

Influence of Feeding Frequency on Growth and Body Composition of *Clarias gariepinus* Burch.

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Abstract: A 6-week growth trial was conducted to examine the influence of feeding rate on growth and body composition in young African catfish, *Clarias gariepinus* (16-18 cm; 14.98-15.81 g). Fish were stocked in indoor 70 L polyvinyl flow-through (1-1.5 L/min) circular tanks and fed experimental diet at different feeding frequencies (two, three and four times a day). Diets were fed in the form of moist cake. Maximum weight increment (0.49 g fish⁻¹ day⁻¹) and live weight gain (132.42%) were obtained in fish fed three times a day. Feeding frequency significantly ($P < 0.05$) influenced the body composition of fish. Whole body protein and fat contents were significantly ($P < 0.05$) higher in fish groups fed three times a day. Starved fish exhibited lowest protein and fat and highest moisture and ash contents in their carcass. It is evident from the results that three feedings a day is optimum for young *C. gariepinus*.

Key words: Body composition; catfish; *Clarias gariepinus*; feeding frequency; growth.

1. Introduction

Frequency of feeding affects fish growth by regulating the availability of diet. Increased feeding frequency has been reported to improve growth in various fish species (Kono and Nose, 1971; Andrews and Page, 1975; Garcia-Galano et al., 2003). Two or three feedings a day have been found sufficient for maximum growth in species such as channel catfish, *Ictalurus punctatus* (Andrews and Page, 1975), rainbow trout, *Salmo gairdneri* (Grayton and Beamish, 1977), and sea bass, *Dicentrarchus labrax* (Tsevis et al., 1992).

Optimum feeding frequency is influenced by various factors such as size and stocking density of fish, type of feed, feeding levels, feed utilization and water quality, etc., in various species (Murai and Andrews, 1976; Machiels and Henken, 1985; Hossain et al., 2001). Feeding frequency and its effect on fish has been studied by various workers in different species like Chinese catfish, *Clarias fuscus* (Buurma and Diana, 1994), channel catfish, *I. punctatus* (Jarboe and Grant, 1996), Korean rockfish, *Sebastes schlegeli* (Lee et al., 2000) and walking catfish, *C. macrocephalus* (Petkam and Moodie, 2001).

The present study was conducted to observe the influence of feeding frequency on growth and body composition of *C. gariepinus*.

2. Materials and Methods

2.1 Source of fish stock/acclimation

Young *Clarias gariepinus*, obtained from local market, were transported to the laboratory in oxygen filled polythene bags, given a prophylactic dip in KMnO₄ solution (1:3000), and stocked in cement cisterns (1m x 1m x 1m) for acclimation. During the period of acclimation, fish were fed with minced meat twice daily. After a fortnight, desired number of fish were taken out and gradually acclimated to casein – gelatin based semi-purified experimental diet (Table 1), over a two week period in high density polyvinyl indoor circular tanks (water volume, 55 L), fitted with flow-through system (1-1.5 L/min). Unconsumed diet, if any, was collected, dried and weighed to estimate the amount of diet consumed by the fish.

2.2 Preparation of experimental diet

Casein-gelatin based semi-purified test diet was prepared by calculating quantities of dietary ingredients on sensitive electronic balance (Precisa-120A). Gelatin was mixed in water in a stainless steel attachment of Hobart electronic mixer, with constant stirring and heated to 80 °C. The mixer bowl was removed from heating, and quantities of casein, dextrin, minerals and α -cellulose were added to it,

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Table 1. Ingredients and proximate composition of experimental diet.

Ingredients	g/100 g (as fed)
Casein (vitamin-free; 84.6% C.P.)*	40.17
Gelatin (87.0% C.P.)*	12.84
Dextrin	25.71
α - cellulose	7.28
Oil mix (2:1 corn and cod liver oil)	8.00
Vitamin mix	1.00
Mineral mix	3.00
Carboxymethyl cellulose	2.00
Proximate composition (%)**	
Crude protein	45.00
Crude fat	8.00
Carbohydrate	26.00
Energy (kcal/100g)	360.50

* Loba Chemie, India and ** Hina (2001)

Table 2. Composition of mineral mixture (Halver, 1989)

Mineral	g/100 g diet
Calcium biphosphate	13.48
Calcium lactate	32.40
Ferric citrate	02.97
Magnesium sulphate	13.70
Potassium phosphate (dibasic)	23.86
Sodium biphosphate	08.72
Sodium chloride	04.35
Aluminium chloride. 6H ₂ O	00.015
Potassium iodide	00.015
Cuprous chloride	00.010
Magnesium sulphate. H ₂ O	00.080
Cobalt chloride. 6H ₂ O	00.100
Zinc sulphate. 7H ₂ O	00.300

and the content blended in the mixer while still in lukewarm state. This was followed by the addition of vitamin mix and oil (2:1 corn and cod liver oil). After blending the mixture, carboxymethyl cellulose was added to it. The prepared diet, upon obtaining a bread dough-like consistency was poured into Teflon-coated pan and placed in a refrigerator to jell. The prepared diet was in the form of moist cake, which was cut into small cubes and stored in refrigerator (-20 °C) in sealed polythene packs until used. The mineral (Table 2) and vitamin premixes (Table 3) were the same as used by Halver (1989).

Table 3. Composition of vitamin mixture (Halver, 1989) and incorporated with oil* (Hina, 2001).

Vitamin	g/100g diet
α - cellulose	8.000
Chloine chloride	0.500
Inositol	0.200
Ascorbic acid	0.100
Niacin	0.075
Calcium pantothenate	0.050
Riboflavin	0.020
Menadione	0.004
Pyridoxine. HCl	0.005
Thiamin. HCl	0.005
Folic acid	0.0015
Biotin	0.0005
α - tocopherol acetate*	0.040
Vitamin B-12 (10mg/500ml H ₂ O)	0.00001 (0.5ml)

2.3 Experimental design/feeding trial

Fish of desired size and number were sorted out from the acclimated lot and stocked in 70 L polyvinyl circular tanks (water volume 55 L), in triplicate. The tanks were supplied with ground water. The water exchange rate in each tank was maintained at 1-1.5 L/min. Prior to feeding faecal matter was siphoned off from the experimental tanks. Feeding level and schedule was chosen after carefully observing the dietary intake as well as feeding behaviour of the fish. The moisture content in the diet was estimated, and the ration level calculated as dry feed to wet fish weight. Mass weight of fish was taken weekly and amount of ration recalculated for subsequent feeding. On the day of weekly measurements, no feed was offered to the fish and the tanks were thoroughly washed with KMnO₄ solution (1:3000). During the trial, the water temperature ranged between 22 ± 2°C.

2.4 Proximate analysis

Proximate composition of fish was analysed using standard techniques (AOAC, 1995). The analysis was carried out in triplicate.

2.5 Estimation of moisture

A weighed quantity of finely ground/homogenized sample was taken in a pre-weighed silica crucible and placed in an oven (100° C) for 24 hr. The crucible containing the dry sample was transferred to the desiccator, allowed to cool and reweighed. This process was repeated till a constant weight obtained.

The loss in weight was expressed as per cent of moisture.

2.6 Estimation of ash

A known quantity of finely powdered sample was taken in a pre-weighed silica crucible and incinerated in a muffle furnace (600 °C) for 3-4hrs till the sample became free of carbon. The crucible containing the incinerated sample was transferred to a desiccator, cooled and reweighed. The quantity of ash was calculated and expressed in percentage.

2.7 Estimation of crude fat

For estimating the crude fat, continuous Soxhlet extraction technique was employed. Petroleum ether (40-60 °C B.P.) was used as a solvent. A weighed quantity of finely ground sample was taken in Whatman fat extraction thimble, cotton plugged, and introduced into the Soxhlet apparatus. A clean dry receiver flask was weighed and fitted to the Soxhlet assembly on a water bath for extraction. Extraction was carried out for 10-12hrs. Thereafter, the receiving flask was removed and kept in a hot air oven (100 °C) to evaporate the solvent traces. The flask was then cooled in a desiccator and reweighed. The amount of fat extracted was expressed in percentage.

2.8 Estimation of crude protein

The technique employed for estimating the crude protein was based on a slight modification of Wong's micro-Kjeldahl method (Jafri et al., 1964). The principle involved digesting a known amount of sample in N-free sulphuric acid, in presence of potassium persulphate used as catalyst, which converts the nitrogenous compounds to ammonium sulphate. This was then treated with Nessler's reagent. The colour developed due to the formation of $(OH)_2NH_2$ was measured by spectrophotometer. The optical density obtained was read off against a standard calibration curve of $(NH_4)_2SO_4$ for nitrogen estimation. To calculate crude protein in the sample, the amount of nitrogen was multiplied with the conventional protein factor (6.25).

0.1g dry powdered sample was taken in a Kjeldahl flask with 5ml of N-free sulphuric acid (1:1), and 5ml potassium persulphate added to it. The volume was raised to 3ml with distilled water. The solution was then nesslerized using Bock and Benedict's Nessler reagent (Oser, 1979), kept at room temperature for 10 min for complete colour development. A blank was prepared in the same manner using distilled water in place of aliquot. The amount of nitrogen was obtained by reading the optical density against the standard calibration curve. The nitrogen value was multiplied with 6.25 to obtain the amount of crude protein. The spectrophotometric measurements were made on microprocessor –

controlled split beam spectronic 1001 spectrophotometer (Milton Roy Company, USA).

2.9 Assessment of metabolizable energy

Metabolizable energy of the diets was calculated using physiological fuel values 3.5, 4.5 and 8.5 kcal/g for carbohydrate, protein and lipid, respectively (Jauncey, 1982).

2.10 Assessment of growth and conversion efficiencies

Calculation of growth parameters and conversion efficiencies were made according to standard definitions (Hardy, 1989; Hanley, 1991).

$$\text{Live weight gain (\%)} = \frac{W_2 - W_1}{W_1} \times 100$$

$$\text{Specific growth rate (\%)} = \frac{\log_e W_2 - \log_e W_1}{D} \times 100$$

Where, W_1 = Initial mass weight (g); W_2 = Final mass weight (g) and D = Duration of the feeding trial (days)

$$\text{Feed conversion ratio} = \frac{\text{Total feed intake (g)}}{\text{Live weight gain (g)}}$$

$$\text{Protein efficiency ratio} = \frac{\text{Live weight gain (g)}}{\text{Total protein intake (g)}}$$

2.11 Statistical analysis

Data were subjected to one-way analysis of variance (ANOVA) followed by Duncan's multiple range test at 0.05% significance level (Duncan, 1955; Sokal and Rohlf, 1981).

3. Results

Results on growth of *C. garipinus* at varying feeding frequencies are given in Table 4. Weight increment ($g\ fish^{-1}\ day^{-1}$), live weight gain (%) and specific growth rate (SGR%) in fish groups fed three times a day were significantly ($P < 0.05$) higher than those starved or fed two / four times a day as also evident from Fig. 1 and 2. The lowest ($P < 0.05$) values for these parameters were noted in starved fish.

Proximate composition of fish fed at varying feeding frequencies is given in Table 5. Significantly ($P < 0.05$) higher carcass protein was obtained in fish fed three times a day. However, fish fed two and four times a day contained comparable ($P > 0.05$) protein content. Starved fish showed lowest carcass protein. Carcass fat was insignificantly ($P > 0.05$) different in fish fed two, three and four times a day. Starved fish exhibited lowest ($P < 0.05$) fat content. Moisture content was highest in starved fish, while it varied insignificantly ($P > 0.05$) in fish fed at different feeding frequencies. Ash content was significantly ($P < 0.05$) higher in starved fish.

Table 4. Growth of *C. gariepinus* fed at varying feeding frequencies

No. of feeding (day ⁻¹)	Total diet fed (g ⁻¹ fish ⁻¹)	Initial average weight (g)	Final average weight (g)	Weight increment (g fish ⁻¹ day ⁻¹)	Live weight gain (%)	Specific growth rate (%)
Starved fish	- ± 0.00	14.98 ± 0.00	10.22 ± 0.00	- 0.11 ^c ± 0.00	- 31.77 ^d ± 0.00	- 0.91 ^c ± 0.00
2 times	28.30 ^a ± 0.04	15.81 ± 0.41	34.87 ± 1.05	0.45 ^b ± 0.02	120.51 ^b ± 0.83	1.88 ^b ± 0.01
3 times	26.73 ^{ab} ± 0.02	15.55 ± 0.33	36.14 ± 0.06	0.49 ^a ± 0.00	132.42 ^a ± 0.64	2.00 ^a ± 0.00
4 times	25.69 ^b ± 0.02	15.52 ± 0.50	33.61 ± 0.62	0.43 ^b ± 0.01	116.55 ^c ± 0.85	1.83 ^b ± 0.03

Results are mean ± SE of triplicate runs; values in each column with same superscript are insignificantly ($P > 0.05$) different.

Table 5. Carcass composition of *C. gariepinus* fed at varying feeding frequencies

Number of feeding (day ⁻¹)	Moisture	Crude protein*	Crude fat*	Ash*
Starved fish	78.43 ^a ± 0.59	70.72 ^c ± 0.03	11.30 ^b ± 0.62	9.31 ^a ± 0.41
2 times	75.46 ^b ± 0.65	72.38 ^b ± 0.07	13.90 ^a ± 0.28	8.59 ^{ab} ± 0.51
3 times	75.11 ^b ± 0.29	72.63 ^a ± 0.07	14.82 ^a ± 0.52	8.26 ^b ± 0.27
4 times	74.32 ^b ± 0.69	72.34 ^b ± 0.03	14.12 ^a ± 0.02	8.10 ^b ± 0.27

*Dry-weight basis; results are mean ± SE of triplicate fish groups; values in each column are insignificantly ($P > 0.05$) different.

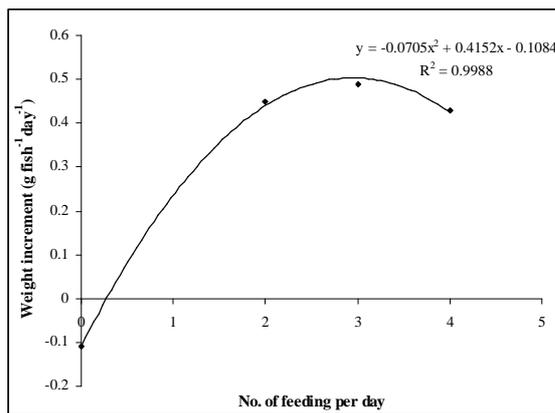


Figure 1. Second degree polynomial relation of weight increment (g fish⁻¹day⁻¹) and varying daily feeding frequency in *C. gariepinus*.

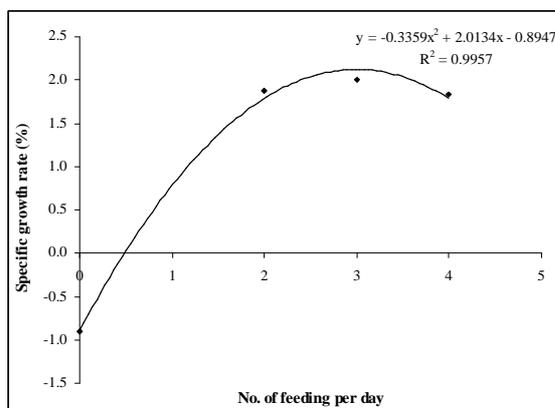


Figure 2. Second degree polynomial relation of specific growth rate (%) and varying daily feeding frequency in *C. gariepinus*.

4. Discussion

C. gariepinus fed three times a day produced best growth. Negative growth was observed in starved fish. The results seem in agreement with the observations made on rainbow trout, *Oncorhynchus mykiss* (Ruohonen et al., 1998). Grayton and Beamish (1977) reported that one or two feedings a day was not sufficient for maximum growth of rainbow trout, *Salmo gairdneri*. In *Catla catla* fingerlings, best growth was noted at a feeding frequency of three times a day (Patra et al., 2001). Increased feeding frequency resulted in better growth in *C. lazera* (Hogendoorn, 1981) and *C. fuscus* (Buurma and Diana, 1994). Feeding frequency for maximum growth of fish may differ with species, fish size, dietary protein and energy content, etc. (Wang et al., 1998). Fixed feeding frequency can greatly reduce wastage of feed and increase the growth rate of fish for a particular stage. Young *C. gariepinus* fed three times a day produced best SGR (%) values. Reports are also available on other species where feeding time markedly affected growth performance, and feeding, tailored to the feeding rhythm, consistently gave better results (Baras et al., 1995; Boujard et al., 1995).

In several fish species studied, increased growth with increasing feeding frequency has been attributed to increased food consumption rather than increased feed assimilation (Grayton and Beamish, 1977; Charles et al., 1984). It has been suggested that this increase is due to gastric emptying taking place before the next meal (Vahl, 1979).

In the present study, maximum carcass protein was obtained in fish fed three times a day. In *C.*

gariepinus, carcass fat increased with feeding levels but declined at a feeding frequency of four times a day. Various workers have reported increased body lipid content in fish with increase in feeding frequency (Grayton and Beamish, 1977; Kayno et al., 1993; Lee et al., 1996). Frequent feeding generally improved fish growth due to increased feed intake that elevated lipid deposition and changed body composition (Grayton and Beamish, 1977). The results revealed starved *C. gariepinus* contained higher moisture and lower fat and protein contents in their carcass. Hogendoorn (1983) found a similar trend in *C. lazera*, and attributed it to catabolism of body fat and protein for energy conversion during starvation.

It may be concluded that feeding young *C. gariepinus* three times a day produced best weight increment ($\text{g fish}^{-1} \text{day}^{-1}$) and SGR (%). Maximum protein and fat, and low ash contents were also obtained in fish receiving three feedings a day.

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