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Influence of Feed, Manure and Their Combination on The Growth of *Cyprinus carpio* (L.) Fry and Fingerlings

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Abstract

Two field experiments, each of 98-day duration, were conducted in outdoor mud- bottomed cement tanks, employing common carp (*Cyprinus carpio*) fry and fingerlings, with a view to study the effect of nutrient inputs (feed, manure and their combination) on different life stages of the fish. The treatments consisted of control (C), only feed (F), only manure (M) and manure plus feed (M+F). Poultry manure was applied in split doses to tanks of manure treatments (M and M+F). Fry (Experiment one) and fingerlings (Experiment two) of average weight 0.67 g and 3.2 g respectively, were stocked seven days after the initial manure application at 4 individuals/m^2. Fish in F and M+F treatments were provided a fish meal based pelleted diet once daily in the morning at 5% of body weight. The most dominant genera of phytoplankton encountered were *Microcystis*, *Anabaena* and *Microspora*, while among the zooplankton, *Keratella* and Nauplii dominated. M+F treatment had the highest plankton biomass (P<0.05). Significant (P<0.05) variation in both phyto and zooplankton dry weight was recorded with respect to the study period. Both feeding and manuring, individually and in combination, significantly improved (P<0.05) the growth of fish. The highest specific growth rate, final fish weight, and gross production were recorded in M+F treatment in both the experiments. The difference in survival among the control and treatments was not significant in experiment one (P>0.05), whereas F and M+F treatments recorded lower survival (P<0.05) in experiment two. Fish production was comparable under feed (F) and manure (M) treatments (P>0.05) in experiment one, but was significantly (P<0.05) different in experiment two. The increment in gross fish production over the control was 103.22 and 119.99% in feed, 77.30 and 59.44% in manure and 162.34 and 175.08% in M+F treatments of the two experiments respectively. Carcass protein, fat and ash contents were significantly (P<0.05) higher in the three treatments as compared to the control. The different digestive enzymes recorded higher values in fish from the three treatments than those of the control.

Keywords: Natural food, poultry manure, life stage, carcass composition, digestive enzymes.

Larva ve Yavru *Cyprinus carpio* (L.)'un Gelişimi Üzerine Beslenme ve Gübre Kombinasyonlarının Etkisi

Özet

Balığın farklı evrelerinde besin girişlerinin (beslenme ve gübre kombinasyonları) etkileri hakkında bir fikir elde edebilmek için 98 gün süresince içinde *Cyprinus carpio* larva ve yavruları bulunan çamur dipli açık çimento tanklarında iki farklı saha denemesi yapılmıştır. Muamele kontrol (C), sadece yem (F), sadece gübre (M) ve gübre artı yem (M+F) 'den oluşmaktadır. Kümes hayvanlarından alınan gübre, gübre muamelesi yapılan (M ve M+F) tanklara bölünmüş dozlarda uygulanmıştır. Larvalar, ortalama 0,67 g (deney 1) ve yavrular, ortalama 3,2 g (deney 2) ilk gübre uygulamasından sonra yedi gün stoklanmıştır (4 birey/m^2). F ve M+F'deki balıklara pellet diyetler vücut ağırlıklarının %5'ine denk gelecek miktarda günde bir kez olacak şekilde sadece sabahları verilmiştir. Fitoplanktonlar arasında en baskın olanlar *Microcystis, Anabaena* ve *Microspora*'dır. Zooplanktonlar arasında ise dominant olanlar *Keratella* ve Nauplii'dir. M+F muamelesi en yüksek plankton biyokütlesine sahipti (P<0,05). Fito ve zooplanktonun her ikisinin kuru ağırlığındaki önemli (P<0,05) varyasyon çalışma boyunca kaydedilmiştir. Yemleme ve gübrelemenin her ikisi de teker teker ve kombinasyonlar halinde balığın büyümesini önemli derecede arttırmıştır (P<0,05). Her iki denemedeki M+F muamelelerinin en yüksek spesifik büyüme oranı, balığın son ağırlığı ve brüt üretim kaydedilmiştir. Hayatta kalma açısından kontrol ve muameleler arasındaki fark deney 1'de önemli (P>0,05) çıkmamaştır; oysa deney 2'de F ve M+F muamelelerinde daha düşük hayatta kalma (P<0,05) oranı kaydedilmiştir. Deney 1'de balık üretimi, yemleme (F) ve gübreleme (M) muameleleri için kıyaslanabilir (P>0,05) fakat deney 2'de önemli derecede farklı bulunmuştur (P<0,05). Brüt balık ürünündeki artış, kontrol grubunda 103,22 ve %119,99; yemde 77,30 ve %59,44; gübrede 162,34 ve %175,08 M+F muamelesinde gözlemlenmiştir. Karkas proteini, yağ ve kül içeriği kontrol grubuyla kıyaslandığında her üç muamelede de önemli derecede yüksek çıkmıştır (P<0,05). Kontrollere göre her üç muamele grubunda da yüksek değerlerde farklı sindirim enzimleri kaydedilmiştir.

Anahtar Kelimler: Doğal yem, kümes hayvanı gübresi, yaşam evresi, karkas kompozisyonu, sindirim enzimleri.

Introduction

Fertilization and supplemental feeding are the two important management measures adopted in the semi-intensive system of carp and tilapia culture in Asia. A number of studies focus on the role of fertilizers in fish production (Garg and Bhatnagar, 2000; Dhawan and Kaur, 2002; Das *et al*., 2005; Sayeed *et al*., 2007; Bwala and Omoregie, 2009) and of supplemental feed in systems receiving fertilizers (Aziz *et al.,* 2002; Virk and Saxena, 2003; Ahmed *et al*., 2005; Waidbacher *et al*., 2006; Manjappa *et al*., 2009; Elnady *et al*., 2010). While supplemental feeding affects fish growth directly, fertilization contributes to growth via the planktonic natural food. In addition to acting as a food for fish, plankton perform other important functions in pond aquaculture: a net producer of dissolved oxygen, which is indispensable for fish growth (Teichert-Coddington and Green, 1993) and the most important sink of ammonia-nitrogen, which is excreted by fish (Hargreaves, 1998; Jiménez-Montealegre, 2001). Jhingran (1991) observed that natural food also supplies certain digestive enzymes that improve the utilization of artificial diets. The FAO/AADCP Regional Expert Consultation has emphasized the need for a greater understanding of the role of natural food organisms in semi-intensive farming based on systems that optimize pond fertilization, in order to bring down the cost of fish production (NACA/FAO, 2000).

The best way to reduce the cost of fish production is to minimize the use of supplemental food; this can be best achieved by exploiting the synergetic interaction between natural food and supplemental feed. According to Moav *et al.* (1977), judicious organic manuring of fish ponds can eliminate the need for supplementary feeding. Increase in production by a given regime of supplementary feeding is of great economic importance, but is difficult to predict whether it is related to the amount of natural food available, the density of stocking or the range of other management variables. In this study, an attempt has been made to compartmentalize the effects of feed, manure and their combination in the monoculture of common carp (*Cyprinus carpio*) fry and fingerlings carried out separately, to get an insight into their contribution to fish growth and production.

Materials and Methods

Experimental Design

The two parallel experiments of 98 day duration each were conducted in outdoor cement tanks of 25 m^2 (5x5x1 m), with a soil base. The treatments consisted of control (C), only feed (F), only manure (M), and manure plus feed (M+F) in triplicate. Poultry manure was applied to manure treatment (M and M+F) tanks initially @ 2000 kg/ha and subsequently at fortnightly intervals @ 200 kg/ha. Common carp fry (mean wt. 0.67 g) in experiment one and fingerlings (mean wt. 3.2 g) in experiment two were stocked in the tanks 7 days after manuring, at 4 individuals/m^2. Fish in F and M+F treatments were provided a fish meal-based pelleted feed (Table 1) once daily at 5% of body weight.

The feed was prepared using finely ground ingredients as per composition shown in Table 1. They were mixed thoroughly with water to make a dough. The dough was then transferred to an aluminum container and steam cooked in a pressure cooker at 15 psi for 15 minutes. Vitamin-mineral mixture was mixed after cooling the dough. Pellets (2 mm diameter size) were prepared by a hand pelletizer and were air dried in an oven at 40°C.

Fish were sampled fortnightly to determine weight and length and the feed quantity was re-adjusted based on the weight recorded at each sampling. On termination of the experiment, all the surviving fish were weighed individually and yield calculated.

Water Quality

Analysis of water quality for temperature, pH, dissolved oxygen (DO), free carbon dioxide CO_2), total alkalinity, phosphate, ammonia, nitrate and nitrite was done at weekly intervals, collecting samples from the experimental tanks between $09.^{00}$ and $10.^{00}$ hr. Water temperature was recorded using a digital thermometer, while pH was measured with a digital pH meter (LI-120, ELICO, India). The rest of the parameters were determined following standard procedures (APHA/AWWA/WEF, 1998). In addition, diurnal monitoring of temperature, pH and DO was done by analyzing water samples once in every 3 hours on a monthly basis (30^{th}, 60^{th} and 90^{th} day).

Qualitative and Quantitative Analysis of Plankton

Phytoplankton and zooplankton samples were collected at weekly intervals for qualitative analysis

Table 1. Composition of formulated feed

Ingredient	(%)
Fish meal	30
Groundnut oil cake	35
Rice bran	21
Tapioca flour	13
Vitamin-mineral Mixture*	1

* Supplevite-M (Each 250 g provides, Vitamin A - 500,000 I.U.; Vitamin D_3 - 100,000 I.U.; Vitamin B_2 - 0.2 g; Vitamin E - 75 units; Vitamin K - 0.1 g; Calcium pantothenate - 0.25 g; Nicotinamide - 1.0 g; Vitamin B_{12} - 0.6 mg; Choline chloride - 15 g; Calcium - 75 g; Manganese - 2.75 g; Iodine - 0.1 g; Iron - 0.75 g; Zinc - 1.5 g; Copper - 0.2 g; Cobalt - 0.045 g) supplied by Sarabhai Chemicals, Baroda, India.

by towing 15 µm and 60 µm nets respectively, across each experimental tank. The zooplankton samples were preserved in LUGOL and phytoplankton in 4% formalin. Further, dry weight of plankton was also determined every fortnight by filtering 100 liters of water from each tank through a plankton net of 15 µm size and drying the filtrate in a hot-air oven at 80°C, till a constant weight was obtained. The quantitative estimation of total plankton was done by the "Direct census method" (Jingran *et al*., 1969).

Proximate Composition

Proximate composition of feed ingredients, feed and fish carcass from experiment one was estimated. Carcass was obtained upon harvest, by collecting five fish each from the triplicate tanks and drying at 80°C to a constant weight. The dried carcass of each group was pooled together and ground. Moisture and ash contents were estimated following AOAC (1995) procedures. Crude protein, fat and fibre contents were analyzed using Kjeltec (Tecator, 1002 distilling unit), Soxtec (Tecator, 1043 extraction unit) and Fibretec (Tecator 1017 hot extractor) systems. Carbohydrate content was calculated as nitrogen free extract (NFE) by the difference method of Hastings (1976). The energy value of each ingredient as well as feed was obtained by multiplying protein, lipid and carbohydrate contents by factors 22.6, 38.9 and 17.2 respectively (Mayes, 1990) and expressed in kJ/g.

Digestive Enzyme Activity

Digestive enzyme activity was analyzed from the gut of representative samples of five fish from each treatment, on termination of experiment one. Analysis of the activity of amylase, protease and lipase in the hepatopancreas and intestine was carried out by the methods of Rick and Stegbaurer (1974), Kunitz (1947) and Bier (1962) respectively.

Fish Growth, Survival and Production Calculation

Specific growth rate (SGR), survival (SR) and production were calculated using the following equations.

SGR (%/d) = [(ln final weight – ln initial weight)/ experimental duration in days] ×100

SR (%) = % of live fish number at harvest.

Production (g) = Mean body weight (g) x Total number of viable fish at harvest.

Data Processing and Statistical Analysis

Mean values of fish growth parameters at harvest, carcass proximate composition and digestive enzymes were compared by one-way ANOVA. All plankton and water quality parameters were subjected to two-way ANOVA with treatment and sampling date as factors. When a main effect was significant, pair-wise comparison of treatment means was done by Duncan's multiple range test (P = 0.05) (Duncan, 1955). All analyses were done using the ANOVA procedure of SAS version 6.12 (SAS Institute Inc., Cary NC 27513, USA).

Results

Proximate Composition of Feed Ingredients and Feed

The highest values for protein, fat and ash were recorded in fish meal and the least values for protein and fat in tapioca flour. Rice bran had the highest fiber content, while tapioca flour had the highest level of NFE. The feed contained 28.87% protein, 5.1% fat and 30.54% NFE (Table 2).

Water Quality

Weekly Analyzed Parameters

The values of water quality parameters monitored weekly ranged as follows. Water temperature: 26.96 to 27.16°C, pH: 7. 93 to 8.08, dissolved oxygen: 8.77 to 10.85 mg/L, free carbon dioxide: 2.12 to 3.23 mg/L, total alkalinity ($CaCO_3$): 74.1 to 86.19 mg/L, phosphate: 0.76 to 1.0 µg/L, ammonia: 9.49 to 10.53 µg/L, nitrate: 2.78 to 3.08 µg/L, nitrite: 1.68 to 2.47 µg/L (Table 3). Alkalinity and phosphate contents were significantly ($P<0.05$) higher in M + F treatment. pH, free carbon dioxide, ammonia, nitrate and nitrite values did not differ ($P>0.05$) between the treatments and the control (Table 3). All the water quality parameters showed

Table 2. Proximate composition (%) of ingredients and feed

Parameter	Fish meal	Groundnut cake	Rice bran	Tapioca flour	Feed
Moisture	6.91±0.17	6.58±0.18	8.40±0.07	9.99±0.42	8.61±0.42
Crude protein	51.83±0.55	37.49±0.05	4.92±0.55	2.46±0.10	28.87±0.10
Fat	10.92±0.06	6.79±0.10	1.63±0.06	0.53±0.10	5.10±0.10
Ash	25.79±0.03	6.50±0.39	17.69±0.03	14.68±0.03	14.68±0.03
Crude fibre	1.90±0.15	10.80±0.21	31.80±0.15	3.60±0.01	12.20±0.01
Nitrogen-free extract	2.65	31.84	35.56	81.68	30.54
Gross energy(kJ/g)	16.41	16.59	7.86	14.81	13.76

Table 3. Water quality parameters (mean ± S.E.) (Pooled data of the two experiments)

Treatment	Temp. (°C)	pH	Dissolved Oxygen (mg/L)	Free CO_2 (mg/L)	Alkalinity (mg/L)	Phosphate (µg/L)	Ammonia (µg/L)	Nitrate (µg/L)	Nitrite (µg/L)
Control	27.14±0.29[a]	7.96±0.10[a]	10.09±0.34[ab]	3.23±0.49[a]	74.10±2.17[b]	0.76±0.08[b]	9.97±0.51[a]	2.78±0.21[a]	2.39±0.70[a]
Feed	27.16±0.28[a]	7.99±0.09[a]	8.77±0.46[c]	2.52±0.47[a]	76.57±2.53[b]	0.86±0.08[ab]	10.53±0.42[a]	3.08±0.21[a]	2.47±0.88[a]
Manure	27.16±0.26[a]	8.08±0.07[a]	9.18±0.48[bc]	2.12±0.51[a]	76.36±2.00[b]	0.93±0.08[ab]	9.49±0.44[a]	2.80±0.18[a]	1.68±0.63[a]
Manure ± Feed	26.96±0.28[a]	7.93±0.08[a]	10.85±0.17[a]	2.23±0.53[a]	86.19±2.06[a]	1.00±0.09[a]	9.94±0.41[a]	2.84±0.17[a]	2.24±0.60[a]

Different superscripts for values in the same column indicate significant (P≤0.05) difference.

significant (P<0.05) variation with sampling day. The interaction effect of treatment and day was significant only for CO_2 (P=0.04). DO, pH, alkalinity, nitrite and ammonia were the lowest on the first day of sampling.

Diurnal Water Samples

Figure 1 depicts the diurnal variation in water temperature, pH and DO values in the experimental tanks. Treatments did not affect diurnal water quality. Temperature, pH and DO were the highest at 15 hr, excepting DO on the 30th day, it being the highest at 18 hr. All the 3 parameters showed a declining trend from 12 hr to 9 hr, when the values were the lowest.

Plankton Biomass

Table 4 quantifies the planktonic species encountered in the tank water on the sampling days. Among phytoplankton, chlorophyceae was comprised of 15 genera, the major ones being *Microspora, Volvox* and *Scenedesmus. C*yanophyceae was represented by 3 genera, *Microcystis* and *Anabaena* being dominant. Chrysophyceae, Bacillariophyceae and Dinophyceae were represented by one genus each. *Diaptomus*, *Cyclops*, *Moina* and *Keratella* were the zooplankton species encountered. In addition, nauplii and insect eggs were also found in good numbers.

The overall phytoplankton population was the highest under M+F treatment (5167.28 cells or colonies/L) and lowest in control (2227.21 cells or colonies/L). The number of green algae was lower as compared to blue-green algae in all the tanks (Table 4). The ratio between cyanophyceae and chlorophyceae was the lowest in control and highest in manure+feed (M+F) tanks. Density of phytoplankton and zooplankton (no/L) was also significantly low in control tanks (Table 5). The density of both phyto and zooplankton was the lowest (P<0.05) on the first day of sampling and highest on the 98th day. The interaction effect of treatment and day was significant for *Staurastrum* (P=0.0197) and nauplii (P=0.007).

Dry weight of phytoplankton was highest under feed (F) treatment and lowest in control. The values in manure (M) and M + F treatments were almost similar (Table 5), whereas highest dry weight of zooplankton was observed under M + F (2.15 mg/L) treatment, followed by manure (1.69 mg/L), feed (1.65 mg/L) and control (0.77 mg/L). Significant (P<0.05) variation in both phyto and zooplankton dry weight was recorded over the experimental period, both being the lowest on zero day and highest on 98th day.

Fish Growth, Survival and Production

The final weight and length of fish in experiments one and two are given in Tables 6 and 7. The highest final weight was observed in M+F treatment in both the experiments. Growth was similar in F and M treatments in experiment one, but significantly (P<0.05) different in experiment two. Fish growth was significantly (P<0.05) poor in the control in both the experiments. SGR values followed the trend of fish growth at harvest.

The overall survival ranged from 52.12% in M treatment to 63.03% in M+F treatment in experiment one, while it was in the range of 61.67% (M+F) to 82.78% (M) in experiment two. However, difference in survival among the control and the treatments was not significant (P>0.05) in experiment one. Net production in this experiment varied from 575.13 (control) to 1508.21 g/tank/98 days (M+F treatment). In F and M treatments production was nearly equal, being significantly (P<0.05) higher from the control and lower from M+F treatment (Table 6). In experiment two, net production was higher compared to experiment one (839.40 to 2309.01 g/tank/98 days) and varied significantly between the control and all the treatments (Tables 7).

Proximate Composition

The proximate composition of fish carcass from experiment one is shown in Table 6. Crude protein, fat and ash contents were significantly (P<0.05) higher in the 3 treatments compared to the control. No difference was found between the crude protein content of F and M treatments and fat content of F, M and M+F treatments.

Digestive Enzyme Activity

Activity of digestive enzymes was higher in fish from the three treatments compared to the control, both in the hepatopancreas and intestine as observed

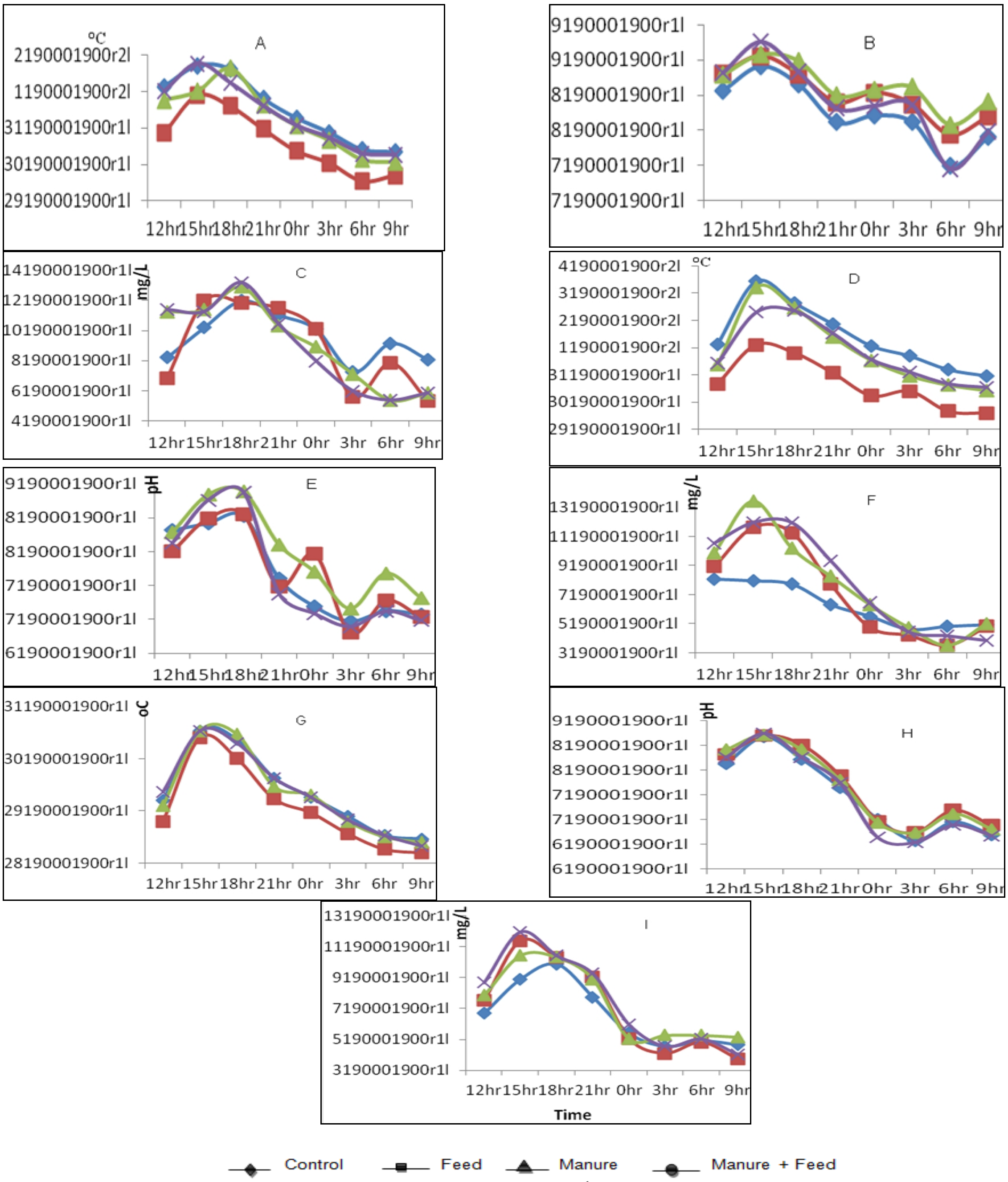

Figure 1. Diurnal variation in water temperature, pH and DO on 30th (A,B and C), 60th (D,E and F) and 90th day (G,H and I respectively) in the experimental tanks (Pooled data of the two experiments).

in experiment one. Highest amylase and lipase activities were recorded in the hepatopancreas and intestine of fish from M+F treatment, while maximum protease activity was found in the intestine of fish from F treatment (Table 8).

Discussion

Water Quality

Average values of water temperature and pH in the different treatments were similar during the experimental period and varied within a narrow range. pH was in the alkaline range throughout the experimental duration, indicating favourable conditions for biological production. Jhingran (1991) observed that carps thrive well in the temperature range of 18.3°C to 37.8°C. According to Farmanfarmain and Moore (1979), aquatic organisms can tolerate a wider range of temperatures, provided that fluctuations are not severe, sudden and of long duration. DO was high throughout the experimental duration and fluctuated between 8.77 (F) and 10.85 mg/L (M+F treatment); this reflects higher photosynthetic activity in manure plus feed treatment. Dissolved oxygen levels improve due to

Table 4. Abundance of phytoplankton species in tank water (cells/colonies/L) during the experimental period.* (Pooled data of the two experiments)

Group/Genus	Control		Feed		Manure		Manure±Feed	
PHYTOPLANKTON								
Chlorophyceae								
Ankistrodesmus	15.09	3.10	11.79	2.77	16.74	4.03	16.65	5.98
Chlorococcum	10.13	2.07	17.54	5.92	12.90	2.52	31.16	15.55
Closterium	3.39	0.53	2.32	0.61	2.72	0.53	0.45	0.18
Gleocapsa	0.04	0.04	1.12	0.46	0.40	0.15	0	0
Golenkinia	3.35	0.98	7.14	2.12	11.52	6.51	3.08	0.93
Hydrodiction	0	0	0.36	0.16	0.13	0.10	0.27	0.10
Menoidium	0	0	0	0	0.40	0.21	0.54	0.33
Microspora	148.68	203.28	330.67	95.78	226.88	57.55	484.64	109.31
Pediastrum	7.01	1.01	24.20	5.82	3.17	0.61	10.31	3.13
Phytoconis	21.12	5.34	21.12	4.97	9.38	2.56	22.99	3.40
Scenedesmus	59.20	24.73	37.81	9.83	75.85	20.18	71.92	24.60
Selenastrum	67.41	18.72	25.94	4.61	78.08	28.80	44.82	10.42
Staurastrum	22.95	26.07	32.37	4.53	28.44	4.10	60.00	26.70
Tetraedron	35.22	10.47	18.84	7.37	7.05	2.34	9.46	1.44
Volvox	89.87	23.83	14.64	2.46	25.54	7.11	49.20	15.95
Cyanophyceae								
Anabaena	541.61	205.71	1654.73	366.36	1550.27	325.36	2213.26	314.90
Merismopedia	92.14	87.44	1.43	0.33	5.40	1.31	3.30	0.85
Microcystis	1099.33	256.83	2117.72	317.21	2155.40	360.98	2139.46	340.12
Chrysophyceae								
Chrysophyxis	3.66	1.16	2.01	0.43	1.47	0.34	1.38	0.35
Bacillariophyceae								
Anomoeoneis	5.71	1.58	10.18	3.26	3.13	0.62	3.17	0.68
Dinophyceae								
Monomastix	1.29	0.38	0.45	0.18	2.28	0.61	1.21	0.31
Total Phytoplankton	2227.21	513.44	4332.37	472.86	4217.14	628.81	5167.28	530.43
ZOOPLANKTON								
Diaptomus	40.18	6.26	52.99	12.06	77.23	15.56	153.70	87.27
Cyclops	23.44	3.98	19.24	2.45	44.46	9.81	23.30	3.62
Moina	13.66	2.98	7.54	1.27	17.37	8.53	12.86	2.16
Keratella	95.49	48.01	287.54	159.98	39.06	12.98	456.25	220.85
Insect eggs	34.37	4.40	48.70	8.61	74.06	16.02	55.80	12.32
Nauplius	106.70	73.96	28.17	8.57	386.61	248.60	123.79	83.01
Total Zooplankton	313.84	90.71	444.20	165.22	638.79	257.65	825.71	258.11

*Numbers are means of 14 samplings. Numbers in italics are standard errors.

Table 5. Plankton biomass and density (± S.E.) in experimental tanks (Pooled data of the two experiments)

Treatment	Dry weight (mg/L)		Density (no/L)	
	Phytoplankton	Zooplankton	Phytoplankton	Zooplankton
Control	0.77±0.14^{b}	0.77±0.24^{c}	2227.21±513.44^{b}	313.84±90.71^{a}
Feed	1.98±0.27^{a}	1.65±0.15^{b}	4332.37±472.86^{a}	444.20±165.22^{a}
Manure	1.73±0.24^{a}	1.69±0.17^{b}	4217.14±628.81^{a}	638.79±257.66^{a}
Manure±Feed	1.70±0.25^{a}	2.15±0.24^{a}	5167.28±530.43^{a}	825.71±258.11^{a}

Different superscripts for values in the same column indicate significant ($P \leq 0.05$) difference.

photosynthesis, while ammonia levels are reduced through assimilation by phytoplankton (Boyd, 1990). Generally, cyprinids are capable of tolerating low oxygen levels of 3 mg/L (Huet, 1972). Highest total alkalinity was recorded in the M+F treatment (86.19mg/L) and the lowest in control (74.1 mg/L). A productive pond is expected to have total alkalinity range of 75-100 ppm (Sinha *et al*., 1985). Total alkalinity was significantly greater where organic fertilization and feeds were applied to ponds (Kumar *et al*., 2005). Alkalinity increases with organic fertilization because bacterially generated CO_2 from manure decomposition dissolves calcium and magnesium carbonate in pond water into calcium and magnesium bicarbonate (Boyd, 1990).

Higher concentrations of ammonia nitrogen are often noticed in fish culture ponds (Edwards, 2008). However, the values of ammonia recorded in the present experiments were low (Table 5). Sugiyama and Kawai (1978) reported that higher concentration

Table 6. Growth parameters (average ± S.E.) of common carp fry under different treatments (Experiment one)

	Control	Feed	Manure	Manure±Feed
Final weight (g)	18.16±1.15[b]	35.86±0.22[a]	36.84±4.50[a]	43.43±2.02[a]
Increment in growth over control (%)	_	97.46	102.86	139.15
Final length (cm)	11.41±0.7[b]	14.05±0.20[a]	12.78±0.50[ab]	13.17±0.10[a]
SGR (%)	2.75±0.09[b]	3.32±0.02[a]	3.32±0.18[a]	3.47±0.07[a]
Survival (%)	57.58±7.57[a]	61.20±14.2[a]	52.12±12.76[a]	63.03±2.09[a]
Production (g/tank/4 months)	575.13±105.72[c]	1168.80±91.85[b]	1019.68±38.95[b]	1508.21±173.35[a]
Increment in production over control (%)	_	103.22	77.30	162.24
Proximate composition of fish (%wet weight)				
Moisture	73.50±2.60[a]	70.19±2.03[a]	68.90±2.99[a]	70.72±1.71[a]
Crude protein	17.21±0.08[c]	19.20±0.05[b]	19.27±0.06[b]	21.60±0.07[a]
Fat	1.08±0.06[b]	1.35±0.02[a]	1.31±0.11[a]	1.36±0.04[a]
Ash	5.78±0.02[c]	6.16±0.01[b]	6.54±0.01[b]	6.13±0.03[a]

Initial weight and length of fry were 0.67±0.06 g and 2.32±0.11 cm respectively.

Table 7. Growth parameters (average ± S.E.) of common carp fingerlings under different treatments (Experiment two)

	Control	Feed	Manure	Manure+Feed
Final weight (g)	19.78±0.82[d]	49.87±1.62[b]	26.94±1.48[c]	56.33±2.75[a]
Increment in growth over control (%)	_	152.12	36.20	184.78
Final length (cm)	9.50+1.14[c]	13.70±1.01[a]	12.25±0.50[b]	14.07±0.81[a]
SGR (%)	1.52±0.03[b]	2.29±0.03[a]	1.77±0.04[b]	2.62±0.27[a]
Survival (%)	70.55±2.94[ab]	68.89±6.83[b]	82.78±0.55[a]	61.67±2.55[b]
Production(g/tank/months)	839.40±63.85[d]	1846.67±107.22[b]	1338.37±70.33[c]	2309.01±141.09[a]
Increment in production over control (%)	_	119.99	59.44	175.08

[1]Initial weight and length of fingerlings were 3.20±0.41 g and 5.22±0.21cm respectively.

Table 8. Gut digestive enzyme activity (± S.E.) in common carp under different treatments (Experiment one)

Treatment	Amylase		Lipase		Protease	
	Intestinal	Hepatop.	Intestinal	Hepatop.	Intestinal	Hepatop.
Control	4.71±0.01[b]	0.50±0[b]	33.67±0.02[c]	30.40±0.02[c]	1.50±0[b]	1.10±0.02[b]
Feed	7.07±0.02[a]	0.85±0.02[a]	42.51±0.09[b]	45.88±0.14[b]	1.53±0.01[b]	1.28±0[a]
Manure	6.51±0.04[b]	0.52±0.01[b]	40.40±0.03[b]	39.77±0.02[b]	1.79±0.01[ab]	1.15±0[b]
Manure±Feed	6.33±0.01[b]	0.58±0.01[b]	47.65±0.02[a]	60.82±0.02[a]	2.17±0.01[a]	1.25±0[a]

Hepatop. = hepatopancreatic
Enzyme activity is expressed in µ moles of product liberated per minute per mg of tissue protein at 28 °C.
Values with the same superscript in each column are not significantly different (P>0.05).

of dissolved oxygen decreases ammonia level through oxidation. Phosphorus was significantly higher ($P<0.05$) in M+F treatment in comparison with the control (Table 3). The higher phosphorus concentration may be associated with the increase in phosphorus produced during the decomposition of organic fertilizer and also from the feed through fish excreta. Both soluble organic phosphorus and orthophosphate are released during the process of organic fertilizer decomposition under aerobic conditions (Wudtisn and Boyd, 2005).

Values of water temperature, pH and DO in the diurnal samples showed no effect of treatments. The increase in the values of these parameters with the progress of day and decrease with the progress of night can be related to the presence and absence of light which affects temperature and also dissolution of oxygen in pond water. Further, photosynthesis during day time is responsible for the higher DO values, whereas consumption of DO by plankton reduced night time DO. Similarly, pH variations can also be correlated with photosynthetic activity.

Plankton Biomass

Significant ($P<0.05$) variation in both phytoplankton and zooplankton dry weight was recorded with respect to study period. The interaction effect of treatment and day was also significant for both phytoplankton and zooplankton. The number of green algae was lower as compared to blue-green algae in all the experimental tanks. In fish ponds, blue green algae constitute greater part of phytoplankton; higher alkalinity, nitrate, ammonia and phosphate favour the multiplication of cyanophyceae (Padmavathi and Veeraiah, 2009). Kulkarni (1992), who studied the effect of distillery waste on plankton and fish production, reported a significant ($P<0.05$) correlation between phosphorus level and blue green algae production. Rahman *et al.* (2008) reported that common carp increased bio-available N and P in the water column and plankton availability was positively correlated with bio-available N and P. The relationship between provision of manure/feed and plankton biomass observed in the present study can be related to the nutrient input. In addition, fish excreta would have contributed to the level of N and P in tank water, particularly towards the later part of the experiment.

Fish Growth, Survival and Production

Feeding and manuring, individually and in combination, improved the growth of fish significantly ($P<0.05$) in both the experiments (Tables 6 and 7); highest final weight was recorded in M+F treatment. Specific growth rate followed the growth trend in both the experiments. While there was no difference in growth of fish between feed (F) and manure (M) treatments in experiment one, it differed significantly ($P<0.05$) in experiment two. Growth under F, M and M + F treatment in experiment one works out to 97.46%, 102.86%, and 139.15% higher respectively over the control. The corresponding values in experiment two are 152%, 36.20% and 184.78%. It is clear that fed treatments (F and M+F) had greater impact on the growth of fingerlings as compared to fry; fingerlings are better equipped in terms of mouth size and digestive enzymes to accept and utilize pelleted diet. Further, a comparison of the final weights of fish from experiment one with that of experiment two, points out to the difference in growth rate due to life stage. The increase in weight of control fish is only 1.62 g, whereas under manure treatment (M), there is a reduction of 9.9 g. As against this, in fed treatments there is an increase of 14.01g (F) and 12.9 g (M+F) (Tables 6 and 7). This shows that the nutrient requirement of fingerlings is not satiated by natural food alone, contrary to that in the case of fry. Boyd (1990) reported a strong positive correlation between fish growth and primary productivity in fertilized ponds without supplementary feeding. Natural food is nutritive and contains 51.1% protein, 27.3% carbohydrate and 7.7% fat, while the calorific value ranges from 6.7 to 23.8 kJ/g (De Silva and Anderson, 1995). It is possible that as the fish grows bigger, it prefers artificial diet when available. Rahman *et al.* (2008) observed that common carp growth, in polyculture with rohu, *Labeo rohita*, was higher in the presence of artificial feed and negatively correlated with natural food availability. They also recorded higher ingestion of benthic macroinvertebrates, copepods and rotifers, and a lower ingestion of phytoplankton by common carp.

In the tanks receiving no supplementary feed, the growth obtained is entirely attributable to the natural food. This applies to the control as well as manure (M) treatment. In experiment one, growth of fish in M treatment was double that of control, reflecting the ability of common carp fry to extensively feed on the available natural food and convert it into flesh. Though the control tanks did not receive any nutrient input during the experiment, the soil bottom of all the tanks used in the present study contained some nutrients accumulated from earlier trials; the effect of these could be considered as equal under all treatments. Common carp as a bottom feeding fish enhances the availability of nutrients to phytoplankton through stirring of the mud bottom (Milstein et al., 2002). Ritvo et al. (2004) demonstrated that common carp by perturbations results in appreciable mixing of the sediment; this mixing would bring out nutrients into circulation, facilitating natural food production.

The difference in survival of fish in the control and treatment tanks was not significant ($P>0.05$) in the first experiment, whereas fed treatments (F and M+F) recorded lower survival ($P<0.05$) in the second experiment. This could be due to some natural mortality of fish in tanks of the two treatments, since water quality was similar in all the treatments, but for higher alkalinity and phosphate levels in M+F treatment. Gross fish production was influenced both by fish weight and survival. Production was the highest in M+F treatment in both the experiments. In experiment one, production was comparable ($P>0.05$) in F and M treatments. The increment in gross fish production over the control was 103.22% in F, 77.30% in M and 162.34% in M+F treatments (Table 6). In experiment two, production was significantly ($P<0.05$) higher in the feed treatment (F) compared to the manure (M) treatment, again indicating the significance of feeding in the case of fingerlings. The corresponding figures of increment for experiment two are 119.99%, 59.44% and 175.08% (Table 7). Abbas et al. (2010) reported highest gross production of carps in the treatment with the combination of organic and inorganic fertilizers and supplementary feeding, compared to combinations of any two of these.

Proximate Composition

Proximate analysis of fish carcass revealed that treatments affected crude protein and fat, both being lowest in control and highest in M + F treatment. However, there was no difference in moisture level among the treatments and control (Table 6). This is indicative of protein accretion and true growth involving an increase in the structural tissue such as muscle and various organs (Fafioye *et al*., 2005). The type of feed ingested and their nutritional quality is known to be one of the main factors affecting fish carcass composition (Reinitz and Hitzel, 1980).

Digestive Enzyme Activity

Higher activity of the different digestive enzymes was recorded in fish from the three treatments compared to the control fish (Table 8). Factors like feeding habit and diet affect the activity of digestive enzymes in fish (Hofer, 1979; Gangadhara *et al*., 1997; Manjappa *et al*., 2009). The lower digestive enzyme activity in control fish can be attributed to the lower food availability.

The results obtained in this study clearly point out to the importance of natural food in fish culture. Growth of fish in experiment one indicates similar potential of poultry manure and the feed provided in inducing growth of common carp fry. In contrast, fish growth in experiment two was significantly better under fed treatments (F and M+F). This shows that nutritional requirement of common carp fingerlings is not fully met by natural food alone, contrary to that of fry. The findings can be used in developing a feeding strategy for fish at different life stages during culture.

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