# Effects of Density on Larval Growth and Spatfall in *Pinctada fucata* (Gould)

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Abstract: The effect of density in larval rearing in the Indian pearl oyster *Pinctada fucata* was studied at the Molluscan shellfish hatchery, Tuticorin. Larval densities from 1 larva to 20 larvae per ml were tested in 4,50 and 500 l of seawater in glass beakers and fibreglass containers. *Isochrysis galbana* was given as food at the rate of 5000 cells from straight hinge stage to umbo stage, 10,000 cells from umbo stage to eyed stage and 15,000 cells/larva/day from eyed stage onwards upto spat settlement. Marginal difference in larval growth was observed between the densities in each experiment. However, an inverse relationship was noted between the densities and larval growth as well as in spatfall. The results indicated that the density at 2-3 larvae per ml was found to be the optimum which gave good growth and spatfall.

#### Introduction

In seed production programmes it is of importance to determine the suitable density in rearing the bivalve larvae so as to achieve faster growth, good survival, early metamorphosis and high spatfall. Several authors used densities ranging from 0.3 to 52 larvae/ml in the bivalve rearing studies (Davis, 1953; Loosanoff et al., 1955; Minaur, 1969; Nishikawa, 1971; Wada, 1973; Walne, 1974; Alagarswami et al., 1989).

In the present study the effect of larval density in 4 l, 50 l and 500 l of water was studies with reference to larval growth, metamorphosis and spatfall.

#### Materials and Methods

A series of experiments were conducted on larval density in containers holding 4 l, 50 l and 500 l of seawater in the shellfish hatchery at Tuticorin (lat. 8° 47′N; long. 78° 08′ E). Five litre capacity glass jars, each holding 4 l of

filtered seawater were used in three experiments. The larval concentrations 1,2,3,4,5 and 10 per ml were tested in the first two experiments and 1 to 5 per ml in the third one. The glass jars were placed in a fibreglass tank containing water to keep the jars cool and were covered by black cloth to avoid dust and algal growth. Two experiments were conducted on larval density in 50 l of seawater in fibreglass tanks  $(76 \times 50 \times 25 \text{cm})$ . The larval densities 1,2,4,6 and 8 per ml were tested in the first experiment and 2,4 and 6 per ml in the second. The effect of larval density was studied in 500 l of seawater in fibreglass tanks (200 x 100 x 50cm) in four experiments. The larval densities tried were 4,5,6 and 7 per ml in the first experiment; 5,10 and 15 per ml in the second and 2,3,4 and 5 per ml in the third and fourth experiments.

The size of larvae along the dorsoventral axis (DVM) was recorded. The growth of larvae was monitored once in two days by measuring fifty larvae. Water was changed in the rearing containers once in two days by siphoning through the nylobolt sieve of 40 and 80µm. The rearing containers were cleaned thoroughly in freshwater at every water change, rinsed with seawater and then used. Isochrysis galbana was given as food at the rate of 500 cells from straight hinge stage to umbo stage, 10,000 cells from umbo stage to eyed stage and 15,000 cells/larva/day from eyed stage onwards upto spat settlement. No aeration was given till the setting of spat occurred. The total spatfall in each density was estimated at the end of experiment.

#### Results

During the experiments in 4 l of seawater, the water temperature varied from 22.0 to 27.8 in the first experiment, 24.0 to 28.0 in the second and 26.8 to 30.0° C in the third experiment. The water temperature in the experimental tanks containing 50 l of water varied from 25.5 to 28.5° C. In 500 l of water the temperature fluctuated between 29.4 and 32.4, 29.8 and 32.8, 27.0 and 32.1 and 27.1 and 29.9°C in the four experiments respectively. The salinity ranged between 33.0 and 38.0 ppt and the pH between 8.0 and 8.2.

Table 1. Larval growth and percentage of spatfall in different densities in 4 l of seawater.

Expt. No.	Larval density (per ml)	Initial mean size of larvae (µm)	Mean growth on the day of first spatfall (µm)	Rate of growth per day (µm)	Day of first Spatfall	Percentageof spatfall
1	1	64.0	184.6	6.35	19th	89.7
	2	64.0	190.0	6.63	19th	99.9
	3	64.0	191.6	6.72	19th	92.1
	4	64.0	180.9	6.15	21st	97.1
	5	64.0	163.3	5.23	21st	71.8
	10	64.0	158.6	4.98	21st	18.9
2	1	60.7	196.4	5.03	27th	44.4
	2	60.7	195.3	4.99	27th	22.7
	3	60.7	200.3	5.17	27th	20.0
	4	60.7	188.8	4.74	27th	30.5
	5	60.7	184.6	4.59	27th	14.2
	10	60.7	181.3	4.47	27th	19.6
3	1	73.0	<b>17</b> 0.0	5.39	18th	13.3
	2	73.0	-	-	18th	45.3
	3	73.0	165.3	5.13	18th	5.3
	4	73.0	148.3	4.18	18th	2.5
	5	73.0	147.0	4.12	18th	0.9

# Larval density in 4 l of Seawater

The mean size of larvae at the time of stocking in experiments 1,2 and 3 were 64.0, 60.7 and 73.0 µm respectively. Highest spatfall of 99.9% was recorded at the density of 2 larvae/ml in experiment 1, 44.4% in 1 larva/ml in experiment 2, and 45.3% in 2 larvae/ml in experiment 3 ( Table 1). The growth was better at densities of 1-3 larvae/ml when compared of higher densities. The data on growth and spatfall in relation to density generally indicated an inverse relationship.

# Larval density in 50 l of Seawater

The mean size of larvae at the time of stocking was 64.8 and 60.3 µm in two experiments. The growth was better at densities of 1-2 larvae/ml except for higher growth at 6 larvae/ml in experiment 2. In both the experiments (Table 2) the highest percentage of spatfall was recorded in 2 larvae/ml concentration.

# Larval density in 500 l of Seawater

The mean size of larvae at the time of stocking was 60.1, 61.2, 67.5 and 62.1µm in experiments 1-4. The rate of growth of larvae is shown in (Table 3). In experiments 3 and 4 larval growth was better at 2 larvae/ml when compared to higher concentration. In the third and fourth experiments the highest spatfall was obtained at 2 larvae/ml being 31.6 and 12.8% respectively.

The first spatfall occurs simultaneously on the same day in all densities in all experiment but the settlement rate was more at lower densities and less at higher densities. At times, the spatfall was earlier in lower densities when compared with higher densities. The spatfall was prolonged even upto 30 days from the date of commencement of spat setting. However more than 50% of spatfall would be over within 4 or 5 days and the rest would settle afterwards.

Table 2. Larval growth and percentage of spatfall in different densities in 50 l of seawater.

Expt. No.	Larval density (per ml)	Initial mean size of larvae (µm)	Mean growth on the day of first spatfall (µm)	Rate of growth per day (µm)	Day of first Spatfall	Percentage of spatfall
1	1	64.8	126.8	3.88	20th	17.6
	2	64.8	141.5	4.79	20th	67.0
	4	64.8	117.8	3.31	23rd	14.3
	6	64.8	89.0	1.51	25th	0.2
	8	64.8	81.0	1.01	25th	0.04
2	2	60.3	148.3	4.19	21st	16.5
	4	60.3	146.3	4.10	21st	3.0
	6	60.3	163.4	4.91	21st	1.5

Table 3. Larval growth and percentage of spatfall in different densities in 500 l of seawater.

Expt. No.	Larval density (per ml)	Initial mean size of larvae (µm)	Mean growth on the day of first spatfall (µm)	Rate of growth per day (µm)	Day of first Spatfall	Percentage of spatfall
1	4	60.1	166.6	6.6	17th	0.72
	5	60.1	139.9	4.99	17th	0.30
	6	60.1	99.4	2.46	17th	0.12
	7	60.1	139.2	4.94	17th	0.14
2	5	61.2	151.0	4.73	19th	11.7
2	10	61.2	150.0	4.67	19th	6.1
	15	61.2	151.1	4.73	19th	5.4
3	2	67.5	174.8	7.15	15th	31.6
	3	67.5	125.9	3.89	15th	19.1
	4	67.5	164.8	6.49	15th	14.1
	4 5	67.5	144.1	5.11	15th	13.6
		62.1	172.5	6.49	17th	12.8
4	2	62.1	144.3	4.84	17th	6.8
	3	62.1	162.7	5.92	17th	8.9
	<b>4</b> 5	62.1	122.0	3.52	17th	1.3

### Discussion

Crowding is one of the important biological factors affecting growth and survival. Loosanoff et al. (1951) studied the effect of overcrowding in clam larvae (Venus mercenaria) and the results indicated that overcrowding might not be dangerous to their survival but the rate of growth was slower at higher concentrations and the time of metamorphosis was correspondingly delayed. The larvae of Mercenaria mercenaria could be grown in concentrations of 50 to 100 per ml of water (Loosanoff and Davis, 1963). Highest concentration of 20 larvae/ml was tried in P.fucata by Wada (1973) and Anuradhakrishnan and Alagarswami (1987). In the present study in P.fucata it was 15 larvae/ml. Alagarswami et al. (1983) experimented the larval concentration upto 28 per ml in *P. fucata* and Minaur (1969) 10-15 per ml in *P. maxima*.

Loosanoff and Davis (1963) reported that on 10th day the mean Mercenaria growth of larvae of mercenaria in a series of cultures containing six, thirteen, twentysix, and fifty two individuals per ml was 162, 156, 151 and 144 µm respectively. Davis (1953) used 0.6, 2.8, 18.5 and 32.9 concentration of larvae of Crassostrea virginica per ml of water and fed them with the same quantity of food. The results showed that there was an inverse relationship between the concentrations and rates of growth of larvae. The data obtained in all the experiments in the present study are comparable to the above. Though there was an inverse relationship between growth and density, a particular range of densities in an experiment showed good growth potential of the larvae. The data in this study indicates that 1-3 larvae/ml was the optimum density for faster growth of larvae. In the seed production programme larval stocking density of 2-3 per ml is recommended for better utilisation of hatchery facility.

A similar trend was also seen in the the densities. spatfall among Anuradhakrishnan and Alagarswami (1987) reported that the percentage of spatfall was 8.8, 6.0 and 1.4 in the densities 5, 15 and 20 larvae/ml respectively. The larvae of Perna indica at an initial stocking density of 5 per ml to 15 per ml showed good settlement in 5 per ml only (Appukuttan et al., 1987). Walne (1956) experimented the larvae of Ostrea edulis in different densities viz., 0.3, 0.9 and 1.3 per ml which gave the spatfall of 67.5, 54.9 and 27.0% in the respective densities. In the present investigation the densities 5, 10 and 15 per ml resulted in the spatfall of 11.7, 6.1 and 5.4 % respectively. Identical results were obtained in other densities also. However, highest spatfall was seen at a particular density in each experiment. In this study highest spatfall of 99.9% was recorded at 2 larvae/ml in experiment 1, 44.4% at 1 per ml in experiment 2 and 44.3% at 2 per ml in experiment 3 in 4 l of water; 67.0% at 2 per ml in experiment 1, 16.5% at 2 per ml in experiment 2 in 50 l of water and 31.6% at 2 per ml in experiment 4 in 500 l of water. Considering the trend of spatfall it is obvious that 2-3 larvae/ml is the optimum density for good spatfall.

A comparison of growth and percentage spatfall of larvae in 4 l, 50 l and 500 l containers show that both these parameters are high in 4 l followed by 50 l and 500 l larval rearing containers. This may be due to minimum disturbance caused to the larvae in small containers during water change when compared to larger containers where the stress factor is probably high as it takes longer time to change the water.

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