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Proximate analysis of local and Thai climbing perch, *Anabas testudineus*

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Abstract
Local and Thai climbing perch (Koi), *Anabas testudineus* is an important food fish and their demand increasing day by day to minimize nutitional deficiency in Bangladesh. In this study the nutritional values of local and Thai koi are determined. Local and Thai koi are rich in protein (15.2-16.3%), lipid (5.3-8.7%) and contained a lower amount of carbohydrate content ranges from 1.6-2.2%. Both the fishes were also rich in mineral contents (total ash content is 1.2-2.1%). The moisture content ranges from 71.8 to 75.3%. Among the minerals, iron (0.13-0.65 mg/100g), calcium (192.3-241.4 mg/100g), phosphorus (48.6-50.8 mg/100g), zinc (0.012-0.231 mg/100g), and cobalt (0.057-0.044 mg/100g) were detected. Iron and zinc were found significantly higher in Thai variety but local variety of *A. testudineus* is significantly richer in calcium content. Data of this study suggest that both climbing perch are rich in nutritional value.

Keywords: *Anabas testudineus*, carbohydrate, lipid, minerals, protein

1. Introduction
Fish is the major protein sources in the diet of the Bangladeshi people. Fish contributes about 60% of the available protein in the diet and the rest 40% protein comes from livestock and poultry. It indicates the importance of fish in contributing to the level of nutrition of the people of Bangladesh [1]. In spite of having large fisheries resources, Bangladesh is facing an acute malnutrition problem due to the shortage of animal protein supply in their diet. The present per capita fish consumption is only about 21 g/day, whereas 38 g/day is the required amount. The nutritional status of local and Thai climbing perch, in order to increase knowledge among the culturist to give more attention in culture and eventually to the common people to compensate any bodily nutrients simply through enlisting in a routine table diet [2, 3].
Local and Thai climbing perch found in Bangladesh are good source of nutrients, especially protein, lipid, and minerals. Protein is an important nutritional component and protein deficiency is the world's most serious human nutritional problem, especially in third world countries like Bangladesh [4]. It is an important food item concerning human health, nutrition and disease prevention [5]. Nutritional analysis of several fish species of different sources had been carried out in many laboratories in the world but nutritional values of locally cultivated fishes remain speculative. Generally, people in Bangladesh are still not very aware of nutritional and medicinal importance of *A. testudineus*. The aim of this investigation was to analyze the nutritional values of climbing perch, with a goal of increasing awareness of the beneficial effects among the consumers.

2. Materials and Methods
For the convenient nutritional analysis, a standard number of local and Thai *A. testudineus* were colleted from different fish market in and around Savar, Dhaka and preserved in ice condition (-20 ^{0}C) for biochemical study. Collected fishes were first of all cleaned and body muscles were carefully separated and dewatered to measure its muscle moisture contents. Then it was preserved in an air tight polyethylene bag for future use of determining other parts of biochemical study. For the determination of nutrient content from local and Thai *A. testudineus*, 5g of each fresh fish muscles was taken with 50ml phosphate buffer and homogenized with a tissue homogenizer (Polytron: PT 1200). Homogenized solution was used as sample for the determination of nutrient content. For the determination of nutrient content of local and Thai Koi were dried with hot air at 40 °C for 48 hours and finely pulverized.

2.1 Moisture determination
Five grams of fish flesh was weighed into a weighed moisture box (A&D company ltd. N 92; P1011656, Japan) and dried in oven at 100-105 ºC and cooled in a desiccator. The process of heating and cooling was repeated till a constant weight was achieved. The moisture content was calculated as following equation [6].
Moisture (%) = (Initial weight- final weight) × 100/Weight of sample

2.2 Determination of total protein
Five grams of grinded fish muscles were taken with 50ml of 1N NaOH and boiled for 30 minutes. The solution was cooled in room temperature and centrifuged at 1000g by a table centrifuge machine (DIGISYSTEM: DSC-200T, Taiwan). The supernatant was collected and total protein content was measured according to the Biuret method [7] with a diagnostic kit (Total Protein: Colourimetric test- Biuret method// Crescent Diagnostics, Saudi Arabia). According to this method, the peptide bonds present in proteins react with Cu^{++} ions in alkaline solutions to form a colored complex of which the absorbance was measured spectrophotometrically at 540nm which was directly proportional to protein content.
For the determination of protein content in fish muscles, 5ml of homogenized fish tissue sample was taken with 50ml of 1N NaOH and protein content was determined as mentioned above.

2.3 Determination of total lipid
Total lipid was determined by slight modified method of Folch *et al.* [8]. Five gram of grinded fish muscles was suspended in 50ml of chloroform: methanol (2:1 v/v) mixture then mixed thoroughly and let stand for 3 days. The solution was filtrated and centrifuged at 1000g by a table centrifuge machine. The upper layer of methanol was removed by Pasteur pipette and chloroform was evaporated by heating. The remaining was the crude lipid. For the determination of lipid content in fish mucles, 5ml of homogenized fish tissue sample was taken with 50ml of chloroform: methanol (2:1 v/v) mixture and lipid content was determined as mentioned above.

2.4 Determination of total carbohydrate
The content of the total carbohydrate was determined by phenol-sulphuric acid method [9]. Carbohydrates were first hydrolyzed into simple sugars using dilute hydrochloric acid. In hot acidic medium glucose was dehydrated to hydroxymethyl furfural. This formed a green colored product with phenol and sulphuric acid and the concentration was measured by spectrophotometer at 490nm.

2.5 Determination of total ash
One gram of the sample was weighed accurately into a crucible. The crucible was placed on a clay pipe triangle and heated first over a low flame till all the material was completely charred, followed by heating in a muffle furnace for about 8 hours at 600 ºC. It was then cooled in a desiccator and weighed. To ensure completion of ashing, the crucible was then heated in the muffle furnace for 1hour, cooled and weighed. This was repeated till two consecutive weights were the same and the ash was almost white or grayish white in color. Then total ash was calculated as following equation [10].
Ash content (%) = Weight of ash × 100/Weight of sample taken.

2.6 Mineral analysis
Total ash was taken for the analysis of mineral contents. 2ml of concentrate HNO_3 was added to the ash and heated for 2 minutes. One drop of hydrogen peroxide was added into the solution to remove turbidity. The solution was then transferred into a volumetric flask and total volume was made 50ml by adding deionized water. This was then used to analyze the contents of calcium (Ca), iron (Fe), phosphorus (P), cobalt (Co), and zinc (Zn) by flame and graphite method with atomic absorption spectrophotometer [11].

2.7 Statistical Analyses
The statistical programs used were Microsoft Excel and Statistical Program for Social Science (SPSS 11.5). All parameters for inter group differences were analyzed by one-way analysis of variance (ANOVA) and then post hoc comparisons, LSD (least significant difference) and DMRT (Duncan's multiple range test) at $P \leq 0.05$ level.

3. Results
3.1 Moisture, protein, lipid, carbohydrate, and ash contents of body muscles of both local and Thai climbing perch
In the present experiment moisture, protein, lipid, carbohydrate, and ash contents from three different size i.e. small (8-10 cm), middle (10-12 cm), and large (12-14 cm) of local and Thai climbing perch, *A. testudineus* were estimated (Fig. 1 to Fig. 5).
Maximum moisture content (75.3%) was recorded in small size of local koi and followed by 75.1%, 73.9%, and 73.7%, respectively in middle size of local koi, small and middle size of Thai koi (Fig. 1). The results indicated that the moisture content of local *A. testudineus* was higher as compared to Thai *A. testudineus*.

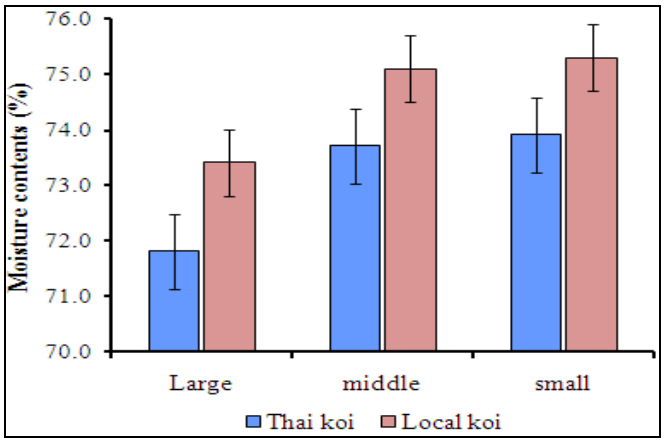

Fig 1: Moisture contents of local and Thai climbing perch (koi), *Anabas testudineus*

Maximum protein content (16.3 g/100g) was recorded in large size of local koi, while lowest protein content (14.4 g/100g) was recorded in middle size of Thai koi (Fig. 2). The results also indicated that the protein content of local *A. testudineus* was higher as compared to Thai *A. testudineus*.

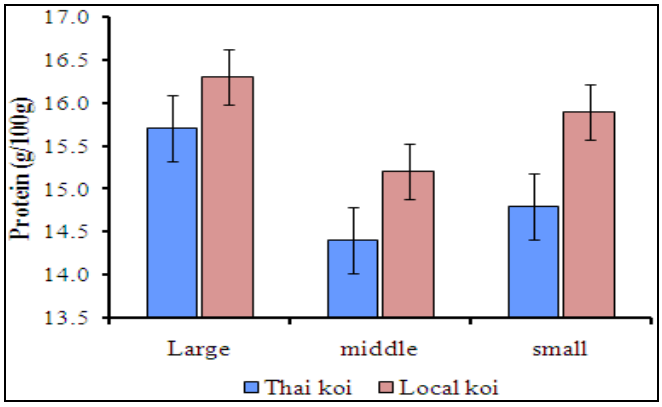

Fig 2: Protein contents of local and Thai climbing perch (koi), *Anabas testudineus*

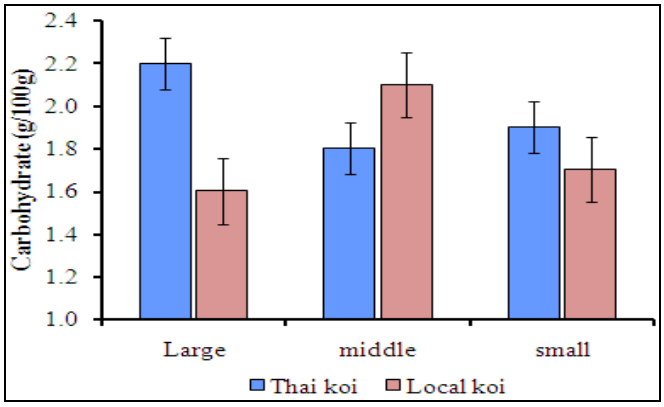

Fig 4: Carbohydrate contents of local and Thai climbing perch (koi), *Anabas testudineus*

Maximum lipid content (8.7 g/100g) was recorded in middle size of Thai koi, while lowest lipid content (5.3 g/100g) was recorded in small size of local koi (Fig. 3). The results indicated that the lipid content of Thai *A. testudineus* was higher as compared to local *A. testudineus*.

Almost similar highest amount of carbohydrate content (2.2 g/100g and 2.1 g/100g) were recorded in large size of Thai koi, and middle size of local koi, respectively, whereas lowest carbohydrate content (1.6 g/100g) was recorded in large size of local koi (Fig. 4). The results indicated that the carbohydrate content of local and Thai *A. testudineus* is almost similar.

Maximum ash content (2.1g/100g) was recorded in large size of local koi. Large size of Thai koi and small size of local koi showed similar amount (1.8g/100g) of ash (Fig. 5).

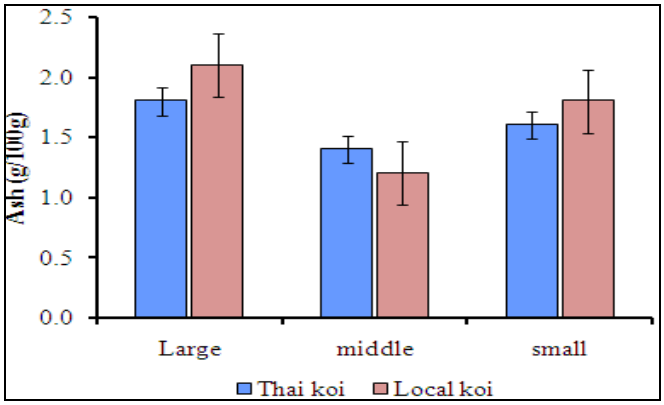

Fig 5: Ash contents of local and Thai climbing perch (koi), *Anabas testudineus*

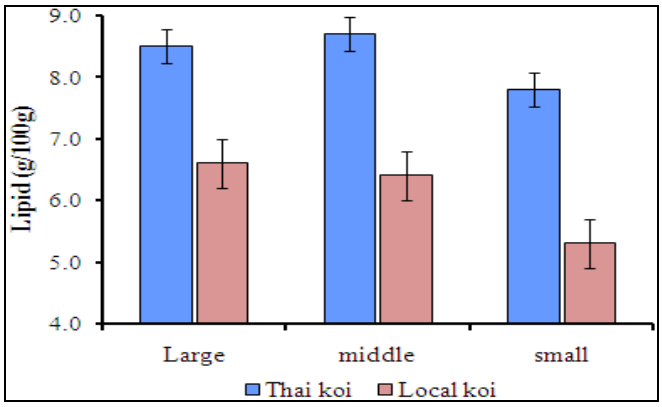

Fig 3: Lipid contents of local and Thai climbing perch (koi), *Anabas testudineus*

4.2 Mineral contents of local and Thai climbing perch (koi), *Anabas testudineus*

Mineral contents i.e. iron, calcium, phosphorus, zinc, and cobalt of local and Thai, *A. testudineus* have been presented in Table 1. Maximum iron content (0.65mg/100g) was recorded in Thai koi as compared to local koi (0.13mg/100g). Maximum calcium content (241.4mg/100g) was recorded in local koi as compared to Thai koi (192.3mg/100g). Highest phosphorus and zinc content (50.8mg/100g and 0.231mg/100g, respectively) was recorded in Thai koi as compared to local koi (48.6mg/100g and 0.012mg/100g, respectively). Maximum cobalt content (0.057mg/100g) was recorded in local koi as compared to Thai koi (0.044mg/100g).

Table 1: Mineral contents of local and Thai climbing perch (koi), *Anabas testudineus*

Variety	Mineral contents (mg/100g)				
	Iron	Calcium	Phosphorus	Zinc	Cobalt
Local	0.13±0.0	241.4±18.4	48.6±5.2	0.012±0.0	0.057±0.0
Thai	0.65±0.1	192.3±15.7	50.8±4.9	0.231±0.0	0.044±0.0

Values are mean ± SD of 5 samples

4. Discussion

Several nutritional parameters were measured for both local and Thai *A. testudineus*. In the present experiment moisture, protein, lipid, carbohydrate, and ash contents were measured from three different size i.e. small (8-10cm), middle (10-12cm), and large (12-14cm) of local and Thai climbing perch. Maximum moisture content (75.3%) was recorded in small size of local koi. The results indicated that the moisture content of local *A. testudineus* was higher as compared to Thai *A. testudineus*. Rubbi *et al.* [12] observed the similar results and reported that moisture of fresh fish varied from 72.1% to 83.6%. Govindon [13] reported that sometime, this variation might be a complex one and it is impossible to distinguish because of many factors interplaying in the process.

The protein content of local koi was higher as compared to Thai *A. testudineus*. On the other hand, the lipid content of Thai *A. testudineus* was higher as compared to local koi. It is believed that gonad developments of almost all freshwater fishes start and proceed rapidly until ovulation with the on-set of monsoon. Al-Habib [14] estimated the protein contents of six fresh water fishes and observed that these fishes contained 11.00-16.75% protein that was well agreed with present findings. Chandrasheker and Deosthale [15] observed that wide variation exist in protein of freshwater fishes and it was 13.5% to17.3%. Thus, it can be said that the studied fish maintain almost similar ranges of protein concentration as found in other freshwater fishes by several scientists. According to Wilson and Halver [16], fish digest protein to obtain free amino acids, which are absorbed from the intestinal tract and used by various tissues to synthesize new protein. Stansby [17] reported that fresh water fish contained 5.0% lipid per gram of body muscle. The above discussion was more or less similar to the findings.

The carbohydrate content of local and Thai *A. testudineus* is almost similar. Maximum ash content (2.1 g/100g) was recorded in large size of local koi. In fish, carbohydrate present in minimum quantity. The nutritional value of carbohydrates varies among fish. Warm water fish can use much greater amounts of dietary carbohydrate than cold-water and marine fish. No dietary requirement for carbohydrates has been demonstrated in fish, however, if carbohydrates are not provided in the diet, other compounds, such as protein and lipids, are catabolized for energy and for the synthesis of various biologically important compounds usually derived from carbohydrates. Thus, it is important to provide the appropriate concentration of carbohydrate in the diet of the fish species being cultured. Enzymes for carbohydrate digestion are apparently present in fish. The enzymes for the major carbohydrate metabolic pathways, such as glycolysis, tricarboxylic acid cycle, pentose phosphate shunt, gluconeogenesis, and glycogen synthesis, have been demonstrated [18].

The ash content refers to the amount of minerals, vitamins and other edible constituents. Rubbi *et al*. [12] reported that the ash of fresh water scaly and non-scaly fishes ranged between 0.85%-5.11% in body muscle. Ahmad and Hassan [19] reported that fresh water fishes generally contained 0.85%-5.11% in fish body muscle. These values more or less supported the present findings.

Maximum iron content (0.65 mg/100g) was recorded in Thai koi as compared to local koi (0.13mg/100g). Maximum calcium content (241.4 mg/100g) was recorded in local koi as compared to Thai koi (192.3 mg/100g). Highest phosphorus and zinc content (50.8 mg/100g and 0.231 mg/100g, respectively) was recorded in Thai koi as compared to local koi (48.6 mg/100g and 0.012 mg/100g, respectively). Maximum cobalt content (0.057 mg/100g) was recorded in local koi as compared to Thai koi (0.044 mg/100g).

Iron is a necessary trace element found in fish. Iron-containing enzymes and proteins, often containing heme prosthetic groups, participate in many biological oxidations and in transport [20].

Calcium is an important component of a healthy diet and a mineral necessary for life. Calcium plays an important role in building stronger, denser bones early in life and keeping bones strong and healthy later in life. Approximately 99 percent of the body's calcium is stored in the bones and teeth. The rest of the calcium in the body has other important uses, such as some exocytosis, especially neurotransmitter release, and muscle contraction. Long-term calcium deficiency can lead to rickets and poor blood clotting and in case of a menopausal woman, it can lead to osteoporosis, in which the bone deteriorates and there is an increased risk of fractures. Fish is an excellent source of dietary calcium for those whose, bodies tolerate it because it has a high concentration of calcium and the calcium in fish is excellently absorbed [21]. However, it is clear that increasing the intake of calcium promote its deposition in the bones, where it is of more benefit in preventing the compression fractures resulting from the osteoporotic thinning of the dendritic web of the bodies of the vertebrae, than it is at preventing the more serious cortical bone fractures that happen at hip and wrist bones in human [22].

Zinc is an essential trace element. It is typically the second most abundant transition metal in organisms after iron and it is the only metal which appears in all enzyme classes. In proteins, Zn ions are often coordinated to the amino acid side chains of aspartic acid, glutamic acid, cysteine and histidine [23]. Thus, it has been called as the brain's dark horse, since it also can be a neurotoxin suggesting zinc homeostasis and plays a critical role in normal functioning of the brain and central nervous system [24].

Cobalt is essential to all animals. It is a key constituent of cobalamin, also known as vitamin B_{12}, which is the primary biological reservoir of cobalt as an ultratrace element. The cobalamin-based proteins use corrin to hold the cobalt. Coenzyme B_{12} features a reactive C-Co bond, which participates in its reactions. In humans, B_{12} exists with two types of alkyl ligand: methyl and adenosyl [25, 26]. According to Rubbi *et al*. [12] the phosphorus contents of koi (*Anabas testudineus*) was 75.2 mg, Bele was 75.22 mg, chapila was 143 mg, foli and kachki were 153.9 mg and 172.3 mg, respectively. The mineral content of fish, unlike its vitamin A content, is apparently not species specific. Therefore, climbing perch are an excellent source of minerals. Both local and Thai koi are rich in protein, lipid and minerals but poor in carbohydrate content. In this study, five minerals i.e. Ca, Fe, P, Zn and Co were detected. Among the minerals iron and zinc were found significantly higher in Thai variety but local koi significantly richer in calcium content.

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