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Spawning and early development of *Loligo vulgaris* Lamarck, 1798, under experimental conditions

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Because of its characteristics and high market price, efforts are being made to introduce the European squid Loligo vulgaris *into aquaculture. The results of previous trials to rear different stages of the squid did not satisfy the needs of commercial culture.*

In this study, three live females of L. vulgaris *were jigged from the shore in front of the Institute of Oceanography and Fishery in Split, Croatia (northern Mediterranean Sea) in May 2002. After a short time in captivity, the squids spawned spontaneously, leaving their strands on the water exchange pipe immersed in the pool. On day 21 after spawning, the first hatchlings were observed. The paralarvae were fed small fish pellets and brine shrimp* Artemia salina *in open circuit aquaria and glass pots, but died after eight days. For economic production and rearing, more sophisticated studies are needed, concentrating on nutritional requirements and balanced diet of early development stages.*

Key words: *Loligo vulgaris*, paralarvae rearing, experimental conditions, fish pellet, *Artemia salina*

INTRODUCTION

The common squid *Loligo vulgaris* Lamarck,1798 is a very interesting species for aquaculture. Even though it has been well studied and there are many papers about its fecundity and life in general, there are still unknown aspects of their life stages.

Experimental trials for rearing different stages of *L. vulgaris* were described by BOLETZKY (1974) who raised *L. vulgaris* to the age of 45 days, HURLEY (1976) who reared *L. opalescems* to 100 days and TURK *et al.* (1986) who succeeded in maintaining *L. vulgaris* for 140 days after hatching. However, the results of these trials were not commercially applicable.

Egg dimensions are 2.0-2.2 x 1.5 x 1.6 mm (LAPTIKHOVSKY, 2000). They hatch into planktonic paralarvae (YOUNG & HARMAN, 1988) which are active swimmers and need a large amount of food to maintain their rapid growth of more than 5% body weight per day (FORSYTHE & VAN HEUKELEM, 1987). The squid's growth is rapid and allometric (KRSTULOVIĆ-ŠIFNER, 2001), but its life span is relatively very short (RAYA *et al.*, 1999).

L. vulgaris has an extended and intermittent terminal spawning period (ROCHA & GUERRA, 1996). Oogenesis proceeds in six histological stages (LOPES *et al.*, 1997), meaning that oocytes grow and develop asynchronously. The relationship between fecundity and the dorsal mantle length is only weakly correlated, meaning that small females are capable of harboring more oocytes than larger ones (GUERRA & ROCHA, 1994).

The estimated duration of planktonic life is about two months (WORMS, 1983). Twenty days after hatching, squids begin to swim in a horizontal position for several minutes, maintain their position against a current for more than 5 minutes and swim several centimeters in pursuit of prey (TURK *et al.*, 1986). As soon as the paralarvae hatch, until the end of their life, they eat a carnivorous diet (BOUCAUD-CAMOU *et al.*, 1985; BOUCHER-RODONI *et al.*, 1987; BOUCAUD-CAMOU & ROPER, 1995, 1998).

The aim of this work was to spawn and raise *L. vulgaris* from hatching through the critical first few days to maturity in different rearing conditions. The paralarvae were fed small fish pellets and brine shrimp *Artemia salina* in open circuit aquaria and glass pots.

MATERIAL AND METHODS

Three live female *L. vulgaris* were jigged from the shore in May 2002 in front of the Institute of Oceanography and Fishery in Split, Croatia (northern Mediterranean Sea). They were placed in a cylindrical concrete pool with a capacity of 3 m^3. The sides of the pool were black and the bottom was white.

The squid were fed European anchovy (*Engraulis encrasicolus*) and European pilchard (*Sardina pilchardus*) cut into 2 cm pieces, small frozen prawns or juvenile fish from a trawl boat *ad libitum* twice a day (early in the morning and eight hours later). This feeding continued throughout the period of strand attaching and afterwards. Feeding must be slow and accurately placed, as squid do not take food from the bottom of the pool. The water flow was 15 l min^{-1}, the temperature ranged 17.6-21.0°C and the salinity ranged 32.9-35.2 ppt. The pool was inside a building and maintained in a natural photoperiod.

After a short time in captivity, the squid spawned spontaneously leaving their strands on the seawater exchange pipe immersed in the pool. On day 21 after spawning, the first hatchlings were observed. Three days later, 30 paralarvae were caught and placed in three glass pots, each with a capacity of 5 l. A few strands with eggs were placed in three 15 l aquaria. Both the glass pots and the aquaria had air pumps. The temperature ranged 19.6-20.6°C and salinity was the same as in cylindrical concrete pool.

During the first five days, the paralarvae were fed smashed fish pellets and, afterwards, freshly decapsulated *A. salina*. Every day, the temperature was measured, the aquaria and pots were cleaned and the dead paralarvae were removed. The tank bottom was cleaned daily by siphoning and, every two days, 2/3 of the sea water was changed.

Several days after the eggs hatched, the female squids died.

Morphometric measurements of the dorsal mantle length and the total wet weight were recorded daily from the dead paralarvae (Fig. 1).

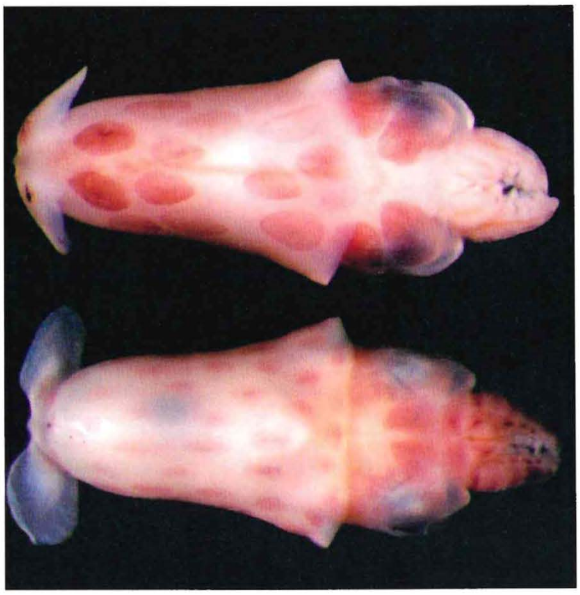

Fig. 1. Dorsal (upper) and ventral (lower) view of three days old Loligo vulgaris

RESULTS

The female *L. vulgaris* spawned in captivity in the cylindrical concrete pool of 3 m^3. The squids in the study were larger than the average Adriatic population; their mean dorsal mantle length was 146 mm compared to the average mean dorsal mantle length of 114.9 mm (KRSTULOVIĆ-ŠIFNER, 2001). The squids acclimated quickly and their food consumption was satisfactory. Life in captivity was easy to maintain but a larger pool might have been better because of the squids' active and rapid movements and frequent changes of direction. No cannibalism or aggressiveness was recorded in the relatively small pool, although cannibalism is not unusual for squids in these conditions (FISHER *et al.*, 1987).

The adult squids were somewhat shy but their predatory instincts during feeding overcame their shyness, resulting in their attacking even the thermometer in their attempts to catch food.

As spawning in *L. vulgaris* is intermittent, the squids laid their strands during a 5-day period. They approached already attached strands carefully and gently to lay the next ones. They moved very close to the end of the pipe where the strands were laid and were obviously disturbed when we approached the strands. Squids usually attach their strands to the bottom where the current is weak but, in this case, the strands were attached to the end of the pipe were the water flow was very high. The simple reason may be that the bottom contained no natural materials such as stones, pieces of wood or floral material to which the strands could be attached. The high infusion of fresh sea water seemed not to bother the squids or the strands. The eggs hatched as frequently in the concrete pool with the high water flow as in the aquaria. In nature, squids attach their strands to a hard surface (RODHOUSE *et al.*, 1990), however, strands can be found from February till June attached to the leaves of *Posidonia oceanica* (KRSTULOVIĆ-ŠIFNER, 2001).

Fertile eggs had a smooth surface, were dark yellow and had a mean diameter of 2.09 ± 0.037 mm.

After the adult squid's natural death, their ovaries were weighed. The mean weight was 4.64 g; the ovaries were empty, loose and yellowish red.

After hatching, the young paralarvae were easily noticed on the surface of the concrete pool by their swimming movements, as their buoyancy control was not yet properly developed. Their mean dorsal mantle length was 3.63 ± 0.29 mm, and total wet weight was 0.0094 ± 0.0005 g.

The hatchlings seldom oriented toward or attacked food organisms that were more than a few millimeters away, so food had to be given to them in their vicinity. Care was taken to administer freshly decapsulated *A. salina* every day. When the mouth diameter of the paralarvae surpassed the length of the *A. salina*, they were given the chance to chase and catch live food when hungry. Food consumption was poor. The paralarvae indeed showed predatory behavior, but the ingested food was not always digested. Fish pellets were mashed to the smallest particles and although the paralarvae swam in the direction of the pellets, chemotactically attracted by organic material, the pellets were not engulfed. It seemed more like a demonstration of their instinct than a real attempt at consumption. The paralarvae died after eight days.

The paralarvae held until the end of the trial in the concrete pool were fed the same diet but did not survive much longer than the paralarvae in the aquaria and in the jars, even though the former had a constant flow of sea water which enabled them to encounter their natural prey and feed as in the open sea.

Weak and dying paralarvae accumulated on the bottom, were slow in movement and the color of their eyes changed from petroleum green to reddish brown.

The conditions in the aquaria and glass pots were optimal for rearing, but the food was inappropriate (NAVARRO & VILLANUEVA, 2000). Nevertheless, live paralarvae survived for eight days after hatching.

DISCUSSION

Because of its biological characteristics (large number of eggs, rapid growth), non-aggressive behavior in captivity and high market price, *L. vulgaris* is one of the most interesting aquaculture species in the Adriatic. In terms of economic and statistical aspects, the FAO recorded a slight drop in the world's squid catch from 1992 to 1997, even though the value in the Adriatic seems to have slightly increased (FAO, 1998)

In the northern and mid-Adriatic, the distribution of squid is random and unequal and varies seasonally. The mean concentration of squid is in costal zones reaching 100 m in depth. Small juveniles, up to 35 mm where sex is difficult to determine, were most frequently caught in September, although they were found in all other periods except March and April. The smallest quantity of squids was caught in the summer (June-July) and the highest harvests were recorded in November when the temperature and salinity were relatively low (KRSTULOVIĆ-ŠIFNER, 2001).

The highest prevalence of fertile individuals (over 75%) in the Adriatic was recorded in February-May, meaning that the main spawning period occurs in winter-spring, but lower levels were also recorded throughout the year except September (KRSTULOVIĆ-ŠIFNER, 2001). The exact spawning months are dependent on abiotic environmental factors, and it can be concluded that in constant well-established rearing conditions, oocytogenesis and spermatoforogenesis can continue throughout the year, insuring the continuous production of paralarvae or, depending on technical factors, induction during a desired season. In these terms, the short life span would not be an obstacle in the rearing of *L. vulgaris*. The squid's growth is allometric, with more pronounced growth in length than in weight, so the final product would reach market size much earlier.

The abiotic factors (temperature, salinity, light) in the trial were the same as those in the open water, so the possibility that environmental stress triggered the mortality is excluded.

TURK *et al.* (1986) biweekly added a mixture of trace elements in the amount of 0.005% of the system volume in their experimental rearing, but only in the case of artificial sea water. This additive helped prevent statolith abnormalities that lead to swimming and orientation problems, extremely important for catching prey. The authors used zooplankton - estuarian or neritic copepods (*Acartia tonsa*, *Labidocera aestiva* and *Centropages velificatus*); mysid shrimps (*Mysidopsis* spp); palaemonid shrimps (*Palaemonetes* spp) and several species of small fish (*Menidia beryllina*, *Poecilia latipima*, *Gambussia affinis*, *Cypriodon variegatus* and *Fundulus* spp) - as food organisms. They were collected from the field several times a week, treated for parasites and pathogens, size-sorted and transferred to holding tanks. The authors succeeded in rearing hatchlings 140 days (dorsal mantle length 75 mm, total wet weight 28.2 g), but the effort spent on collecting and preparing live food would disqualify their technique for use in commercial circumstances. Further, their mortality was very high, with the best survival being 10%.

BOLETZKY (1979) stated that his rearing trials did not require a lot of working effort and that thorough tank cleaning or other maintenance was not performed on a daily basis during a 75-day period. A good side effect was the growth of red and green algae on the walls of the tank that prevented the abrasions typical of hatchlings reared in glass aquaria.

VILLANUEVA (1994, 2000) elaborated on the rearing of the decapod crab zoeae *Pagurus prideaux* as food for paralarvae. VIDAL *et al.* (2002) proposed that hatchlings be fed enriched *Artemia* nauplii during the first 30 days to achieve a higher survival rate. In our case, swimming nauplii attracted paralarvae and predation was successful however the survival rate was low, suggesting the possibility that other factors were involved in the mortality.

It is easy to maintain adult individuals in captivity as they naturally take fish food. However, food for rearing *L. vulgaris* paralarvae is a bottleneck to the introduction of this species in aquaculture. There are a number of papers about the nutritional requirements of cephalopods (NAVARRO & VILLANUEVA, 2000) but, at this stage, their results are achievable only on an experimental scale. For wider production and economically worthwhile rearing, more sophisticated studies must be made.

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Mriješćenje i rani razvoj *Loligo vulgaris* Lamarck, 1798, u eksperimentalnim uvjetima

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SAŽETAK

Uslijed osobina Europske lignje *Loligo vulgaris* i u svezi s njenom visokom tržišnom cijenom, postoji značajan napor za uvođenje ove vrste u akvakulturu. U prošlosti su postojali eksperimentalni pokušaji uzgoja različitih stadija Europske lignje, ali rezultati ne mogu zadovoljiti komercijalni uzgoj.

Tri žive ženke *L. vulgaris* uhvaćene su "peškafondom" s obale u svibnju, nasuprot Instituta za oceanografiju i ribarstvo u Splitu. Nakon kratkog vremena provedenog u zatočeništvu, lignje su se spontano izmrijestile, ostavljajući vrpce s jajima na cijevi za upust svježe morske vode, koja je bila uronjena u bazen. 21 dan nakon mriješćenja uočena je prva mlađ.

Paralarve su hranjene malim ribljim peletom i slanišnim škrgonošcem *Artemia salina* u akvarijima i staklenim posudama, ali su uginule nakon osam dana. Za širu produkciju i ekonomsku isplativost uzgoja, trebalo bi provesti detaljnije studije s naglaskom na prehrambene potrebe i uravnoteženost ishrane ranih razvojnih stadija.

Ključne riječi: *Loligo vulgaris*, uzgoj, eksperimentalni uvjeti, riblji pelet, *Artemia salina*