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Captive Breeding and Rearing of Fry and Juveniles of Cherry Barb (*Puntius titteya* Deraniyagala), a Highly Threatened Endemic Fish Species in Sri Lanka

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ABSTRACT. *Puntius titteya (Cherry barb) is a highly threatened endemic fish species, found in shaded shallow streams and rivulets of Sri Lanka. In this study, spawning of P. titteya did not occur in indoor and outdoor tanks even with hypophysation. However, captive breeding was successful in flowing water systems (without hormones) due to the availability of high Total Dissolved Solids. They were found to be multiple spawners exhibiting perennial breeding behaviour. Average fecundity was 226-284 eggs/fish/ spawning,, with an egg size of 849-1149 µm. Duration of development stages were egg to hatchling 36-48 h, hatchling to fry stage 2-3 days, fry to juvenile stage 45-60 days and adult stage 150–210 days at 26-28°C. Of the four tested diets for fry stage, Formulated Feed 1 (FF1), Artemia nauplii and microworm produced a significantly higher growth than plankton diet ($P<0.05$). Fry exhibited a higher preference for live feeds at their initial stage and they could be weaned from live feed to formulated feed at 42 days of age without causing a reduction in growth and survival. Plankton fed juveniles exhibited significantly higher growth when compared with those fed with dried swine liver and Formulated Feeds (FF1 and FF2) ($P<0.05$). Dried swine liver (66.3% crude protein) was found to be the most suitable artificial feed type for nurturing of juvenile stage producing significantly higher growth than FF1 (46.3% crude protein) and FF2 (45.1% crude protein).*

INTRODUCTION

The endemic fish *Puntius titteya* Deraniyagala (Cherry barb) is found in shaded shallow streams and rivulets of Sri Lanka. The species is identified as a highly threatened species in the National Red Data List (IUCN, 2000). However, under Fisheries and Aquatic Resources Act No 2 of 1996, this fish species is classified as only restricted (not prohibited) for exportation. The decline in *P. titteya* population over the years was caused mainly by over exploitation of more colourful varieties by the aquarium trade, pollution of their natural habitats and deforestation (Pethiyagoda, 1994; 1999).

The adults are omnivorous and feed on detritus, green algae, diatoms, dipterans and animal matter (De Silva *et al.*, 1977; Moyle and Senanayake, 1984; Wickramanayake and Moyle, 1989). In the aquaria they feed on fine ellets and freeze-dried *Tubifex* spp. and it was stated that the wild caught fish are better to feed on brine shrimps during the first few weeks (Pethiyagoda, 1991). The rearing of fry in the

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aquarium conditions poses many problems, particularly when *Daphnia* spp. like live feeds are not available (Pethiyagoda, 1991).

Since *P. titteya* is under highly threatened status, it is vital to conserve them from further declining in their population. Apart from the habitat rehabilitation, according to Senanayake and Moyle (1982), translocation is the possible and the most effective process of conserving a number of fishes in the natural habitats. Therefore, captive breeding and rearing of the young prior to reintroduction are very important tasks for conservation. However, no successful methods are available for captive breeding. Information on their reproductive biology and breeding behaviour is scanty. The objectives of the present study were to identify various methods for captive breeding, to determine a suitable feed among formulated feeds, live feeds (*Artemia* nauplii, microworm, phytoplankton and zooplankton) and animal liver that can be used in the nurture of young stages of *P. titteya*.

MATERIALS AND METHODS

Preparation of test diets and feeding

Two formulated feeds, *Artemia* nauplii, microworms, dried swine liver and planktons were tested to find the most suitable feed for the fry and juveniles. Formulated Feed 1 (FF1) was prepared according to Sundarabarathy *et al.* (2001) and the Formulated Feed 2 (FF2) was prepared adopting the same procedure of FF1 replacing fish meal with dried swine liver. *Artemia* cysts (origin: Great Salt Lakes, Utah, USA) were hatched, using the method described by Bengston *et al.* (1991) and fed as Instar 1 nauplii. Microworms (*Anguillula silusae*) were cultured and separated using the method described by Parameshwaran *et al.* (2001). The possibility to use swine liver was also investigated as an alternative high protein feed due to its low price (Rs. 25/kg) and high availability. Dried swine liver was processed by boiling chopped swine liver for 10 min, and drying in an oven at a temperature of 50°C for a period of 12 h and grinding to a particle size of 0.25 – 1.5 mm. Planktons were obtained by filtering water from a fishpond using 15 µm plankton net. Phytoplankton consisted mainly species of *Euglena*, *Chlamydomonas*, *Volvox*, *Pandorina*, *Microspora*, *Stichococcus*, *Coelastrum*, *Scenedesmus*, *Netrium*, *Closterium*, *Cosmarium*, and zooplanktons mainly rotifers, copepods and cladocerans.

Proximate analysis was done for formulated feeds FF1, FF2 and dried swine liver, following AOAC (1990) (Table 1). Protein was analyzed by Kjeldahl method (N x 6.25), fat by Sohxlet extraction method, moisture by drying in an oven overnight at 105°C and ash by burning samples in a muffle furnace at 600°C overnight. Gross energy of diets was determined by a Ballistic bomb calorimeter. Feeding was done twice a day at 9:00 and 16:00 h *ad libitum* for 6 days per week. The proximate compositions of the tested diets were compared using One-way ANOVA and the means were compared using Duncan's Multiple Range test at 95% significance level.

Collection of brood stock from the natural habitat

Mature males and females of *P. titteya* were collected from a stream in *Ambagamuwa* village, *Ginigathena* and the experiments were conducted at the Department of Animal Science, University of Peradeniya. These fish were used for fecundity studies as well as for the breeding experiments.

Table 1. Proximate composition of Formulated feed 1, Formulated feed 2 and Dried swine liver.

Component	Dried swine liver	Formulated feed 1	Formulated feed
Crude protein (%)	66.3 [a]	46.3 [b]	45.1 [b]
Crude fat (%)	2.6 [a]	0.12 [b]	0.2 [b]
Crude fibre (%)	0.003 [b]	6.2 [a]	0.04 [b]
Dry matter (%)	97.6 [a]	93.0 [a]	95.5 [a]
Ash (%)	3.8 [b]	13.6 [a]	5.8 [b]
Moisture (%)	2.4 [c]	7.0 [a]	4.5 [b]
Energy (cal g^{-1})	163.7 [a]	62.5 [b]	70.8 [b]

Values within rows followed by different superscripts are significantly different ($P<0.05$).

Study of egg diameter distribution and fecundity

Since this fish species is an endemic and threaten species in Sri Lanka, only ten mature brooders were sacrificed for the fecundity studies. They were preserved in 3% formaldehyde for a period of three weeks. Total length, body weight (without gut and gonads) and gonad weight of each specimen were measured. Maturity of ovaries was evaluated according to De Silva (1973) and ovaries in Stage V were preserved in Gilson's fluid (Bagenal and Braum, 1978) and kept in the dark for one week. At the end of one week, each ovary was washed with distilled water and the total number of eggs was estimated. Diameter of eggs was determined using a graduated micrometer eye-piece. Gonado-Somatic Index [GSI = (Gonad weight/Body weight) x 100] was calculated according to De Silva *et al.* (1985). Fecundity was measured as the number of ripening eggs (yolked eggs) per female prior to the next spawning period as Bagenal and Braum (1978).

Acclimatization and selection of brooders

Mature males and females were acclimatized for a period of 4 weeks in indoor glass tanks (30 x 60 x 45 cm^3; water depth: 30 cm) at a stocking density of 10 individuals per tank. Each tank was provided with gravel filter and a constant aerator. They were fed with both formulated feed and *Artemia* nauplii. Since the males do not readily extrude milt, mature males with bright red colored fins and body were selected for the experiment. The best females for the broodstock were selected by observing release of yolked eggs when slight pressure was applied on to the abdomen. Body weight and the total length of females were recorded as the body dimensions are shown to be related to subsequent spawning.

Breeding under captive conditions

Captive breeding experiments were conducted in indoor glass tanks, outdoor fibreglass tanks and flowing water system. Each of the experiments given below was repeated five times at two-month intervals to increase the number of replicates.

Experiment 1. Breeding under indoor conditions

Four indoor glass tanks with a size of 60 x 30 x 45 cm^3 were used as spawning tanks. Bottoms of the spawning tanks were covered with fine sand mixed with mud for a thickness of 5 cm and were filled with dechlorinated water to a depth of 15 cm and allowed one day to settle the mud and silt particles. Subsequently *Hydrilla* spp. bushes were placed at the bottom. Females and males at a ratio of 1:2 were introduced to each tank at 1600 h and their breeding behaviour was observed. Tanks were provided with illumination for 12 h/day.

Experiment 2. Breeding under outdoor conditions

Four fiberglass tanks with a size of 30 x 60 x 30 cm^3 were used for spawning and was kept at a shady place. Rest of the conditions was provided as in the indoor experiment.

Experiment 3. Breeding under flowing water conditions

A concrete tank was constructed with a size of 90 x 50 x 60 cm^3 and placed at a shady place below the water level of a fishpond, which was used as the water source. A continuous water supply was given to the running water system from the fishpond and the water was allowed to leave from the system through an out-let continuously, resulting an artificial stream. Rest of the conditions was provided as in the indoor experiment.

Water quality parameters in the indoor, outdoor and flowing water breeding experiments were measured using following methods. Temperature, Total dissolved solids and Specific conductivity were measured by a portable conductivity meter. The pH was measured with a standard pH meter and Dissolved Oxygen was estimated titrimetricaly by Winkler's method (Golterman *et al.*, 1978). The water quality parameters in different systems were analyzed using One-way ANOVA and the means were compared using Duncan's Multiple Range test at 95% significance level.

Determination of duration of life cycle from hatchling up to sexual maturity

Time taken for eggs to hatch, hatchling to fry, fry to juvenile stage and adults was determined, and recorded in order to estimate the life cycle. The juveniles were observed at monthly intervals for the development of bright red colour on the body and especially on the fins of males and for the release of eggs from the females in order to determine the age of maturation.

Dietary treatments for fry stage of *Puntius titteya*

A feeding experiment was conducted for 49 days with microworms, *Artemia* nauplii, plankton and FF1 (Formulated Feed 1) as test diets. Twelve indoor glass tanks were used for the experiments (30 X 30 X 45 cm^3; water depth, 30cm). A batch of 240 uniform sized seven-day old fry with a mean length of 8 mm ± 0.02 SE, were selected and they were randomly divided into 12 groups and 20 individuals per tank (three replicates for each treatment). As the fry stage was very sensitive, only Total

Length (TL) was measured in order to estimate the growth. TL of 10 randomly selected fry of each tank was measured at weekly intervals, i.e. 7th, 14th, 21st, 28th, 35th, 42nd and 49th day, by placing them in a glass petri-dish with water and subsequently placing the petridish on a graph paper (with squares of 1 mm^2), and the length of fry was measured by counting the squares.

Dietary treatments for juvenile stage of *Puntius titteya*

Sixty-day old juveniles (mean length: 18 mm ± 0.04 SE; mean weight: 80 mg ± 0.03 SE) were tested with four diets: FF1 (Formulated Feed 1), FF2 (Formulated Feed 2), Dried SWL (Dried swine liver) and plankton for 84 days, adopting the procedure for fry stage. Total Length and the weight of all juveniles were measured fortnightly, i.e. 14th, 28th, 42nd, 56th, 70th and 84th day. A transparent calibrated ruler was used to measure the TL of juveniles up to the nearest milimeter. Fish were blotted on a tissue paper to remove the water on its body surface and kept in a small net bag in order to measure the Body Weight (BW) up to the nearest 0.0001g using an electronic balance.

Estimation of growth parameters

Weight (and length) gains were measured by the difference between final wet weight (length) and initial wet weight (length) following Chiu (1989).

Specific Growth Rate corresponding to Weight (SGR -W) was calculated as

$$SGR\text{-}W = \frac{\ln W_t - \ln W_0}{d} \times 100$$

Similarly Specific Growth Rate corresponding to Length (SGR -L) was found by

$$SGR\text{-}L = \frac{\ln L_t - \ln L_0}{d} \times 100$$

Where: ln = Natural Logarithm; W_t = Mean final live weight; W_0 = Mean initial live weight; L_t = Mean final live length ; L_0 = Mean initial live length; d = Growth interval in days (Purchase and Brown, 2001).

Survival rate was expressed as a percentage of the number of fish introduced into each tank and number of mortality. Results of feeding experiments, length gain, weight gain, SGR – L and SGR – W data of different growth stages and feeding treatments were compared using Two – way Analysis of Variance procedure at 95% significance level, using SAS package. Interaction between time and feed was also tested for significance. Duncan's Multiple Range Test was used to compare the treatment means.

RESULTS AND DISCUSSION

Fecundity and egg diameter distribution in the ovaries

The ovaries were classified into Groups I, II and III according to the size of eggs. Fig. 1 shows the frequency distribution of egg diameter of each fish. Multiple

peaks in the egg diameter-frequency distribution suggest that *P. titteya* are multiple (batch) spawners in which successive batches of eggs become mature as the previous ones are released. In *P. titteya* except the reserve oocytes, (diameter > 399 µm) the ovary consisted of three sizes of yolked eggs (Group I). This situation suggests that *P. titteya* spawns more than once during breeding season. In Group II and III, largest eggs with a mean diameter of 897 µm were absent which may be due to release of eggs on or just before the time of sampling.

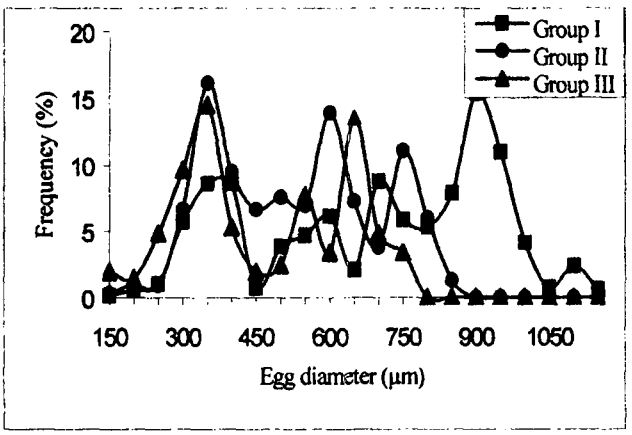

Fig. 1. Frequency distribution of egg diameter of *Puntius titteya*.

Chandrasoma (1996) mentioned that 85% of the total eggs of *P. nigrofasciatus* are reserve oocytes. However, the present study showed that only 30% of the total eggs were reserve oocytes in *P. titteya*. Fecundity (the number of eggs about to spawn) was between 226-284 (Table 2). However, during the laboratory examination, mean number of eggs, which were released, was between 180-200 (Table 3). According to De Silva *et al.* (1985), fecundity of *P. titteya*, which were found in the Kalu Ganga, ranged between 28–201. The lowest value of 28 obtained for fecundity was probably due to their nature of multiple spawning patterns. Furthermore, Bagenal and Braum (1978) showed that there would be a great variability in the number of eggs in fish of the same length, weight and age. However, it could only be possible to predict the relationship between fish length and fecundity by taking a large number of samples. Since *P. titteya* is listed as a highly threatened species (IUCN, 2000) it is not advisable to kill a large number to study their fecundity.

Moreover according to De Silva *et al.* (1985) the Total Length of the *P. titteya* at maturity ranged from 2.1 to 4.0 cm in the *Kalu Ganga* system. However, the maximum total length was found to be 5.4 cm in the present study. It was also observed that they spawn 4–5 times annually under captive conditions. Therefore, it could be suggested that the *P. titteya* exhibit a year round breeding pattern.

Spawning behaviour of *Puntius titteya* under captivity

Out of the three systems supplied to *P. titteya*, i.e. indoor tanks, outdoor tanks and running water, spawning was only observed in the running water system. After introduction into running water system, males and females exhibited active swimming movements and chasing behaviour (males chasing females) on the following day in the morning around 07.00 am. Females tended to expel 3-4 eggs on the *Hydrilla* bushes and males fertilized them. It was observed that the females did not lay eggs continuously but serially. When the abdomen of the female became slender, the parents were removed from the breeding tank in order to prevent predation on the eggs.

Table 2. Sexual maturity, gonadal and egg size parameters of *Puntius titteya*.

Parameter	Mean ± SD
Total Length at maturity (cm)	4.7 ± 0.61
Body Weight at maturity (g)	1.24 ± 0.47
Gonadal Weight (g)	0.16 ± 0.12
Gonado- Somatic Index	17.63 ± 8.47
Fecundity (No of eggs at Stage V)	260 ± 24.00
Mean Egg Diameter (μm)	
Large	897 ± 122.47
Medium	729 ± 79.05
Small	574 ± 64.55

Table 3. Spawning performance of *Puntius titteya*.

Performance / spawner[1]	Mean ± SD
Number of eggs laid at 1st spawning	200 ± 35.2
+Number of eggs laid at 2nd spawning	150 ± 25.41
Percentage of fertilized eggs	76 ± 7.4
Percentage of hatchlings	63 ± 8.4
Length of hatchling (mm)	4.5 ± 0.5
Weight of hatchling (mg)	0.4 ± 0.12

[1] Total length: 4.0– 4.6 cm; body weight: 1.0–1.23 g.

Eggs hatched within 36–48 h after fertilization at 26–28 °C and the crawling movements could be detected from the hatchling with the yolksac immediately after hatching. Yolk sac was absorbed within 2–3 days and the colourless fry unlike their adults, tended to swim within the upper water column. They became red in colour like their parents within two weeks. The breeding pairs of the broodstock exhibited spawning again in 48 – 60 days (Table 3). Although brooders in the indoor tanks and

outdoor tanks were kept for a month by changing the water level and feeding with *Artemia* nauplii, they failed to spawn under those conditions.

Owing to the small size of *P. titteya*, like other fish species such as goldfish (*Carassius auratus*), the stimulation of spawning was not possible using hypophysation procedure (Vallipuram and Edirisinghe, 1996), which could damage the body of the fish and cause high degree of stress. Therefore, the possible method for stimulation of spawning of *P. titteya* is through supplying their preferred environmental conditions. Running water, which was supplied from the fishpond causing continuous change in the contents of dissolved solids, specific conductivity as well as the rate of flow, would have stimulated the spawning under the flowing water system. Furthermore, a comparatively higher total dissolved solids content and conductivity level were found in the flowing water system (Table 4). De Silva *et al.* (1985) have suggested that the spawning pattern of *P. titteya* is related to the rainfall pattern of their natural habitats which may bring about changes in water quality parameters specifically dissolved solids, water currents and water depth.

Table 4. Water quality parameters (mean ± SD) in spawning tanks[1].

Parameter	Running water	Indoor tanks	Outdoor tanks
pH	$7.9^{a} \pm 1.5$	$7.5^{a} \pm 1.2$	$7.6^{a} \pm 1.0$
Total dissolved solids (mg l^{-1})	$199.8^{a} \pm 78.54$	$77.2^{b} \pm 5.9$	$75.7^{b} \pm 6.4$
Conductivity (µs)	$345.7^{a} \pm 84.21$	$119.6^{b} \pm 8.9$	$110.4^{b} \pm 5.2$
Dissolved Oxygen (mg l^{-1})	$7.9^{a} \pm 1.2$	$9.1^{a} \pm 1.6$	$8.0^{a} \pm 2.7$
Temperature (°C)	$25.3^{a} \pm 2.7$	$24.8^{a} \pm 0.2$	$26.4^{a} \pm 1.2$

[1]Values within rows followed by different superscripts are significantly different ($P < 0.05$).

Among the different treatments (indoor, outdoor and flowing water) which were replicated five times, spawning was observed only in four of the replicates belonging to the flowing water system, indicating that flowing water is necessary for spawning. The other factors such as the water currents and fluctuations in the dissolved solids as well as conductivity also may have contributed to the induction of their spawning behaviour. Recovery period for the next spawning was shorter (7-8 weeks) in *P. titteya* when compared to 15 weeks for *P. nigrofasciatus* (Chandrasoma, 1996). The reason for the difference may be that about 70% of the total eggs are yolked eggs in *P. titteya* while in *P. nigrofasciatus* only 15% of the total were with yolk (Chandrasoma, 1996). Thus, *P. titteya* seems to need less recovery period for the maturation of reserve oocytes compared to *P. nigrofasciatus*.

Life cycle of *Puntius titteya*

The fry of *P. titteya* attained the juvenile stage in 45–60 days and reached a total length of 1.2 – 1.8 cm. The release of yolked eggs from females was observed in

180–210 days. Males developed their characteristic bright red colour in 150–180 days specially on the fins, indicating their age of maturity (Table 5).

Effects of test diets on growth of fry stage of *Puntius titteya*

The results of ANOVA revealed that the growth parameters, length gain and SGR–L of fry were significantly affected by the test diets ($P<0.05$) over the period of 49 days (Table 6). Of the 4 tested diets, FF1 and *Artemia* nauplii diets produced significantly higher length gain and SGR–L while they were lowest for plankton diet ($P<0.05$). This may be partly due to the absence of preferred plankton varieties of fry in the plankton diet provided. However, the fry tested on all four types of diets produced a high percentage of survival. Therefore it could infer that even though fry fed on plankton diet showed comparatively lower growth, the planktons could still be used for the rearing of fry.

Table 5. Important phases in the life cycle of *Puntius titteya*.

Phase	Duration
Egg to hatchling	36 – 48 h
Hatchling to fry stage	2 – 3 days
Fry to juvenile stage	45 – 60 days
Adult : Male (size: 2 – 2.5 cm)	150 –180 days
Female (size : 2.5 – 3 cm)	180 – 210 days

Table 6. Growth performances of *P. titteya* fry on test diets.

	Mean[1] ± SE			
	FF1	*Artemia*	Microworm	Plankton
Initial length (mm)	$8.0^a \pm 0.01$	$8.0^a \pm 0.01$	$8.0^a \pm 0.01$	$8.0^a \pm 0.01$
Final length (mm)	$18.5^a \pm 0.36$	$17.9^a \pm 0.64$	$16.1^b \pm 0.48$	$13.5^c \pm 0.45$
Length gain (mm)	$9.4^a \pm 0.07$	$9.2^a \pm 0.03$	$7.9^b \pm 0.05$	$5.7^c \pm 0.03$
SGR - L	$3.16^a \pm 0.16$	$3.01^a \pm 0.14$	$2.73^b \pm 0.13$	$2.17^c \pm 0.10$
Survival %	99	100	98	98

[1] Values within rows followed by different superscripts are significantly different ($P<0.05$).

There was a significant interaction between the diets and the time period with respect to growth performance ($P<0.05$). The fry fed with *Artemia* and microworm exhibited a higher Length Gain and SGR-L when compared with FF1 diet, up to 28 days (Fig. 2). However, from 35 days, these parameters were higher with FF1 than that with *Artemia* and microworm (Fig. 2). Therefore fry of *P. titteya* could be weaned from

live feeds (*Artemia* and microworm) at 42 days of age (7 days old fry were used in the experiment) and introduce formulated feeds without causing a reduction in growth and survival. The present findings indicate that just as the larval forms of other fish, *P. titteya* fry also show a higher preference for live feeds at their initial stage. However, the cost of production for *Artemia* is Rs. 156.25 per 25 g while it is Rs. 35.00 per 1 kg of microworms (Sumith Kumara *et al.*, 2002). Therefore, rearing of fry stage with microworm would be more profitable. After 42 days of age they could be switched on to any other suitable formulated feed.

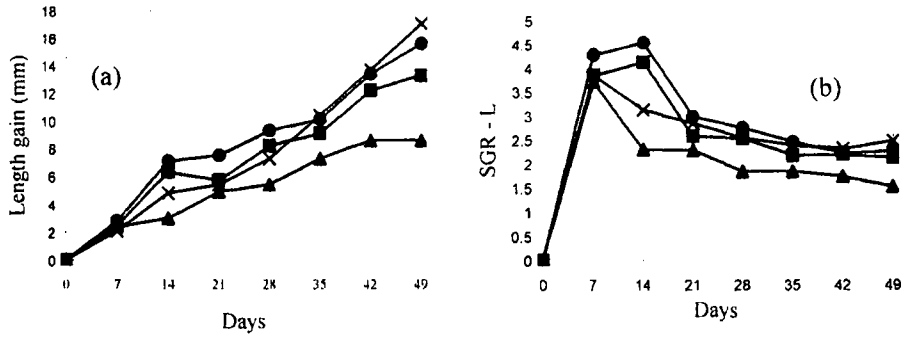

Fig. 2. (a) Length gain (mm) and (b) Specific Growth Rate corresponding to length (SGR – L) of *P. titteya* fry for 49 days of experimental period (age 7 to 56 days) fed with test diets; ■= Microworm; ●= *Artemia* nauplii; ▲= Plankton; × = FF1.

Furthermore, Chandrasoma (1996) has stated that fry stage of *P. nigrofasciatus* could be reared up to 18 days of age with zooplankon such as *Brachionus* spp. and *Moina* spp., and *Artemia* nauplii whereas the formulated diets are suitable from 18 days onwards. According to Kaiser *et al.* (2003), fry stage of goldfish (*Carassius auratus*) could be weaned from *Artemia* to artificial diets by Day-24. The present findings showed that unlike *P. nigrofasciatus* and goldfish, *P. titteya* takes a longer period of 42 days of age to switch on to formulated diets.

Effects of test diets on growth of juvenile stage of *Puntius titteya*

The proximate analysis revealed that dried swine liver contained higher protein and energy content than that of Formulated Feeds FF1 and FF2 (Table 1). Length Gain, Weight Gain, SGR–L and SGR–W of juveniles were significantly different among the different test diets ($P < 0.05$) (Table 7). The highest growth was exhibited with the plankton diets and the lowest growth with FF2 diet (Fig. 3). Results also confirm that unlike fry of *P. titteya*, the juveniles grow better on the plankton diet. Of the three artificial diets, the highest mean length gain, mean weight gain, SGR–L and SGR–W were exhibited by dried SWL and lowest performance by FF2 fed juveniles (Table 7 and Fig. 3).

Table 7. Growth performances of *Puntius titteya* juveniles fed on test diets; SE: Standard Error.

	Mean[1] ± SE			
	Plankton	Dried SWL	FF1	FF2
Initial length (mm)	15.0 ± 0.12	15.4 ±0.03	15.0 ± 0.05	15.2 ± 0.01
Initial weight (mg)	91.3 ± 0.10	91.0 ± 0.10	90.1± 0.08	89.2 ± 0.12
Final length (mm)	27.0^{a} ± .06	24.9^{b} ±0.04	22.7^{c} ± 0.04	21.90^{c} ± .02
Final weight (mg)	250.2^{a} ± .01	180.2^{b} ± 0.01	140.6^{c} ± 0.01	130.5^{c} ± 0.01
Length gain (mm)	7.0^{a} ± 0.07	2.9^{b} ± 0.04	2.1^{c} ± 0.04	1.9^{c} ± 0.03
Weight gain (mg)	162.8^{a} ± 0.02	79.4^{b} ± 0.01	46.1^{c} ± 0.02	41.1^{c} ± 0.01
SGR -L	0.70^{a} ± 0.03	0.28^{b} ± 0.01	0.21^{c} ± 0.03	0.17^{c} ± 0.02
SGR - W	2.73^{a} ± 0.03	1.09^{b} ± 0.01	0.88^{c} ± 0.02	0.71^{c} ± 0.02
Survival %	99	99	100	97

[1] Values in the same row followed by different superscripts are significantly different ($P < 0.05$).

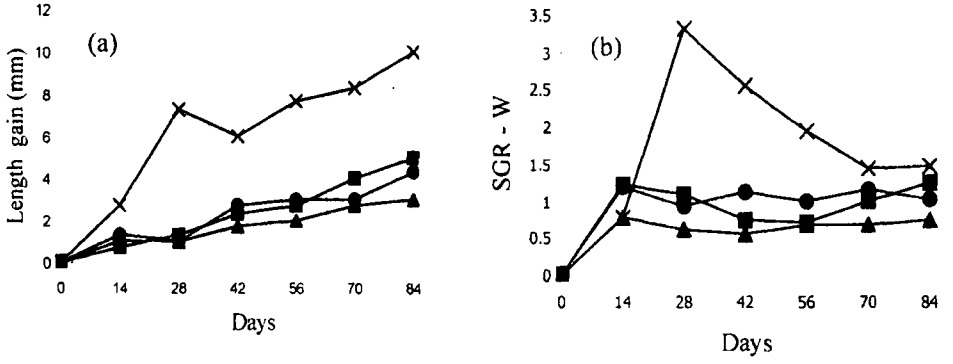

Fig. 3. (a) Length Gain (mm), (b) Weight gain (mg), (c) Specific Growth Rate corresponding to Length (SGR–L) and (d) Specific Growth Rate corresponding to Weight (SGR–W) of *Puntius titteya* juveniles for a experimental period of 84 days (age 60 to 144 days) fed with test diets; ■= FF1; ●= Dried SWL; ▲= FF2; ×= plankton.

CONCLUSIONS

Captive breeding of *P. titteya* can be performed successfully using flowing water systems under laboratory and outdoors conditions using both live and formulated feeds. *P. titteya* is a multiple spawner and exhibit a perennial spawning pattern with a recovery period of 2 months. It attains maturation within one year of age. Like most of the fish, *P. titteya* fry stage also shows a higher growth with live food at their initial stage. However, they could be weaned from live feed to a suitable formulated feed at an age of 42 days without causing a reduction in growth and survival. Although a higher growth of juveniles was exhibited with plankton diet, they could be reared effectively with suitable formulated diets. Furthermore dried swine liver could be used

successfully to feed the young stages of *P. titteya*. Present findings could be used to rear young stages of *P. titteya* under laboratory conditions, which are important for *ex situ* as well as *in situ* conservation.

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