FEEDING BEHAVIOR, FOOD CONSUMPTION, GROWTH, AND RESPIRATION OF THE SQUID *LOLIGO OPALESCENS* RAISED IN THE LABORATORY

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ABSTRACT

The squid *Loligo opalescens* was raised in the laboratory to a maximum age of 100 days on a diet of *Artemia* nauplii and adults. Newly hatched squid (2.7 mm mantle length) readily attacked *Artemia* nauplii (length 0.7 mm), *Artemia* adults (length 5 mm), copepods (length 1 mm), and larval fish (length 4 mm). Feeding rates varied between 35 and 80% of squid body weight per day. Growth rate was highly variable in different individuals, ranging from 0.5 to nearly 4.5 mm mantle length per month. Respiration rates were obtained at 15°C for squid of three different ages and at 10°, 15°, and 20°C for 1-day-old squid.

The squid *Loligo opalescens* Berry is a common pelagic predator off the west coast of North America from British Columbia to Baja California. Because a fishery exists for this species, considerable information is available concerning adults in the spawning schools (Fields 1965), but little is known about the early life stages. In a paper on larval squid abundance off California, Okutani and McGowan (1969) found few *L. opalescens* in their samples; and McGowan (1954) reported that despite considerable effort he could not catch newly hatched *L. opalescens* over the spawning grounds.

To obtain information on the early life history, I reared *L. opalescens* in the laboratory. Several workers have succeeded in rearing decapod cephalopods, but all of the species they used tend to be closely associated with the bottom (Choe 1966, three species of *Sepia*, the squid *Sepioteuthis lessoniana*, the sepiolid *Euprymna berryi*; LaRoe 1971, *S. sepioidea*; Boletzky et al. 1971, four species of *Sepiola* and two species of *Sepietta*; Arnold et al. 1972, the sepiolid *E. scolopes*). Attempts to raise pelagic species such as *Loligo opalescens* have met with little success (Fields 1965; Arnold et al. 1974). Workers have attributed their failure to lack of food and to infections. I describe here a simple technique for rearing early stages of *L. opalescens* and present data on the growth, respiration, and food requirements of *L. opalescens* reared for 100 days in the laboratory.

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MATERIALS AND METHODS

Five groups (referred to as groups 1 through 5) of squid have been reared, three (1 through 3) of which will be described in detail in this report. Eggs were collected from a water depth of 20 m off La Jolla, Calif., and were maintained in circulating seawater at about 13°C. The young squid were transferred to the rearing tanks after they had hatched. Fields (1965) and McGowan (1954) have described the methods of egg deposition and structure of the egg masses in detail.

The rearing tanks were cylindrical (122 cm diameter, 36 cm deep) and made of black fiber glass. Tanks were illuminated by fluorescent lights which had a cycle of 18 h light, 6 h dark. During the dark period, lights in other rooms of the aquarium building provided a source of dim light. The tanks were immersed in water baths which kept the temperature within the tanks between 15° and 17°C. Squid were transferred to the rearing tanks with a beaker. Squid in groups 2 and 3 were counted during transfer. In group 1, the number of squid was estimated after the squid were in the tank. Groups 1 and 2 began with 300 squid; group 3 began with 250. The water in the tanks was noncirculating. Each tank was aerated by a gently bubbling air supply. The squid in group 1 were transferred to a holding tank on day 62 and on day 76, and on each day their tank was drained, cleaned, and refilled. Tanks 2 and 3 were both similarly cleaned on day 49. Dead food was removed from the bottom of all tanks with a siphon, and small amounts of seawater were added to maintain a constant volume.

During the first 4 wk the squid (groups 1 through 3) were fed newly hatched brine shrimp, *Artemia salina*, nauplii which were kept at densities ranging from 1 to 20 nauplii/ml. After this time, small adult brine shrimp were added (average length 5.4 mm; range 2.5 to 8.0 mm) and were the major source of nourishment for the remainder of the rearing period. In groups 4 and 5, small adult *Artemia* as well as nauplii were used as food during the first 4 wk.

Squid were measured using an optical micrometer on a dissecting microscope. Measurements are of dorsal mantle length (measured dorsally from the tip of the tail to the farthest anterior point on the mantle). Mantle length is less variable than a measurement of total length, which depends upon the degree of stretch of the arms and tentacles. To make possible conversions to total length, measurements were made of both dorsal mantle length (ML) and total length (to tips of arms, not tentacles) (TL) on 35 juvenile animals, and the average ratio ML/TL was 0.62 ± 0.014 (±2 SE). Measurements are all on freshly dead unpreserved animals. For weight measurements, squid were rinsed in distilled water and oven dried at 60°C to a constant weight.

Respiration measurements were made using a Warburg constant volume respirometer with respiration vessels kept at constant temperature in a water bath. The respiration vessels contained from 2 to 30 squid and were kept in constant motion by gentle shaking.

Estimates were made of the number of squid surviving at intervals throughout the study. The number of squid alive on any day was the average of three counts taken of live animals in the tank.

Daily observations were made of the feeding behavior of the squid. At various times throughout the day, a squid was selected and observed for about 5 min. The number of feeding attempts and successful captures of prey were recorded.

RESULTS

Survival

Mortality in all of the tanks was initially high (Figure 1). This is similar to what LaRoe (1971) found in rearing *Sepioteuthis sepioidea*. LaRoe speculated that the high initial mortality was due to insufficient quantities of food. This probably was not the case in my studies, as a large amount of food was continually available at this stage. Some of this mortality could have been caused by squid which did not initiate feeding. Fields (1965) found that *L. opalescens* which did not appear to be feeding lived up to 10 days and still had some internal yolk reserves left at the end of this time. From 30 to 60 days mortality was low, but after 60 to 70 days mortality again increased. It is possible that the brine shrimp did not provide an adequate diet for squid older than 60 days.

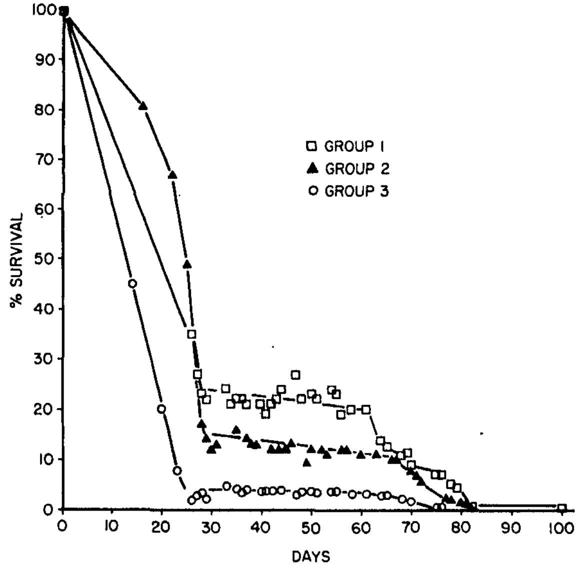

FIGURE 1.—Estimated percent survival of *Loligo opalescens* in the rearing tanks. Group 1 started with 300 squid; group 2, with 300 squid; and group 3, with 250 squid.

Feeding Behavior

Attack

The attack of a young *L. opalescens* is similar to that described for adult *Loligo* (Fields 1965), *Sepioteuthis* (LaRoe 1970), and *Sepia* (Messenger 1968). Messenger divided the *Sepia* attack into three motor patterns: attention, positioning, and seizure. These three patterns may also be used to describe the attack of young *L. opalescens*. During attention, the squid orients toward a particular prey. The arms and tentacles are extended in front of the squid and form a tight cone which is pointed toward the prey. Color changes such as those noted for *Sepioteuthis* (LaRoe 1970) and *Sepia* (Messenger 1968) were not observed.

After the squid oriented toward a particular prey, it approached the prey until it was within attacking distance (positioning). This distance was not constant. At times there was no clear separation between the attention and positioning patterns. LaRoe (1970) suggested that the positioning approach is an example of an aggression-fear conflict. This appears to be the case in *Loligo*. The young squid would sometimes flee rapidly after closely approaching a large prey.

The prey was usually captured with the tentacles (seizure), although occasionally the arms alone were used. The arms were used to maneuver the food toward the mouth. At times a new attack began while the squid was holding other prey in the arms.

LaRoe (1970) reported that for *Sepioteuthis sepioidea* physical fights over food were rare. This was not true for young *L. opalescens*. Fighting between squid was never observed when prey was small (brine shrimp nauplii), but if the prey was large and could not be completely enclosed within the arms, other squid would often chase the one which caught the food and try to take the food away from it. Often several (in one case, four) squid held on to the captured prey and all fed on it. The prey would be tugged about until one squid pulled it away from the others. This behavior occurred even when there was an abundance of prey in the tank. This attack on captured prey at times allowed small squid to eat larger prey organisms than they could normally subdue alone.

Prey Selection

Unlike *Sepioteuthis* (LaRoe 1971), young *L. opalescens* were not extremely selective as to the type and size of prey they would attack. Within a few days after hatching, the young *Loligo* (2.7 mm ML) readily attacked *Artemia* nauplii (0.7 mm long), *Artemia* adults (5 mm long), copepods (1 mm long), and larval fish (4 mm long). Occasionally, squid attacked and ate dead prey (e.g., dead *Artemia* dropped into the tank), but usually the food had to move before it was attacked. An exception to this was that the squid attacked fish larvae which appeared to be motionless in the water.

When the squid were 17 days old, nine squid from group 2 were placed in a small cylindrical container (8 liters of water) to determine whether a food size preference existed in *Loligo*. The food used was *Artemia* nauplii (0.6 to 0.8 mm long) at 10/ml and small adult *Artemia* (2 to 4 mm long) at 0.2/ml. After the squid were added, I recorded the number of attacks until a prey was captured and the type of prey being attacked. If no prey was captured in 20 min, I selected another squid. At this age, the squid attacked both large and small prey. During the 164 min of observation, 23 nauplii were attacked (9 actually captured) and 30 adults were attacked (8 actually captured). These results are different from those given for *Sepioteuthis sepioides* (LaRoe 1971). That squid only attacked food species in a very limited size range. Within several days, *Sepioteuthis* would cease to attack the prey it had previously eaten and would only attack larger prey. This seemed to occur when the squid were 1 to 1½ times as large as their prey. Although *Loligo* captured both large and small prey with about equal frequency, a preference may exist for larger prey as their density in the container was much lower.

An experiment was run with group 1 when the *L. opalescens* were 49 days old. In this case the choice was between two different prey species of approximately the same size. Two thousand 2-day-old chub mackerel, *Scomber japonicus*, larvae were added to one of the rearing tanks where the squid had been feeding on *Artemia* adults. There were approximately 2,000 *Artemia* in the tank. The same method was used to record feeding as in the previous experiment. Observation time in this case was 69 min. The squid showed a high incidence of attacks on fish larvae (52 attacks, 6 captures) even though the success rate was much lower than when attacking *Artemia* (4 attacks, 3 captures). This may indicate a preference for fish larvae, but without further experiments it is impossible to say whether this is true.

Feeding Success

The ability of the squid to successfully complete an attack sequence depended on the size and species of prey and the age and experience of the young squid. Figure 2 is a record of the percent of successful attacks on *Artemia* nauplii as a function of the age of the squid. Each point is an average from the squid observed during that day. The number of squid observed per day ranged from 5 to 11, with the total daily observation time ranging from 25 to 55 min. The attack efficiency increased with the age of the squid, but a number of prey were still being lost even after 3 wk. LaRoe

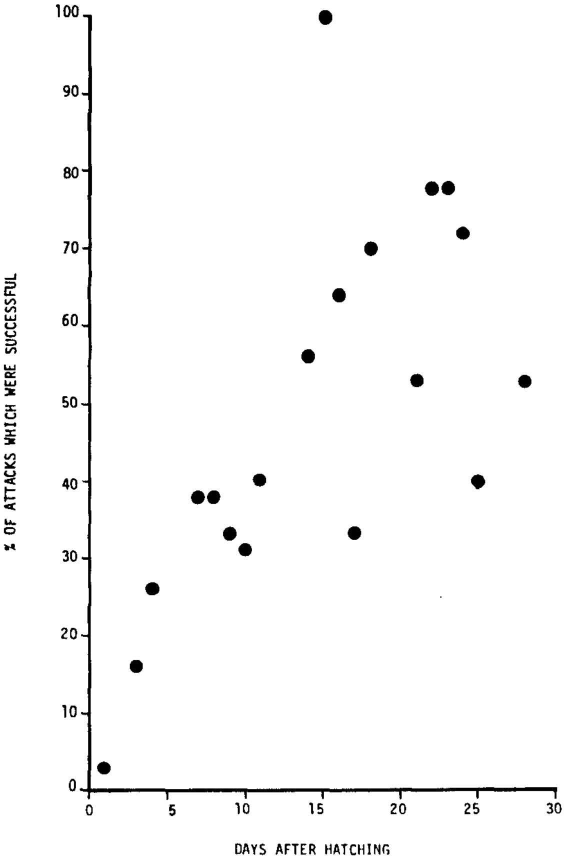

FIGURE 2.—Percent of attacks on *Artemia* nauplii which were successful as a function of the age of the squid.

(1970) found that for *Sepioteuthis*, the majority of the prey were lost because the squid were unable to judge the attack distance. In my experiments, most unsuccessful attacks occurred because the prey managed to escape after being initially struck. Some of the variability in success rates may have been due to different motivational states of the squid.

Feeding Rates

Several methods were used to determine the food ration of the developing squid. When the squid fed on nauplii, feeding rates were determined at irregular intervals by choosing a squid and watching it for 5 min to determine the number of *Artemia* nauplii consumed during this period. All of the observations accumulated during a given week were combined. For each week, I calculated the food eaten over a 24-h and 18-h feeding period. The squid captured prey when the overhead lights were off, but it was not possible to establish how much was eaten. When adult *Artemia* was the primary source of nourishment, record was kept of the approximate number of food organisms introduced to the tank and their average weight. There is some error introduced here because some of the brine shrimp died and were not consumed. The average weight of the squid during each week was obtained from the growth data and length-weight relationships presented in the next section. Average weight of *Artemia* adults was 0.3 mg (obtained from six random samples of 10 to 20 individuals each) and average weight of nauplii was 0.002 mg (John R. Hunter pers. commun.). Food consumption is shown in Table 1.

TABLE 1. — Estimated feeding rates (percent body weight eaten per day) of squid in rearing tanks. Each value is average for all values for a given week. Values through week 4 are based upon observed short-term feeding rates on *Artemia* nauplii and are given for assumed 18- and 24-h feeding periods. Subsequent values are based on counts of *Artemia* adults consumed in tanks 1 and 2.

	Nauplii		Adults	
Week	18 h	24 h	Tank 1	Tank 2
1	46	60	—	—
2	46	61	—	—
3	47	63	—	—
4	37	50	—	—
5	—	—	—	—
6	—	—	36	45
7	—	—	67	80
8	—	—	48	51

One short-term experiment was performed to examine the feeding rate of 36-day-old squid on yolk-sac larval anchovies. Five squid were placed with 100 anchovy larvae in 8 liters of water and were left for 285 min. At the end of this period 58 larvae had been eaten. This gives a feeding rate of 2.4 larvae/squid·hour. Theilacker and Lasker (1974) gave the average weight of a larva of this size as 0.022 mg. Using this information and the average weight of the squid, a feeding rate of 0.028 mg anchovy/mg squid·h is obtained.

Growth

Since the number of squid being reared was small, specimens were not sacrificed for growth measurements alone. Every time a squid died, it was immediately measured. These measurements constitute the majority of the points on the growth curve shown in Figure 3. The points indicated by the ×'s are measurements which were made on squid that had been selected while alive

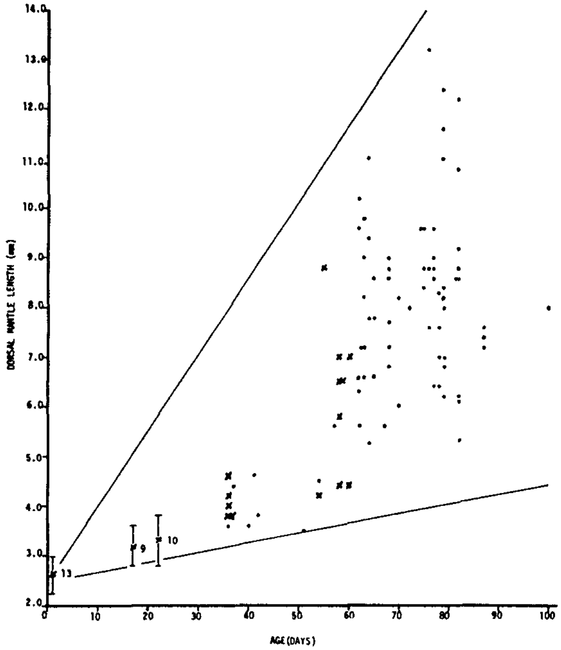

FIGURE 3.—Size data for *Loligo opalescens*. A dot denotes measurement made on squid which had died, and × denotes measurement made on squid that had been selected while alive to give an indication of the size range of individuals in the tanks. For days 1, 17, and 22, the numbers of squid measured, means, and ranges are given. The upper solid line gives a constant growth rate of 4.5 mm/mo. The lower one gives a rate of 0.5 mm/mo.

to give an indication of the full size range of squid in the tank. Since the squid were not randomly sampled during this time, Figure 3 cannot be taken to give an average growth rate for the population, but it does give an indication of the range of growth rates. There was a large difference in the rates of growth of individuals. Maximum growth rates were nearly 4.5 mm/mo (upper line in Figure 3). Minimum growth rates were 0.5 mm/mo (lower line in Figure 3).

The linear regression equation for the log length-log weight relationship for the developing squid is log weight (mg) = −1.22 + 2.37 log length (mm) with little scatter around the regression line.

Respiration

Measurements were taken of the oxygen consumption of young *L. opalescens* using a Warburg respirometer and a constant temperature water bath. Measurements were taken at 15°C for squid of three different ages and at 10°, 15°, and 20°C for 1-day-old squid (Table 2). Average oxygen consumption values are as follows: 1 day, 10°C, 1.5 μl O_2/mg squid·h; 1 day, 15°C, 2.5 μl O_2/mg squid·h; 1 day, 20°C, 3.5 μl O_2/mg squid·h; 3 wk, 15°C, 3.5 μl O_2/mg squid·h; 8 wk, 15°C, 3.7 μl O_2/mg squid·h. These measurements may be artificially high because of the crowding which occurred in the small respiration vessels. It was observed, however, that the oxygen consumption tended to decrease (at a given temperature) with increasing number of animals present in the same vessel. It is possible that these lower rates occurred because some of the animals became moribund in the crowded conditions. But this is not likely, since the respiration rates remained constant over the course of the 2-h experiments.

To compare these measurements to those made by other investigators, conversion factors had to be obtained to transform dry weight to wet weight. The ratio wet/dry was calculated for nine juvenile squid and gave a mean of 5.4 ± 0.21 (±2 SE). Wet weights were calculated by placing the squid on the weighing pan, blotting it with filter paper, weighing it at measured time intervals, and extrapolating the line obtained to zero time.

The previous rates expressed in terms of wet weight are: 0.28, 0.46, 0.65, 0.65, and 0.69 μl O_2/mg squid·h. These values are similar to those obtained by LaRoe (1971) for 2- and 6-day-old *Sepioteuthis sepioidea* (0.64 μl/mg·h at 23°C) and with the figure of 0.60 μl/mg·h for adult *L. pealei*, calculated from data in Redfield and Goodkind (1929).

TABLE 2. — Oxygen consumption rates for *Loligo opalescens*. Respiration vessels had a volume of 18 ml and contained approximately 5 ml seawater. The duration of the experiments was 2 h.

Temp. (°C)	*N*	Age of squid (days)	Number of squid/vessel	Range of oxygen consumption (μl O_2/mg squid (dry wt)·h)
10	3	1	10-30	1.4-1.6
15	3	1	8-25	2.1-3.6
20	3	1	10-21	3.2-3.8
15	1	21	10	3.5
15	2	56	2-3	3.5-3.9

DISCUSSION

It is extremely difficult to assess the role which an animal such as *L. opalescens* plays in the California Current ecosystem. Estimates of population size of adults are very poor because of the difficulties involved in sampling large active animals. Fisheries statistics are not particularly helpful because the catches come mainly from a few locations. It has been possible to get some field information on the diet of the adult squid (Fields 1965) but these data are completely lacking on such necessary information as feeding rates.

It appears to be equally difficult to obtain information on young *L. opalescens* from field samples. The young squid have well-developed eyes and are very sensitive to vibrations. Therefore, even the young are likely to be able to avoid many nets. Okutani and McGowan (1969) published data on the abundance of young *L. opalescens* (size range 3.5 to 7 mm dorsal ML) taken in net tows during the California Cooperative Oceanic Fisheries Investigations cruises in 1954 to 1957. In their report, however, they emphasized the problems involved in sampling the young squid and stressed that the abundances given probably should only be used to compare relative abundances of different species. They found that *L. opalescens* was the third most abundant species of larval squid present in their samples, but that its abundance was quite low when compared to the most common fish larvae present (e.g., 0.008 times the abundance of northern anchovy, *Engraulis mordax*).

If the role of a young *L. opalescens* as a predator is to be evaluated, it is necessary to know the type of prey which it eats. Fields (1965) has determined the diet of the adult squid from an examination of stomach contents, but to my knowledge no one has done a similar study on the very small squid. From the laboratory results presented in this paper, it appears that young *L. opalescens* must be considered as predators on a wide range of prey types and prey sizes. They are capable of preying on species ranging in size from 0.7 to 7 mm and they readily attack prey species ranging from brine shrimp adults and nauplii to copepods and larval fish. McGowan (pers. commun.) has found that they also successfully attack the mysid *Metamysidopsis elongata*.

It is also possible to use the data presented here to estimate a feeding rate for the young squid. The respiration data can be used to calculate the amount of food a young squid would need to sustain itself. The respiration rate of the squid in the rearing tanks can be taken as 3 μl O_2/mg dry wt·h. An average value for the caloric value of oxygen consumed is 5×10^{-3} cal/μl of O_2. Therefore, a newly hatched squid (2.7 mm ML, weighing 0.625 mg) would use 0.22 cal for respiration alone in 24 h.

It is possible to determine how many prey items of different types of prey would satisfy this requirement. A newly hatched *Artemia* nauplius is the equivalent of 0.0096 cal (John Hunter pers. commun.). Therefore, a newly hatched squid would need 23 *Artemia* nauplii per day. If the squid were instead feeding on newly hatched northern anchovies, it would need a total of 2 anchovy larvae per day (using a value of 5 cal/mg, weight of larva = 0.022 mg; Theilacker and Lasker 1974). Similar calculations can be made for older squid. A squid 7 mm ML (~2 mo old, 6 mg) would consume 225 nauplii or 20 anchovy larvae simply to meet its metabolic needs. The actual amount of food consumed per day was appreciably more than this, averaging about 50% of body weight per day. At this rate, a newly hatched squid would consume 150 nauplii or 14 anchovy larvae per day, while a 7-mm squid would consume 1,500 nauplii or 135 anchovy larvae per day.

Data on feeding rates and abundance could be used to calculate the impact that young squid might have on populations of potential prey items, but before such calculations can be meaningful, more information must be known about the ability of the squid to locate sources of food. *Loligo opalescens* was only one hundredth as abundant as the most common fish larvae (Okutani and McGowan 1969). But with feeding rates of 15 to 135 larvae per day, young squid could potentially have a large impact on such populations if they concentrate on this type of food and if they have effective means of finding such prey. Laboratory observations indicate that larval fish may be a preferred food, and the squid do occur in areas where larval fish are common. Okutani and McGowan found that *L. opalescens* was most common in the upper 40 m, and this is the stratum where the highest abundance of northern anchovy larvae occur (Ahlstrom 1959).

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