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CAPTIVE REARING OF *HIPPOCAMPUS KUDA* AND ISSUES OF SEAHORSE CONSERVATION IN INDIA

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ABSTRACT

Captive breeding and rearing of *Hippocampus kuda,* one of the most dominant species of seahorses was done to determine the optimum conditions of rearing, which would give the highest survival, and growth of juveniles. A diet of enriched *Artemia* and mixed marine copepods was found to be the best feed combination during a rearing period of 60 days. A stocking density of 3 juveniles per litre was found to be the optimum, which gave the highest mean increment of growth in length and weight, as well as the highest mean production per litre. The juveniles of *H. kuda* were found to grow equally well in salinities ranging from 15 to 35 ‰. The highest mean length and weight were obtained at 30 ‰ while rearing for 14 days. Illumination of the rearing tank had a significant effect in improving the early juvenile survival of *H. kuda*. Rearing tanks, which were partially covered to improve the feeding efficiency of *Artemia* nauplii, registered the highest mean survival and growth. The results of the captive breeding and rearing trials indicate that the seahorses represent a potential candidate for aquaculture in India.

INTRODUCTION

Seahorses are bony fishes (teleosts), and belong to the family Syngnathidae. They are found worldwide, usually in shallow coastal, tropical and temperate seas. Seahorses are abundant in the Indo Pacific, in waters less than about 20 m deep, but also in shallow rock pools, and depths of over 150 m from where they are trawled. There are about 32 species of seahorses all over the world (Lourie *et al*., 1999), which belong to one genus *Hippocampus*. Most seahorses are marine, although some species such as *H. capensis* live in estuaries (Whitfield, 1995). The general shape of the seahorse is easily recognisable, but species identification is difficult. Members of the same species may differ in appearance because seahorses can change colour and grow skin filaments to blend with their surroundings.

Seahorses are also unique in their reproductive behaviour, in which the males become pregnant. This is one of the most extreme examples of paternal care. The male fertilizes and broods the eggs produced and deposited into its pouch by the female during courtship and mating, and in turn delivers the hatchling, after a long period of pregnancy and labour. Most species of seahorses studied so far exhibit unique sexual fidelity and form faithful pair bonds (Lourie *et al*., 1999), in which case one male and one female mate repeatedly and exclusively giving up opportunities to interact with non-partners.

India has a long history of trade on seahorses, which mostly originate in the southeast coast, particularly Tamil Nadu. Limited quantities of seahorses are also reported to be collected from

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the coasts of Kerala, Maharashtra and Karnataka. Six species of seahorses were reported from theIndia has a long history of trade on seahorses, which mostly originate in the southeast coast, particularly Tamil Nadu. Limited quantities of seahorses are also reported to be collected from the coasts of Kerala, Maharashtra and Karnataka. Six species of seahorses were reported from the Palk Bay coast of Tamil Nadu, where a target fishery was in existence involving divers collecting seahorses along with sea cucumbers and chanks, catering to an organised trade for export (Salin *et al.*, 2004). There existed a well-established target fishery for seahorses along the Palk Bay coast whereas the landings mostly come as trawl by-catch in the Gulf of Mannar region and Kerala coast. Salin *et al.* (2004) also reported two species from Kerala coast, though there was no organised fishery and trade. Most of the catch is dried and exported to Singapore, Hong Kong and Malaysia, apart from their local consumption in limited quantities in folk medicine for treatment of asthma, fitz etc.

According to MPEDA (2003), about 2.53 tonnes of seahorses worth Rs. 15 lakh were exported from India during 2000-01, mainly to Singapore, UAE and Hong Kong. This has increased to 4.5 MT during 2001-02, worth Rs. 27 lakh, Chennai being the major port of activity. Obviously, the UAE is only a transit point, and most of the Indian export is destined to Singapore or Hong Kong, countries with a sizeable population of ethnic Chinese communities.

The major quantity of the trade on seahorses in India originates from the south Tamil Nadu coast, especially the Palk Bay and Gulf of Mannar areas, and the Kerala coast. Despite the existence of a flourishing trade, the study of Indian seahorses attracted little interest till recently. The number of species available, their biology, suitability to aquaculture and larval rearing, all deserve serious consideration. Since most of the seahorses in trade are wild-caught, considerable efforts including captive breeding and juvenile rearing also need to be invested to conserve the dwindling seahorse fishery. Aquaculture of seahorses should be taken up to meet the growing demand and to ease the fishing pressure on seahorses.

MATERIALS AND METHODS

Three feeding regimes of freshly hatched and enriched *Artemia* nauplii (INVE Thailand), mixed marine copepods and freeze-dried Cyclop-eeze (Argent Laboratories, USA) were tested for rearing the hatchlings obtained from wild, pregnant males of *H. kuda*. Untwisted strands of a polypropylene rope, weighted at the bottom using a hard piece of coral stone were provided as hold fast to the growing seahorse fry in glass tanks (45 litres).

In the first treatment (T_1), enriched *Artemia* nauplii alone was fed to the hatchlings, in the second treatment (T_2), enriched *Artemia* nauplii + copepods, and in the third treatment (T_3), enriched *Artemia* nauplii + freeze-dried Cyclop-eeze were given, using 4 replicates for each treatment. Feeding with newly hatched *Artemia* nauplii was started on the day of stocking of the hatchlings twice daily at 7 hours and 16 hours during the first week. From second week onwards, daily feeding was split into three times at 6 hours, 12 hours and 18 hours, with enriched *Artemia* nauplii given at 12 hours and the rest two feedings of copepods or Cyclop-eeze. The percentage survival of the hatchlings was noted after 7th, 14th, 21st, 28th and 60th day of rearing and the mean length and weight measurements were taken just after hatching and after 7th, 14th, 28th and 60th days of rearing.

The percentage growth and the specific growth rate over the experimental period were calculated. The results were statistically analysed to find out the optimum feed combination that ensured maximum survival and growth.

The *Artemia* nauplii were fed to the hatchlings at 2 g for 300 hatchlings which was later increased based on demand. The *Artemia* nauplii harvested from the hatching tanks were enriched (as per the recommended procedure) using Super Selco (INVE Thailand), which is a commercially available *Artemia* enrichment medium. Marine copepods collected from the seawater storage tanks and high saline brackish water ponds were sieved through fine mesh nets of 1000, 500 and 250 µ size in that order. Cyclop-eeze is a freeze-dried copepod organism reported to contain one of the highest levels of the carotenoid pigment, astaxanthin and essential fatty acids, and is being used widely as a substitute for the live feed *Artemia* in the larval rearing of many marine finfish and shellfish (Lieberman (2001). Cyclop-eeze was reported to contain 17,000-18,000 organisms per gram and the size measured about 800 µ, which is bigger than that of the freshly hatched *Artemia* nauplii (420 - 530 µ). The predation rate was found to be 7 Cyclop-eeze h^{-1}. Therefore 1 g Cyclop-eeze was fixed to begin with, for feeding 300 seahorse hatchlings per day, and later the quantity adjusted based on demand.

Six treatments of stocking density of the hatchlings of *H. kuda* namely 1 L^{-1}, 2 L^{-1}, 3 L^{-1}, 4 L^{-1}, 5 L^{-1} and 6 L^{-1} were tested with three replications for each treatment. The percentage survival, and the production of juveniles per litre for each treatment of stocking density were calculated after a rearing period of 60 days. The increment in length and weight of the juveniles were also noted. The results obtained were statistically analysed to determine the optimum stocking density. Seven salinity levels of 5 ‰, 10 ‰, 15 ‰, 20 ‰, 25 ‰, 30 ‰ and 35 ‰ were tested to determine the optimum salinity of rearing the juveniles of *H. kuda*. The feeding of the hatchlings was done with newly hatched *Artemia* nauplii during the first week and later with marine copepods and enriched Ar*temia* nauplii. The percentage survival and the growth in length in weight obtained under different salinity conditions after rearing for 14 days were statistically analysed to determine the best salinity.

Early hatchlings of seahorse *H. kuda* were seen to have a peculiar problem of entrapping air bubbles, leading to death while trying to feed from the water surface. The major live feeds for the seahorse viz. *Artemia* nauplii and copepods, are highly phototactic, and normally congregate at the water surface, especially near to the walls of the container. Since the seahorses have a unique feeding habit of sucking in the prey in a sudden and strong inhalant current, it usually results in ingesting air bubbles while feeding at the water surface leading to buoyancy problems, and the seahorses that float on water surface often die. This is one of the main factors affecting the initial survival of seahorse hatchlings.

Three treatments were tested in the experiment with 5 replicates for each, and was conducted in aquarium glass tanks (20 litres). The tanks were kept open in diffused sunlight inside an area roofed with transparent FRP sheets, and the tanks under the first treatment (T_1) were left uncovered. The set of tanks in second treatment (T_2), were covered with a black plastic sheet, from top of the tank up to 10 cm down from the water mark, leaving a gap of about 10 cm from the bottom of the tank uncovered so as to get 'illuminated' from the diffused sunlight in the room. In the third treatment (T_3), the set of tanks were totally covered with black plastic sheet throughout the rearing period so as to cut off any light into the experimental rearing tank. At the end of 14 days of rearing under standard conditions, the percentage survival and growth was calculated and the results analysed statistically.

RESULTS

The percentage survival of the newly hatched young ones of seahorses after rearing period of 60 days using different feeds are provided in Fig 1. The highest mean survival of 56% was obtained in the second treatment (T_2) in which the young ones were fed with enriched brine shrimp nauplii and mixed marine copepods. This was followed by *Artemia* alone (T_1) with a survival of 43%. The treatment (T_3) in which *Artemia* and freeze-dried Cyclop-eeze were used, gave the lowest survival of 21 % at the end of 60 days of rearing. The results of the length and weight measurements of the juveniles on 7^{th}, 14^{th}, 28^{th} and 60^{th} days of rearing on a diet of *Artemia* and mixed marine copepods (T_2) are given in Fig 2. The juveniles attained a maximum growth of 49.4 mm and 316.14 mg after 60 days of rearing. The percentage growth from 7^{th} to 60^{th} day was computed as 99.1 % at a mean daily weight gain of 5.91mg per day. The mean specific growth rate (SGR), which denotes the percentage increase in body weight per day for a period of 7 to 60 days of rearing, was determined as 8.89 %.

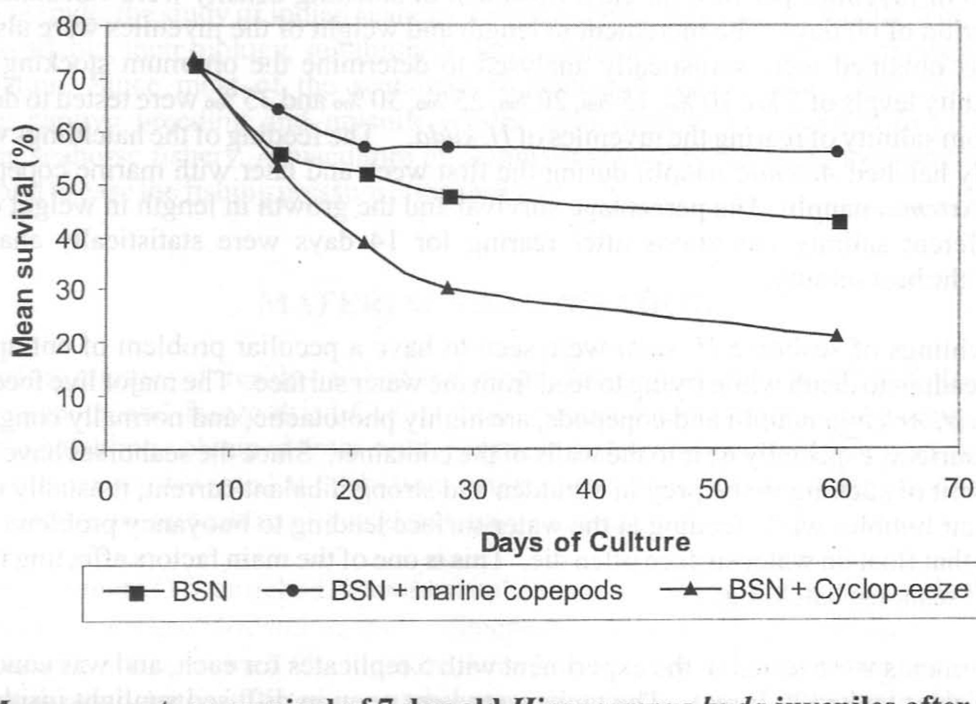

Fig.1 Mean percentage survival of 7 day old *Hippocampus kuda* juveniles after 60 days (post hatching) of rearing on three feed combinations of enriched brine shrimp nauplii (BSN) alone (T_1), BSN + marine copepods (T_2) and BSN +freeze-dried Cyclop-eeze (T_3)

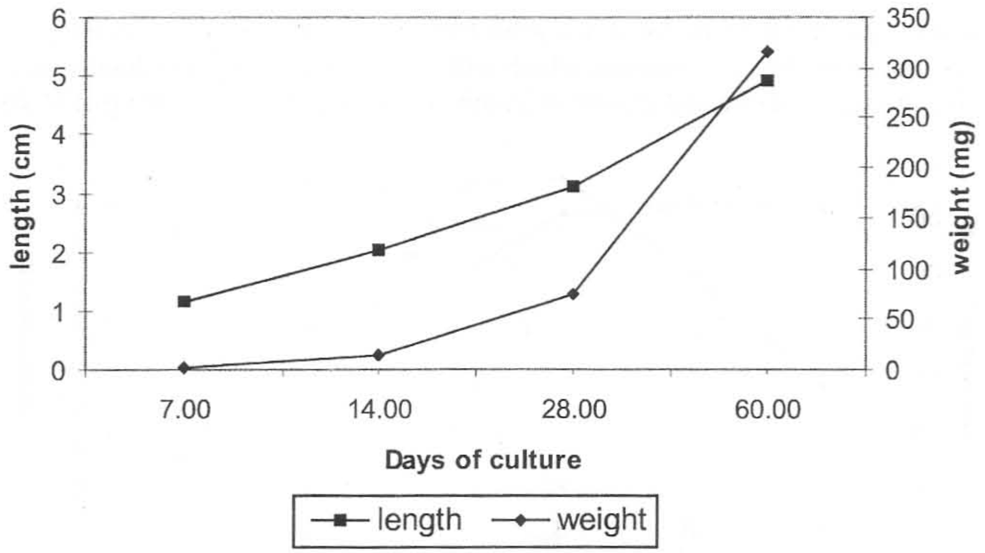

Fig. 2 Growth in length and weight attained by juvenile *Hippocampus kuda* after 60 days of rearing by feeding enriched *Artemia* and marine copepods

The ANO of the percentage survival indicated that the treatments had significant effects (P<0.05) on the survival of seahorse hatchlings, except on the first and second weeks of rearing, and that the three feed combinations were significantly different except during the first seven days of rearing. Thus it could be inferred from the results that the second treatment (using enriched *Artemia* and marine copepods), which gave the highest mean survival of 56 % after 60 days of rearing was the best feed combination for rearing the *H. kuda* juveniles. The highest decline in juvenile survival occurred during the first 7 days of rearing.

The results of the experiment to determine the optimum stocking density for rearing the juveniles of *H. kuda* are presented in Fig 3 and Fig 4. The mean survival of the juveniles reared for 60 days under the six treatments of stocking densities were highest (52.59 %) at a density of 2 L^{-1}, whereas the maximum production per litre was achieved at 3 L^{-1}. The highest mean increment in length was obtained at 1 L^{-1} (44.5 mm), followed by 2 L^{-1} (44 mm), 3 L^{-1} (43.67 mm) and 4 L^{-1} (39.33 mm). The highest mean increment in weight of the juveniles was obtained at 2 L^{-1} (318.8 mg), closely followed by 1 L^{-1} (314.87 mg), 3 L^{-1} (312.83 mg) and 4 L^{-1} (300.07 mg).

The analysis of variance of the data showed that the stocking densities tested had significant effects (P<0.05) on the survival rates and growth in length and weight of *H. kuda* reared under captivity. The densities of 5 L^{-1} and 6 L^{-1} had significantly lower survival and growth compared to all other treatments. The densities of 1 L^{-1}, 2 L^{-1} and 3 L^{-1} were identical in the percentage survival and growth in length, whereas in the case of growth in weight, only the densities of 1 L^{-1} and 2 L^{-1} were identical, and all other treatments were significantly different from one another.

The production of juveniles per litre is an important consideration in the viability of a commercial hatchery operation. In the present study, although highest percentage of survival was obtained at the stocking density of 2 L^{-1}, the maximum production per litre (1.5) was obtained at the density of

3 L^{-1}. The production per litre obtained was only 1.05 at 2 L^{-1}. Therefore, a stocking density of 3 juveniles per litre could be the optimum, which will give the maximum production per litre, and significantly high survival rate and growth in length and weight for early rearing of *H. kuda*.

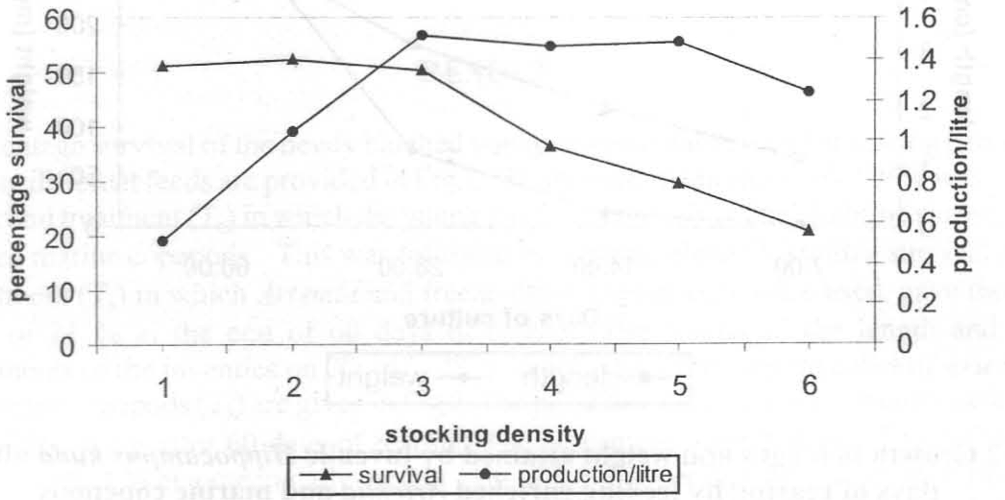

Fig. 3 Effect of stocking density on the survival and production/litre of hatchlings of *Hippocampus kuda* reared for a period of 60 days

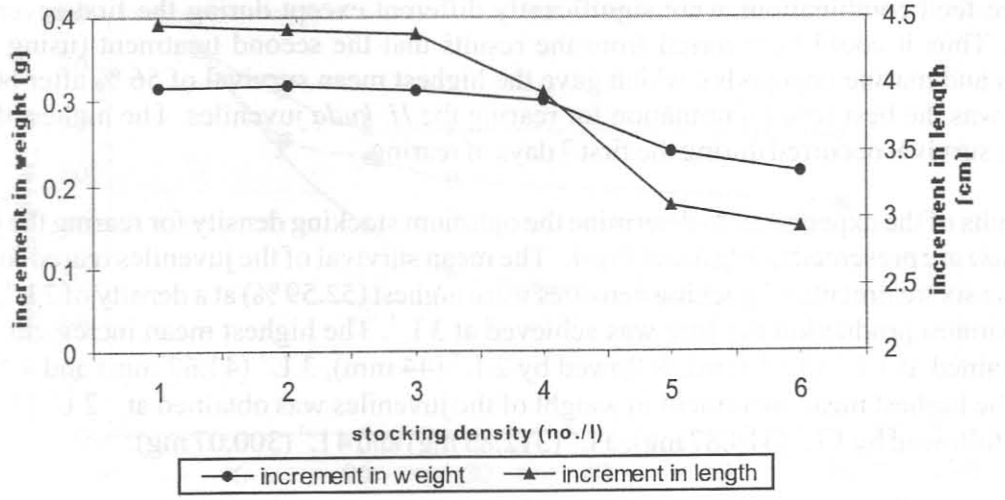

Fig. 4 Effect of stocking density on the increment of growth in length and weight of the hatchlings of *Hippocampus kuda* reared for 60 days

The percentage survival and growth of the juveniles of *H. kuda* reared for 14 days under different salinities are summarised in Fig.5 and Fig.6 A maximum survival of 60.56 % was obtained for 30 ‰, followed by 35 ‰ (59.23 %), 25 ‰ (58.34 %), 20 ‰ (55 %), and 15 ‰ (53.89 %). The lowest survival rates of 2.78 % and 16.67 % were obtained for 5 and 10 ‰ respectively. A maximum mean length of 28.7 mm at 30 ‰ followed by 27.4 mm at 35 ‰ and 22.6 mm at 25 ‰ was achieved. The weights gained were identical at 30 ‰ (39.1 mg) and 35 ‰ (39 mg), which were followed by 25 ‰ (34.3 mg). The mean length gained by juveniles reared at 15 and 20 ‰ salinity were similar (18.1 mm and 18.6 mm, respectively), whereas the weight gained was more for 15 ‰ (29.5 mg) than that for 20 ‰ salinity (25.0 mg).

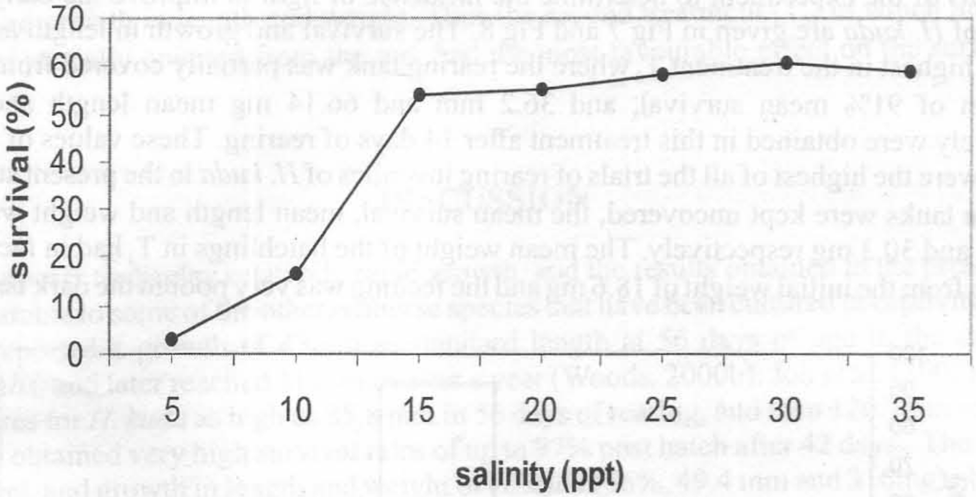

Fig. 5 Effect of salinity on the percentage survival of the hatchlings of *Hippocampus kuda* reared for 14 days

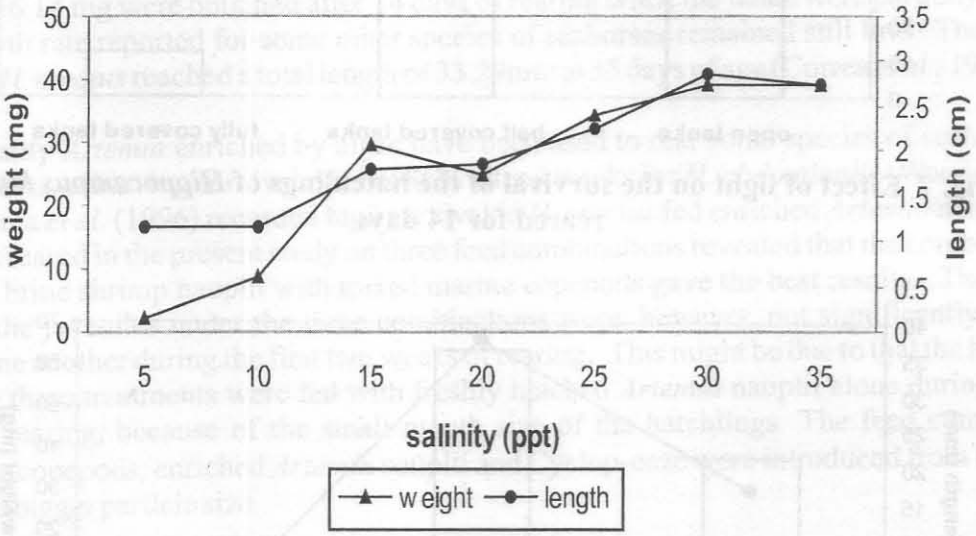

Fig. 6 Effect of salinity on the growth, in length and weight of the hatchlings of *Hippocampus kuda* reared for 14 days

The analysis of variance of the data on percentage survival, and growth in length and weight of the juveniles after a rearing period of 14 days revealed that the salinity of rearing water had significant effect ($p<0.05$) on the survival and growth of the juveniles. The salinities of 5 ‰ and 10 ‰ were significantly different from all the other salinities tested. There was no significant difference among 15 ‰, 20 ‰, 25 ‰, 30 ‰ and 35 ‰ tested, in terms of the percentage survival and the mean weight gained. However, the mean length of the juveniles reared in 5 ‰, 10 ‰, 15 ‰ and 20 ‰ were identical.

The results of the experiment to determine the influence of light to improve the early juvenile survival of *H. kuda* are given in Fig 7 and Fig 8. The survival and growth in length and weight were the highest in the treatment T_2 where the rearing tank was partially covered from top. The maximum of 91% mean survival, and 36.2 mm and 66.14 mg mean length and weight, respectively were obtained in this treatment after 14 days of rearing. These values of the initial survival were the highest of all the trials of rearing juveniles of *H. kuda* in the present study. In T_1 where the tanks were kept uncovered, the mean survival, mean length and weight were 55 %, 21.4 mm and 30.3 mg respectively. The mean weight of the hatchlings in T_3 had in fact shown a reduction from the initial weight of 18.6 mg and the feeding was very poor in the dark tanks.

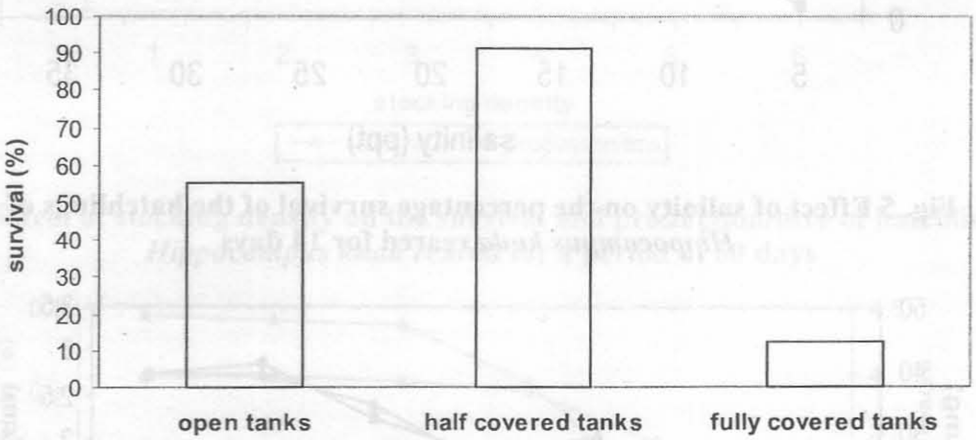

Fig. 7 Effect of light on the survival of the hatchlings of *Hippocampus kuda* reared for 14 days

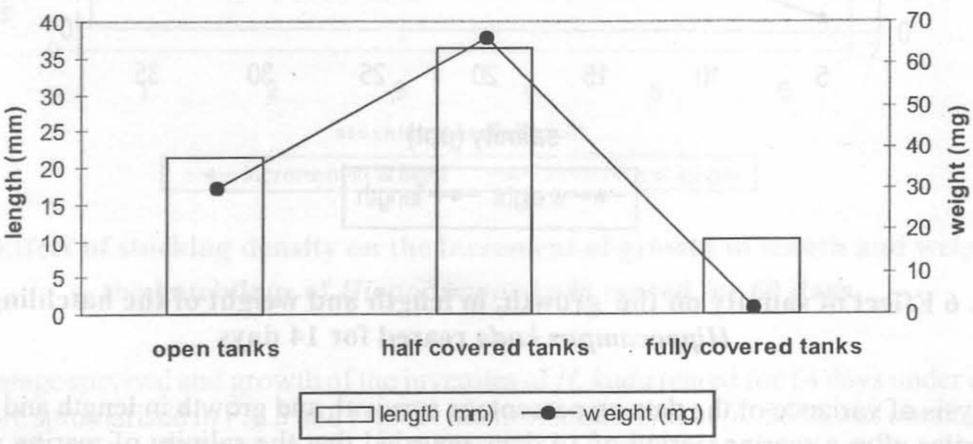

Fig. 8 Effect of light on the growth, in length and weight of the hatchlings of *Hippocampus kuda* reard for 14 days

The analysis of variance of the data on percentage survival, and growth in length and weight of The analysis of variance of the results has shown that illumination of the rearing tank had a significant effect ($P<0.05$) on the juvenile survival, and growth in body length and wet weight. All the three treatments were significantly different from one another in relation to the percentage survival and growth in length and weight. Thus the second treatment (T_2) in which the rearing tanks were partially covered from the top, had the most favourable effect on the survival and growth.

DISCUSSION

H. kuda appears to display relatively rapid growth, and the results obtained in the present study are comparable to some of the other seahorse species that have been cultured in captivity. Woods (2000a) reported a growth of 43mm in standard length at 56 days of age in the case of *H. abdominalis,* and later reached 110.7mm after a year (Woods, 2000b). Job *et al.* (2002) obtained growth rates for *H. kuda* as high as 85.8 mm in 56 days of rearing, and then 120.7mm in 98 days. They also obtained very high survival rates of up to 97% post hatch after 42 days. The results of the survival, and growth in length and weight of *H. kuda* (56%, 49.4 mm and 316mg) obtained in the present study after 60 days of rearing without light manipulation was similar to that obtained for Woods (2000a) for *H. abdominalis*. The survival of 97% obtained for *H. kuda* by Job *et al.* (2002) was unusually high. However, in the present study a survival of 91 % and growth of 36.2 mm and 66.14 mg were obtained after 14 days of rearing when the tanks were partially covered. The growth rate reported for some other species of seahorses remained still low. The Atlantic seahorse *H. erectus* reached a total length of 33.29mm at 35 days of age (Correa *et al.*, 1989).

Good quality *Artemia* enriched by algae have been used to rear some species of seahorse with great success, achieving survival rates of 86% after 4 weeks for *H. abdominalis* (Woods, 2000b). Also Correa *et al.* (1996) recorded high survival in *H. erectus* fed enriched *Artemia* nauplii. The juveniles reared in the present study on three feed combinations revealed that the combination of enriched brine shrimp nauplii with mixed marine copepods gave the best results. The survival rates of the juveniles under the three combinations were, however, not significantly different among one another during the first two weeks of rearing. This might be due to that the hatchlings in all the three treatments were fed with freshly hatched *Artemia* nauplii alone during the first week of rearing, because of the small mouth size of the hatchlings. The feed combinations involving copepods, enriched *Artemia* nauplii and Cyclop-eeze were introduced from the 8th day due to the bigger particle size.

The feeding of seahorses are peculiar in that they prefer live moving prey alone, and they have a unique aversion to nonliving foods (Vincent, 1996), although they have recently been reported to have weaned to artificial diets (Vincent, 1995a; Baldassano; 1996 Giwojna, 1996a; Giwojna, 1996b). The larvae and juveniles of most marine fish species require live feed with a high nutritional content. Freshly hatched nauplii of the brine shrimp *Artemia* have been widely used as a live feed in the larval rearing of marine and freshwater fish and prawns. However, the quality of *Artemia* nauplii is affected by various factors such as the strains, locality and the season of harvest (Hsu *et al.*, 1970). The nutritional value of *Artemia* is marked by the absence of omega-3 polyunsaturated fatty acids (PUFAs), which are essential to the diet of marine fishes and crustaceans. Since the brine shrimp nauplii lack important nutritional requirements of the fish larvae, such as essential fatty acids, enrichment using PUFAs was resorted to in the present study. Sargent *et al.* (1997) reported that enriching *Artemia* nauplii with commercial preparations that are high in PUFAs make them more suitable for feeding young fish. Super Selco is a proven commercial product being used in many commercial fish and prawn hatcheries for enrichment of the *Artemia* nauplii before feeding to the larvae. Payne and Rippingale (2000) had also used Super Selco in the enrichment of *Artemia* nauplii during rearing of the seahorse juveniles.

The survival of the seahorse hatchlings reared in the present study on a feed combination of enriched *Artemia* and mixed marine copepods were significantly greater than that when enriched *Artemia* was given alone, or when enriched *Artemia* was supplemented with an inert feed such as freeze-dried Cyclop-eeze. Copepods were found to be a more efficacious diet than even enriched *Artemia* for rearing *H. subelongatus* juveniles (Payne and Rippingale, 2000). This supports the general assertion that *Artemia* are not an appropriate mono-diet for juveniles of some seahorse species (Lunn and Hall, 1998). Payne and Rippingale (2000) suggested that the copepod nauplii were better assimilated than *Artemia,* and this apparent difference in prey digestibility probably accounted for much of the difference in growth and survival between the two diets.

Tipton and Bell (1988) observed that in the sea, the diet of adult dwarf seahorses *H. zosterae* consisted mainly of copepods. Liang (1992) emphasized that pelagic copepods are indispensable for rearing seahorse fry. Also the marine planktonic copepods are reported to have high nutritional value. Raymont (1983) and Raymont *et al.* (1971) observed that the *Calanus* spp. contained 10-40% protein, 12-47% lipid, 3% chitin and 3% ash (calculated as dry weight). The high PUFA content of copepods make them suitable as a diet for young fish (Watanabe *et al.*, 1983). Payne and Rippingale (2000) reported that the early growth and survival of seahorse *H. subelongatus* were significantly greater when fed with copepod nauplii. The survival rates obtained by Payne and Rippingale (2000) were slightly higher than the mean survival rate of 56% obtained in the present study for the feed combination of enriched *Artemia* and copepods. However, they found that when enriched *Artemia* alone was used the survival rates were down to just above 20%, which was quite low when compared with the mean survival obtained in the present study, which was 43% when enriched *Artemia* nauplii alone was fed to the juveniles.

The copepods used in the present study contained heterogeneous species and were not enriched with any microalgae, as against the unispecies culture of calanoid copepod *Gladioferens imparipes* enriched with microalgae *Isochrysis galbana* fed to the seahorse fry by Payne and Rippingale (2000). The mixed culture of copepods in the present study contained a majority of calanoid copepods such as *Eucalanus* spp., *Euchartia* spp. *Centropages* spp., *Labidocera* spp., *Acartia* spp., *Rhinocalanus* etc.; cyclopoid copepods such as *Oithona* spp., *Copilia* spp., *Sapphirina* spp. etc. and harpacticoid copepods such as *Enterpina* spp. and *Macrosetella* spp. that naturally occurred in seawater and were allowed to bloom. There was no control on the microalgae developed in seawater that facilitated the proliferation of copepods. This might be the reason for a slightly lower survival of the juveniles in the present study than that obtained by Payne and Rippingale (2000).

One of the main bottlenecks in establishing economically viable and biologically successful seahorse aquaculture is the provision of sufficient quantities of nutritionally rich live food. In their natural habitat, seahorses are visual predators that target live prey such as copepods, amphipods, mysid shrimp, and caridean shrimp (Reid, 1954; Tipton and Bell, 1988). In captivity, aquarists, researchers and commercial aquaculturists have relied heavily on cultured live foods such as brine shrimp, copepods, mysids and amphipods, as well as collecting wild foods such as various assemblages of zooplankton (Correa *et al.*, 1989; Lockyear *et al.*, 1997; Wilson and Vincent, 1998; Hilomen-Garcia, 1999; Payne and Rippingale, 2000). Culturing the large quantities of live food required by seahorses in commercial culture could prove difficult and costly. Costs of establishing and maintaining a healthy live food culture system might ruin the economic feasibility of a commercial venture of seahorse rearing, and therefore alternative diets to live feed need to be investigated and tested (Woods, 2000a). Mixed feeding of live and non-live diets is a commonly used strategy to help wean larval fish onto non-live or manufactured foods and has been shown to enhance larval growth and survival beyond that achieved by feeding either types of food alone (Drouin *et al.,* 1986; Ehrlich *et al.,* 1989; Rosenlund *et al.,* 1997; Daniels and Hodson, 1999).

Seahorses have not generally been reared on artificial foods in commercial culture, due to difficulties in getting them to accept nonliving foods. However, there are general reports of commercial seahorse culturists using artificial foods to some degree, such as shrimp and fishmeal based diets (Chen, 1990; Forteath, 2000). Frozen foods such as frozen mysids and copepods are also utilised to feed seahorses (Garrick-Maidment, 1997; Forteath, 2000). The ability of seahorses to utilise nonliving foods in the rearing of *H. kuda* has important implications for the commercial culture of this species, as the use of non-live foods can dramatically reduce material and labour costs, that in turn potentially increases the economic viability of a commercial operation. Nonliving foods also provide for a more predictable and reliable food source.

The freeze-dried Cyclop-eeze used in the present study is a very popular larval feed in crustacean hatcheries, and ornamental fish rearing. Marco (2003) reported the use of Cyclop-eeze in the diet of adult *H. zosterae*, and feeding at various stages in the development of juvenile *H. abdominalis*, *H. kuda*, *H. erectus*, *H. breviceps*, the Dusky pipefish *Syngnathus floridae*, and the Northern pipefish *Syngnathus fuscus*. In the present study, one-week-old juveniles of *H. kuda* could be successfully weaned onto the freeze-dried Cyclop-eeze, though the results were not better compared to that of the live feeds tested. The lower survival of hatchlings fed *Artemia* and Cyclop-eeze might be due to the comparatively lower acceptability of the hatchling towards it, being a dead feed. Given the relative difference in the digestion of *Artemia* and Cyclop-eeze, copepods could be a clearly more efficient diet compared to the former two. Given the almost ubiquitous presence of copepods in the sea it is likely that seahorses have evolved to recognize them as prey and to benefit nutritionally from them. However, there is presently no such a convenient and effective alternative diet to *Artemia* for feeding during the initial stages, the mouth size of the hatchlings being very small.

On a commercial point of view, the highest survival alone is not the only criterion that determines the success of a hatchery operation; the production per litre has a more meaningful significance than the percentage survival. In the present study, the production of seahorse juveniles obtained per litre was also considered in view of its commercial application. Although the best growth and survival was obtained at a stocking density of 2 L^{-1}, the one that yielded the highest production per litre was 3 L^{-1}. However, the survival and growth, particularly in weight, was significantly reduced at stocking densities of 4 and 5 L^{-1}. Hence the stocking density of 3 L^{-1} was assumed to be the optimum density that is commercially viable, although the survival rates were higher at lower densities of 1 and 2 L^{-1}.

The juveniles of *H. kuda* reared in the present study were found to grow equally well in salinities ranging from 15 to 35‰. This is promising since it opens up the possibility of culturing seahorses in brackish water as well. The only earlier report of the successful rearing of *H. kuda* under different salinities was by Chaladkid and Hruangoon (1996) in Thailand, who obtained the highest survival rate of 60.67% while rearing the juveniles at 15‰ compared to 20, 25, 30 and 35‰. The survival rate obtained in the present study was slightly less (53.86%) at 15‰, but the growth in length and weight were encouraging. Chaladkid and Hruangoon (1996), however, obtained the lowest survival rates of juveniles at 30‰ (31.34%), which is contrary to the findings in the present study in which 30 ‰ was found to be the optimum salinity for the rearing of *H. kuda,* registering the highest mean survival of 60.56%. Chaladkid and Hruangoon (1996) obtained better survival rates of 52.67%, 34% and 36.67% at 20, 25 and 35‰ respectively. The corresponding survival rates obtained in the present study were 55%, 58.34% and 59.23%. However, there was no statistically significant difference in terms of the percentage survival and the mean weight gained by the juveniles among the salinities of 15, 20, 25, 30 and 35‰, suggesting that *H. kuda* could be grown successfully in the salinity range from 15 to 35‰. Hatching was also found to occur in a range of salinity 5 to 50‰.

In the present study a survival of 91% and growth of 36.2 mm and 66.14 mg were obtained after 14 days of rearing when the tanks were partially covered. The increments in weight achieved by the juveniles were much higher than that achieved while rearing without light manipulation, although, the length attained by the juveniles were less. Visibility of the prey is important in the efficient feeding in larval rearing since It is established that vision is the primary sense used in prey detection and capture in many teleost fish (Blaxter, 1980). Therefore, characteristics of the prey (like shape, colour, movement etc.) when viewed against different backgrounds by fish can affect their feeding efficiency, and consequently their growth and survival (Naas *et al.,* 1996; Martin Robichaud and Peterson, 1998; Planas and Cunha, 1999). This suggests that small *Artemia* nauplii are visually differentiated from their background more effectively by the seahorse juveniles in clear containers. However, Naas *et al.* (1996) considered black tanks to be the best choice for larval fish rearing as compared with a white environment, since it would serve to avoid 'wall trapping' due to various phototactic responses of the larvae.

In the present study, hatchlings immediately after release from the brood pouch of the male were seen to rise to the water surface and gulp in air for a while. This is probably important for the hatchlings of seahorses, as they appear to initially inflate their swim bladder through the ingestion of air at the water surface. Non-inflation of the swim bladder in fish may result in dysfunctional buoyancy and atrophy of the swim bladder (Battaglene and Talbot, 1990; Martin Robichaud and Peterson, 1998). Lawrence (1998) attributed the main cause of mortality in juvenile seahorses, *H. angustus* to poor swim bladder inflation as a result of an oily surface film that prevented juveniles from initially inflating their swim bladders. Hyperinflation of the swim bladder has also been reported in fish which has been attributed to excessive air ingestion (Nash *et al.,* 1977) that may hinder normal swimming, or in extreme cases, result in floating fish and increased mortality. The floatation of the hatchlings, which was one of the major reasons for mortality during most of the experiments in the present study, was considerably reduced by using the partially covered tank for rearing.

CONSERVATION

Seahorses depend on their habitats to survive. Seahorses are vulnerable to degradation of their preferred seagrass, mangrove, and coral reef habitats in addition to their depletion due to fishing. Thus attempts to diminish the impact of the trade on seahorse populations might be ineffectual unless protection and management are also sought for their vital sea grass, mangrove and coral reef habitats. These are highly productive ecosystems that are being degraded and lost through dredging, dumping and polluting, silting, clearing, felling, dynamiting and cyanide fishing (Hatcher *et al.*, 1989). Pro-active measures should be implemented to ensure the rejuvenation of these areas so as to make them suitable for seahorses and other marine fauna.

Any attempt of conservation of seahorses should also take into consideration the unique features of their fishery. Seahorses are caught by artisanal or subsistence fishers, or as a by-catch of trawling, although target fishery is in existence in several areas, including India. Large scale mechanised fisheries for seahorses are unknown, and are unlikely ever to be viable, because these fishes are patchily distributed, they recolonise slowly, and are often found in areas (coral reefs or mangroves) that are difficult to access with fishing gear. However, incidental by-catch from shrimp trawling and other forms of net fishing is an important contributor to the seahorse trade in many areas including India. Given the economic dependence of a large section of the fishermen population on seahorses, any effort on regulating their exploitation should be done with caution. The ban on the fishery and trade in India is no way near to any solution to the plight of seahorses since a full-fledged study is yet to be attempted to delineate the population dynamics of seahorses in the Indian waters.

Salin *et al.* (2004) reported that although the seahorses in the south Tamil Nadu coast were subjected to relatively heavy fishing pressure, the numbers had not come down alarmingly over the years, and in fact the individual size of dried seahorse had gone up from 2.5g in 1995 (Vincent, 1996) to 3.68g in 2001, Salin *et al.* (2004). Marichamy *et al.* (1993) and Vincent (1996) had reported that the annual catch of seahorses to be about 3.6-4.8 tonnes in 1992 and 3.6 tonnes (1.5 million numbers) in 1995 while Salin *et al.* (2004) estimated the annual landings of seahorses from southern Tamil Nadu coast as 9.75 tonnes (2.65 million numbers), which meant that there had been a substantial increase in the seahorse landings. Therefore, the conservation efforts should be more realistic in that the stakeholders in trade as well as the fishery do not suffer. An exhaustive study on the population and fishery characteristics of seahorses in India should first be undertaken so as to determine a sustainable yield level. Any further efforts on conservation shall be based on this study. Conservation and trade should go in consonance and one should not be at the expense of the other.

The ban on collection of seahorses and sea cucumbers in India in 2001 has put thousands of fishermen who subsisted on these resources for their livelihood into great hardship. The conservation and management of Indian seahorses are currently limited by the absence of data on the abundance and distribution of seahorse species in the region (Sreepada *et al.*, 2002). Thus any serious attempt of conservation should include the popular strategies of fisheries management, such as closed seasons, and or delineating marine sanctuaries that would serve as flourishing grounds for seahorses. Regulating or prohibiting fishery of seahorses during the peak-breeding season could be considered as one such option. This might have a beneficial effect on sustainability of the stock.

A novel community based management programme for sustainable seahorse fishery has been implemented in the Philippines (Vincent and Pajaro, 1997). As part of the world's first seahorse conservation and management project at Handumon village in the Central Philippines, a three-pronged strategy was developed, based on a socio-economic study, which included protective measures, fishery modification and enhancement efforts. A 33 ha, no-exploitation sanctuary and an adjacent traditional fishery zone was established, and enforced at Barangay. The pregnant males caught are placed in sea cages from where the newborn young escape before the male was sold. The fishers also re-seed areas depleted of seahorses and had begun developing seahorse culturing skills.

These are some of the strategies that could be considered in India. Aquaculture should also be undertaken so that seahorses of different species could be reared under captivity and used for sea ranching in their habitats. Aquaculture of seahorses could also be effectively used to fulfil the requirements of the trade. It has already been proved by various studies in different parts of the world, and the present study in India that seahorses could be successfully reared under captivity. Therefore, it is imperative that the regulation of seahorse fishery aimed at its conservation has to be realistic, and any such effort disregarding the subsistence fisherfolk would be futile, and far from its objective.

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