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Effect of starvation on the proximate composition of freshwater fish *Anabas testudineus* under laboratory condition

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Abstract
The present experiment was conducted to study the impact of feed starvation in fish muscle of *Anabas testudineus* fingerling for a period of 15th weeks from the 4th December, 2016 to 18th March, 2017. The initial values of moisture, protein, lipid and ash were 80.73%, 14.10%, 2.05% and 1.20% in, D1, D2, D3 and control respectively. After termination of the experiment the corresponding moisture values were 73.42%, 73.52%, 74.43% and 74.87%, 73.33% in D1, D2, D3, D4 and control, respectively. Crude protein levels were 19.06%, 18.88%, 18.72%, 18.12%, and 19.25% in, D1, D2, D3 and D4, control respectively. The corresponding lipid contents of muscle were 2.64%, 2.56%, 2.03%, 1.90% and 2.86%. The corresponding total ash values were 2.42, 2.34, 2.08, 2.04, and 2.61 in D1, D2, and D3and D4 control respectively. There was no significant difference in moisture content and protein content among all the treatments during the period of investigation. There was significant difference in crude lipid content among D3 ($P<0.05$) and D4 ($P<0.05$) treatments during the period of experiment. In the ash content there was no significant difference between control and D1as well as control and D2. However, there was significant difference in D3 ($P<0.05$) and D4 ($P<0.05$) compared to control during the period of experiment.

Keywords: Feed starvation, *Anabas testudineus* fingerling, moisture, protein, lipid and ash content

1. Introduction
Feeding schedule involving starvation, re-feeding cycles could be a promising feed management option in aquaculture (Azodi *et al.*, 2016) [4]. A period of starvation may arrest the growth of animals under natural and aquaculture conditions and may even cause negative growth. However, after removal of starvation stress, individuals often undergo a period of rapid growth, a phenomenon known as compensatory growth. Similarly, compensatory population growth can be defined as the recovery of animal populations by an increase in population growth rate that exceeds the normal rate at re-feeding after a period of starvation (Marimuthu *et al.*, 2010) [16]. In captive condition fish species undergo periods of starvation because of solving water quality problems, reduce handling stress and mortality due to disease outbreak, temperature changes and also for saving feed to increase farm profit (Caruso *et al.*, 2011) [5]. Food nutrients include water, carbohydrates, proteins, fats, vitamins and minerals amongst others. Fishes are known to provide protein, fat and vitamins which are of great benefit to human health (Johansen *et al.*, 2010) [13]. Thai climbing perch (*Anabas testudineus*, Thai variety) is indigenous to Thailand. This species with accessory respiratory organ can be farmed at high stocking density and in relatively poor water quality. 'Thai variety *A. testudineus* was first brought into Bangladesh by Talukdar farm at Rupganj for commercial aquaculture, it was first introduced in Bangladesh from Thailand in 2002 (Kohinoor *et al.*, 2012) [14], which is recognized as a distinct strain of *A. testudineus*. It contains easily digestible protein, fat of very low melting point and many essential amino acids making them an ideal nutritious food. Its muscle protein content is 19.50%. It contains physiologically available iron and copper essentially needed for haemoglobin synthesis (Saha, 1971) [18]. This species is considered as a valuable item of diet for sick and convalescent. The fish contains high values of physiologically available iron and copper essentially needed for hemoglobin synthesis. Some authors have reported on the proximate composition of fish species (Gokoglu *et al.*, 2004; Fawole *et al.*, 2007; Turkkan *et al.*, 2008; Weber *et al.*, 2007) [10, 9, 20, 23].

However, information on proximate composition of some fish species under feed restriction is scanty. Thus, there is still a need to research on the proximate composition of such fish species. The aim of this study was to determine the moisture, protein, fat, and ash contents of freshwater fish *Anabas testudineus* under laboratory condition which is readily available and consumed in West Bengal.

2. Materials and Methods

2.1 Site of Experiment

The present study was conducted in the Department of Fisheries Resource Management of Faculty of Fishery Sciences under West Bengal University of Animal and Fishery Sciences at Chakgaria campus of South 24 Parganas district in West Bengal. The experiment was conducted for a period of 15th weeks from the 4th December, 2016 to 18th March, 2017.

2.2 Preparation of experiment

The experiment was carried out in 15 glass aquaria (60x30x 30cm 3) and each aquarium on an average twelve number of fingerlings was kept. The aquaria were cleaned thoroughly using scrub and then dried for a week. They were filled with good quality tap water up to a depth of 20cm.

2.3 Proximate Composition of formulated diet

Proximate composition such as moisture, crude protein, crude fat and total ash of formulated diet were analyzed immediately after preparation. The proportion of different ingredients used for preparation of formulated diet is presented in Table 1.

Table 1: Proportion of ingredients used in formulated diet

Ingredients	Formulated diet (gm)
Fish meal	400
Ground nut oil cake	400
Wheat flour	20
Rice polish	150
Vitamin-mineral mixture	10
Soya bean oil	20ml

2.4 Feeding protocol

Fishes of four treatments (Starvation) were feed under deprivation protocol and were designed as D_1, D_2 and D_3 and D_4 respectively. Here fishes were starved for one day, two days and three days and four days respectively in each week towards end and then fed at the rate of 5 percent body weight for rest of days of the week. Thus the corresponding feeding cycles were (One day starvation + 6 days feeding), (2 days starvation + 5 days feeding) and (3 days starvation + 4 days feeding) and (4 days starvation + 3 days feeding) for treatments D1, D2 and D3 and D4 respectively. This experiment continued for the period of 105 days and terminated on 106th day. Water quality parameters such as temperature pH, alkalinity, hardness and dissolved oxygen contents of such aquarium were monitored at weekly intervals following standard methods (APHA. 1995) [2].

2.5 Proximate composition of fish muscle

Proximate composition of body muscle of *Anabas testudineus* fingerlings was estimated during the commencement and termination of the experiment. Muscle samples were collected from the trunk portion of the fishes avoiding bony parts. It was dried in hot air oven to obtain dry matter. Sample was homogenized by hand homogenizer for estimation of protein in calorimetric method. Samples were analyzed for crude fat, moisture and total ash following the standard procedure (AOAC, 1995) [2].

3. Results and Discussions

The result of proximate composition of the flesh of *Anabas testudineus* fingerlings at the beginning, and at the end of the experiment are presented in Table.2 as well as in Fig: 1, Fig: 2 and Fig: 3 respectively. The initial values of moisture, protein, lipid and ash were 80.73%, 14.10%, 2.05% and 1.20% in, D1, D2, D3, D4 and control respectively. After termination of the experiment the corresponding moisture values were 73.42%, 73.52%, 74.43% and 74.87, 73.33 in D1, D2, D3, D4 and control, respectively. Crude protein levels were 19.06%, 18.88%, 18.72%, 18.12%, and 19.25% in, D1, D2, D3 and D4, control respectively. The corresponding lipid contents of muscle were 2.64%, 2.56%, 2.03%, 1.90% and 2.86%. The corresponding total ash values were 2.42, 2.34, 2.08, 2.04, and 2.61 in D1, D2, and D3and D4 control respectively. There was no significant difference in moisture content and protein content. There was significant difference in crude lipid content among D3 ($P<0.05$) and D4 ($P<0.05$) treatments during the period of experiment compared to control. However, there was no significant difference between the treatments control and D1 as well as control and D2. In the total ash content there was no significant difference between control and D1aswell as control and D2. However, there was significant difference between control and D3 ($P<0.05$) as well as control and D4 ($P<0.05$) during the period of experiment. Artificial feeding influence biochemical characteristics of fish flesh. In present study, there was significant marked difference in biochemical characteristics of fish flesh at termination of the experiment. It was found that moisture content declined in fish over initial values. There was an increase in protein content of fish over the initial values.

Table 1: Proximate composition of fish muscle under feed deprivation protocol

Treatments	Moisture	protein	Fat	Ash
Initial	80.73±2.03	14.10±0.56	2.05±0.11	1.20±0.12
C	73.33±2.50	19.25±0.68	2.86±0.16	2.61±0.19
D_1	73.42±0.08	19.06±0.66	2.64±0.13	2.42±0.14
D_2	73.52±0.03	18.88±0.54	2.56±0.14	2.34±0.15
D_3	74.43±0.07	18.72±0.41	2.03±0.17	2.08±0.11
D4	74.87±0.06	18.12±0.48	1.90±0.18	2.04±0.17

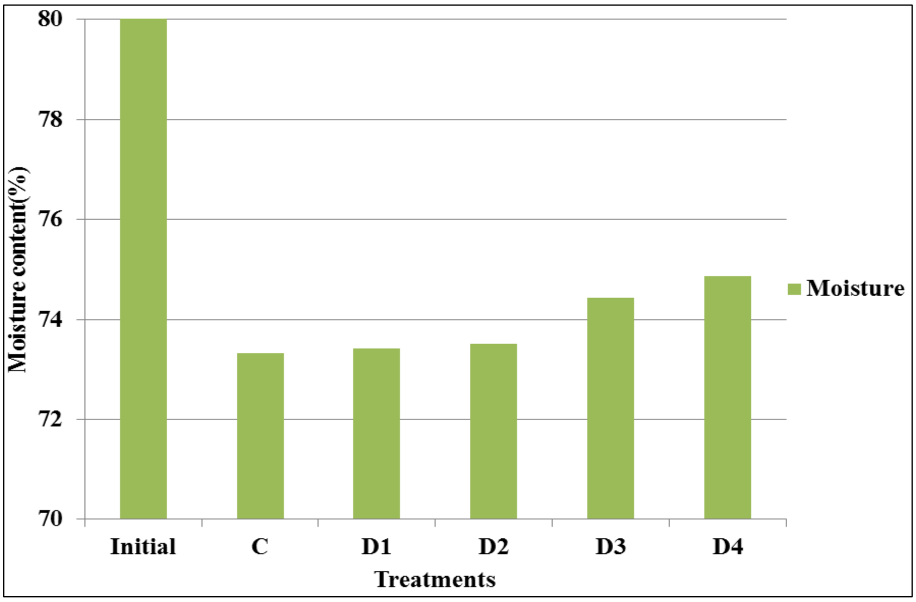

Fig 1: Moisture content in feed deprivation protocol

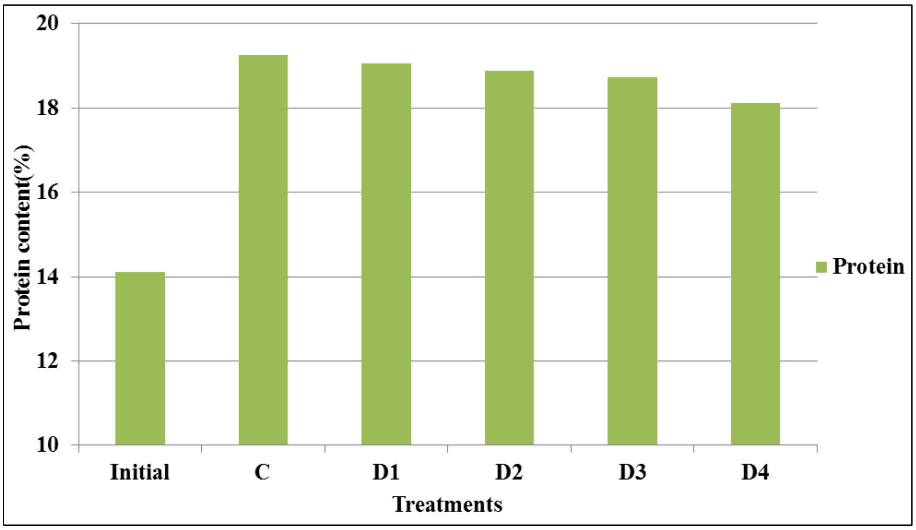

Fig 2: Protein content in feed deprivation protocol

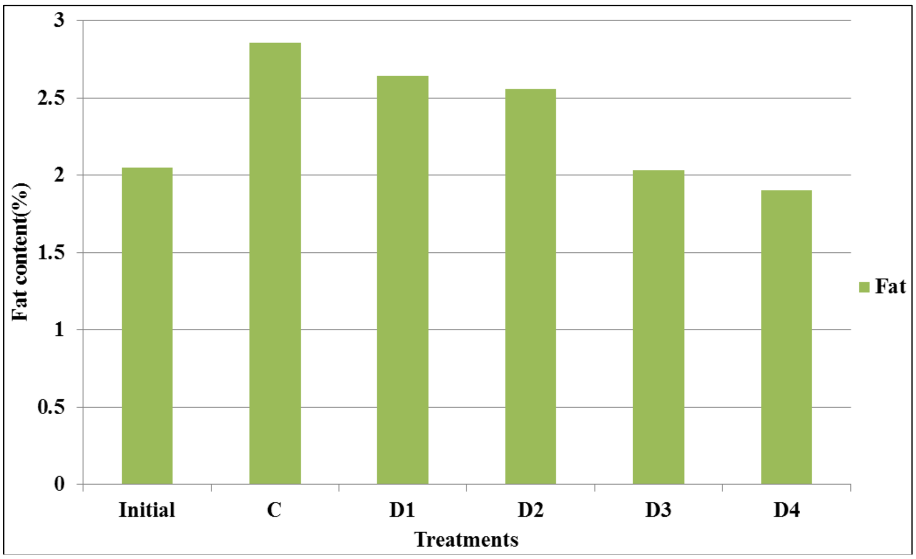

Fig 3: Fat content in deprivation protocol

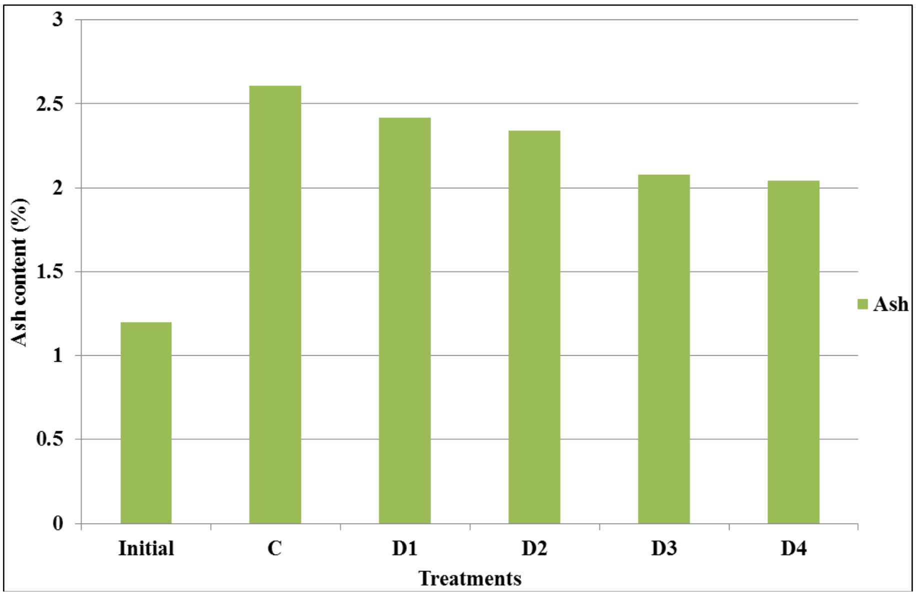

Fig 4: Ash content in deprivation protocol

During starvation essential processes are maintained at the expense of accumulated energy reserves which of course, results in the progressive depletion of body tissue (Love, 1980) [15]. Starvation results in tissue hydration (Jobling, 1993; Wang, 2009; Miglavs and Jobling, 2010) [12, 22, 17]. This plays a role in the limitation of the loss of even the maintenance of wet body weight during starvation (Love, 1980) [15]. Starvation results in significant decrease in lipid contents of carcass and viscera. Following depletion of liver lipid stores, lipid content in perivisceral adipose tissue is utilized along with a pexial muscle glycogen. In fishes, lipid is stored in the liver viscera and muscles. Lipids are broken down early in starvation, and often constitute the main energy source for maintenance of fish during over starvation. Eroldogon *et al*. (2008) [7] reported that the cycles of restricted feeding regime had no significant influence on the moisture, lipid and ash content in juvenile gilthead Sea bream but the body protein content in R6 (50% restricted feeding for six days and then re-fed for six days to apparent satiation) was less than that of control, S1 (Starvation for one day than re-fed for two days to apparent satiation) and R2 (50% restricted feeding for two days and then re-fed for two days to apparent satiation). Cui *et al*. (2006) [6] observed that there was no significant difference (P>0.05) in whole body protein content between control and deprived fish whereas fish deprived for two, three and four weeks had a lower whole body lipid and higher ash and moisture content than the control (P<0.05) in gibel caup *Carassius auratus gibelio*. No significant difference in proximate commotion in gibel carp was seen experiencing cycles of food deprivation and re-feeding (Zhu *et al*., 2004); Ali *et al*., 2006) [25, 1] reported that feed cycling did not have any detectable effect on ash, lipid, protein and organic content in *Labeo rohita* whereas moisture content was significantly different among the treatments. Wang *et al*. (2000); Wang *et al*. (2009) [21, 22] reported there were no significant differences in moisture, crude lipid, crude protein, ash content between the control and fish deprived for one, two and four weeks in hybrid tilapia. Ash content was found not significant (P>0.05) among all the treatments in his experiment on effect of feed cycling on growth and survival of *Catla catla*. Xie, *et al*. (2001) [24] found that crude protein and crude lipid content of muscle in fish in S1 (one day feed deprivation in 1 week duration) and C (feed regularly) were significant higher (P<0.05) than other treatments deprived for two, three and four days in 1 week experiment, while significant differences was found on moisture and ash content (P<0.05). Eslamloo *et al*. (2012) [8] found that, the total body protein, fat, moisture, ash and energy contents varied between T3 fish (eight days starvation followed by 24 days re-feeding) and the other treatments T1 (two days starvation followed by six days re feeding) T2 (four days starvation followed by 12 days re-feeding) and control (fed twice daily). Tian *et al*. (2003) [19] reported that there was no significant difference among S1 (food deprivation for 4 days after satiation feeding till 64 days) S2 (food deprivation for 8 days) and control in the content of moisture, lipid, protein, ash and energy (P>0.05). But the re-feeding, period metabolic rates of juvenile tongue sole increased rapidly the fat content for the starved for 32 days (S4) and these increases were directly proportional to the length of the starvation period.

4. Conclusion

There was no significant difference in moisture content and protein content among all the treatments during the period of investigation. There was significant difference in crude lipid content among D3 (P<0.05) and D4 (P<0.05) treatments during the period of experiment in the ash content there was no significant difference between control and D1as well as control and D2. However, there was significant difference in D3 (P<0.05) and D4 (P<0.05) compared to control during the period of experiment. Muscle nutrients in these two treatments D1 and D2 also exhibited similarity with the control. However there was significant difference in muscle characteristic in the fish of treatment D3 and D4 compared to control in treatments indicating lack of full compensation in growth. It indicated that feed restriction of treatment D1 and D2 can compensate the growth and thus reduce the feed consumption considerably.

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