experiments this year in which we exposed larvae to a wider range of food concentrations appear to consume oil in inverse proportion to the food concentration. Complete results of these experiments are still being analyzed.

Survival -

Larval survival began to decline (Figures 4 and 5) once larvae relied on either oil alone (as in the starved condition) or on oil and food combined to meet metabolic requirements. The highest mortality rates occurred as total oil consumption approached (days 14 to 22). At the end of the experiment on day 29, the percent surviving was directly related to the amount of food. The more food available the higher was the larval survival rate. Starved larvae survived to day 29 in the 1976 experiments and in the current 1977 experiments starved larvae were still alive after 34 days.

The point-of-no-return, the irreversible time at which starved larvae are unable to feed if food is presented, was never reached. Larvae starved one week and fed survived at 57% by day 29 and those starved two weeks survived 24%.

## Discussion

The presence of oil globules in fish eggs and larvae is ecologically and phylogenetically widespread (see Russell, 1977 for marine fish examples). Some fishes have numerous droplets as Solea solea and Trachinus vipera while others like the striped bass have a single large globule. Although present in many other fishes nowhere in the literature have we found it in such large amounts. One problem is there are few papers describing the chemical composition of fish eggs. For comparison Salmo gairdneri (irideus) had 10.7% of its egg dry weight in oil (Hartman et al, 1947) and California sardine (Sardinops caerulea) eggs were found to have 13.0% (Lasker, 1962). The oil globule was first thought to have an important role in flotation (Cunningham, 1889). This was soon disproved when eggs without oil globules were found to have similar specific gravities (Russell, 1976). It is our conclusion that the oil globule serves a role of providing a high energy source in a compact form. The caloric content of oil was found to be much higher than that of yolk, 9291 versus 6917 calories/gm. In this way larvae have a large amount of energy available to meet metabolic needs in a form which does not require a large volume.

Utilization of the oil, which is characterized as mainly carotenoid-pigmented glyceride fats, is usually last (Smith, 1957; Blaxter, 1969). Proteinaceous yolk was consumed prior to feeding. This shows a more efficient use of the animal's resources since the yolk energy contains less energy per unit volume and weight.

Feeding larvae consumed their oil globule at faster rates than did starved larvae (Figures 1, 2 and 3). Evidently starved larvae were able to metabolically adjust to the absence of food, thereby conserving the only energy source available to them. Starved larvae did not grow during the experimental period of 29 days. Once food was given, however, as we found in the point-of-no-return experiments, the larvae proceeded to grow at normal rates.

Results of the point-of-no-return experiments and the long survival of starved larvae up to 29 days reveal a metabolic flexibility within striped bass larvae which has high survival value. This is especially important when one considers that striped bass larvae use estuaries as nursery grounds. These are generally ecosystems with highly variable environments. The oil globule provides the larvae with a reserve energy source upon which the animal can rely when food supplies are insufficient.

Table 1. Mean and range values of physical/chemical conditions in striped bass larval feeding experiments

| | Mean | Range |
|---|---|---|
| Temperature | 18.4 | 16.9 - 19.4 |
| Dissolved oxygen | 8.4 | 7.7 - 9.1 |
| pH | 7.8 | 6.9 - 8.2 |

## Literature Cited


Blaxter, J. H. S. 1969. Development: eggs and larvae. In W. S. Hoar and D. J. Randall (editors) Fish Physiology. Volume III, pp. 178-252.

Bonn, E. W., W. M. Bailey, J. D. Bayless, K. E. Erickson, and R. E. Stevens. 1976. Guidelines for striped bass culture. Publ. of the Striped Bass Committee of the Southern Division, American Fisheries Society, p. 103.

Cunningham, J. T. 1889. Studies of the reproduction and development of teleostean fishes occurring in the neighborhood of Plymouth. J. Mar. Biol. Ass. U. K., 1, 10-54, Pls. I-VI.

Hartman, M., F. Medem, F. Graf, R. Kuhn, and H. Bieleg. 1947. Untersuchungen über die Befruchtungsstoffe der Regenbogenforelle. Z. Naturforsch. 2, 330-349.

Lasker, R. 1962. Efficiency and rate of yolk utilization by developing embryos and larvae of the Pacific sardine Sardinops caerulea (Girard). J. Fisheries Res. Bd. Can. 19, 867-875.

Mansueti, R. 1958. Eggs, larvae and young of the striped bass, Roccus saxatilis. Maryland Dept. of Res. and Ed., Contrib. No. 112, p. 35.

Smith, S. 1957. Early development and hatching. In M. E. Brown (editor) The physiology of fishes. Academic Press Inc., New York. pp. 323-359.

Stevens, D. E. 1977. Striped bass (Morone saxatilis) year class strength in relation to river flow in the Sacramento-San Joaquin Estuary, California. Trans. A.F.S., 106(1):34-42.

Russell, F. S. 1976. The eggs and planktonic stages of British Marine fishes. Academic Press, New York, London, San Francisco. p. 524.

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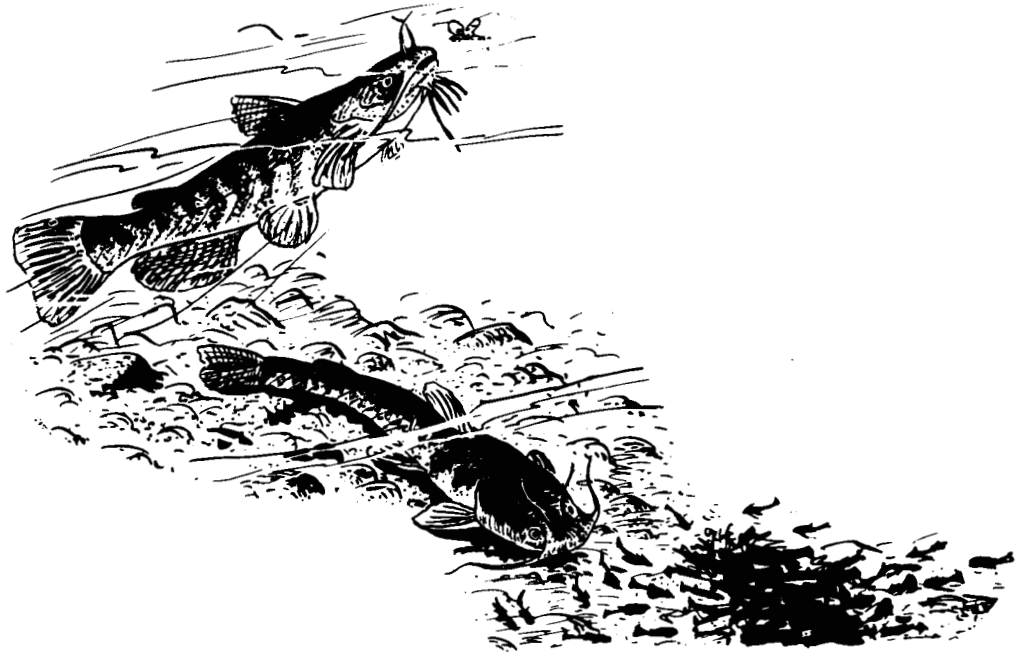
Figure 1. Dry weights (mg) of tissue and oil globules of striped bass larvae starved for 25 days.

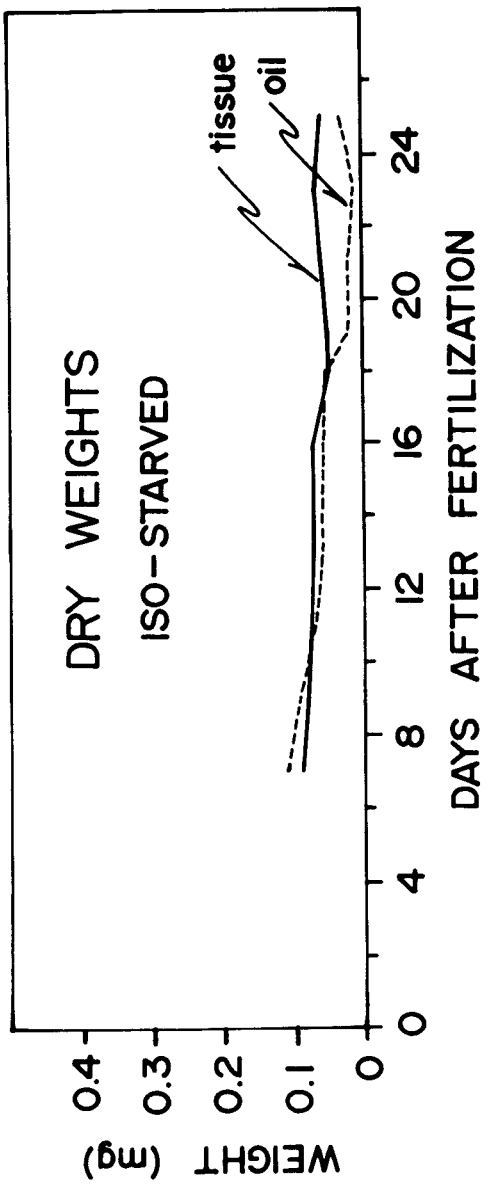
Figure 2. Dry weights (mg) of tissue and oil globules of striped bass larvae fed Artemia salina in 3 different concentrations.

Figure 3. Oil globule volumes ($mm^3$) of striped bass larvae reared in 3 different concentrations of Artemia salina and starved.

Figure 4. Mean percent survival of starved striped bass larvae reared for 29 days after fertilization.

Figure 5. Mean percent survival of striped bass larvae fed 3 different concentrations of Artemia salina for 29 days.




DRY WEIGHTS
ISO-STARVED
tissue
oil
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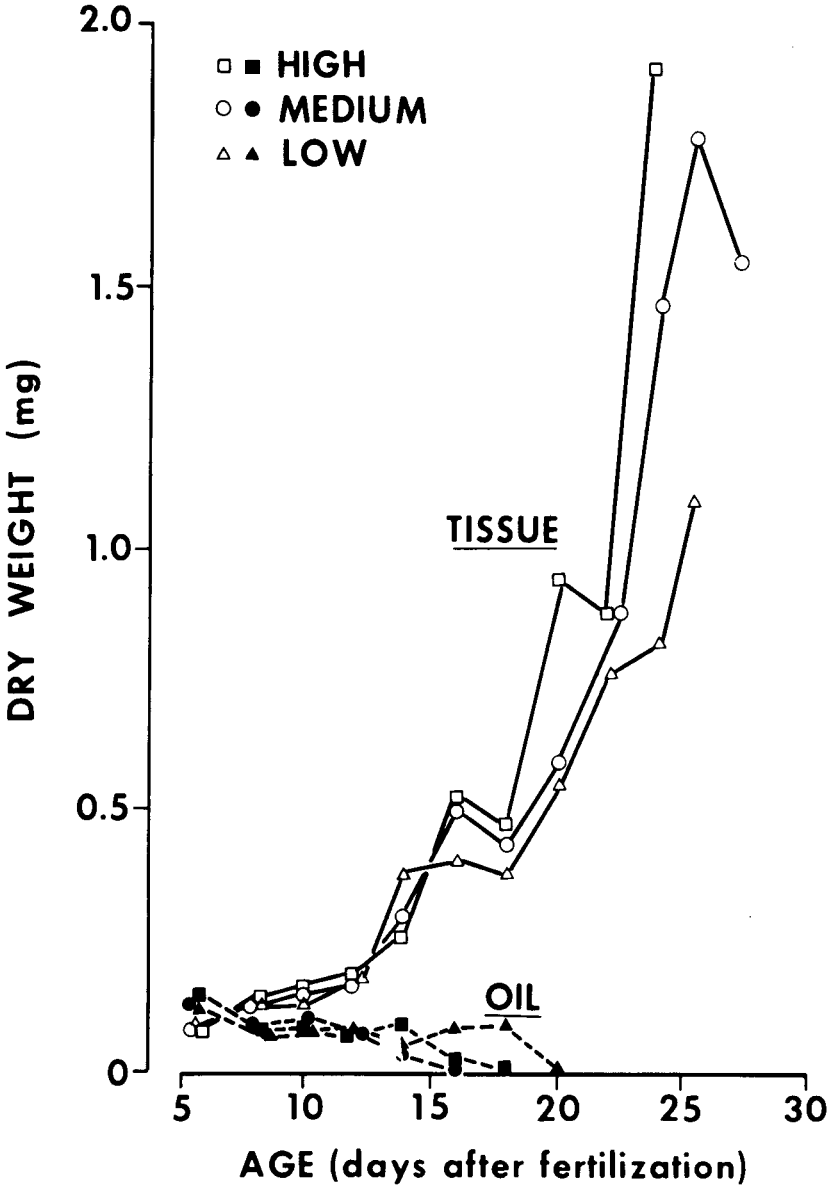
0.2
0.1
0
0
4
8
12
16
20
24
DAYS AFTER FERTILIZATION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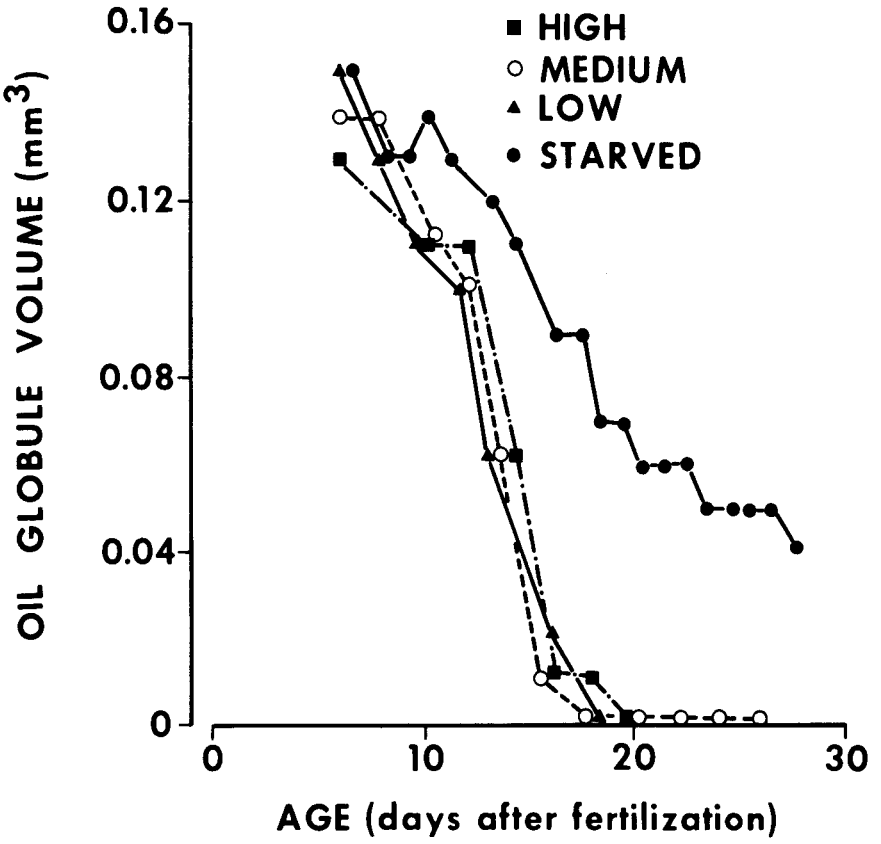
1.0
0.5
0
DRY WEIGHT (mg)
HIGH
MEDIUM
LOW
TISSUE
OIL
5
10
15
20
25
30
AGE (days after fertilization)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HIGH
MEDIUM
LOW
STARVED
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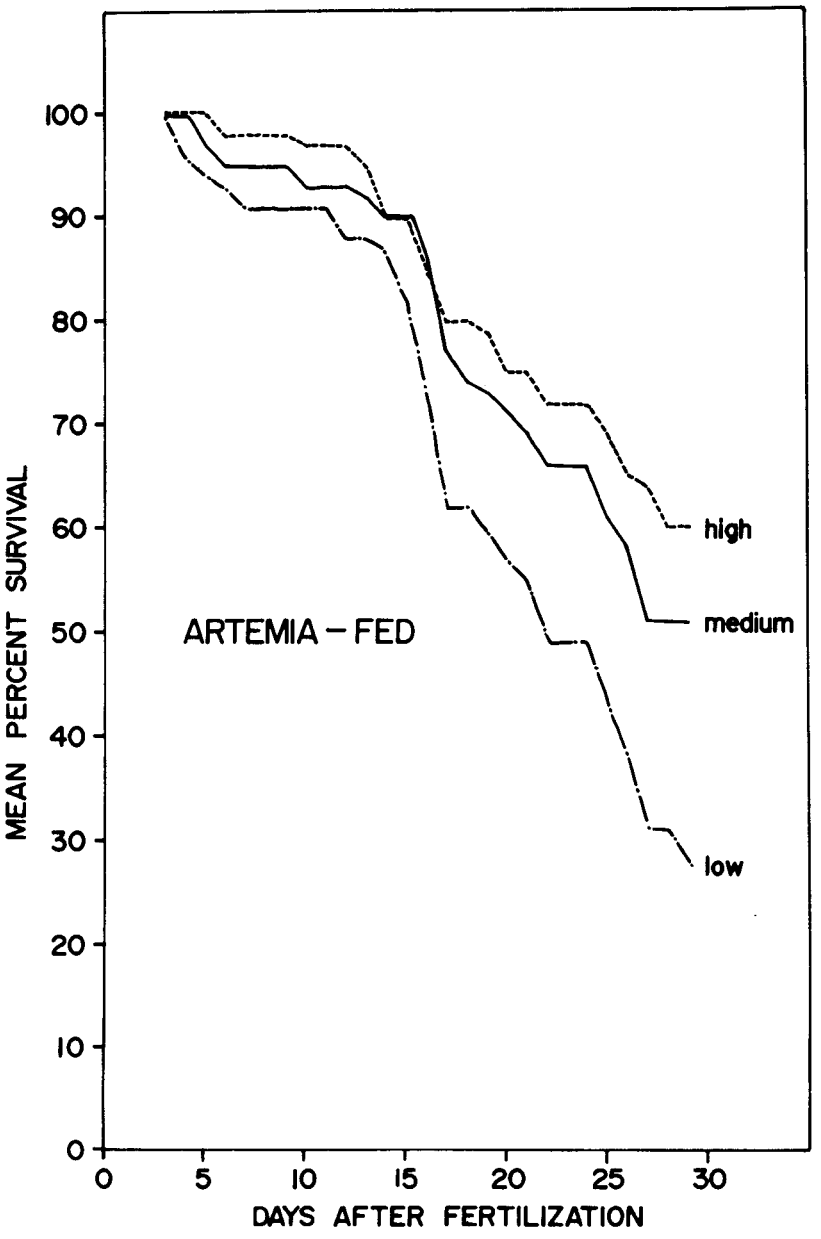
10
20
30
AGE (days after fertilization)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10
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ARTEMIA – FED
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medium
low
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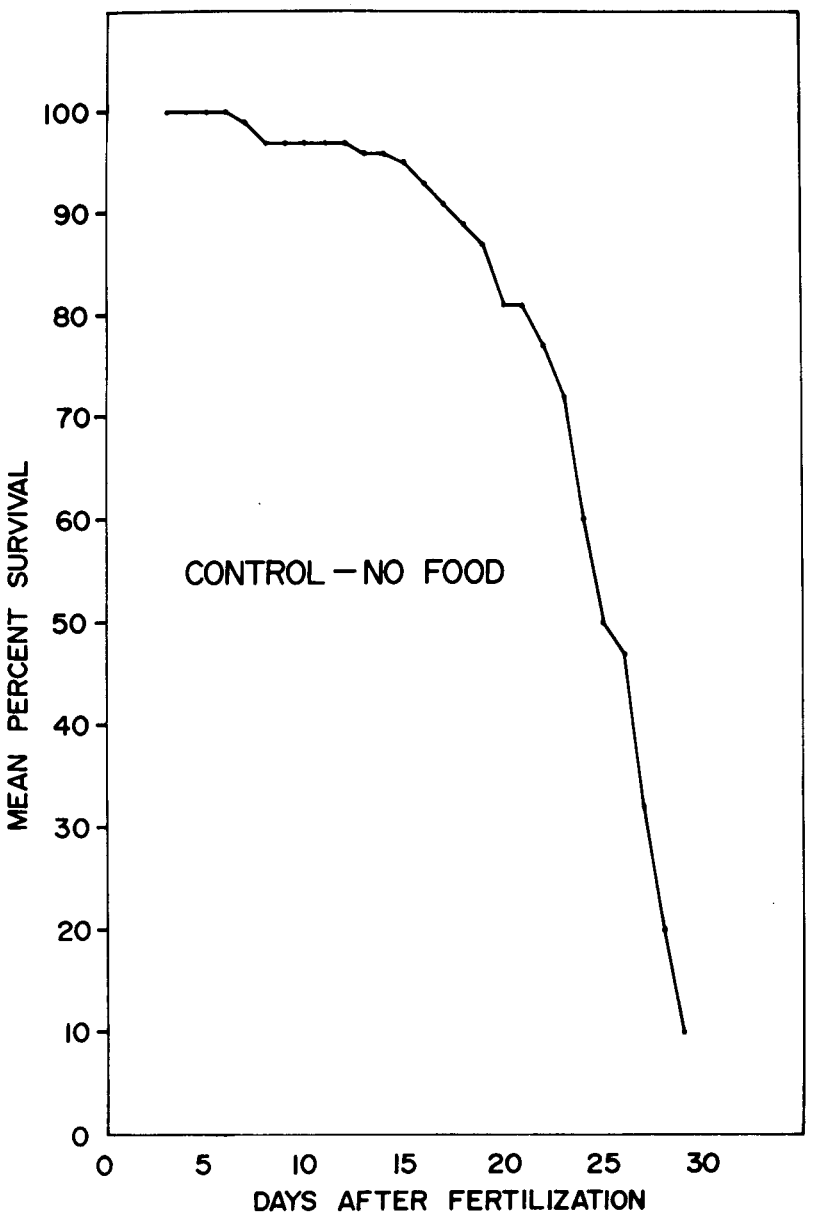
10
15
20
25
30
DAYS AFTER FERTILIZATION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MEAN PERCENT SURVIVAL
CONTROL — NO FOOD
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5
10
15
20
25
30
DAYS AFTER FERTILIZATION