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## Optimization of stocking density and feeding ration for rearing of stunted *Labeo rohita* fingerlings in cages

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### Abstract

**Aim:** To rear stunted *Labeo rohita* fingerlings in cages and determine the optimum stocking density and feeding ration for better growth performance of fishes.

**Methodology:** Eight-month-old stunted fingerlings of *Labeo rohita* (14.65cm/38.23g) were stocked in floating net square cages at different stocking densities (10, 15, 20 and 25 fishm<sup>-2</sup>) and fed with different feeding ration (3%, 4%, 5% and 6% of body weight), further reared for 330 days and fed twice a day with commercial floating pellets with 25% crude protein. The study followed 4\*4 factorial design and were triplicated for each treatment and level.

**Results:** The study found a decrease in the final body weight, average body weight gain, and specific growth rate with increasing stocking density. The highest final body weight (732.64 g), body weight gain (694.30 g) and specific growth rate (0.89 % day<sup>-1</sup>) were recorded in 10/m<sup>2</sup> stocking density with feed ration of 6% body weight. In contrast to this, fish reared in higher stocking density (25/m<sup>2</sup>) exhibited poor feed conversion ratio. Fish reared in lower stocking density (10/m<sup>2</sup>) and fed with 6% feeding ration displayed an enhanced feed conversion ratio, feed efficiency ratio and protein efficiency ratio which indicated that rearing of stunted rohu in intensive cage culture at lower stocking density (10/m<sup>2</sup>) with higher feeding ration (6%) increase their feed utilization capacity.

**Interpretation:** Rearing of stunted rohu in intensive cage culture is possible, however, the stocking density and feeding ration should be 10/m<sup>2</sup> and 6%, respectively, compared to traditional cage practices, in order to reduce stress and to get optimum production.

**Key words:** Cage culture, Feeding ration, Fingerling, *Labeo rohita*, Stocking density

Optimization of stocking density and feeding ration for rearing of stunted *Labeo rohita* (Hamilton, 1822) fingerlings in cages.

Experimental site: *Dimbhe* Reservoir, *Ambegaon*, Pune district Maharashtra, India



Stunted fingerlings were stocked with average weight 38.23 ± 1.90 g and length 14.65 ± 0.3 cm



Highest weight gain (694.30 g) and SGR (0.89 % day<sup>-1</sup>) were achieved with stocking density - 10/m<sup>2</sup> and 6% of feeding ration

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## Introduction

Reservoirs are man-made impoundments that are constructed by obstructing the surface flow of rivers/streams. It forms an important resource for fish production besides holding up economic growth through various cultural and ecological services. In India, the total area of reservoirs is estimated to be 3.51 million ha (CWC, 2016; Sarkar and Mishal, 2017). Indian reservoirs are recognized as “sleeping giant” for the fisheries development, since the resources are untapped, and it has a huge potential to attain a higher fish yield. Management and intensified culture practices in reservoirs can help to increase the fish production. Cage culture is an intensified way of culturing aquatic organisms in the open water bodies such as reservoirs, open seas, etc. The advantages of cage culture in these water bodies include flexibility in utilizing resources, simplified culture practices and harvesting, and multi-use of water resources (Karnatak and Kumar, 2014).

The growth of cage farming in India got momentum during 2010–2012 with funding support from the National Fisheries Development Board, National Mission on Protein Supplementation, Rashtriya Krishi Vikas Yojna which installed cages in the reservoirs of Jharkhand, Chhattisgarh, Madhya Pradesh, and Maharashtra. At present, there are more than 6000 floating cages made of bamboo, galvanized iron, high density polyethylene installed in inland open water bodies (DAHD, 2018). The main problem in intensive cage culture practice is identification of suitable candidate species. Several researchers have studied the culture possibility of carps in cages by changing the variables such as growth, survival, feed utilization, stocking density, feeding ration, etc., especially using catla (Govind *et al.*, 1988), rohu and common carp (Kohli *et al.*, 2002), Indian major and minor carps (Bhattacharjya *et al.*, 2008), *Labeo rohita* fry (Biswas *et al.*, 2015) and carp fry (Sarmah, 2017). Generally, healthy carp fry at 12–15 mm and fingerling of 100–150 mm are preferred for raising in cages in Indian reservoirs (Das *et al.*, 2004). However, the growth and survival of fish stocked in cages is slow. Therefore, the farmers and institutions are in need of quality seed of carps which can survive and grow well in intensive cage culture practices. The stunted fingerlings or yearlings of Indian Major carps are produced by rearing normal fingerlings at higher stocking densities than usual with sub-optimal level of feeding and fertilization for 8–12 months (Ravi *et al.*, 2012).

This reduces the normal growth of fish and results in stunted seed due to less food availability and overcrowding. When these stunted yearlings are stocked in production ponds, under normal rearing conditions, they show compensatory growth, resulting in faster growth than normal fingerling. The capacity of an animal to grow rapidly after a period of reduced growth is often termed as compensatory growth (Jobling *et al.*, 1994; Jobling and Kaskela, 1996; Melard *et al.*, 1998; Ali *et al.*, 2003; Jobling, 2010). Before starting the commercial level of stunted seed rearing in cages, a right candidate species is to be identified and its optimum stocking density and feeding ration

need to be standardized, in order to maximize the production and profitability. Rohu (*Labeo rohita*) is one of the Indian major carps and is considered as the tastiest among the major carps. It is commonly cultured in the Asian countries, especially in the Indian sub-continent (FAO, 2020). Therefore, this preliminary study was conducted to explicate optimization of stocking density and feeding ration for rearing of stunted rohu in open water cage culture.

## Materials and Methods

**Experimental design and rearing of fish:** The present study followed a factorial design (4x4) and carried out for 330 days in floating cages installed in a perennial Dimbhe Reservoir, Ambegaon, Pune district Maharashtra, India. Eight-months-old stunted rohu (14.65 cm/ and 38.23 ± 1.90 g) were procured from private freshwater fish seed farm, Bharuch district, Gujarat, India. Fish were carefully transported in 1000 l capacity of syntax water tank and oxygen level was maintained at saturation level during transportation using an oxygen cylinder. At experimental site, fishes were disinfected with KMnO<sub>4</sub> solution and released into cages for acclimatization for 15 days. Fishes were stocked in 48 cages (3 m x 3 m x 3 m) at different stocking densities such as 10, 15, 20 and 25 m<sup>2</sup> and fed with different feeding ration such as 3, 4, 5 and 6% of body weight. Stocked fishes were further reared for 330 days and fed commercial floating fish feed (Crude protein 25%) twice a day in morning (9.00 hrs) and evening (17.00 hrs). The cages were cleaned manually, weekly once, for proper exchange of water to avoid attachment of any barnacles and algae.

**Water quality analysis and growth sampling:** The water quality parameters of water inside the cages were assessed every month following the standard methods of APHA (2017). Growth sampling was carried out at monthly interval. In each sampling, individual cages were lifted above the water to catch fish. From each cage, 30 fish (n=30) were collected to measure the length and weight. The following fish growth parameters were calculated using the standard formulas.

$$\text{Weight gain (\%)} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}} \times 100$$

$$\text{Feed conversion ratio} = \frac{\text{Feed consumption on dry weight basis}}{\text{Body weight gain on wet weight basis}}$$

$$\text{Protein efficiency ratio} = \frac{\text{net weight gain on wet weight basis}}{\text{protein fed on dry matter basis}}$$

$$\text{The survival (\%)} = \frac{\text{Total fish harvested}}{\text{Total fish stocked}} \times 100$$

**Statistical analyses:** Statistical analysis of different growth parameters was analyzed by Two-way analysis of variance (ANOVA) using SPSS version 22.0 for windows (SPSS Inc., Chicago, IL, USA). Tukey's range test was used for post hoc comparison of mean values (P<0.05) between different treatment groups. Statistical significance (P for the test) was set at P<0.05.

**Table 1:** Water quality parameters recorded in cages reared with stunted rohu fingerlings during the experimental period

Parameters	March – June	July - October	November- February
Temperature (°C)	28-30	23-29	19-24
Dissolved Oxygen (mg l <sup>-1</sup> )	6.3-6.4	7.4-6.8	6.5-7.3
pH	7.4-7.5	7.3-7.5	7.3-7.6
Total Alkalinity (mg l <sup>-1</sup> )	62-78	56-67	36-54
Total Hardness (mg l <sup>-1</sup> )	67-84	58-72	38-58
Transparency (cm)	133-139	100-105	130-135
Total Suspended Solids (mg l <sup>-1</sup> )	0.051-0.054	0.059-0.062	0.052-0.056
Ammonia-Nitrogen (mg l <sup>-1</sup> )	0.28-0.32	0.15-0.23	0.17-0.24
Nitrite-Nitrogen (mg l <sup>-1</sup> )	0.02-0.03	0.01-0.02	0.01-0.02
Nitrate-Nitrogen (mg l <sup>-1</sup> )	0.06-0.09	0.05-0.08	0.05-0.08
Phosphate (mg l <sup>-1</sup> )	0.02-0.07	0.01-0.03	0.01-0.05
Free Carbon dioxide (mg l <sup>-1</sup> )	Nil	Nil	Nil
Biological Oxygen Demand (mg l <sup>-1</sup> )	36-64	30-47	35-56
Chemical Oxygen Demand (mg l <sup>-1</sup> )	28-40	26-32	29-38

## Results and Discussion

The temperature was found to be in the range of 19 to 30°C during the experimental trial with highest and lowest values recorded from May-June and November to February, respectively. The range of dissolved oxygen was recorded 6.3 to 7.4 mg l<sup>-1</sup> with the minimum level observed during pre-monsoon time (May-June). The pH values were within the range of 7.3 to 7.6 throughout the experimental period. The total alkalinity during the study was in the range of 36 to 78 mg l<sup>-1</sup> and the maximum (78 mg l<sup>-1</sup>) and minimum (36 mg l<sup>-1</sup>) values were observed during pre-monsoon (May-June) and post-monsoon (November-February) periods, respectively. Ammonia was observed from 0.15 to 0.32 mg l<sup>-1</sup> and the highest and lowest values were recorded during monsoon (July-October) and pre-monsoon season (November-February). The recorded water quality parameters such as temperature, pH, dissolved oxygen, total alkalinity, ammonia, etc., inside the cages installed in Dimbhe reservoir (Table 1) were within the optimum range for growth and survival of rohu (*Labeo rohita*) fingerling as reported in previous studies (Banerjee, 1967; Huet, 1972; Bettoli et al., 1985; Boyd and Tucker, 1998; Bhatnagar and Devi, 2013). Furthermore, previous study conducted on this reservoir also reported similar range of water quality parameters which further confirmed conducive environment of this reservoir for rohu cage culture (Mane et al., 2017).

The present trial found significant difference ( $P < 0.05$ ) in the final body weight among the treatments at the end of the experiment (Table 2). The highest final body weight (732.64 g), weight gain (694.30 g) and SGR (0.89% day<sup>-1</sup>) were recorded in fish reared at low stocking density 10 fish m<sup>-2</sup> and fed with the highest feeding ration (6% body weight). Fish raised in higher stocking density (25 fish m<sup>-2</sup>) and fed with lower feeding ration (3% body weight) exhibited a lower body weight (211.29 g), body weight gain (172.96 g) and SGR (0.52% day<sup>-1</sup>). The two-way ANOVA analysis clearly showed a significant interaction ( $P < 0.05$ ) between stocking density and feeding ration followed in the

present study on final body weight, weight gain and specific growth rate. In the present study, lower stocking density (10 fish/m<sup>2</sup>) of stunted *L. rohita* in cages showed better growth performance in terms of higher final body weight, weight gain and specific growth rate which indicates that at lower stocking density stunted fish has better growth potential. On the other side, higher stocking density (25 fish/m<sup>2</sup>) negatively affected the growth performance of stunted rohu. In general, at higher stocking densities, individual fish gets reduced space which affects the social interaction and increases stress. In stressed condition, fish spends more energy for stress mitigation which reduces the normal growth of fish (Biswas et al., 2015; Ofor and Afia, 2015). Similar growth pattern was reported in catla (Sukumaran et al., 1986; Govind et al., 1988) *Labeo rohita* (Kohli et al., 2002; Chattopadhyay et al., 2013; Biswas et al., 2015) hybrid catfish (Ofor and Afia, 2015) catla and rohu (Kohli, 2002; Mane et al., 2017).

In fish, the optimal rate of feeding plays a pivotal role which determines the growth potential (Mihelakakis et al., 2002; Cho et al., 2007). The present study found a direct relationship between growth rate and feeding ration which indicated that increased rate of feeding increased the growth performance of stunted rohu. Similarly, stunted rohu and feed restricted Nile tilapia, under optimal feeding conditions, displayed a better growth performance (Das et al., 2008; Limbue and Jumanne, 2014). Stunted fish, in post-stunting phase or optimal feeding condition, exhibited an accelerated body growth as a compensatory growth response (Ali et al., 2003). The reason for faster growth of stunted rohu is increased availability of nutrients which increases the feed consumption and results in better body weight gain. In general, in stunted fish, food availability in post-stunting phase profoundly affects the compensatory growth. Similar values were previously reported in tropical fish, *Clarias gariepinus* (Marimuthu et al., 2011) and *Colossoma macropomum* (Silva et al., 2007). Therefore, the increased availability of nutrients with the increased feed ration could be the reason for better growth of fish reared in higher feeding ration.

**Table 2:** Growth performance, feed utilization and survival of stunted *L. rohita* fingerlings reared in cages under different stocking densities and feeding ratios

Factors effect	Final weight (g)	Weight gain (g)	Specific growth rate (% day <sup>-1</sup> )	Feed conversion ratio	Feed efficiency ratio	Protein efficiency ratio	Survival (%)
<b>Effect of stocking density</b>							
10	657.85 <sup>a</sup>	619.54 <sup>a</sup>	0.86 <sup>a</sup>	1.73 <sup>d</sup>	0.58 <sup>a</sup>	2.43 <sup>a</sup>	100.00 <sup>a</sup>
15	349.39 <sup>b</sup>	311.08 <sup>b</sup>	0.67 <sup>b</sup>	3.20 <sup>c</sup>	0.31 <sup>b</sup>	1.31 <sup>b</sup>	98.27 <sup>b</sup>
20	254.78 <sup>c</sup>	216.47 <sup>c</sup>	0.57 <sup>c</sup>	4.59 <sup>b</sup>	0.22 <sup>c</sup>	0.93 <sup>c</sup>	97.87 <sup>c</sup>
25	243.80 <sup>d</sup>	205.49 <sup>d</sup>	0.56 <sup>d</sup>	5.90 <sup>a</sup>	0.17 <sup>d</sup>	0.71 <sup>d</sup>	95.67 <sup>d</sup>
SEM	0.68	0.69	0.001	0.04	0.003	0.01	0.07
P-value	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<b>Effect of feeding rate</b>							
3	310.16 <sup>d</sup>	271.83 <sup>d</sup>	0.61 <sup>d</sup>	3.68 <sup>b</sup>	0.34 <sup>a</sup>	1.41 <sup>a</sup>	97.30 <sup>b</sup>
4	355.22 <sup>c</sup>	316.84 <sup>c</sup>	0.65 <sup>c</sup>	3.63 <sup>b</sup>	0.34 <sup>a</sup>	1.40 <sup>a</sup>	98.29 <sup>a</sup>
5	409.19 <sup>b</sup>	370.99 <sup>b</sup>	0.69 <sup>b</sup>	3.77 <sup>b</sup>	0.32 <sup>b</sup>	1.35 <sup>b</sup>	98.10 <sup>a</sup>
6	431.26 <sup>a</sup>	392.92 <sup>a</sup>	0.71 <sup>a</sup>	4.33 <sup>a</sup>	0.29 <sup>c</sup>	1.20 <sup>c</sup>	98.11 <sup>a</sup>
SEM	0.68	0.69	.001	0.04	0.003	0.01	0.07
P-value	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<b>Stocking density* Feeding rate</b>							
10*3	548.79 <sup>d</sup>	510.46 <sup>d</sup>	0.81 <sup>c</sup>	1.62 <sup>m</sup>	0.62 <sup>a</sup>	2.57 <sup>a</sup>	100.00 <sup>a</sup>
10*4	630.78 <sup>c</sup>	592.39 <sup>c</sup>	0.85 <sup>b</sup>	1.66 <sup>m</sup>	0.60 <sup>b</sup>	2.51 <sup>b</sup>	100.00 <sup>a</sup>
10*5	719.20 <sup>b</sup>	681.00 <sup>b</sup>	0.89 <sup>a</sup>	1.71 <sup>l</sup>	0.58 <sup>c</sup>	2.43 <sup>c</sup>	100.00 <sup>a</sup>
10*6	732.64 <sup>a</sup>	694.30 <sup>a</sup>	0.89 <sup>a</sup>	1.91 <sup>k</sup>	0.52 <sup>d</sup>	2.19 <sup>d</sup>	100.00 <sup>a</sup>
15*3	268.19 <sup>j</sup>	229.86 <sup>j</sup>	0.59 <sup>h</sup>	3.10 <sup>l</sup>	0.32 <sup>e</sup>	1.35 <sup>e</sup>	97.28 <sup>c</sup>
15*4	312.77 <sup>g</sup>	274.39 <sup>g</sup>	0.64 <sup>f</sup>	3.12 <sup>l</sup>	0.32 <sup>e</sup>	1.33 <sup>f</sup>	98.77 <sup>bc</sup>
15*5	393.14 <sup>i</sup>	354.94 <sup>i</sup>	0.71 <sup>e</sup>	3.19 <sup>l</sup>	0.31 <sup>ef</sup>	1.31 <sup>g</sup>	99.26 <sup>ab</sup>
15*6	423.47 <sup>e</sup>	385.13 <sup>e</sup>	0.73 <sup>d</sup>	3.37 <sup>h</sup>	0.30 <sup>f</sup>	1.24 <sup>h</sup>	97.78 <sup>cd</sup>
20*3	212.36 <sup>n</sup>	174.03 <sup>n</sup>	0.52 <sup>j</sup>	4.06 <sup>g</sup>	0.25 <sup>g</sup>	1.03 <sup>i</sup>	98.15 <sup>cd</sup>
20*4	244.48 <sup>l</sup>	206.09 <sup>l</sup>	0.56 <sup>j</sup>	4.04 <sup>g</sup>	0.25 <sup>g</sup>	1.03 <sup>i</sup>	97.22 <sup>cd</sup>
20*5	268.23 <sup>j</sup>	230.03 <sup>j</sup>	0.59 <sup>h</sup>	4.43 <sup>f</sup>	0.23 <sup>h</sup>	0.94 <sup>j</sup>	97.59 <sup>cd</sup>
20*6	294.07 <sup>h</sup>	255.72 <sup>h</sup>	0.62 <sup>g</sup>	5.84 <sup>c</sup>	0.17 <sup>h</sup>	0.72 <sup>k</sup>	98.52 <sup>c</sup>
25*3	211.29 <sup>n</sup>	172.96 <sup>n</sup>	0.52 <sup>j</sup>	5.93 <sup>b</sup>	0.17 <sup>h</sup>	0.70 <sup>l</sup>	93.78 <sup>e</sup>
25*4	232.86 <sup>m</sup>	194.48 <sup>m</sup>	0.55 <sup>k</sup>	5.69 <sup>b</sup>	0.18 <sup>i</sup>	0.73 <sup>k</sup>	97.19 <sup>cd</sup>
25*5	256.18 <sup>k</sup>	217.98 <sup>k</sup>	0.58 <sup>i</sup>	5.74 <sup>d</sup>	0.17 <sup>h</sup>	0.73 <sup>k</sup>	95.56 <sup>d</sup>
25*6	274.87 <sup>i</sup>	236.52 <sup>j</sup>	0.60 <sup>h</sup>	6.22 <sup>a</sup>	0.16 <sup>i</sup>	0.67 <sup>m</sup>	96.15 <sup>cd</sup>
SEM	1.36	0.34	0.003	0.08	0.006	0.03	0.10
P-value	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Values (mean; n=30) in the column with different superscripts differ significantly ( $p < 0.05$ ). SEM – Standard error of mean

Feed conversion ratio values were found to be lower in low stocking density (1.62) and high in higher stocking density (6.22) with feeding ration of 3% and 6%, respectively. Significantly higher feed efficiency ratio and protein efficiency ratio values were exhibited by fish raised in lower stocking density fed with different feeding rations, except 6% feeding ration. Overall, no significant ( $P > 0.05$ ) difference was observed among 3, 4 and 5% feeding groups. However, significant difference was found among different stocking densities for feed efficiency ratio and protein efficiency ratio values. Statistically, a significant interaction ( $P < 0.05$ ) was found between stocking density and feeding ration.

Feed conversion ratio and feed efficiency ratio are the feed intake parameters used to determine the growth rate and feed utilization capacity of cultured fish (Amin *et al.*, 2005). The

feed conversion ratio in the present study showed a direct relationship, *i.e.*, the increase in stocking density and feeding ration increased the feed conversion ratio values. Contrast to this, feed efficiency ratio and protein efficiency ratio values showed inverse relationship, *i.e.*, increase in stocking density and feeding ration decreased the protein efficiency ratio and feed conversion ratio values. The increased feed conversion ratio in higher stocking densities and feeding ration may be due to stress, and the fish might have spent the extracted feed energy for stress mitigation (Liti *et al.*, 2006; Mensah *et al.*, 2013). The result is in agreement with the previous studies conducted in turbot fry (Devesa, 1994; Sahin, 2001). Protein efficiency ratio is used to determine how well the fish had utilized the crude protein present in the diet. In the present study, protein efficiency ratio of fish was negatively affected by increased stocking density and feeding

ration. The stunted fish reared in lower stocking density (10 fish m<sup>-2</sup>) exhibited higher protein efficiency ratio probably due to the absence of crowding stress which might have helped them to efficiently utilize protein (25% crude protein) present in the given diet. Similar to stocking density effect, increased feeding ration (6%) significantly decreased the protein efficiency ratio value and this may be due to overfeeding of fish where a quantity of feed remains uneaten which increase the value of protein efficiency ratio. Similar observations were reported in tilapia (Essa and Nour, 1988; Cruz and Ridha, 1991) and rohu fry (Biswas et al., 2015). The cent percent survival of fish was observed in low stoking density (10 fish m<sup>-2</sup>) and it decreased as the stocking density increased. Significantly lower survival (93.78%) was recorded in higher stocking density (25 fish m<sup>-2</sup>) fed with low feeding ration (3% of body weight). At lower stocking density and higher feeding rate, the survival percent was found better. In contradictory, higher stocking density did not affect the survival of silver catfish (Kpogue, 2013; Tossavi et al., 2016).

However, the present study registered a better survival rate in all treatments which could be due to the fact that stunted seeds are too hardy and survive in stressed condition (Ali et al., 2003). Further more, few studies stated that favorable ecological conditions, also, play a major role in survival of fish in captive conditions (El-Sherif and El-Feky, 2009; Tossavi et al., 2016). Favorable environmental conditions of the study site could be the possible reason for better survival of fish in cages. Hence, Dimbhe reservoir can potentially be used to improve the production of fish through cage culture practices by stocking stunted seed.

The stunted rohu in cages exhibited a better growth performance at lower stocking density and higher feeding ration, therefore, the results suggest that stocking density of 10 fish m<sup>2</sup> with a feeding ration of 6% body weight would be optimum for rearing stunted rohu in cages. From economic point of view, feeding ration of 5% body weight is cost-effective at 10 fish m<sup>2</sup> stocking density.

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### Add-on Information

**Authors' contribution:** V.K. Paswan: Carried out the study and prepared the manuscript; K.D. Rawat: Conceptualization of research, guidance and manuscript correction; P.P. Srivastava, M.D. Aklakur: Co-guidance and manuscript correction; C. Prakash: Co-guidance, data analysis, , S. Saseendran: Assisted during sampling; R.S.S. Lingam: Assisted during sampling, data analysis and paper drafting.

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