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# INFLUENCE OF PREY (*ARTEMIA* NAUPLII) DENSITIES ON THE SURVIVAL, GROWTH AND FEED CONSUMPTION OF SPOTTED SCAT SCATOPHAGUS ARGUS LARVAE, AN INDO-PACIFIC ORNAMENTAL FISH REARED UNDER CONTROLLED CONDITION

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### ABSTRACT

Spotted Scat (*Scatophagus argus*) is a popular brackish water Ornamental fish but currently sourced largely from the wild. To optimize the conditions on the food ration to be supplied and rearing with live feed, an experiment was conducted to evaluate the survival and growth with different rations of *Artemia* nauplii under laboratory conditions. 10-day post-hatch (DPH) hatchery produced larvae of Scat with a mean length of  $4.07\pm0.00$  mm were stocked (a) 10 fish L<sup>-1</sup> in 50 L fiber tanks filled with 30-L of filtered sea water and fed with five different prey densities of 4, 8, 12, 16 and 20 nauplii  $m\Gamma^{-1}$  day<sup>-1</sup>. An experiment was conducted for 15 days and was performed with six replicates. Survival of *S.argus* larvae improved linearly (80% to 100%) with feeding density. The higher feeding level of 20 nauplii  $m\Gamma^{-1}$  day<sup>-1</sup> showed a significant (p<0.05) increase in growth (13.6±0.08 mm/138.3±2.70 mg) of *S. argus* larvae, compared to the lower densities of 4, 8, 12 and 16 nauplii  $m\Gamma^{-1}$  day<sup>-1</sup>. The foraging and their feeding behavior of larvae were observed regularly and the performance indices were documented. The present findings are a first step towards the development of a production protocol for *S. argus* larvae and suggests a reliable consumption pattern with reference to the feed availability and its effect on their growth.

Keywords: Growth performance, Artemia feeding density, Spotted scat larvae.

## 1. INTRODUCTION

Spotted scat, *Scatophhagus argus* is a popular aquarium species all over the world due to its pigmentation. It is prized as a food fish in countries throughout south and South East Asia due to its high protein content and delicacy [1]. It is popularly known as the 'India discuss', and it is in high demand as an ornamental fish across the world including Singapore, Indo-Pacific islands and South East Asia region. Because of its importance in aquaculture, seed availability could not meet the needs of the aquarium industry [2]. To meet the demand, development of commercially, effective sustainable technologies for induced breeding and scat larviculture will be a promising step towards scat rearing.

For a successful economic aquaculture production, reliable and regular availability of larvae is important [3]. The transition from an endogenous to an exogenous food supply is a crucial period to early larval growth and survival. In fish larvae culture, the selection of appropriate feed and adequate feeding strategy is proportionately more important [4]. Prey size, type, and

density are critical factors for successful larval rearing in hatcheries and are required for effective live feed management. In hatcheries, feeding fish larvae with *Artemia* nauplii significantly increases their growth, development and survival rate [5]. *Artemia* nauplii fed fish showed an elevated level of EFA, resulting in improved pigmentation and metabolic activity. They can also be used to improve the quality of aquaculture plant effluent water. Adoption of an optimal feeding regime/schedule is one of the crucial strategies to be followed to meet the balanced nutrient requirements of the larvae during mass rearing. The bottleneck persists in the majority of cultured species.

Inadequate feeding practices during fish larval culture result in inefficient nutrient absorption, reducing growth performance, high mortality rate, immune-suppressed state and water quality in the growing environment [6-8]. Thus, establishing an optimal consumption rate and intervals for fish larvae is critical as it has a direct impact on survival and growth performance, food schedule, manpower optimization and productivity [9, 3]. Understanding that healthy aquaculture practices demand attentive monitoring and maintaining prey densities, this study aims to optimize *Artemia* nauplii densities and to investigate the economics of larvae production. However, due to the lack of standardized protocols for controlled larviculture, feeding trials are currently being conducted.

## 2. MATERIAL AND METHODS

### 2.1. Scatophagus argus larvae

The experiment was carried out at a Scat hatchery, CIBA, Muttukadu experimental site. *S. argus* larvae were obtained from hormonal induced broodstocks (male: female 150-350 g body weight and 160-210 mm total length) and maintained in 500 litre FRP tanks (salinity  $30.0\pm1.0$  ppt, temperature  $27\pm1^{\circ}$ C).

### 2.2. Ethics Statement

We strictly followed the rules and regulations of current Animal Welfare Laws, India and also assured that no animals were stressed or harmed in our research. Since the experimental fish *S. argus* is not endangered, the state regulations of the Indian Wildlife Protection Act of 1972 does not apply to experiments conducted with this fish.

### 2.3. Experimental design

Three hundred numbers of randomly distributed 10 DPH S. argus larvae were maintained in 50 L fiber tanks filled with 30-L of filtered seawater (to prevent the entry of other live feed and detritus) under natural photoperiod and acclimated for 7 days before the initiation of the feeding trial experiment. In six replicates, five feeding densities were tested: 4, 8, 12, 16 and 20 nauplii *m<sup>1</sup>* 'day'. The initial length  $(4.07\pm0.02 \text{ mm})$  and weight  $(3.67\pm0.02 \text{ mg})$  of 30 larvae were recorded before the commencement of feeding. To achieve high prey concentrations, newly hatched Artemia nauplii (Great salt lake, USA) were washed in sea water and concentrated in a 5 l tank. 1 ml of concentrated suspension was made into 100 ml by diluting with 99 ml sea water, from which 1ml was taken to have the nauplii count. Finally, the volume of nauplii suspension required for each experimental tank was calculated. The water temperature was maintained at  $27\pm1^{\circ}$ C. Air stones were placed in the centre of each tank to promote a homogeneous distribution of prey.

All larvae were fed once a day (8:00 am) to allow a precise estimation of the feed consumption rate. Every morning, a sample not less than 500 ml was taken from each tank at three different depths and the numbers of residual *Artemia* nauplii were counted. Dead larvae, left-

over food and faeces were siphon-cleaned an hour before each feeding, and approximately 30-40% of the water volume in each tank was renewed daily to ensure that no residual nauplii remained. The feed consumption rate was calculated by the following,

Feed consumption rate (%) = No. of nauplii given on  $1^{st}$  day - No. of nauplii remaining in the next day. Similarly the feed consumption rate was calculated and documented during the entire experimental period.

## 2.4. Survival rate

The survival rate (%) was calculate as [10], Survival (%) = {(Nf - Ni) / Ni} X 100 Where, Nf = Final number of larvae, and Ni = Initial number of larvae.

## 2.5. Growth performance

Thirty swim-up larvae from each tank were sampled at 3day intervals of the experiment. Each larva was anesthetized (0.1ppm Phenoxyethanol) and their length (mm) and weight (mg) was measured.

Specific growth rate in length (SGR  $_{(L)}$ ) was calculated as [11],

SGR  $_{(L)}$  (% day<sup>-1</sup>) = {(Final length (mm) - Initial length (mm))/ Days of rearing} X 100

Specific growth rate in weight (SGR  $_{(W)}$ ) was calculated as, SGR  $_{(W)}$  (% day<sup>-1</sup>) = {(Final weight (mg) - Initial weight (mg))/ Days of rearing} X 100

## 2.6. Water quality characteristics

The water quality parameters such as dissolved oxygen, temperature (°C), salinity (ppt), alkalinity and pH were monitored at the morning during the sampling period, (Hana Instruments, USA). Ammonia and nitrite was estimated by using water testing kit (NICE chemicals).

### 2.7. Statistical analysis

Data were analyzed by a one-way analysis of variance (ANOVA), followed by Duncan's multiple range test at p<0.05 significant level (SPSS 17.0). The percentage of data was normalized through arcsine transformation.

### 3. RESULTS

### 3.1. Survival rate (%)

Our present study ensures that an optimal feeding density plays an important key role for a higher survival rate in *S. argus* larviculture. Evidently, our experimental fish fed with different prey density of 12 (96.7 $\pm$ 5.01%), 16 (100 $\pm$ 0.00%) and 20 (100 $\pm$ 0.00%) nauplii *ml*<sup>-1</sup>day<sup>-1</sup> for 15 days showed a significant increase (*p*<0.05) in

survival rate compared to lower prey density of 4 (80.0 $\pm$ 8.71 %) and 8 (81.1 $\pm$ 7.84 %) nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (Fig. 1).



Fig. 1: Survival rate (%) of *S. argus* larvae reared at different prey densities (nauplii  $m\Gamma^{1} \text{day}^{-1}$ ) during 15 days of the experiment (Mean±SE).

#### 3.2. Growth performance

The prey density i.e. the number of *Artemia* nauplii affected the growth performance of the fish larvae significantly (Table 1). The growth parameter final mean length (mm) showed a significant increase in larvae fed with the highest food density of 20 nauplii  $ml^{-1}$ day<sup>-1</sup>

 $(13.6\pm0.08 \text{ mm})$  and it was moderately lesser in 12 nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (11.7±0.04 mm) and 16 nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (12.1±0.13 mm). Meanwhile, the prey density 4 nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (7.6±0.04 mm) and 8 nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (9.5±0.26 mm) were recorded with the lower value compared to the other higher densities. Similarly, the other parameter final mean weight (mg) also showed a significant increase in 20 nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (138.3±2.70 mg) compared to the 4 nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (41.9±1.41mg) and 8 nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (63.1±1.21mg) and moderate increase compared to 12 nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (91.8±2.24 mg) and 16 nauplii  $m\Gamma^{1}$ day<sup>-1</sup> (106.0±4.13 mg).

There is a gradual and moderate increase in SGR (L) (% day<sup>-1</sup>) proportionate to prey densities of 4, 8, 12, 16 and 20 nauplii  $ml^{-1}$ day<sup>-1</sup>. In 20 nauplii  $ml^{-1}$ day<sup>-1</sup> (0.64 $\pm$ 0.02 % day<sup>-1</sup>), the SGR (L) was found to be the highest and the lowest in 4 nauplii  $ml^{-1} day^{-1}$  (0.24±0.01 % day<sup>-1</sup>). The other prey densities 8, 12 and 16 nauplii  $ml^{-1}$ day<sup>-1</sup> showed  $0.36\pm0.22$ ,  $0.51\pm0.39$  and  $00.53\pm0.10$  % day<sup>-1</sup> respectively. Both Specific growth rate in length and weight were positively correlated to prey density (Table 1). SGR  $_{(W)}$  (% day<sup>-1</sup>) of larvae showed a reliable trend that when prey density levels were increased there was a significant (p < 0.05) increase in the growth. The highest SGR (W) was found in 20 nauplii ml<sup>-1</sup>day<sup>-1</sup>  $(24.2\pm0.10 \text{ \% day}^{-1})$  and lowest was in 4 nauplii  $m\Gamma'$  day<sup>-1</sup>  $(16.2\pm0.20 \% \text{ day}^{-1})$ . The length and weight gain showed statistically significant differences (p < 0.05) between various prey densities (Table 1).

Table 1: Growth performance (Mean $\pm$ SD) of *S. argus* larvae reared at different prey densities (nauplii  $m\Gamma^{1}$ day<sup>-1</sup>) after 15 days of the experiment.

Growth Performance	Prey Densities (nauplii ml <sup>-1</sup> day <sup>-1</sup> )				
Indices	4	8	12	16	20
Final Mean Length (mm)	$7.6 \pm 0.04^{\circ}$	$9.5 \pm 0.26^{d}$	$11.7\pm0.04^{\circ}$	$12.1 \pm 0.13^{b}$	$13.6\pm0.08^{a^*}$
Final Mean Weight (mg)	$41.9\pm1.41^{\circ}$	63.1±1.21 <sup>d</sup>	$91.8 \pm 2.24^{\circ}$	$106.0 \pm 4.13^{b}$	$138.3 \pm 2.70^{a^*}$
SGR (L) (% day <sup>-1</sup> )	$0.24 \pm 0.01^{\circ}$	$0.36 \pm 0.22^{\circ}$	$0.51 \pm 0.39^{b}$	$0.53 \pm 0.10^{b}$	$0.64 \pm 0.02^{a^*}$
SGR $_{(W)}$ (% day <sup>-1</sup> )	$16.2\pm0.20^{\circ}$	$18.9 \pm 0.11^{d}$	$21.4\pm0.16^{\circ}$	$22.3\pm0.33^{b}$	$24.2\pm0.10^{a^*}$
Length Gain (mm)	$3.53 \pm 0.15^{\circ}$	$5.43 \pm 0.19^{d}$	$7.63 \pm 0.23^{\circ}$	$8.03 \pm 0.11^{b}$	$9.53 \pm 0.09^{a^*}$
Weight Gain (mg)	38.23±1.81 °	$59.43 \pm 1.10^{d}$	88.13±2.29°	102.33±3.79 <sup>b</sup>	$134.63\pm2.12^{a^{*}}$

\* Data are Mean $\pm$ SD. Different superscripts indicate statistical differences (p<0.05).

### 3.3. Consumption rate (%)

The feed consumption rate (%) of *S. argus* larvae during experimental period is shown in Fig. 2. A significantly (p<0.05) higher consumption rate was observed with a prey density of 20 nauplii  $ml^{-1}$ day<sup>-1</sup>, while a lower rate was observed with 4 nauplii  $ml^{-1}$ day<sup>-1</sup>. Statistical differences were observed among different prey densities (p<0.05). Based on these results, a feeding

protocol with *Artemia* nauplii was proposed for the *S*. *argus* larvae rearing from 10 to 25 DPH larvae (Table 2).

#### 3.4. Water analysis

The water parameters such as dissolved oxygen  $(5.06\pm1.01\text{ppm})$ , temperature  $(28.71\pm0.58^{\circ}\text{C})$ , salinity  $(27.38\pm0.15\text{ppt})$ , alkalinity  $(150.2\pm2.32 \text{ ppm})$ , pH  $(7.5\pm0.14)$ , Ammonia  $(0.10\pm0.02 \text{ ppm})$  and nitrite

 $(0.04\pm0.01\text{ppm})$  were recorded at three days of intervals during the study period. The results of the water parameters between the treatments were found to be insignificant (p>0.05).



Fig. 2: Consumption rate (%) of S. argus larvae reared at different prey densities (nauplii  $m\Gamma$  <sup>1</sup>day<sup>-1</sup>) during 15 days of the experiment (Mean $\pm$ SE).

Table 2: Feeding schedule suggested for rearing *S. argus* larvae after 10 days of hatching based on the consumption pattern of *Artemia* nauplii

Days of Culture (DPH)	Prey density suggested (nauplii ml <sup>-1</sup> day <sup>-1</sup> )
10-13	4
14-16	8
17-19	12
20-21	16
22-25	20

### 4. DISCUSSION

The larval phase is a critical stage for small marine ornamental fish. The successful development of a hatchery is primarily dependent on the sustainable culture of larvae. The characters of short length, small size and non-functional mouth of fish larvae [12], limit their ability to feed successfully during the first days after hatching [13]. The most important feeding strategy for fish larval culture is to provide appropriate prey size and to maintain prey density at an optimal level. Studies show that there is a positive correlation between prey densities and feed intake among fish species [3, 11, 14, 15] and ontogenic development stages [16, 17].

The density of prey on larval fish varies according to fish species, visual acuity, size and feeding behaviour [18]. In

the present study, the survival rate was observed higher on the initial day in all the treatments. During the sequential period, the survival rate started to decline with food concentrations. It clearly shows the feeding pattern of *S. argus* larvae. Low survival of *S. argus* larvae at lower prey densities was probably due to a lack of food supply and need to spend more time chasing, capturing and ingesting the prey. Similar results have been observed in Pejerrey larvae [10], Greenback flounder [15], Striped cat fish [4] and Northeast Arctic cod [19] whereas, low survival rates at high prey densities were observed in Sea bream [20] and Australian bass [21], suggesting that overfeeding of rearing systems may create a stressful environment for the larvae.

In the case of growth, the best results for body weight and length were for the *S. argus* larvae fed at 16 and 20 nauplii  $ml^{-1}$ day<sup>-1</sup>, respectively. In relation to growth performance, both SGR in weight and Mean Growth rate in length were positively correlated with prey density. The better growth in higher prey density is due to an increase in prey intake as prey density increases. Similar findings have been made by Claramunt and Wahl [22], Hoxmeier et al. [23], Abe et al. [3] in larvae of Bluegill *Lepomis machrochirus* and Walleye *Sander vitreus*, as well as Golden pencil fish *Nannostomus beckfordi*. On the other hand, *S. argus* larvae fed at 4 and 8 nauplii  $ml^{-1}$ day<sup>-1</sup> had lower growth, indicating partial feed deprivation and need of more energy to swim in search of their prey [11, 24].

The present study involves a thorough investigation of feed consumption rate and supply of required nutrients to optimize the feeding strategy of S. argus larval rearing. Some studies have estimated feed consumption rate in fish larvae including Hippoglossus hipoglossus [25], Morone saxatilis and Morone americana [26], Pleuronectes ferruginea [27], Paralichthys dentatus [28], Seriola lalandi [29], Trachinotus ovatus [30] and Heros severus [11]. The feed level should be adapted to the needs and consumption of larvae at varying ages so that food is not wasted, the larvae are not malnourished and the rearing water is not contaminated [31]. The consumption rate of S. argus larvae increased with age. Increasing consumption rate of larvae with a gradual increase at different prey densities has been observed for many species [23, 32, 33].

Expensive live feed production is one of the major issues in fish hatcheries, though a sincere effort is made to substitute them by microparticulate diets [34]. The global decrease in *Artemia* cyst production and the increase in aquaculture production have resulted in increased demand for Artemia cysts and a push-up in their prices, increasing the production costs of fish larvae [35]. In conclusion, the development of optimal food management for S. argus can improve larval performance and reduce production cost. In our present investigation, the hatching rate of the Artemia strain is 1,65,000 nauplii g<sup>-1</sup> cysts and our findings suggest that if the feeding strategy proposed in this paper is adopted, around 1,000 S. argus larvae can be produced for 150 g of Artemia cysts. Our study ascertains that live feed plays an important role in survival and growth performance of S. argus larvae but it strongly establishes that an optimum supply of Artemia nauplii will cut down the overuse and wastage of them eventually reducing the production cost of live feed. Our feed concentration and corresponding results justify the above statement assuring that adapting our feeding strategy can increase the production of *S*. *argus* at low cost.

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#### Conflict of interest

The authors declare no conflict of interest.

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