# Impact of different feeds on cannibalism in early larval stages of Koi carp (*Cyprinus carpio*)

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# ABSTRACT

Koi carp (*Cyprinus carpio*) is one of the commercially important ornamental fishes and reared in cottage industry for domestic and export market in many countries. In hatchery larval rearing, provision of suitable feed and control of cannibalism are the prime factors which govern the growth and survival of this species. In the present study, growth and survival of early larval stages of Koi carp were evaluated by feeding them with different zooplankton like rotifer (*Brachionus plicatilis*), cladoceran (*Ceriodaphnia reticulata*), copepod (*Apocyclops dengizicus*), mixture of these three zooplankton and the pelletized feed. Feeding trials were conducted in triplicate for three weeks. Results indicated higher survival and growth performance of larvae fed with rotifer, copepod and mixed zooplankton than those fed with cladoceran and pellet feed. Cannibalism is predominantly observed in the larvae ranging in length from 9.00 to 16.00 mm. Higher percentage of cannibalism was recorded in pellet- and cladoceran-fed larvae than in other feeds experimented. To improve the economics of Koi carp culture in hatcheries, use of rotifer (*B. plicatilis*) and copepod (*A. dengizicus*) feed is recommended that provides better growth and survival performance as well as less cannibalism.

# 1. INTRODUCTION

The cannibal behavior is common in the fish and it is considered one of the main causes of mortality in the larviculture[1]. Cannibalism is interaspecific predation [2] and takes place at various ages; however, it is more intense among fast-growing larvae and juveniles than among adults [3]. In certain cultivable teleost species, there may be population loss ranging from 15 to 90% due to sibling cannibalism [4]. It was reported that there are many factors such as environmental, behavioural, genetic and physiological which affect cannibalism in the teleost species [5]. Cannibalism influencing factors in the early fish larvae include availability of food, live food or pelletized feed, population density, photoperiod and turbid condition[6].

Cannibalism is common in freshwater fish and wide spread among marine species [7]. Valerio and Barlow[8] opined that absence of cannibalism in a group of fishes should be considered as exceptional. The review of cannibalism in teleost by Polis [9] reveals that in many species population structure, life history, competition for mates and resources, and behaviour might be the influencing factors in nature. In the case of hatchery seed production of finfish one of the factors is cannibalism, as most of commercially important fish show sibling cannibalism, there should be adequate research focussed towards causes, effects and control of cannibalism so as to achieve higher survival rates of their larvae in captive rearing. Several factors appears to influence cannibalism under natural and rearing conditions and these factors differ in different species as reported in fishes such as Esox lucius (L.)[10]. Stizostedion vitreum (Mitchill)[11], Clurius gariepinus (Burchell) [4]. Common carp and Koi carp larvae [12-15], Pomatomus saltatrix L.[16]., Asian sea bass Lates calcarifer (Bloch)[17-19], Common snook Centropomus undecimalis, [20]. and orange-spotted grouper Epinephelus coioides, [2]. Larviculture of Koi carp is a difficult task for fish farmers since they are carnivorous, piscivorous and cannibalistic during larval stages. Considering the fact that the live food is one of the influencing factors of cannibalism in the early developmental stages of fishes, it is necessary to assess suitable species of zooplankton for growth and survival of different fish larvae so as to minimize cannibalism. This article evaluates cannibalism in Koi carp larvae fed on three different zooplankton species, mixed feed of these plankton and a commercial pelletized feed.

## 2. MATERIAL AND METHODS

#### 2.1 Larviculture

Mature Koi carp (one female and two male) were introduced in cement tank in a fish farm (Kolathur, Chennai, India) to spawn the fish by natural breeding.Parent Koi carp were removed after breeding and eggs were allowed to hatch. After hatching,5day-old Koi carp larvae were brought from fish farm to laboratory with least disturbance and they were acclimatized for one day with rotifer live feed in the wet lab and then used for the experiments.

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On the seventh day after hatching, duplicate groups of Koi carp larvae were transferred to the 100-l larval rearing FRP tanks. In each tank 100 larvae (mean total length 9±0.13 mm and wet weight  $0.02\pm0.0$  mg) were introduced in 50 l of water. The larvae were fed with the following four live food diets: the rotifer, Bracionus plicatilis; the cladoceran, Ceriodaphni reticulata; the cyclopoid copepod, Apocyclops dengizicus; mixed zooplankton diet, (B. plicatilis, C. reticulata and A. dengizicus in equal volume) and the pelletized feed (Professional Koi Food, Excel Pond, China). The feed was given twice a day at 8.00 and 16.00 hours to satiation with not less than 100 live organism/larvae/meal [21]. Experiment was conducted for 21 days to quantify cannibalism in koi carp larvae following the method of Van Dammi et al. [22]. All the fish from each aquarium were removed at the end of every week and counted for recording cannibalism. Fishes injured by the cannibals and that died of natural causes were examined daily. To calculate the rate of cannibalism, the difference in fish numbers between each count was reduced by the recorded natural mortality. During the course of rearing, the number of cannibals was estimated. The sediment (faeces and dust) in the tanks were siphoned from the bottom and also 30% water was replenished from rearing tank with least disturbance to the fish larvae. Physicochemical parameter was maintained in the following range during the experimental period; water temperature 25± 1°C, pH 7.85±0.11, Salinity 6±0.14 ppt and DO 4.1±0.21. Fish were sampled at the end of every week of the experimental period for evaluation of growth and survival. Length and weight of 20 individual fish collected from rearing tanks at random for every feed were measured. Length, weight gain, survival and specific growth rate and cannibalism were calculated by employing the following equations:

**Length gain of larvae (mm) =** Average final larvae length-Average initial larvae length

Weight gain of larvae (g) = Average final larvae weight -Average initial larvae weight

Specific growth rate ( $\% \text{ day}^{-1}$ ) =

<u>Final fish larvae weight -Initial fish larvae weight ×100</u> Experimental period

Survival rate (%) = <u>No. of fish larvae survived ×100</u> No. of fish larvae introduced Cannibalism rate (%) = 100- (Survival rate% + Observed mortality %)

The experimental data was analyzed using one-way ANOVA and followed by Duncan Post Hoc test by using IBM SPSS software version 19 at P < 0.05% level confidence.

## **3. RESULTS**

#### 3.1 Growth

The Koi carp larvae readily consumed all the feeds and one way ANOVA showed significant differences in length and weight of the fishes when fed with different feeds at P< 0.05 level (Table 1). Duncan Post Hoc test recoded pronounced variations with regard to length and weight of larvae fed different feeds during different weeks of experimental period (Table 2). Larval total length (7.55 $\pm$ 0.14 mm) and wet weight gain (0.030 $\pm$ 0.001g) were significantly high in *B. plicatilis* and *A. dengizicus* (7.19 $\pm$ 0.3 mm and 0.028 $\pm$ 0.001 mg, respectively) diet compared to other diets provided. The least length of 4.05 $\pm$ 0.17 mm and weight gain of 0.007 $\pm$ 0.00 g was recorded in pelletized feed. Larvae fed rotifer live feed (*B. plicatilis*) showed higher specific growth rate (1.33%) than other diets. Lowest specific growth rate (0.03%) was recorded in pellet fed larvae.

#### 3.2 Survival and mortality

Koi carp larvae fed on live feeds viz. *B. plicatilis, A. dengizicus,* mixed zooplankton diet and *C. reticulata* showed significantly high survival rate 80%, 75%, 71.6% and 70%, respectively, and low survival rate (50%) was recorded in pelletized diet during 21 days rearing period. Significant differences were recorded in the survival rate of the larvae with different feeds. Observed mortality also showed variation in larvae fed with pelletized feed and live feeds (Table 1).

#### 3.3 Cannibalism

In the present study cannibalism in Koi carp larvae was observed from 8 day post hatch (dph) to 23 dph and larval length during this period ranged between 9. 0 to 16.0 mm. Cannibals swim vigorously around the tank, ignoring live feed and targeting their siblings. Though cannibalism was recorded in all the feeds experimented, it was significantly lower in fish fed with B. plicatilis (19.67%), A. dengizicus (24.33%) and mixed zooplankton diet (27.33%) during 10 dph to 15 dph. Cannibalism was high in fish fed with crushed pellet feed (48.66%) and C. reticulata (29.67%) (Table 1). Generally, cannibals attack the abdomen and tail region of the prey (Figs.1 and 2). It was observed that during the first week of experimental period, mouth size of the cannibal was smaller than the tail size of the prey, in such condition shooter attack and injures the yolk sac and tail region. During the second and third week mouth of the cannibal develops bigger (diameter of 990 to 1080µm) than the tail height of the prey (765 to  $855\mu m$ ) and consumes the whole prey by engulfing it from the tail region (Fig. 2).



Fig. 1: Injured tail of Koi carp prey in cannibalism.



Fig. 2: Cannibal engulfing its prey in Koi carp cannibalism.

The mean length and width of rotifer, *B. plicatilis* (210  $\mu$ m and 154  $\mu$ m) might be of suitable size for Koi carp early larvae. Easier digestion and suitability of nutrient content of this feed might be the reason for maximum growth and survival and least cannibalism of Koi carp larvae in this feed. The length and width of adult cyclopoid copepod, *A. dengizicus* was 964  $\mu$ m and

459  $\mu$ m, respectively and also availability of their different naupliar stages ranging in length from 200 to 964  $\mu$ m and width of 150 to 459  $\mu$ m along with high nutrient content might support higher growth, lower cannibalism and high survival. Availability of suitable sized prey in mixed zooplankton might be attributed to the better growth, survival and low cannibalism of Koi carp larvae in this feed.

The size of the cladoceran, *C. reticulate* (429  $\mu$ m in length 265  $\mu$ m in width,) along with its circular body covered by shell might cause slow ingestion and digestion of this feed which might be the reason for comparatively low growth, low survival and high cannibalism in this feed than other live feeds. Minimum growth, survival and maximum cannibalism were recorded in artificial pellet which could be attributed to the preference of live feed than the pelletized feed by the Koi carp larvae. In the absence of live feed in their environment, they tend to develop cannibalistic behavior and indulge in predation of their own sibling.

Table. 1: Growth perform	ance of Koi carp (Cyprinus co	arpio) larvae fed with different fee	eds during 21 days rearing pe	eriod (Mean ± SE)
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	B. plicatilis	C. reticulata	A. dengizicus	Mixed diet	(Pellet feed)
Initial length of fishes (mm)	9.05±0.13	9.05±0.13	8.95±0.13	9±0.14	8.95±0.13
Final length of fishes (mm)	16.60±0.27	14.83±0.29	1614±0.43	15.23±0.39	13±0.30
Gain in length (mm)	7.55±0.14	5.78±0.16	7.19±0.3	6.23±0.25	4.05±0.17
Initial weight of fishes (g)	0.020±0.001	$0.020\pm0.001$	$0.019 \pm 0.001$	$0.021 \pm 0.001$	0.020±0.001
Final weight of fishes (g)	$0.050 \pm 0.002$	0.040±0.003	0.047±0.002	$0.044 \pm 0.002$	0.027±0.001
Gain in weight (g)	$0.030 \pm 0.001$	$0.020 \pm 0.002$	0.028±0.001	$0.023 \pm 0.001$	$0.007 \pm 0.00$
Specific growth rate (SGR) (%)	$1.33 \pm 1.14$	0.72±0.59	1.09±0.91	0.10±0.05	0.03±0.02
Survival rate (%)	80±5.00	70±7.63	75±5.00	71.6±6.66	50±10.40
Observed mortality (%)	0.33±0.00	0.33±0.00	0.67±0.00	1.07±0.57	1.33±0.66
Cannibalism (%)	19.67±5.33	29.67±7.63	24.33±4.70	27.33±6.35	48.66±10.89
Cumiounism (70)	17.07-5.55	27.01±1.03	27.33±7.70	21.33±0.33	+0.00±10.07

Table. 2: Growth performance of Koi carp (*Cyprinus carpio*) larvae fed with different feeds during different weeks of rearing period (Mean ± SE).

	Parameters	B. plicatilis	C. reticulata	A. dengizicus	Mixed Zooplankton	(Pellet feed)
0 Week	Initial length of fishes (mm)	9.05±0.13	9.05±0.13	8.95±0.13	9±0.14	8.95±0.13
	Initial weight of fishes (g)	$0.020 \pm 0.001$	0.020±0.001	$0.019 \pm 0.001$	$0.021 \pm 0.001$	$0.020 \pm 0.001$
1Week	Length of fishes during 1 <sup>st</sup> week (mm)	11.06 <sup>ab</sup>	11.18 <sup>ab</sup>	11.12 <sup>ab</sup>	10.59 <sup>ab</sup>	9.79 <sup>aa</sup>
	Fish length gain during 1 <sup>st</sup> week (mm)	2	2.65	2.16	1.58	0.83
	Weight of fishes during 1 <sup>st</sup> week (g)	0.028 <sup>bb</sup>	$0.022^{ba}$	0.023 <sup>ba</sup>	$0.022^{ba}$	0.021 <sup>ba</sup>
	Fish weight gain during 1 <sup>st</sup> week (g)	0.008	0.002	0.004	0.001	0.001
	Specific growth rate during 1 <sup>st</sup> week (SGR) (%)	3.614	1.914	2.928	0.01	0.01
	Survival rate during 1 <sup>st</sup> week (%)	90	85	85	85	70
	Observed mortality during 1 <sup>st</sup> week (%)	1	1	0	1	2
	Cannibalism during 1 <sup>st</sup> week (%)	9	14	15	14	28
2 <sup>nd</sup> Week	Length of fishes during 2 <sup>nd</sup> week (mm)	16.33 <sup>cc</sup>	13.69 <sup>cb</sup>	15.71 <sup>cc</sup>	14.62 <sup>cb</sup>	12.56 <sup>ca</sup>
	Fish length gain during 2 <sup>nd</sup> week (mm)	5.28	2.52	4.60	4.03	2.77
	Weight of fishes during 2 <sup>nd</sup> week (g)	$0.052^{de}$	0.033 <sup>db</sup>	$0.042^{dc}$	0.037 <sup>db</sup>	0.021 <sup>da</sup>
	Fish weight gain during 2 <sup>nd</sup> week (g)	0.024	0.011	0.019	0.015	0
	Specific growth rate during 2 <sup>nd</sup> week (SGR) (%)	0.342	0.157	0.285	0.20	0.014
	Survival rate during $2^{nd}$ week (%)	75	65	70	65	45
	Observed mortality during 2 <sup>nd</sup> week (%)	0	0	2	2	2
	Cannibalism during 2 <sup>nd</sup> week (%)	25	35	28	33	53
3 <sup>rd</sup> Week	Length of fishes during 3 <sup>rd</sup> week (mm)	16.60 <sup>ed</sup>	14.83 <sup>eb</sup>	16.14 <sup>ec</sup>	15.23 <sup>eb</sup>	$13^{ea}$
	Fish length gain during 3 <sup>rd</sup> week (mm)	0.270	1.14	0.43	0.620	0.45
	Weight of fishes during 3 <sup>rd</sup> week (g)	$0.056^{\text{gc}}$	$0.040^{gb}$	0.047 <sup>gb</sup>	0.045 <sup>gb</sup>	$0.027^{ga}$
	Fish weight gain during 3 <sup>rd</sup> week (g)	0.004	0.007	0.005	0.008	0.006
	Specific growth rate during 3 <sup>rd</sup> week (SGR) (%)	0.05	0.10	0.07	0.11	0.08
	Survival rate during 3 <sup>rd</sup> week (%)	75	60	70	65	35
	Observed mortality during 3 <sup>rd</sup> week (%)	0	0	0	0	0
	Cannibalism during 3 <sup>rd</sup> week (%)	25	40	30	35	65

Note: Superscripts denotes subsets in Duncan Post hoc test significant at 0.05 level

#### 4. DISCUSSION

The requirement of huge quantity of live feed in aquaculture particularly in ornamental fish culture is indispensible one to rear the larvae of many fishes from endoexogenous to exclusively exogenous feeding stages. Provision of suitable live feed during this stage is vital and determines the rate of survival and cannibalism in Koi carp larvae. Even though, artificial feed supported the larval rearing of Koi carp larvae this feed is not preferred by the early stage larvae. According to Govoni et al. [23] assimilation efficiency of gut may be lower in larvae than in the adult fishes, due to the lack of morphological and functional stomach in larvae. It is also observed that with the increase in age and growth, the Koi carp prefers large sized prey than the smaller ones. Further, cannibalism can be reduced at different larval stages by providing prey of suitable size to the growing Koi carp. Fukuhara [24] reported that in grouper, mass mortality resulted in the larval stages measuring 8-50 mm in length due to cannibalism. Survival rate was reported to be very poor in earlier stage of Koi carp life cycle [15]. In the present study cannibalism in Koi carp was predominant when fish were at 9-15 mm in body length. Cannibalism is a behavior manifested under conditions of food shortage and in the larviculture their main cause is associated to food availability [1]. Cyprinid larvae are known to prefer natural food items such as free living protozoa and rotifers, and larger planktonic organisms like cladocerans and copepods at fry and fingerling stage [25]. Present study clearly showed that Koi carp larvae mostly preferred natural food than artificial feed. Santamaría and Santamaría [26] reported that rotifers are suitable for the first feeding larvae of many ornamental fishes due to their small size, slow swimming and nutritional quality The results of present study also revealed that rotifers (B. plicatilis) is suitable live feed for the first feeding Koi carp larvae as this feed promoted better growth, survival and also prevented cannibalism. Copepods have been used to rear the larvae and fry of many finfish and shellfish [27] and also reported to be an advantageous initial feed in fish larviculture [28]. Malnutrition is one of major responsible factors for failure in larval growth and survival [27]. The larvae fed with copepods exhibited better growth and survival than those fed artificial diets [29]. The results of present study also revealed that copepod (A. dengizicus) is suitable live feed for the first feeding Koi carp larvae due to suitable size, attractive jerking movement and high nutritional values which promote the growth, survival and reduced sibling cannibalism. The cladoceran Ceriodaphnia is preffered as live food organism by early larval fish due to smaller size than other cladocerans [30-32]. However, it is observed that even though C. reticulata can be used as live feed for Koi carp larvae culture, this feed showed lower growth and survival than rotifer and cyclopoid copepod feeds. Nevertheless, provision of C. reticulata during post larval stage may support better growth and survival of Koi carp larvae. Many factors such as genetic, behavioural, environmental and physiological appear to cause cannibalism in fishes. Genetic factors appear to influence the size variation within the coeval

sibling. It is the genotypic variations in the individuals which might cause the different growth rate [33, 8] and larger egg size leading to larger larvae at hatching [9]. Up on having the advantage of larger initial size through genetic set up, the larger individuals of the cohort develop aggressive behavior, while vulnerable or weaker larvae with physiological stress become subject of cannibalism. Similar to this view Koi carp larvae showed size variations in their eggs, hatchlings, and different larval stages which might also influence cannibalism. Giles et al. [10] while reporting cannibalism in the larvae of walleye, Stizostedion vitreum, also expressed similar view. Pomeroy [34] also observed that the presence of larger potential prey can stimulate the development of cannibalism in larvae. In Clarius gariepinus, Hecht and Appelbaum [4] observed that food availability, population density and refuge were significant environmental factors that affected the rate and extent of cannibalism. In Lates calcarifer, stocking density, water quality, light intensity and frequency at which alternative prey is offered determines the rate of cannibalism [33], while Katavic et al. [35] reported that cannibalism can be reduced by increasing the feeding frequency. Cannibalism which is prevalent in the early larval stage of Koi carp showed steep decline after 28 dph probably due completion of ontogeny and attainment of functionality of many systems like digestive system, receptors and locomotory system. Advanced larval forms and juveniles avoid cannibalism by developing schooling behavior.

# 5. CONCLUSION

The present study reveals that better growth and survival of Koi carp larvae can be achieved using rotifer and copepod live feed. Further, these feeds reduce cannibalism in the critical larval stages and enhance their survival. Hence for Koi carp, live feed of zooplankton (*B. plicatilis* and *A. dengizicus*,) are recommended as initial larval feed and use of this indigenous live feed might have a positive impact on economy of the ornamental fish industry.

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