Seasonal Variationsin Proximate, Amino Acid and Fatty Acid Profile of Wild and Cultured *Anabas testudineus*(Bloch, 1795)

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CERTIFICATE

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ABSTRACT

Throughout the year, fish are subjected to considerable environmental changes and fluctuations in availability and compositions of feed that will affect their proximate, amino acids and fatty acid compositions.The purpose of this study was to clarifythe seasonal variations in proximate, amino acid and fatty acid profile of wild and cultured *Anabas testudineus*.The proximate compositions, total amino acid and fatty acid profile were determined by AOAC, HPTLC and GC respectively. Theresultsindicated that proximate (Moisture, Ash, Protein and Lipid), amino acid and fatty acid profile varied significantly ($p<0.05$) between wild and cultured fishes depends on season.For both cultured and wild *Anabas testudineus,*amount of protein was higher in post-monsoon than in pre-monsoon period.The fat and fatty acid as well as protein and amino acid contents of the investigated fish species showed a significant ($p<0.05$) seasonal dependency. Among the 14 recorded amino acids (Essential Amino Acid-6, Non-essential Amino Acid-8), the major amino acid in the total crude protein were histidine, methionine, isoleucine, lysine, glutamic acid and glycine. The findings of the present study also showed that unsaturated fatty acids represented 59.88-60.98%and 60.12-61.32% of the total fatty acids, whereas saturated fatty acids accounted for 39.02-40.11% and 37.56-39.86% for wild and cultured koi fish depending on seasons.The major fatty acids from wild and cultured *Anbastestudineus*were oleic acid (18:1), palmitic acid (16:0), linoleic acid (18:0), stearic acid, palmitoleic acid (16:1*)* andmyristic acid (14:0).Fatty acid profile resulted that some fatty acid (Lauric acid (C12:0), Pentadecyclic acid (C15:0), Arachidic acid (C20:0), Caproic acid (C6:0), Enanthic acid (C7:0), Caprylic acid (C8:0)) were not detected in cultured fish but present in wild investigated fish.Most nutritionally important docosahexaenoic acid (DHA, C22:6 n-3) was dominant in wild fish fillets instead of linoleic acid (C18:2 n-6) in the farmed fish. The ratio of Essential Amino Acid (EA)/Non-Essential Amino Acid (NEA)was higher in cultured fish of post-monsoon seasons. It is found that fatty acid composition and n-3/n- 6 is quite high in wild koi fish in post-monsoon and it is preferable for human diet than cultured fish. Therefore, a comprehensive study on cultured and wild fishes is required for the further confirmation of the result of present study.

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LIST OF ABBREVIATIONS AND SYMBOLS

DoF	Department of Fisheries
GDP	Gross Domestic Product
EPA	Eicosapentaenoic Acid
DHA	Docosahexaenoic Acid
EFA	Essential Fatty Acid
PUFAs	Polyunsaturated Fatty Acids
BCSIR	Bangladesh Council of Scientific and Industrial Research
AOAC	Association of Official Analytical Chemists
GC	Gas Chromatography
HPLC	High Performance Liquid Chromatography
HPTLC	High Performance Thin Layer Chromatography
FAMEs	Fatty Acid Methyl Esters
SPSS	Statistical Package for the Social Sciences
ANOVA	Analysis of Variance
%	Percentage
+	Plus
-	Minus
±	Plus-Minus
×	Multiplication
>	Greater than
<	Less than
≈	Approximately equal
=	Equal to
kg	Kilogram
p	Level of Significance
Ca	Calcium
Mg	Magnesium
K	Potassium
Na	Sodium
P	Phosphorus
Fe	Iron

S	Sulphur
Cl	Chloride
Cu	Copper
Mn	Manganese
Br	Bromine
I	Iodine
Sr	Strontium
Hg	Mercury
Cd	Cadmium
Mo	Molybdenum
Co	Cobalt
Ba	Barium
Al	Aluminium
Zn	Zinc
Ni	Nickle
HCl	Hydrochloric acid
H_2SO_4	Sulfuric acid
NaOH	Sodium hydroxide
NH_3	Ammonia
Na_2SO_4	Sodium sulfate
H_2O	Water
NH_4	Ammonium
H^+	Hydrogen Ion
H_2BO_3	Dihydrogen borate
H_3BO_3	Boric acid

1 INTRODUCTION

1.1 Background

Bangladesh has vast fishery potentials within her boundaries and the territorialand economic zones in the sea.Fish and fisheries are inseparable from the culture and heritage of Bangladesh. The country is a transitional zone of flora and fauna, because of its geographical settings and climatic characteristics. She is cross cut with an impressive network of rivers totaling and about a quarter of the country is inundated each year (Brammer,2004).The lives of Bangladeshis centre around and fish and water. It is natural that the water resources of the existing extent and magnitude should harbor and support populations of a large variety of vertebrate and invertebrate aquatic living organisms.

Fish is an irreplaceable animal-source food in the diet of millions and accounting for approximately 60% of animal protein intake at 18.1 kg consumed per person per year and frequency of consumption, far exceeding that of any other animal-source food (Belton *et al.,* 2014). Fisheries is now the most valuable agricultural activities in Bangladesh, playing a substantial role in nutrition, food security, employment and income generation, trade, source of foreign exchange earnings and above all poverty reduction in the economy. Fisheries sector plays a very important role in the national economy having a share 3.69 percent in national GDP, almost one-fourth (22.60%) in agricultural GDPand 2.01% to the country's export earnings. This sub sector also provides about 60% of the animal protein intake (FRSS, 2015). In 2013-2014, the contribution of inland capture and culture fisheries to total production were 28.07% and 55.15% respectively(DoF, 2015). At present Bangladesh is ranked 4^{th} in inland capture fisheries production and 5^{th} in aquaculture production all over the world.Over the last two decades, inland aquaculture of indigenous and exotic carp species, tilapia, pangas and Thai/Vietnam koi expanded massively. Besides, new interest grew in farming of indigenous species like koi, singh, magur, pabda, gulsha,mola etc. because they are getting scarce on open water areas but have high market demand and better contribution to household level nutrition supply (FRSS, 2015).

Anabas testudineus, a small fish of inland waters, *testudineus* means "turtle like". The fish is frequently found on tree tops and also found hanging from trees or living in water filled slits of a palm tree. So they are commonly called as climbing perch .This fish widely distributed throughout south and south East Asia. Its common name in Bengali is

Koi. The contribution of the *Anabas testudineus* both the indigeneous and Thai/Vietnam variety to the national fish production is about 4-5% (DoF, 2015).The fish is popular to the people from the ancient time because it is a very tasty and nutritious. Once upon a time koi fish found in a plenty in the canal, small rivers, swamp, inundation land of Bangladesh. Koi fish is one of the most popular and tasty fish among the all fish of Bangladesh. This fish is also very precious now-a-days especially wild one.

Koi is a local fish breed of Bangladesh. Nowadays commercial koi fish farming in pond is very popular to meet increasing demand. Its farming is very easy, and profitable. The main benefits of cultivating koi fish because of its commercial and nutritional importance. Some points are listed below in this regard.

- ❖ Demand of koi fish is very high; as a result its market value is comparatively higher always;
- ❖ Rich in n-3 fatty acid which is essential for human being;
- ❖ Able to survive in adverse environment and mortality rate is very low;
- ❖ Can be cultivated in greater density;
- ❖ Can be cultivated in small pond or cage andrelatively in small capital,
- ❖ Become suitable for sale comparatively in short time, within 3-4 months;
- ❖ Economically profitable and can be cultivated multiple times per year;
- ❖ Diseases are less in koi fish than other fish breed;
- ❖ Pellet feed for koi fish can be made in own house;
- ❖ Koi fish is mainly insectivorous. For this reason koi fish can be cultivated by providing insects, small fish, toad minnow, snail, oyster flesh etc.

1.1.1 Fish Nutrients

Fish is one of the most important foods and is valued for its nutritional qualities. It includes essential fatty acids, amino acids and some of the principal vitamins and minerals in sufficient amounts for healthy living. Fish oil has been utilised by humans for hundreds of years to help relieve medical problems (Hunter and Roberts, 2000).

Fish oil is one of the most important natural sources of polyunsaturated fatty acids including eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), which have been proven to have useful effects on human health (Saoud*et al.,*2008,Rafflenbeul, 2001). In addition, fish oil is a rich source of vitamins including vitamin A, D, E, and K, which are soluble in oil and must betaken on a regular basis because of their key roles in human health and metabolism (Kinsella, 1987).

Fish lipids have assumed great nutritional significance owing because of its protective role against thedevelopment of cardiovascular disease. Greater protein and lipid content represents higher energy density (Dempson*et al*., 2004).

A regular consumption of Essential Fatty Acid (EFA: EPA+DHA) may play an important role in the prevention of cardiovascular diseases and neural disorders (Lauritzen*et al.,* 2001, Kris-Etherton*et al.,* 2002). It has beneficial effects on bone formation and metabolism (Watkins *et al.,* 2003), and decreases high blood pressure and cholesterol, prevents certain cardiac arrhythmias, lowers the occurrence of diabetes, and appears to reduce symptoms associated with rheumatoid arthritis. Arnett (2000) reported that during pregnancy and lactation an adequate intake of DHA and EPA is particularly important. During this time DHA and EPA must be supplied with food because human body cannot synthesize these essential fatty acids.

The composition of nutrients in fish varies greatly from species to species and also from individual to individual depending on age, sex, environment and season (Huss, 1995).

1.1.2 The Major Components of Fish Muscle

1.1.2.1 Moisture

The main constituent of fish flesh is moisture, which usually accounts for about 80 per cent of the weight of a fresh fish fillet. Whereas the average water content of the flesh of fatty fish is about 70 per cent.Generally the range moisture content varies between 60-80% in many fish (Love, 1970, Murraand Burt, 2001).

The water in fresh fish muscle is tightly bound to the proteins in the structure in such a way that it cannot readily be expelled even under high pressure. After prolonged chilled or frozen storage the proteins are less able to retain all the water, and some of it, containing dissolved substances, is lost as drip. Thus Moisture content is an important measure of fish quality, as the flesh naturally has high water content.

1.1.2.2 Protein

Generally, fish is the excellent source of protein, because of the amino acid composition and degree of digestibility. Protein consists of amino acids. The amount of protein in fish muscle is between 15-20 per cent (Murraand Burt, 2001).

The proteins in fish muscle tissue can be divided into three groups:

- Structural proteins (actin, myosin, tropomyosin and actomyosin) which constitute 70-80 percent of the total proteincontent(Nowsad, 2007).These proteins are soluble in neutral salt solutions of fairly high ionic strength (0.5 M).
- Sarcoplasmic proteins(myoalbumin, globulin and enzymes) which are soluble in neutral salt solutions of low ionic strength (<0.15 M). This fraction constitutes 25-30 percent of the protein(Nowsad, 2007).
- Connective tissue proteins (collagen), which constitute approximately 3 percent(Nowsad, 2007)of the protein in teleosts and about 10 percent in elasmobranch (compared with 17 percent in mammals).

1.1.2.3 Lipid

Lipids are the generic names assigned to a group of fat soluble compounds found in the tissues of animals. Lipids are broadly classified asfats, phospholipids, sphingomyelins,waxes andsterols.

1.1.2.4 Fats

Fats are the fatty acid esters of glycerol and are the primary energy depots of animals. These are used for long-term energy requirements during periods of extensive exercise or during periods of inadequate food and energy intake. Fat content of fish varies with the species (Exler, 1975). Variation is also caused by feeding, size, age, season, physiological status and geographical location (Stansby, 1954). Fat deposition is usually below the skin, around the belly wall and after along lateral line. Fat are made of different kinds of fatty acids. Those fatty acids differ in the amount and arrangement of the carbon and hydrogen atoms.

1.1.2.5 Fatty acid

Fatty acid is a carboxylic acid with a long aliphaticchain, which is either saturated or unsaturated. Most naturally occurring fatty acids have a chain of an even number of carbon atoms, from Fatty acids are usually derived from triglycerides or phospholipids. When they are not attached to other molecules, they are known as "free" fatty acids. Fatty acids are important sources of fuel because, when metabolized, they yield large quantities of ATP.

1.1.2.6 Amino acid

Amino acids are the chemical substances that make up protein. All proteins, are chains of chemical units linked together to make one long molecule. These units are called amino acids. There are about twenty types of amino acids. Some of them are essential and some are non-essential. Human body can not synthesize essential amino acids and it must take from the diet for the maintenance of good health. Essential amino acids are isoleucin, lysine, tryptophan, valine, leucine, threonine, phenylalanine, methionine.Fish protein is an excellent source of these amino-acids especially lysine and methionine are generally found in high concentrations in fish proteins, in contrast to cereal and meat proteins(Chandrashekar and Deosthale, 1993).

1.1.3 The minor components of fish muscle

1.1.3.1 Carbohydrates

The amount of carbohydrate in fish muscle is generally too small. In white fish the amount is usually less than 1 percent, but in the dark muscle of some fatty species it may occasionally be up to 2 per cent. Somemolluscs, however, contain up to 5 per cent of the carbohydrate glycogen (Love, 1970).

1.1.3.2 Vitamins

These include a range of substances widely different in character that must be present in the diet, even if only in minute quantities. It requires not only to promote good health but also to maintain life itself. Fish does provide a well-balanced supply of minerals in a readily usable form.Vitamins can be divided into two groups, those that are soluble in fat, such as vitamins A, D, E and K, and those that are soluble in water, such as vitamins B and C. All the vitamins are necessary for good health.

Water-soluble vitamins in fish mainly present in the skin. But in the liver and the gut water soluble vitamins are more uniformly distributed, and the flesh usually contains more than half the total amount present in the fish. The roe is also a good source of these vitamins.The mineral and vitamin content of fish is not markedly affected by careful processing or by preservation, provided storage is not very prolonged.

1.1.3.3 Minerals

Minerals mainly concentrated in fish bone. Some elements such as boron, fluorine, bromine, lithium, strontium are present in greater concentration in both marine and fresh water fish. Generally fish muscle contains 1-2% minerals.

The principal minerals are Ca, Mg, K, Na, P, Fe, S, Cl, Cu, Mn, I, Br. Some trace elements also present in small quantity. Such trace elements may be Sr, Hg, Cd, Mo, Co, Ni, Al, Ba, Zn etc.

1.2 Rationale

The main part of fish consumed by human is generally muscle. Muscle contains protein, lipid, carbohydrate, moisture, ash, minerals etc. Due to the known variation of protein, lipid, moisture, ash, dry matter proximate analysis is done. Factors affecting fish proximate composition can be either endogenous or exogenous. The endogenous factors are genetically controlled and are associated with the life cycle of fish. Throughout the year, fish are subjected to considerable environmental changes and fluctuations in availability and compositions of feed that will affect their proximate muscle composition. Many other exogenous factors (temperature and salinity) may also affect the proximate and fatty acid compositions. The composition of fish muscle also varies greatly depending on species, sexual cycle, age, feed, stage of maturity, environment, season, organs and muscle location.A few quantitative studies have also indicated that body constituents and energy resources vary with seasonal life cycles (Puwastien*et al.,* 1999).

However, the flesh protein contents less influenced by external feeding since it is mainly dependent on intrinsic factors such as thefish species, culture environment, and size (Shearer, 1994). It is widely believedthat the wild fish acceptability is better than thatof farmed fish. Cultured fish is provided with nutrient rich foods in addition to natural productivity in the pond.

Captured fish on theother hand has to depend totally on natural food for its sustenance. These variations have direct bearing onbody composition, health status and growth of fish. Body composition is therefore, a true reflector of itsfeeding habits and type of food availability (Ashraf *et al.*, 2011). Tadros*et al.* (2005) reported that majorpercentage of lysine is found in fish fillets.Importance of understanding the body composition of fish in relation to growth andreproduction has long been recognized. Proximate composition is used as an indicator of fish quality and it varies with diet, feed rate, genetic strain and age (Austreng and Refstie, 1979).

Fish lipids have been recognized as being beneficial for human health. Many studies have been carried out on the metabolism and function of polyunsaturated fatty acids (PUFAs) in general and on the levels and ratios of n-3 and n-6 fatty acids in particular. Long chain polyunsaturated fatty acids (PUFAs) like linoleic acid, arachidonic acids, α-linolenic acid, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are essential fatty acids (EFA's) cannot be synthesized by humans and required in the diet (Hunter and Roberts, 2000, Sushchik*et al.*, 2007). Today it is known those n-3 fatty acids or a balancedn–3/n-6 ratio in the diet, are essential for normal growth and development and may play an important role in the prevention and treatment of coronary artery disease, diabetes, hypertension and cancer. It is thus important, for human health, to increase the consumption of fish or fish, which are rich in polyunsaturated fatty acids of the n-3 family and poor in polyunsaturated fatty acids of the n-6 family.

Various fish species do not provide the same nutrient profile depending on the seasons and cultured environment. A good number of works has already been done on proximate composition of different freshwater and marine fishes (Mazumder*et al.,*2008, Stansby, 1954,Hossain*et al.,* 2015). Moreover Onyia*et al.*(2013) made a thorough study on variation on proximate composition depending on cultured environments.Commercial feed trials and its effect on proximatecomposition is done several researchers (Brahney*et al.*, 1981, Aursand*et al.,* 1994,Badiani*et al.,* 1996).AgainYeganeh*et al.*(2012), Hassan *et al.*(2010),Sigurgisladottir and Palmadottir (1993)worked on fatty acid and amino acid profiles and their distribution of different habitat fish.

But there is no combinedwork which compares seasonal variation of proximate composition, fatty acid and amino acid profile depending on cultural environment.In view of these facts, the aim of this study is to determine seasonal variations of the proximate, fatty acid composition and n-3/n- 6 ratios and amino acid profile of wild and cultured *Anabas testudineus.*

1.3 Objectives

The overall objective of the thesis work is to know theseasonal variations in proximate, fatty acid and amino acid profile of wild and cultured *Anabas testudineus.*

To fulfill the main objective specific objectives are-

i. to know the length-weight relationship of wild and cultured *Anabas testudineus;*
ii. to analysis proximate composition of wild and cultured *A. testudineus;*
iii. to determine total fatty acid composition of wild and cultured *Anabas testudineus* and
iv. to determine amino acid profile of wildand cultured *A. testudineus.*

2 MATERIALS AND METHODS

2.1 Experimental Fish

The experimental fish,*Anabas testudineus* is grayish green in color and has brown fins. It grows up to 9 inches and is a very hardy fish, due to the presence of accessory respiratory organ. It is known to survive for 8 years in captivity (Flower, 1925). Basically it is a carnivorous fish. Body is covered by ctenoid scales. Lateral line sense organ is identified by the black spots as conspicuous one at the base of the caudal fin.

*Anabastestudineus*can survive out of water for about 6-10 hours (Hughes and Singh, 1970). During dry seasons, the fish burrows in the mud and is in resting phase. The fish travel in troops on the ground, during early morning and at times of rain storm. This is a migratory fish, migrating from one pond to another during rainy season for spawning.

(a) **(b)**

Figure 1: (a) Wild *Anabas testudineus*, (b) Cultured *Anabas testudineus*

Anabas testudineus (koi fish) was selected for the purpose of experiment dueto several reasons. The most common reasons are as follows.

- The climbing perch have a good market demand
- It has more nutritious value

- They can tolerate high stocking density
- They can tolerate poor oxygen level
- Comparatively moderate in market price

2.1.1. Scientific Classification of *Anabas testudineus*

Kingdom : Animalia

Phylum : Chordata

Class : Actinopterygii

Order : Perciformes

Suborder : Anabantoidei

Family : Anabantidae

Genus : *Anabas*

Species : *A. testudineus*(Bloch, 1795)

2.2 Collection of FishSample and Study Period

Both the wild and cultured koi fishes were collected fromJessore district, Bangladesh. Samples were collected in two season; pre-monsoon (March-May)and post-monsoon(September-November). Wild koi fish was collected from Jhapabaor. JhapaBaor is located at the Rajgonj bazaar of Monirampurupazila inJessore district. The shape of the Baor is almost oval. Just has a mouth at the western side. GPS Coordinate of the JhapaBaor is (22°59'1.34"N, 89° 9'36.39"E). Considering the other baors from Jessore district, this one uses to have water more or less at every season. As a result many indigenous fish species like Koi, Puti, Shing. Mola, Dhela, Chapila, Tengra, Kankilaare available thought the year. It plays an important role in economy and biodiversity.The cultured koi (Thai koi) fish were collected from Matri Fish Hatchery and Integrated Farm, Monirampur, Jessore. After collecting the sample it was transported to Aquatic Laboratory of Department of fisheries, University of Dhaka by insulated ice box. The samples were refrigerated until analysis is done.

Plate-1

Sample Collection area

A. JhapaBaor, Jessore, Bangladesh (Wild Koi fish collection area)

B. Matri fish Hatchery and Integrated Farm, Chachra, Jessore, Bangladesh (Cultured Koi fish collection area)

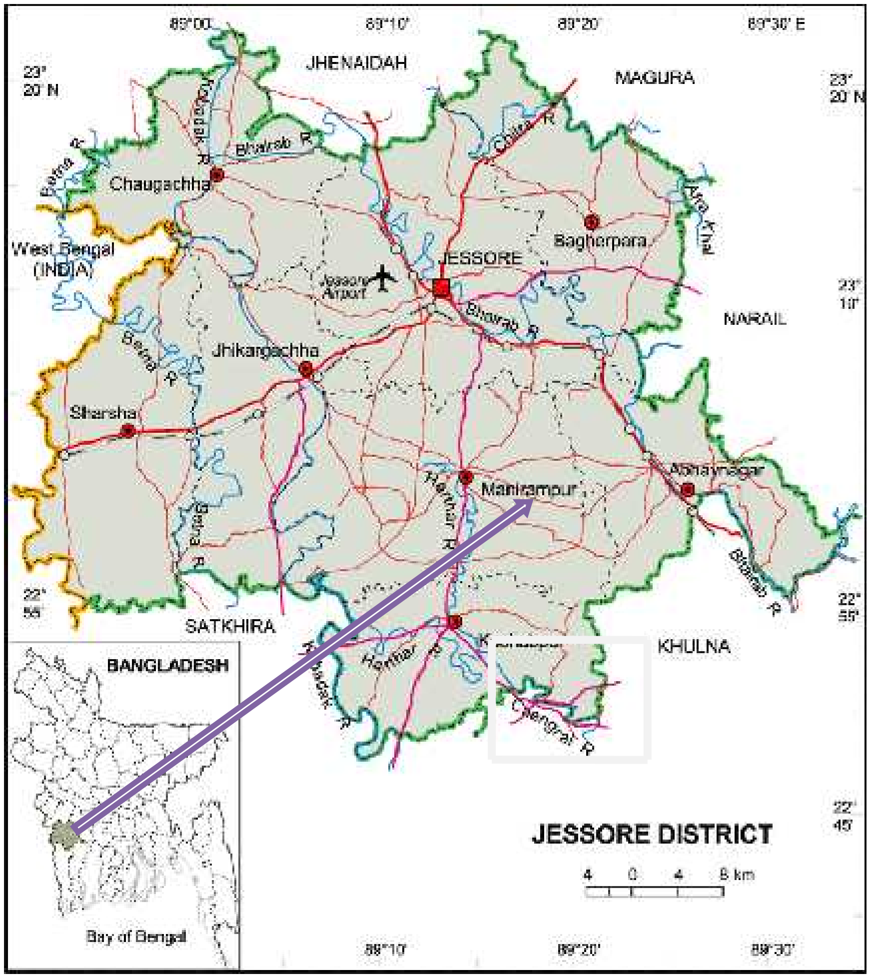


Figure 2: Map of study area

2.3 Sample Analysis

The sample analysiswas carried out in Aquatic laboratory of Fisheries Department,University of Dhaka, and BCSIR(Bangladesh Council of Scientific and Industrial Research), Dhaka, Bangladesh.

2.3.1 Determination of Length-Weight Relationship

The length-weight relationships were determined separately for sample fish, *Anabas testudineus*collected in two different seasons by the linear equation Y = a + bX, where X= independent variable, Y = dependent variable and 'a' and 'b' are constants. For this purpose, the observed values of individual fish were measured and regression analysis was carried out to find out the correlation of coefficient (r) for length and weight. The variation in wild and cultured *Anabas testudineus*was calculated using ANOVA (Analysis of covariance).

2.4 Determination of Nutritional Compositions

The proximate composition of the fish muscle was determined by various methods. Like, Moisture content will be determined by Oven drying method (AOAC, 1995);ash content by Gravimetric method; Crude protein content by using Micro-Kjeldhalmethod, lipid content by Folch method (1957). Amino acid profile and Fatty acid profile were done respectivelyby High Performance Liquid Chromatographic (HPLC) and Gas Chromatographic (GC) analysis.

2.4.1 Determination of Moisture

Principle

Moisture content determined as loss of weight and it was done by oven drying method (AOAC, 1995).

Procedure

Dried the empty Aluminium foil cup (15 minutes) and transferred to the desiccators to cool then weighted

⇩

About (3-5) g sample was taken in each foil

⇩

Weighted the foil with sample and noted

⇩

Dried the sample for overnight (16 hours) at 102^0C

⇩

Removed the dish from oven

Calculation

The percentage of moisture was calculated by the following equation:

$$\text{Moisture (\%)} = \frac{\{(w_1 - w_0) - (w_2 - w_0)\}}{(w_1 - w_0)} \times 100$$

Where,

Weight of the foil cup = W_0

Weight of the foil+ wet sample = W_1

Weight of the foil + dry sample = W_2

Dry matter = 100 - Moisture

$$\text{Moisture factor} = \frac{100 - \text{Moisture}}{100}$$

2.4.2 Determination of ash

Principle

Organic matter is burned off as low temperature as possible and the remaining inorganic material is called the ash content

Procedure

Dried the empty crucibles and transferred to the desiccator then weighted

⇩

5g of sample is transferred in the desiccator and dried overnight at 102^0C.

⇩

Sample is dried in the burner.

⇩

Placed the crucibles inside the muffle furnace, as near to the center as possible and ash overnight at 550^0C.

⇩

Crucibles and cool in a desiccator then reweighted it.

Calculation

Ash content of the fresh sample (%) = $\left(\frac{W2-W0}{W1-W0} \times 100\right) \times$Moisture factor

Where,

Weight of dry crucible = W_0

Weight of dry crucible + dry fish sample = W_1

Weight of drycrucible + ash = W_2

2.4.3 Determination of Protein

Principle

The Protein content may be calculated by multiplying the nitrogen value by 6.25. Non-Protein Nitrogen may present in the material. So, it is called crude protein. Crude protein content was determined by Micro-Kjeldhal method.

The Principle of Micro-Kjeldhal method based on the conversion of nitrogen of Protein into $(NH_4)_2SO_4$

$$\text{Protein} + H_2SO_4(\text{conc.}) \rightarrow H_2SO_4$$

In digestion chamber NH3 liberates by addition of excess NaOH and Steam distillation of NH3 Produced into saturated boric acid.

$$(NH_4)_2SO_4 + 2NaOH \rightarrow 2NH_3 + Na_2SO_4 + 2H_2O$$

$$NH_3 + H_3BO_3 \rightarrow NH_4 + H_2BO_3^-$$

Determination of the NH3 liberated by the back titration to the endpoint with saturated HCl. An indicator in the pH of 5-6 is required Phenopthelin can be used as an indicator.

$$H_2BO_3^- + H^+ \rightarrow H_3BO_3$$

Calculation

The Percentage of nitrogen was calculated from the formula.

$$\text{Nitrogen} = \frac{(S-B)\times A\times 14\times C\times 100}{\text{Weight of sample}}$$

Where,

S = Titration reading for sample

B = Titration reading for Blank

A = Strength of 0.01 N HCl

C = Digest taken for distillation (dilution factor) ≈ 20

Procedure

0.5g dry powdered sample was taken in a kieldhal flask

⇩

1g digestion mixture and 25ml concentrated H_2SO_4 was added in kjeldhal flask

⇩

Kjeldhal flask was set in the digestion chamber for 2.5-3hr unitl the solution becomes colorless

⇩

Digested product was transferred to 100 ml volumetric flask and was made up to 100 ml with distilled water

⇩

At this time heat was produced. so cooled it in the refrigerator for decreasing the temperature at room temperature

⇩

Three new kjeldhal flask and three new conical flask was taken

⇩

5 ml diluted solution, 10ml NaoH, 150 ml distil water was added in kjeldhal flask

⇩

Conical flask 5 ml boric acid and 2 drop Phenopthaline was taken

⇩

Transferred the Kjeldhal flask and conical flask in the kjeldhal distillation unit

⇩

Kjeldhal flask was boiled for 30 minute and condensed water vapour was collected in the conical flask

⇩

Titrated the solution in the conical flask against 0.01 N HCluntil the color becomes pink

⇩

The same procedure was be done for the blank sample

2.4.4 Determination of Lipid content

Principle

The lipid content was determined by extraction with a mixture of chloroform: methanol (2:1). The mixture was allowed to stand overnight and lipid portion was transferred to a weighted beaker and heated to dryness. Amount of lipid content was calculated from the difference of two weights

Procedure

5 g dry, homogenized fish muscle was taken in a conical flask

⇩

Then added chloroform: methanol (2:1) mixture until the sample submerged

⇩

Kept it over night

⇩

Then filtered it through a filter Paper and collected into a Pre weighted conical flask

⇩

Conical flask was kept in the water bath until the solution was fully evaporated and only oil remains in the flask

⇩

Finally weighed the conical flask.

Calculation

$$\text{Percentage of lipid} = \frac{W_2 - W_1}{weight\ of\ sample} \times 100 \times Moisture\ factor$$

Where,

W_2 = Dry weight of conical flask

W_1 = Weight of empty Conical flask

Plate-2

Determination of Moisture and ash

A. Preparation of sample for moisture content.

B. Sample in the oven for overnight drying

C. Sample in the Muffle furnace to convert into ash

D. Weighing the crucible for ash determination.

Plate-3

Determination of Crude Protein

A. Sample preparation for Protein determination in Kjeldhal flask.

B. Protein distillation chamber

C. Titration for Protein determination

D. Samples ready for protein determination

Plate-4

Determination of crude lipid

A. Preparation of Sample for lipid content with chloroform: Methand (2:1)

B. Filtration of sample after keeping overnight

C. Evaporation in the water bath

D. Sample ready for lipid determination

2.4.5 Amino acids determination

Principle

The profile of amino acids was done following high performance thin layer chromatographic (HPTLC) method(Anomynous, 1993). The HPTLC profile of amino acids in the muscle of the koi fish was analyzed.

Procedure:

0.5g fish muscle was pasted with 50ml 6N HCl by mortar pestle

⇩

Solution was filtered

⇩

Hydrolyzed approximately for 22-24hr in a hydrolyzer apparatus

⇩

HCl was removed from filtrate by evaporating in a water bath

⇩

Solution was volume up to 25ml in a volumetric flask by 0.1N HCl. This stock solution was used for determining amino acids

⇩

Solution was passed through micro filter (0.45mm) to remove any foreign particle if present

⇩

2-3mm of sample is placed in auto injector of HPLC

⇩

Standard amino acids were analyzed simultaneously

⇩

Based on the retention time and peak area of the standard amino acids, each amino acid in the unknown sample was identified

2.4.6 Fatty acids determination

Principle

Fat and fatty acids are extracted from fish by hydrolytic method. Fat is extracted into ether, and then methylatedto fatty acid methyl esters (FAMEs).FAMEs are quantitatively measured by gaschromatography. The profile of fatty acids was done following gas chromatographic (GC) method (Nichols *et al.,* 1995). Fattyacids were obtained from lipids by saponification using NaOH dissolved in methanol H_2O mixture (hydrolysiswith alkali).

The flow chart below represents typical procedure for analysis of fatty acid.

Fat

⇩

Hydrolysis

⇩

Methylated to FAMES

⇩

GC quantification

Procedure

Sample was methylated into fatty acid methyl ester using HCl and methanol mixture, which can be easily identified by gas chromatography

⇩

Fatty acid methyl ester was separated using mixture of hexane andanhydrous diethyl ether

⇩

For the organic phase aqueous NaOH was used as base wash and the upper organic layer was separated

⇩

(2-3) µl of sample was injected and analyzed using Gas chromatography, with capillary column and flame ionization detector

⇩

The chromatogram was used for calculation

⇩

Standard fatty acids were analyzed simultaneously

⇩

Based on the retention time and peak area of the standard fatty acids, each fatty acid in the unknown sample was identified

2.5 Statistical analysis

Statistical analysis was performed with the Statistical Package for the Social Sciences (SPSS) v. 20.0 software package (SPSS, SAS Institute Inc. Gary, USA) and Microsoft office excel 2007. The data were analyzed to determine the descriptive statistics such as Mean, Standard Error of Mean, Standard Deviation, Minimum and Maximum value. Multiple comparisons were done with Tukey HSD test with one way ANOVA (Analysis of Variance) at 5% level of significance.

Plate-5

Sample preparation before run in HPLC (Amino Acid Profile)

A. Sample preparation for Amino Acid Profile

B. Sample in the heating mental for hydrolysis

C. Dilution of sample

D. Micro-filter (0.45mm)

Plate-6

Sample run in HPLC (Amino Acid Profile)

A. Sample Passed through microfilter to remove any tiny foreign particle

B. Sample ready to run in HPLC.

C. Chemicals used to run sample in HPLC

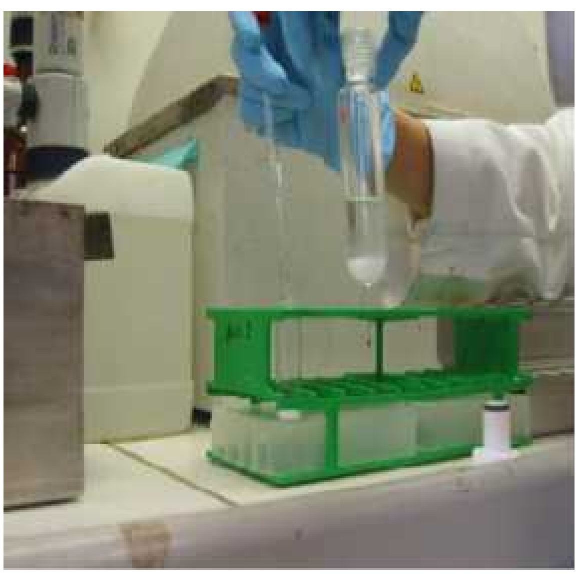
D. Sample in HPLC

Plate-7

Sample preparation before run in GC (Fatty Acid Profile)

A. Extraction of fat from lipid.

B. Methylation of sample for fatty acid profile



C. Collection of top layer after separation

D. Sample ready in vial for GC analysis

Plate-8

Sample run in GC (Fatty Acid Profile)

A. Injection Syringe (UL)

B. Gas Chromatography

C. Sample ready for fatty Acid analysis

D. Injecting (0.2-0.3) µl sample in injection pore of GC

3 RESULTS

3.1 Length-Weight Relationships of Wild and Culture *Anabas testudineus*

For the wild *Anabas tuestudineus,* the total length and weight were ranged from 7.10 to 10.30 cm and 20.31 to 28.84 g respectively collected in pre-monsoon period (March to May), whereas the total length and weight ranged from 9.8 to 13.80 cm and 26.82 to 32.61 g respectively collected in post-monsoon period (September-November)(Appendix A-Table 1). The mean total length for *Anabas testudineus* was calculated as 8.72±1.08 cm and the mean total weight calculated as 25.11± 2.87 g (n=10) collected in per-monsoon, whereas mean total length was calculated as 11.99±1.38 cm and the mean total weight calculated as 30.00± 2.18 g collected in post-monsoon season (Appendix A-Table 2).

In case of cultured *Anabas testudineus,* the total length and weight were ranged from 7.80 to 10.60 cm and 33.81 to 42.76 g respectively collected in pre-monsoon period, whereas the total length and weight ranged from 7.10 to 13.80 cm and 41.56 to 52.84g respectively collected in post-monsoon period (Appendix A-Table 1). The mean total length for *Anabas testudineus* was calculated as 9.25±0.93 cm and the mean total weight calculated as 38.21± 3.06g collected in pre-monsoon time, whereas mean total length was calculated as 10.49±1.91 cm and the mean total weight calculated as 47.90± 4.62 g (n=10) collected in post-monsoon period (Appendix A-Table 2).

The value of correlation coefficient, r = 0.992 and 0.985 (Figure 3)for wild *Anabas testudineus* collected in two different times as pre-monsoon and post-monsoon respectively, and the correlation coefficient values implies that there is was a strong positive association between the length and weight of the samples. In case of cultured *Anabas testudineus*, the value of correlation coefficient, r = 0.973 and 0.963 (Figure 4) collected in pre-monsoon and post-monsoon period respectively, and the correlation coefficient values implies that there is also a strong positive association between the length and weight of the samples. The findings of the present study also revealed that, there were significant differences ($p<0.05$) in length-weight relationship of wild and cultured koi fish, collected in two different time period.

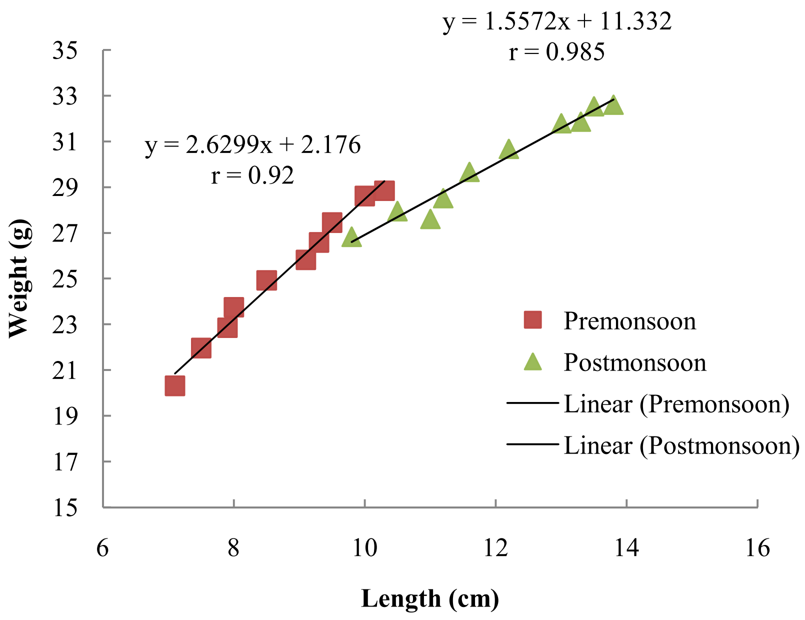


Figure 3: Length-Weight relationship of the wild *Anabas testudineus* collected in two different seasons (Pre-monsoon and Post-monsoon)

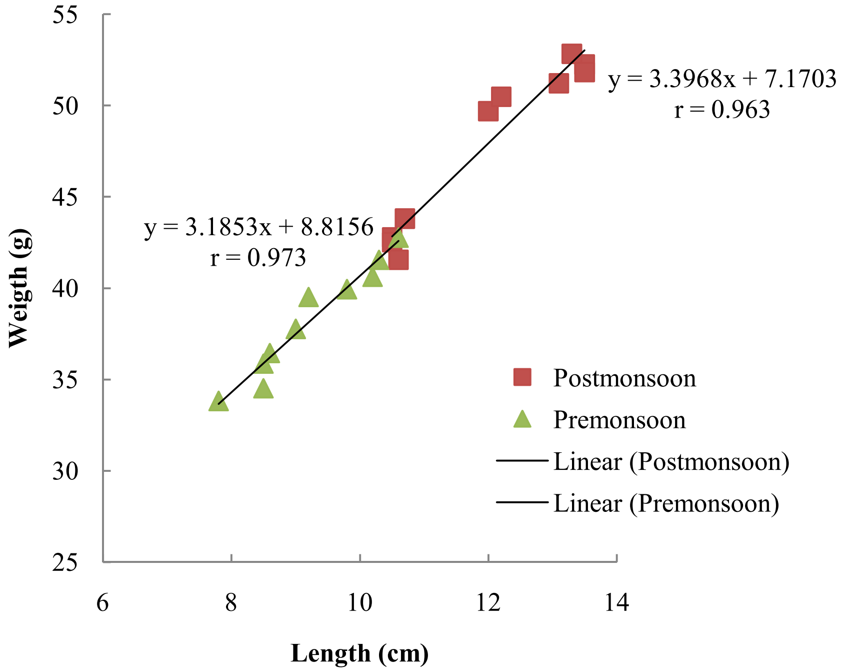


Figure 4: Length-Weight relationship of the cultured *Anabas testudineus* collected in two different seasons (Premonsoon and Postmonsoon)

3.2 Nutritional Composition

A comparative study was undertaken to determine the moisture, ash, protein, lipid, amino acid and fatty acid profile in wild and cultured koi fishes of Jessore district, Bangladesh. Depending on the season, results showed a wide variation in proximate, amino acid and fatty acid profile of wild cultured koi fish.

3.2.1 Moisture content

In our present study seasonal variation is observed in moisture content of both wild and cultured Koi fish. Mean moisture content (Mean±SEM) of the wild *Anabas testudineus*was 79.62±0.29% and 75.27±0.05%, collected in pre-monsoon and post-monsoon season whereas in cultured fish the amount of moisture content in both season was 74.04±0.18% and 73.91±0.16% respectively (Appendix B-Table 3). Seasonally the highest value was found in pre-monsoon wild (79.62±0.29%)koi fish and the lowest value is in cultured fish (73.91±0.16%)of post-monsoon season (Appendix B-Table 3). In both season the moisture content ranged between 73.06-80.02-% (Appendix B-Table 4).

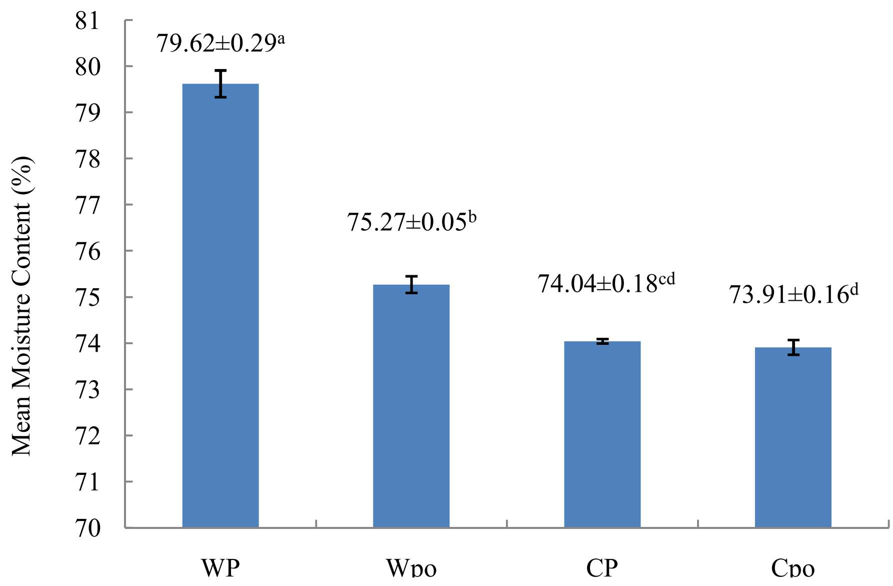


Figure 5: Variation of moisture content (%) of wild and cultured fishes in pre-monsoon and post-monsoon. Different lettersdenote significant differences (p< 0.05) and same letter denotes no significance (p>0.05). (WP=Wild_Premonsoon, Wpo=Wild_Postmonsoon, CP=Cultured_Premonsoon, Cpo=Cultured_Postmonsoon).

Resultaslo represented that moisture content differed significantly (p <0.05) in both season (pre-monsoon and post-monsoon) between wild and cultured koi fish. Wild fish contains higher amount of moisture than cultured fish (Figure 5). Significant difference (p < 0.05) also observed when compare wild fish of both season (Appendix B-Table 6). Wild fish in pre-monsoon season contains higher amount of moisture (Figure 5). But no significant difference (p> 0.05) in moisture content was observed between cultured fish of both seasons (Appendix B-Table 6).

3.2.2 Ash content

Inpre-monsoon, it was found that thewild and cultured koicontains3.62±0.02% and 2.50±0.03% ashrespectively on the contrary in post monsoon the amount was 3.02±0.01% and 2.90±0.03% (Mean±SEM) (Appendix B-Table 3). Ash content was found to vary from (2.37-3.71) %(Appendix B-Table 4). Maximum ash content was recorded in pre-monsoon wild (3.62±0.02) % and the lowest value was found in post-monsoon cultured (2.50±0.03) % koifish(Appendix B-Table 4).

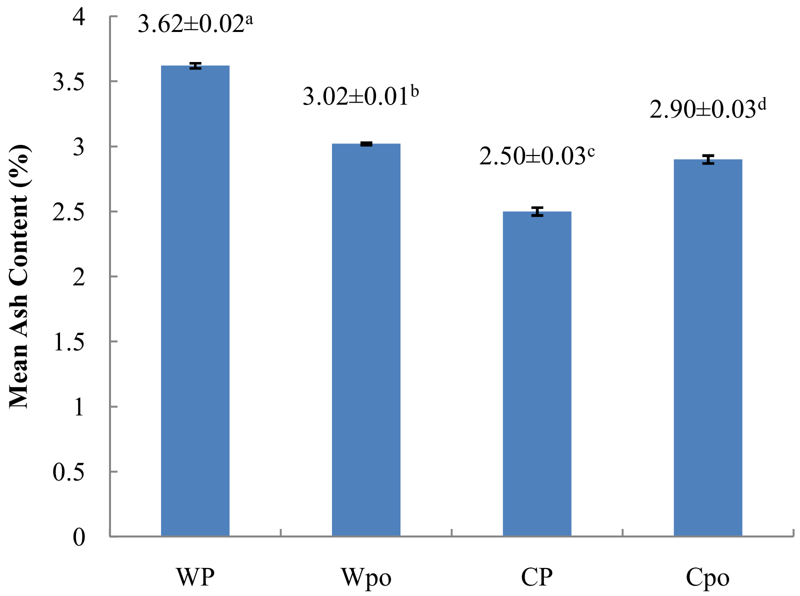


Figure 6: Variation of ash content (%) of wild and cultured fishes in pre-monsoon and post-monsoon.Different lettersdenote significant differences (p < 0.05).

Analysis of the data revealed thatsignificant difference is observed(p<0.05) in both season (pre-monsoon and post-monsoon) between wild and cultured *Anabas*(Appendix B-Table 6). Wild fish contains higher amount of ash when compared with cultured fish (Figure 6). Significant difference (p<0.05) also observed between wild fish of both season(Appendix B-Table 6) as wild fish contains higher amount of ash in post-monsoon than in pre-monsoon (Figure 6). When compare percentage amount of ash in cultured fish of both seasons italso showed significant difference ($p < 0.05$) (Appendix B-Table 6).Cultured fish contains higher amount of ash in post-monsoon than in pre-monsoon (Figure 6).

3.2.3 Crude Protein Content

Findings described that mean crude protein content of wild fishes in pre-monsoon and post-monsoon was respectively 12.13±0.01% and 14.74±0.02% (Appendix B-Table 3).On the other hand cultured fish contains 16.04±0.02 % of crude protein in pre monsoon and 17.92±0.02 % in post-monsoon period (Appendix B-Table 3). The highest value was recorded in post-monsoon cultured fish (17.92±0.02%) and the lowest value in wild fish (12.13±0.01%) of pre-monsoon (Appendix B-Table 1). Seasonally protein content may vary ranged between 12.09-17.96% (Appendix B- Table 4).

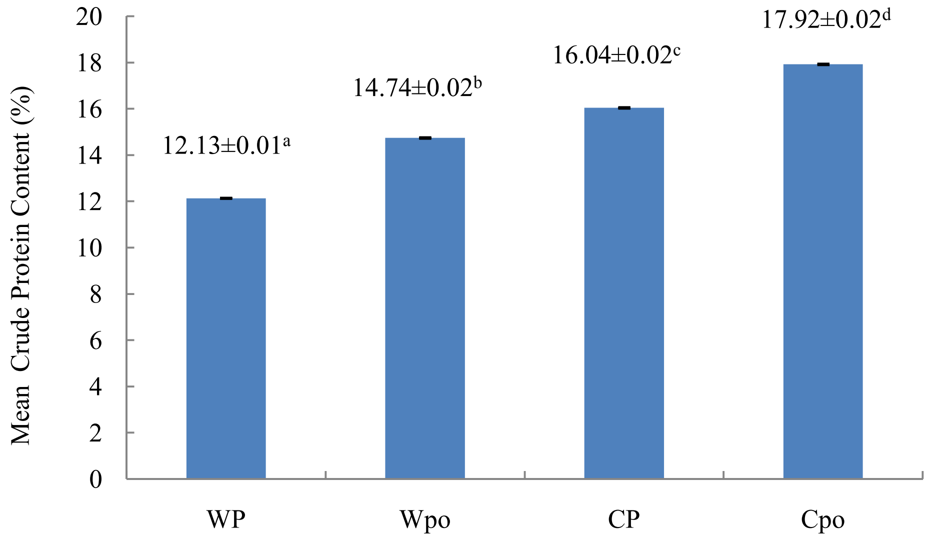


Figure 7: Variation of protein content percentage (%) of wild and cultured fishes in pre-monsoon and post-monsoon.Different lettersdenote significant differences ($p < 0.05$).

Result of crude protein content stated that significant difference ($p < 0.05$) remains in wild fish of both season (Appendix B_ Table 6). In post-monsoon period the amount of protein content is higher than pre-monsoon (Figure 7). Analysis of the data also illustrated that significant difference ($p< 0.05$) was observed in cultured fishesof both season(Appendix B-Table 6). Cultured fish of postmonsoonperiod contained higher amount of crude protein than in pre-monsoon (Figure 7). Again significance difference ($p< 0.05$)was observed betweenboth seasons (pre-monsoon and post monsoon) and between cultured and wild fish (Appendix B-Table 6). Percentage amount of protein is higher in cultured than in wild fish of both season (Figure 7).

3.2.4 Lipid Content

Result of lipid content narrated that lipid content may vary between 3.56-5.61 % in pre-monsoon and post-monsoon (Appendix B-Table 4).Findings described that in pre-monsoon mean lipid content was 3.82±0.02% and 5.14±0.01% in wild and cultured fish respectively on the contrary in post monsoon the amount were 3.61±0.02% and 5.56±0.01% respectively Appendix B-Table 3). Among these result the highest value of lipid content was marked in cultured fish of post monsoon season (5.56±0.01) % as the lowest was in wild fish of pre monsoon (3.61±0.02) % (Appendix B-Table 4).

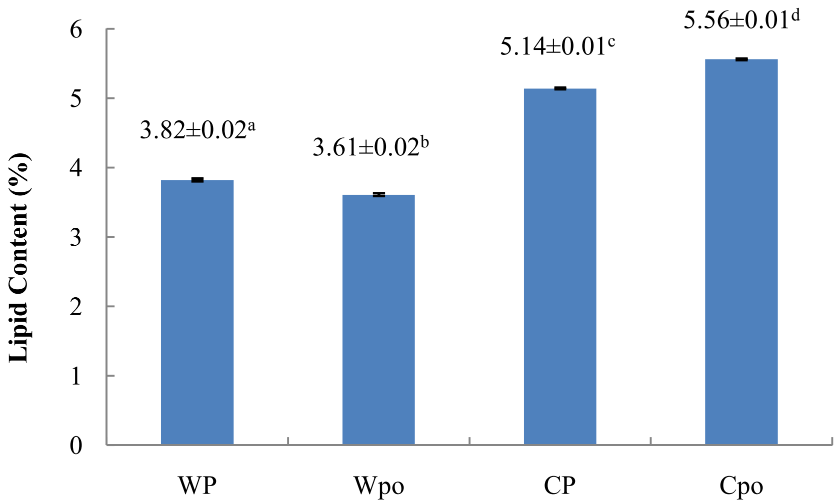


Figure 8: Variation of lipid content percentage (%) of wild and cultured fishes in pre-monsoon and post-monsoon. Different lettersdenote significant differences ($p < 0.05$).

The amount of lipid content exposed that significant difference ($p < 0.05$) resides in wild and cultured fish of both season (pre-monsoon and post-monsoon) (Appendix B-Table 6). Total amount of lipid is higher in cultured fish than wild (Figure 8). Data also expressed that significant difference ($p < 0.05$) is observed in wild fish of both season (Appendix B-Table 6). In pre-monsoon the amount of lipid content is higher (Figure 8). Moreover both seasons cultured fish revealed significant difference ($p < 0.05$)(Appendix B-Table 6). Results appeared that lipid content is higher in post monsoon cultured fish than pre-monsoon.

3.3 Amino Acids

The profiles of amino acids and have also been analyzed to assess and differentiate nutritional quality of cultured and wild *Anabas testudineus*. Amount of amino acid in protein varied seasonally in both wild and cultured fish.

3.3.1 Threonine

Findings described that in pre-monsoon amount of threonine was 0.277±0.003% and 0.437±0.003% (Mean±SEM) in wild and cultured fish respectively on the contrary in post monsoon the amount were0.320±0.01% and 0.460±0.023% (Appendix C-Table 7). Among these result the highest value was marked in cultured fish as the lowest was in wild fish in both season (Appendix C-Table 7).

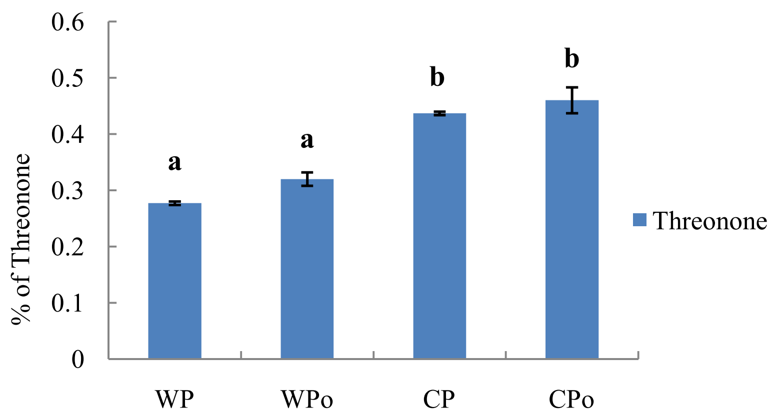


Figure 9: Variation of Threonine (%) in wild and cultured fishes of both seasons (WP=Pre-monsoon wild; WP_o=Post-monsoon wild and CP=Pre-monsoon cultured; CP_o=Post-monsoon cultured). Different lettersdenote significant differences ($p < 0.05$) and same letter denotes no significance ($p>0.05$).

In addition, significant difference ($p < 0.05$) resides in wild and cultured fish of both seasons (pre-monsoon and post-monsoon)but irrespective of the season there were no significance difference (p> 0.05) within the wild and cultured koi fishes (Figure 9, Appendix C-Table 10).

3.3.2 Valine

Findings described that content ofvaline in wild *A. testudineus*in pre-monsoon and post-monsoon was respectively 0.090±0.012% and 0.100±0.006 %. On the other hand cultured fish contains 0.110±0.01% and 0.140±0.017% valinein pre-monsoon and post-monsoon (Appendix C-Table 7). The highest value was recorded in post-monsoon cultured fish and the lowest value in pre-monsoon wild fish but there remains no significant difference (*p*>0.05) in the amount of valine in both seasons and culture habits (Figure 10, Appendix C-Table 10).

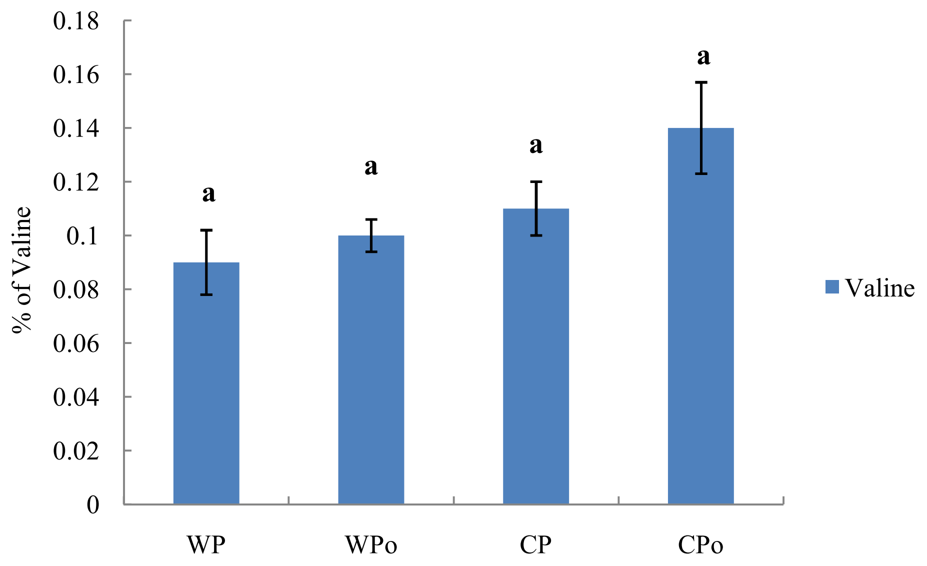


Figure 10: Variation of valine (%) in wild and cultured fishes of both seasons. Same letter denotes no significance (p>0.05)

3.3.3 Methionine

Amount of Methionine in wild fish0.313±0.017 % and 0.350±0.012 % in pre-monsoon and post-monsoon whereas in cultured fish the amount was 0.457±0.013% and 0.490±0.017% respectively. Significant difference was observed (p < 0.05) between wild and cultured *A.testudineus*of both season but on significant difference (p>0.05) is observed between wild fish (Figure 11) but there was no significant difference (p>0.05) in amount of methionine for both wild and cultured fishes based on season (Figure 11, Appendix C-Table 10).

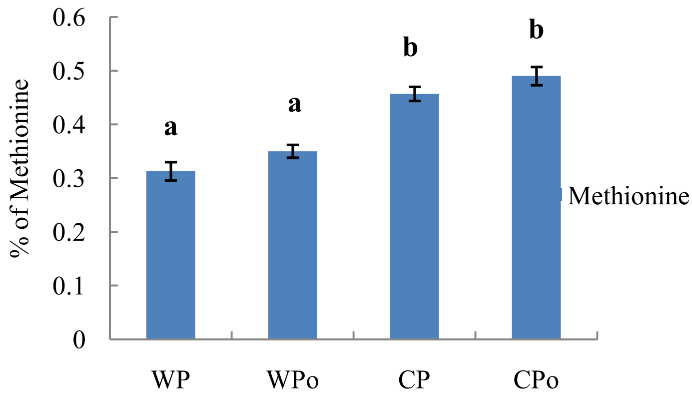


Figure 11: Variation of Methionine (%) in wild and cultured fishes of both season..Different lettersdenote significant differences (p < 0.05) and same letter denotes no significance (p>0.05)

3.3.4 Isoleucine

Mean Isoleucine content of the wild *Anabas* was 0.527±0.019% and 0.620±0.023%, collected in pre-monsoon and post-monsoon season whereas in cultured fish the amount was 0.8433±0.029% and 0.907±0.020% respectively (Appendix C-Table 7). Result illustrated that significant difference (p< 0.05) lies in wild and cultured fish of both season (pre-monsoon and post-monsoon) (Figure 12). Data also expressed that both seasons cultured and wild fish do not show significant difference (p> 0.05) among themselves in amount of isoleucine content (Figure 12, Appendix C-Table 10).

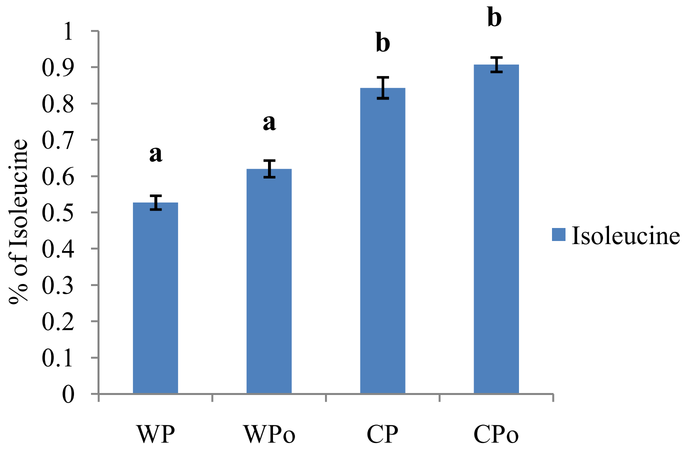


Figure 12: Variation of Isoleucine (%) in wild and cultured fishes of both seasons. Different lettersdenote significant differences (p< 0.05) and same letter denotes no significance (p>0.05)

3.3.5 Leucine

Inpre-monsoon wild and cultured fish illustrated that they contain 0.133±0.012% and ash0.170±0.01% leucinerespectively on the contrary in post monsoon the amount was 0.170 ± 0.012^{ab} % and 0.210 ± 0.006^{b} % (Appendix C-Table 7). Result expressed that significant difference (*p*> 0.05) exists between pre-monsoon wild and post-monsoon cultured koi fish(Figure 13). NoSignificant difference (p < 0.05) liesbetween wild fish of both season as well as between cultured of both season (Figure 13). Again no significant difference (*p*< 0.05) remains in post-monsoon wild and pre-monsoon cultured.

3.3.6 Lysine

In pre-monsoon wild and cultured koi fishcontains 0.300±0.015% and 0.367±0.007% lysinerespectively andin post monsoon the amount was 0.370±0.012% and 0.440±0.017% Appendix C_ Table 7). Among these result the highest value was marked in cultured fish of post monsoon season aswell as the lowest value was in wild fish of pre monsoon and they showed significant difference (*p*<0.05) (Figure 14). Again significant difference (*p*< 0.05) remains between wild fish of both season and cultured fish of both seasons (Figure 14). No Significant difference (*p*> 0.05) lies between post-monsoon wild and pre-monsoon cultured fish (Figure 14).

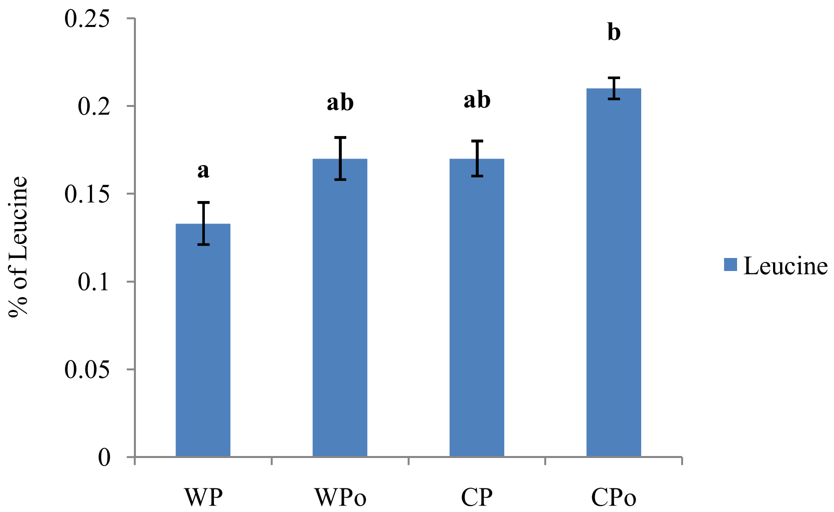


Figure 13: Variation of Leucine (%) in wild and cultured fishes of both season..Different lettersdenote significant differences (p < 0.05) and same letter denotes no significance (p>0.05)

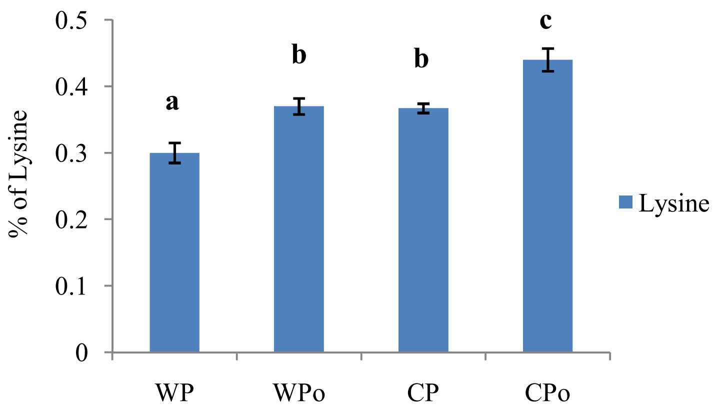


Figure 14: Variation of Lysine (%) in wild and cultured fishes of both seasons..Different lettersdenote significant differences (p< 0.05) and same letter denotes no significance (p>0.05)

3.3.7 Aspertic Acid

In pre monsoon wild and cultured experimental fish amount of Asparticacid was 0.737±0.007% and 0.890±0.01 % whereas in post-monsoon the amount was0.760±0.017% and 0.930±0.006% respectively (Appendix C-Table 7). Result described that amount of Aspartic acid is higher in cultured fish than in wild (Figure 15). Significant difference is observed (p< 0.05)among wild and cultured fish of both season but on significant difference (p>0.05) is observed between both seasons wild fish (Figure 15, Appendix C-Table 10).

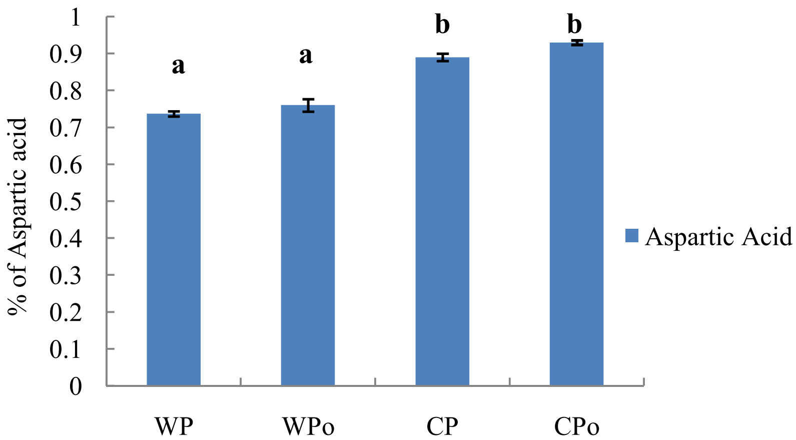


Figure 15: Variation of Aspartic acid (%) in wild and cultured fishes of both season. Different lettersdenote significant differences (p< 0.05) and same letter denotes no significance (p>0.05)

3.3.8 Serine

Cultured koi fish contains 0.360±0.023% and 0.490±0.006% serine in both pre-monsoon and post-monsoon whereas in the case of wild fish the amount was 0.323±0.009 % and 0.463±0.003% (Appendix C-Table 7). Our findings concluded that significant difference is observed ($p < 0.05$) between wild and cultured fish of both season (Figure 16). But no significant difference ($p>0.05$) is observed between wild fish of pre-monsoon and post-monsoon season and it is also applicable for both monsoon cultured fish (Figure 16, Appendix C-Table 10).

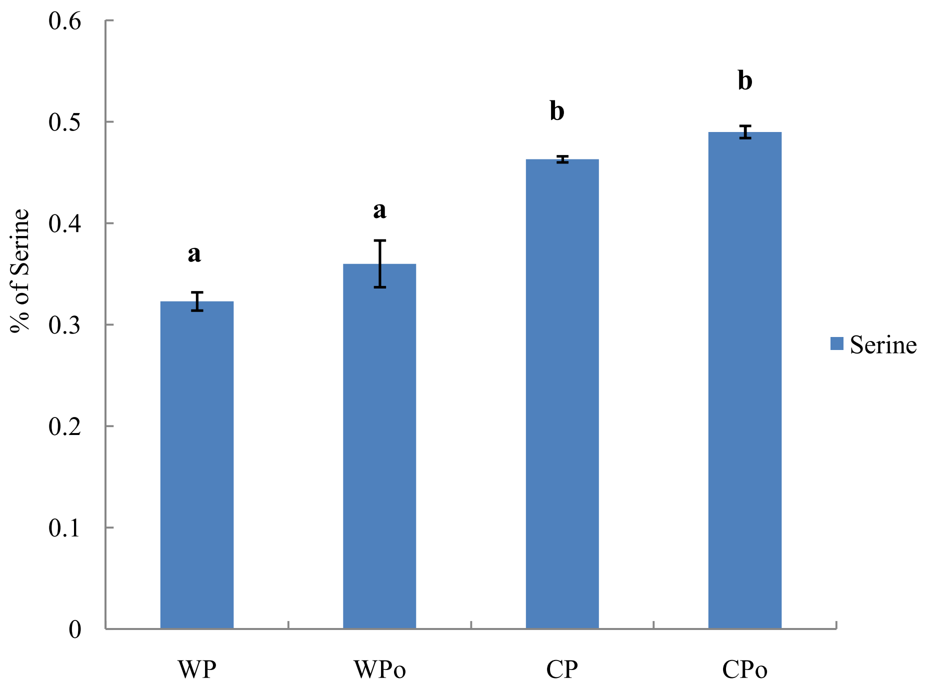


Figure 16: Variation of Serine (%) in wild and cultured fishes of both season. Different lettersdenote significant differences ($p< 0.05$) and same letter denotes no significance ($p>0.05$)

3.3.9 Glutamic Acid

Findings described that in pre-monsoonamount of Glutamic Acid was 0.990±0.015% and 1.147±0.0150 % in wild and cultured experimental samplerespectively on the other hand in post monsoon the amount was 1.040±0.023% and 1.260±0.017% (Appendix C-Table 7). Among these result the highest value was marked in cultured fish in post-monsoon as well as the lowest was in wild fish of pre-monsoon and they expressed Significant difference ($p< 0.05$)(Figure 17). Data also expressed that no significant difference ($p>$

0.05) is observed in both seasons wild fishbut significant difference (p< 0.05)lies between cultured fish of pre-monsoon and post-monsoon(Figure 17, Appendix C-Table 10).

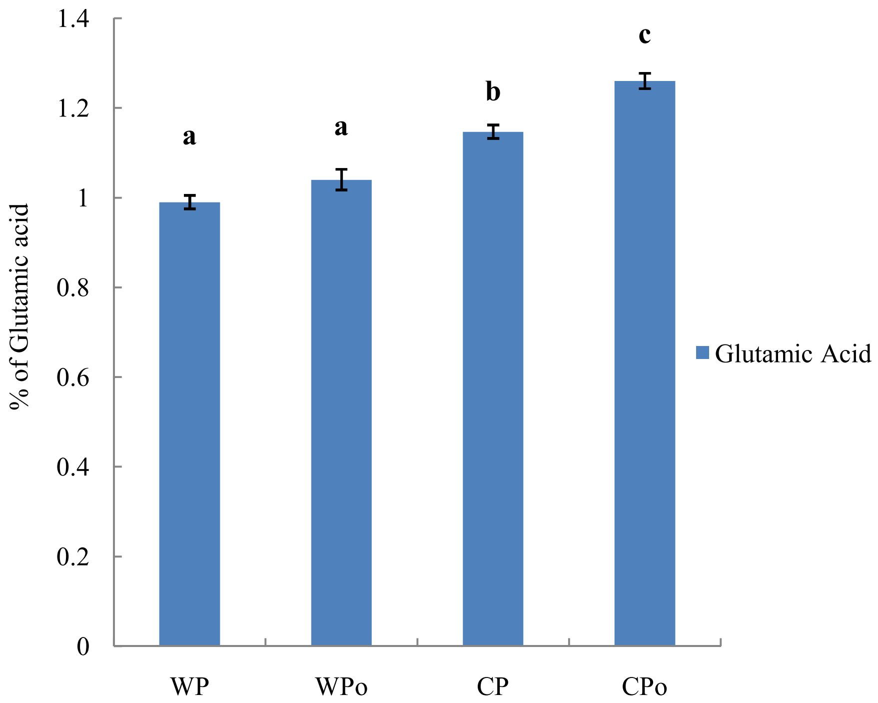


Figure 17: Variation of Glutamic acid (%) in wild and cultured fishes of both seasons. Different lettersdenote significant differences (p< 0.05) and same letter denotes no significance (p>0.05)

3.3.10 Glycine

In both cultured and wild koi fish Glycine content varied based on season (Pre-monsoon and post-monsoon). Findings described that amount of glycine in wild fishes in pre-monsoon and post-monsoon was respectively 1.290±0.015 % and 1.380±0.012% (Appendix C-Table 7). Moreover cultured fish contains 1.450±0.012 % glycine in pre-monsoon and 1.580±0.0% in post-monsoon (Appendix C-Table 7). The highest value was recorded in post-monsoon cultured fish and the lowest value in wild fish of pre-monsoon. Result represented that glycine content differed significantly (p<0.05) in both season (pre-monsoon and post-monsoon) between wild and cultured koi fish(Figure 18).Significant difference (p < 0.05) also observed when compare wild fish of both season (Figure 18). Again both seasons (pre-monsoon and post-monsoon) cultured fish exhibits significant difference (p < 0.05) in glycine content (Figure 18).

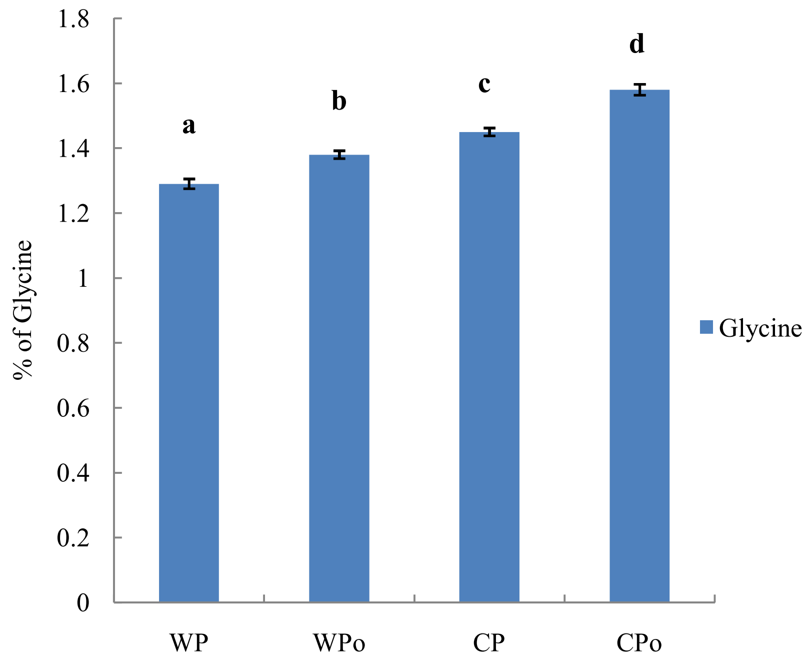


Figure 18: Variation of Glycine (%) in wild and cultured fishes of both seasons. Different lettersdenote significant differences (p < 0.05)

3.3.11 Alanine

Findings described that amount of alanine in koi fish inboth season pre-monsoon and post-monsoon was 0.397±0.012%and0.420±0.017 % respectively. On the contrary the amount was 0.490±0.01 % and 0.550±0.029% in cultured koi fish (Appendix C-Table 7). Among the findings the highest value was recorded in post-monsoon cultured fish as well as the lowest value in wild fish of pre-monsoon and significant difference (p< 0.05) exists between them (Figure 19). No Significant difference (p> 0.05) lies between wild fish of both seasons as well as between cultured of both seasons (Figure 19). Again no significant difference (p< 0.05) remains between post-monsoon wild and pre-monsoon cultured (Figure 19, Appendix C-Table 10).

3.3.12 Histidine

Cultured experimental fish contains 1.823±0.015 % and 1.870±0.006% histidine in both pre-monsoon and post-monsoon whereas in the case of wild fish the amount was 2.023±0.048% and 2.240±0.023 % (Appendix C-Table 7). Our findings concluded that significant difference is observed (p < 0.05) between cultured fish of both season (Figure 20). Again among cultured and wild koi fish significant difference is observed (*p*< 0.05) (Figure 20). But no significant difference (*p*>0.05) is observed in histidine content in wild fish of both seasons (Figure 20).

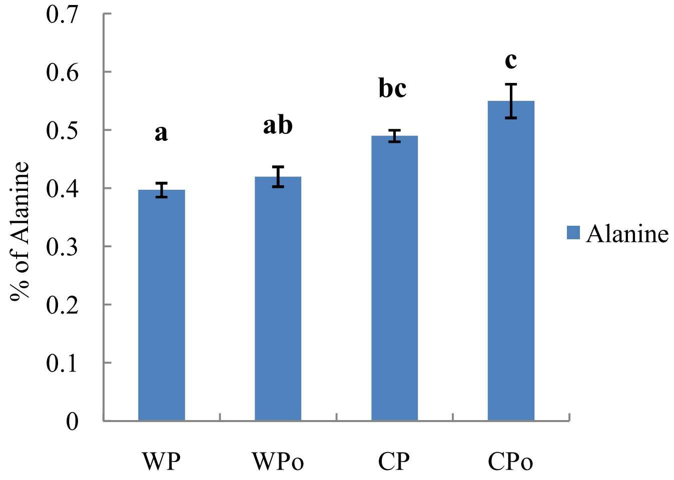


Figure 19: Variation of Alanine (%) in wild and cultured fishes of both season. Different lettersdenote significant differences (*p*< 0.05) and same letter denotes no significance (*p*>0.05)

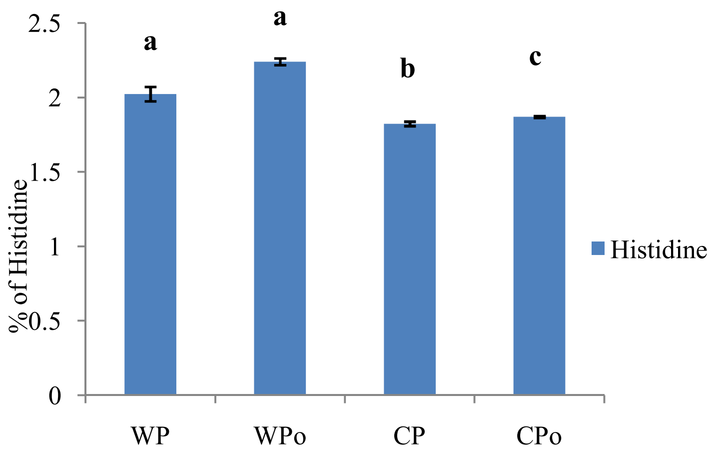


Figure 20: Variation of Histidine (%) in wild and cultured fishes of both season. Different lettersdenote significant differences (p < 0.05) and same letter denotes no significance (*p*>0.05)

3.3.13 Tyrosine

Inpre-monsoon wild and cultured koi fish illustrated that they contain 0.383±0.009 % and 0.473±0.007 % respectively on the contrary in post monsoon the amount was

0.440±0.012% and 0.550±0.023% (Appendix C_ Table 7). Result narrated that significant difference (p< 0.05) exists between pre-monsoon wild and post-monsoon cultured koi fish(Figure 21). No significant difference (p> 0.05) lies between wild fish but both seasons (pre-monsoon and post-monsoon) cultured koi fish exhibit significant difference (p< 0.05) (Figure 21). Moreover, there alsoremains significant difference(p<0.05)between post-monsoon wild and post-monsoon cultured koi fish in the amount of tyrosine content (Appendix C-Table 10).

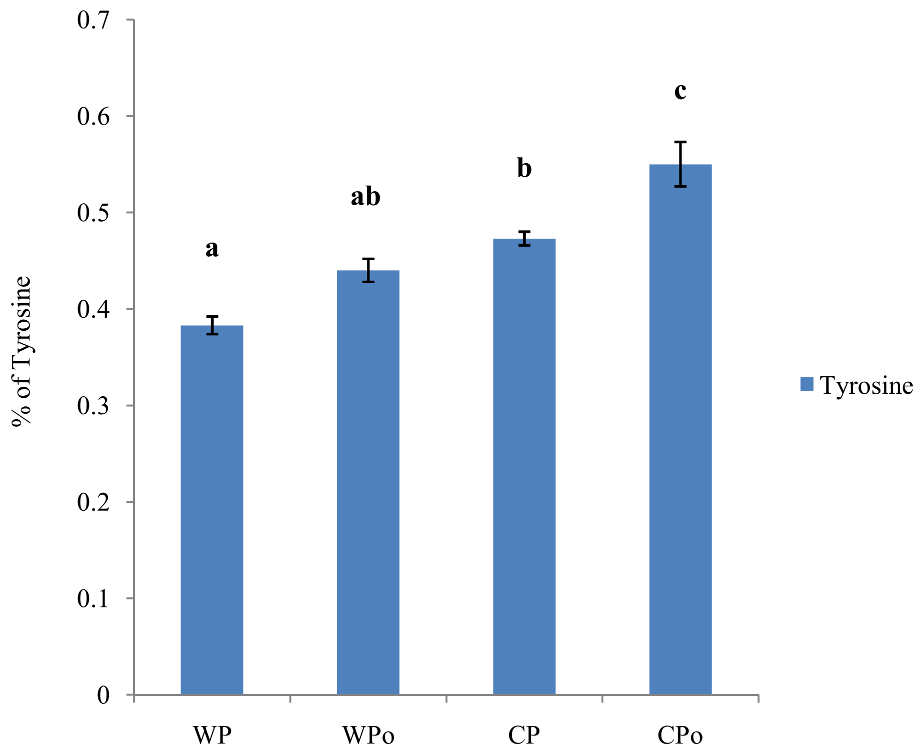


Figure 21: Variation of Tyrosine (%) in wild and cultured fishes of both seasons. where,WP=Pre-monsoon wild; WP_o=Post-monsoon wild and CP=Pre-monsoon cultured; CP_o=Post-monsoon cultured.Different lettersdenote significant differences (p < 0.05) and same letter denotes no significance (p>0.05)

3.3.14 Arginine

Mean Arginine content of the wild *Anabas testudineus*was 0.407±0.009% and 0.47±0.006%, collected in pre-monsoon and post-monsoon season whereas in cultured fish the amount was 0.52±0.015% and 0.58±0.023% respectively (Appendix C-Table 7). Result illustrated that significant difference (p< 0.05) lies between pre-monsoon wild and both season (pre-monsoon and post-monsoon) cultured koi fish (Figure 22). Data also expressed that no significant difference (p> 0.05) is observed between both seasons(pre-monsoon and post-monsoon)cultured fish (Figure 22). Again both seasons wild fish do not show any significant difference (p> 0.05) among themselves(Figure 22, Appendix C-Table 10).

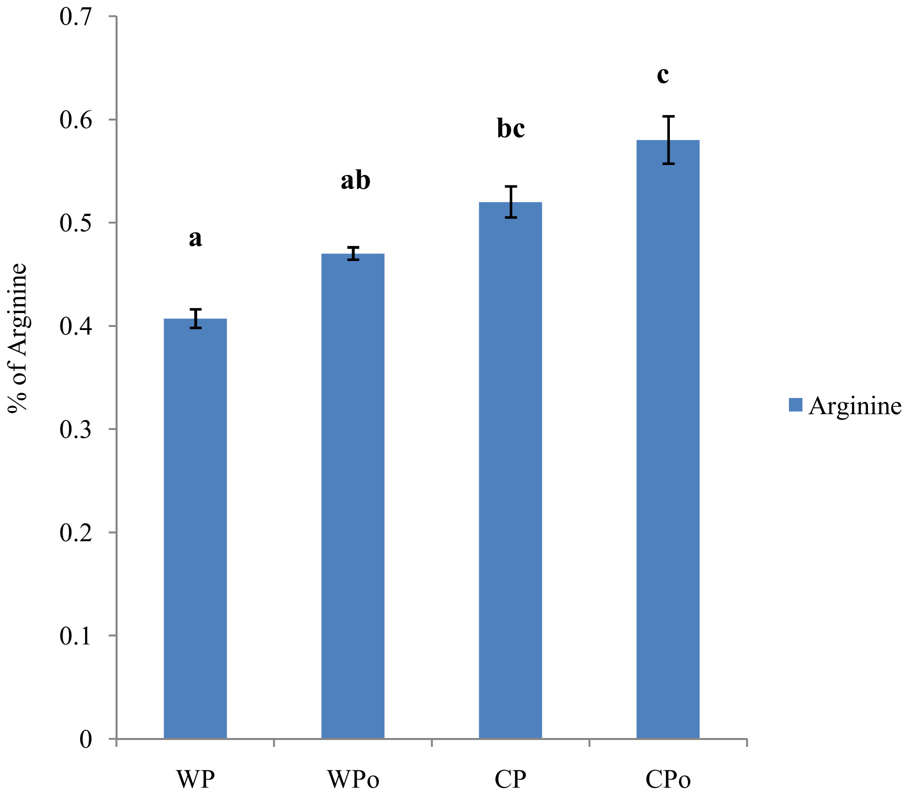


Figure 22: Variation of Arginine (%) in wild and cultured fishes of both season. where,WP=Pre-monsoon wild; WP_o=Post-monsoon wild and CP=Pre-monsoon cultured; CP_o=Post-monsoon cultured.Different lettersdenote significant differences ($p < 0.05$) and same letter denotes no significance ($p>0.05$)

3.4 Fatty Acids

Fatty acids are mainly two types. They are Saturated Fatty Acid (SFA) and Unsaturated Fatty Acid. The total SFA content of lipids were 39.865% in cultured and 40.119% in wild Koi fish collected in pre-monsoon but SFA contents were 39.021 % and 37.569%respectively in both wild and cultured fishes of postmonsoon seasons. Result expressed that wild *Anabas* contains 59.881% and 60.984% unsaturated Fatty acid in pre-monsoon and post-monsoon. Again the amount of unsaturated Fatty acid in both season (pre-monsoon and post-monsoon) was 60.115% and 61.322% in cultured fish.

3.4.1 Saturated Fatty Acid (SFA)

Palmitic acid (C16:0) was the primary saturated fatty acid (SFA). Result showed that wild koi fish contains 29.377±0.002% and 26.138±0.001% (Mean±SEM) palmitic acid in pre monsoon and post-monsoon as in cultured koi fish the amount were 30.277±0.147 % and 27.91±0.004% (Table 1).Among these result, the highest value was marked in cultured fish in pre-monsoon as well as the lowest was in wild fish of post-monsoon and they expressed Significant difference ($p< 0.05$)(Appendix D-Table 13). Data also expressed thatsignificant difference ($p< 0.05$) is observed between wild fish of both seasons as well as cultured fish of both season. But no significant difference (p >0.05)lies between cultured and wild fishes of post-monsoon (Appendix D-Table 13).

Amount of Myristic acid (14:0) is quite high among SFA. Findings described that in pre-monsoonMyristic acid content were1.630±0.012% and 1.524±0.003% in wild and cultured fish respectively, on the contrary in post-monsoon the amount were1.595±0.002% and 0.854±0.003 % (Table 1). Significant difference ($p< 0.05$) resides in wild and cultured fish of both seasons (pre-monsoon and post-monsoon). Total amount of myristic acid is higher in wild fish than cultured. Data also expressed that significant difference ($p < 0.05$) is observed between wild fish of both season. Moreover both seasons cultured fish revealed significant difference ($p < 0.05$) (Table 1).

Table 1: Saturated Fatty Acid composition (%) of wild and cultured koi fish of two different seasons.Values in the same row having the different superscript are significantly different (p<0.05)

Fatty acid composition	Wild (%)		Cultured (%)	
	Pre-monsoon	Post-monsoon	Pre-monsoon	Post monsoon
Saturated fatty acids	**40.119**	**39.021**	**39.865**	**37.569**
Caproic acid (C6:0)	0.077 ± 0.001^{a}	0.6730 ± 0.002^{b}	ND	0.1809 ± 0.001^{d}
Enanthic acid (C7:0)	0.075 ± 0.002^{a}	0.6108 ± 0.002^{b}	ND	0.345 ± 0.001^{c}
Caprylic acid (C8:0)	0.0578 ± 0.001^{a}	0.44 ± 0.023^{b}	ND	0.228 ± 0.005^{c}
Capric acid (C10:0)	0.0183 ± 0.001^{a}	0.1920 ± 0.002^{b}	0.0507 ± 0.001^{c}	0.133 ± 0.004^{d}
Lauric acid (C12:0)	0.054 ± 0.002^{a}	0.176 ± 0.001^{b}	ND	ND
Myristic acid (14:0)	1.630 ± 0.012^{a}	1.595 ± 0.002^{b}	1.524 ± 0.003^{c}	0.854 ± 0.003^{d}
Pentadecyclic acid (C15:0)	0.0820 ± 0.001^{a}	0.252 ± 0.001^{b}	ND	ND
Palmitic acid (C16:0)	29.377 ± 0.002^{b}	26.138 ± 0.001^{a}	30.277 ± 0.147^{c}	27.91 ± 0.004^{a}
Stearic acid (C18:0)	8.550 ± 0.012^{a}	8.245 ± 0.001^{b}	8.013 ± 0.008^{c}	9.027 ± 0.004^{d}
Arachidic acid (C20:0)	0.198 ± 0.001^{a}	0.226 ± 0.002^{b}	ND	ND
Behenic acid (C22:0)	ND	0.072±0.001	ND	ND
Margaric acid(C17:0)	ND	0.1281±0.001	ND	ND

Stearic acid (C18:0) is also a major SFA. Findings described that amount of stearic acid in wild fishes in pre-monsoon and post-monsoon was respectively 8.550±0.012 % and 8.245±0.001 %. On the other hand cultured fish contains 8.013±0.008% in pre monsoon and 9.027±0.004 % in post-monsoon Result narrated that significant difference ($p < 0.05$) is found between wild and cultured fish of both season (pre-monsoon and post-monsoon). Data also described that significant difference ($p < 0.05$) is observed between wild fish of both season (). Again significant difference ($p < 0.05$) both seasons cultured fish(Table 1and Appendix D-Table 13).

Capric acid (C10:0) is present in both wild and cultured fish but the amount is minor.Wild koi fish contains 0.0183±0.001% and 0.1920±0.002% capric acid in pre monsoon and post-monsoon as in cultured koi fish the amount was 0.0507±0.001% and 0.133±0.004% Result described significant difference ($p < 0.05$) among wild and cultured fish of both seasons. Data also expressed significant difference ($p < 0.05$) in between wild fish and between cultured fish of both season ((Table 1 and Appendix D-Table 13).

Among other SFA Caproic acid (C6:0), Enanthic acid (C7:0), Caprylic acid (C8:0) werenot detected (ND) in cultured fish in pre-monsoon but present in wild fish. In wild fish the amount were0.077±0.001%, 0.075±0.002% and 0.0578±0.00 % in pre-monsoon and in post-monsoon the amount was 0.6730±0.002%, 0.6108±0.002% and 0.44±0.023% respectively (Table 1). In post-monsoon these SFA are present in in cultured fish and the amount was 0.1809±0.001 %, 0.345±0.001% and 0.228±0.005%.in the case of all those fatty acid result narrated that Significant difference ($p < 0.05$) is found between wild fish of both season (pre-monsoon and post-monsoon). Moreover significant difference ($p < 0.05$) among wild and cultured fish of both seasons(Table 1).

Lauric acid (C12:0), Pentadecyclic acid (C15:0),Arachidic acid (C20:0) is not detected in cultured fish. In wild fish the amount was respectively 0.054±0.002%, 0.0820±0.001%, and 0.198±0.001% in pre-monsoon. In post monsoon the amount was 0.176±0.001%, 0.252±0.001% and 0.226±0.002% in wild *A. testudineus*(Table 1). Data described that Significant difference ($p < 0.05$) is found between wild fish of both season (pre-monsoon and post-monsoon) (Appendix D-Table 13).

Behenic acid (C22:0) and Margaric acid(C17:0) was not detected in cultured fish as well as in pre-monsoon wild fish. The acid is only present in post-monsoon wild fish and the amount was 0.072±0.001%, and 0.1281±0.001%.

3.4.2 Unsaturated Fatty Acids

3.4.2.1 Monounsaturated Fatty Acids (MUFAs)

Amount of Oleic acid (C18:1) in pre-monsoon wild and cultured fish illustrated that they contain 43.107±0.001% and 42.351±0.004 % respectively on the contrary in post monsoon the amount was 33.864±0.104% and 44.994±0.004% (Table 2). Analysis of the data revealed that significant difference is observed ($p< 0.05$) in both seasons (pre-monsoon and post-monsoon) wild and cultured koi fish. Significant difference ($p< 0.05$) also observed between wild fish of both season. When compare percentage amount of Oleic acid in cultured fish of both seasons they also showed significant difference ($p < 0.05$) (Appendix D-Table 13).

Palmitoleic acid (C16:1) and Eicosenoic acid (C20:1) present in both wild and cultured fish.Mean amount of Palmitoleic acid (C16:1)in wild *Anabas testudineus*was % 4.782±0.001 and 4.616±0.004 %, collected in pre-monsoon and post-monsoon season whereas in cultured fish the amount was 2.707±0.005% and 1.609±0.030% respectively (Table 2). wild koi fish contains 0.747±0.002% and 0.857±0.002% Eicosenoic acid (C20:1) in pre monsoon and post-monsoon and in cultured koi fish the amount was 0.281±0.001^{c} % and 0.435±0.002% (Table 2). In both case results described significant difference ($p< 0.05$) is found among wild and cultured fish of both season(Appendix D-Table 13). Data also expressed that significant difference ($p< 0.05$) lies between wild fish and between cultured fish of both season.

Vaccenic acid (C18:1) was not detected in cultured fish but present in wild fish. In pre-monsoon and post-monsoon amount of vaccenic acid was 0.357±0.004% and 0.145±0.001% in wild fish. Result described significant difference ($p < 0.05$) is among wild fish of both season (Table 2).

Table 2: Unsaturated Fatty Acid composition (%) of wild and cultured koi fish in two different seasons. Values in the same row having the different superscript are significantly different ($p<0.05$)

Unsaturated fatty acids	**59.881**	**60.984**	**60.115**	**61.322**
Mnounsaturated fatty acids (MUFA)	**48.993**	**39.482**	**45.339**	**47.038**
Palmitoleic acid (C16:1)	4.782 ± 0.001^{a}	4.616 ± 0.004^{b}	2.707 ± 0.005^{c}	1.609 ± 0.030^{d}
Oleic acid (C18:1)	43.107 ± 0.001^{a}	33.864 ± 0.104^{b}	42.351 ± 0.004^{c}	44.994 ± 0.004^{d}
Vaccenic acid (C18:1)	0.357 ± 0.004^{a}	0.145 ± 0.001^{b}	ND	ND
Eicosenoic acid (C20:1)	0.747 ± 0.002^{a}	0.857 ± 0.002^{b}	0.281 ± 0.001^{c}	0.435 ± 0.002^{d}
Polyunsaturated fatty acids (PUFA)	**10.888**	**21.502**	**14.776**	**14.284**
Hexadecadienoic acid (C16:2)	0.103 ± 0.003^{a}	0.503 ± 0.003^{b}	ND	ND
Hexadecatrienoic acid (HTA) (C16:0)	1.33 ± 0.000^{a}	0.744 ± 0.002^{b}	ND	ND
Linoleic acid (C18:0)	3.384 ± 0.003^{a}	10.414 ± 0.006^{b}	11.923 ± 0.002^{c}	10.028 ± 0.002^{d}
Linolenic acid (C18:0)	1.613 ± 0.005^{a}	3.567 ± 0.008^{b}	0.8255 ± 0.003^{c}	1.383 ± 0.002^{d}
Eicosadienoic acid (C20:2)	0.321 ± 0.002^{a}	0.466 ± 0.000^{b}	ND	ND
Eicosatrienoic acid (C20:3)	0.322 ± 0.000^{a}	0.527 ± 0.002^{b}	0.396 ± 0.004^{c}	0.583 ± 0.015^{d}
Arachidonic acid (C20:4)	1.238 ± 0.001^{b}	2.173 ± 0.034^{c}	0.646 ± 0.001^{a}	1.195 ± 0.032^{b}
Eicosapentaenoic acid (EPA) (C20:5)	0.062 ± 0.000^{a}	0.162 ± 0.001^{b}	ND	0.329 ± 0.001^{c}
Docosatrienoic acid (C22:3)	0.444 ± 0.003^{a}	0.621 ± 0.002^{b}	ND	0.257 ± 0.002^{c}
Adrinic acid (C22:4)	0.344 ± 0.002^{a}	0.335 ± 0.006^{a}	ND	0.371 ± 0.000^{b}
Docosahexaenoicacid (DHA) (C22:6)	0.971 ± 0.003^{a}	1.408 ± 0.010^{c}	0.971 ± 0.000^{a}	0.138 ± 0.000^{b}
Docosapentanoic acid(DPA) (C22:5)	0.756 ± 0.004^{a}	0.582 ± 0.006^{b}	0.014 ± 0.000^{c}	ND
n-3/n-6	0.83	0.82	0.7	0.67

3.4.2.2 Polyunsaturated Fatty Acids (PUFAs)

Result described that amount of Linoleic acid (C18:0) in wild fishes in pre-monsoon and post-monsoon was respectively 4.384±0.003% and 10.704±0.006 % (Table 2). On the other hand cultured koi fish contains 11.923±0.002%Linoleic acid in premonsoon and 10.028±0.002% in post-monsoon (Table 2). Findings narrated that significant difference (p< 0.05) exists among pre-monsoon and post-monsoon wild and cultured koi fish(Appendix D-Table 13). Again Significant difference(p<0.05) remains between wild fish as well as between cultured koi fish in the amount of Linoleic acid content.

Cultured *Anabas* contains 0.527±0.002 % and 0.583±0.015 % Eicosatrienoic acid (C20:3)in both pre-monsoon and post-monsoon whereas in the case of wild fish the amount was 0.322±0.000 % and 0.396±0.004 % (Table 2). In wild fishes amount of Linolenic acid in pre-monsoon and post-monsoon was respectively 1.613±0.005% and 3.567±0.008% (Table 1). On the other hand cultured koi fish contains 0.8255±0.003 % Linolneic acid in pre monsoon and 1.383±0.002% in post-monsoon. Mean amount of Arachidonic acid (C20:4) in wild koi fish in both season pre-monsoon and post-monsoon was 1.238±0.001%and 2.173±0.034 % respectively on the contrary the amount was 0.646±0.001 % and 1.195±0.032% in cultured koi fish (Table 2).Wild *Anabas* contains 3.384±0.003% and 10.414±0.006% Linoleic acid (C18:0) in both pre-monsoon and post-monsoon whereas in the case of cultured fish the amount was 11.923±0.002% and 10.028±0.002 % (Table 2). In the case of all those fatty acid our findings concluded that significant difference is observed (p < 0.05) between cultured koi fish of both season Again among cultured and wild koi fish significant difference is observed (p < 0.05). Moreover significantdifference (p< 0.05)is also observed between cultured koi fish of both seasons (Appendix D-Table 13).

Findings described that in pre-monsoon amount of Docosahexaenoicacid (DHA) (C22:6)was 0.971±0.003% and 0.971±0.000 % in wild and cultured fish respectively on the contrary in post monsoon the amount was 1.408±0.010 % and 0.138±0.000% (Table 2). Significant difference (p < 0.05) resides between cultured fish of both seasons (pre-monsoon and post-monsoon). Data also expressed that both seasons cultured and wild fish show significant difference (p < 0.05) among themselves. No Significant difference (p>0.05)lies between cultured and wild fish of pre-monsoon (Appendix D-Table 13).

Amount of Eicosapentaenoic acid (EPA) (C20:5) in wild fish was 0.062±0.000% and 0.162±0.001% in pre-monsoon and post-monsoon .Whereas in pre-monsoon cultured fish EPA was not detected but in post-monsoon EPA was detected and the amount was 0.329±0.001%. Significant difference is observed ($p< 0.05$) between wild fish depending on season. Significant difference is also observed ($p< 0.05$) between wild fish based on seasons (Table 2).

In both season pre-monsoon and post-monsoon cultured koi sample Hexadecadienoic acid (C16:2) andHexadecatrienoic acid (HTA) (C16:0) was absent but present in wild (Table 2). Inpre-monsoon wild fish findingsillustrated that they contain0.103±0.003 %Hexadecadienoic acid in pre-monsoon and 0.503±0.003% in post monsoon. Amount of HTA was 1.33±0.000 % and 0.744±0.002% in pre-monsoon and post-monsoon wild fish (Table 1). Result expressed that both fatty acid have significant difference ($p< 0.05$) between pre-monsoon wild and post-monsoon wild koi fish(Appendix D-Table 13).

Docosatrienoic acid (C22:3)and Adrinic acid (C22:4) were not detected in pre-monsoon cultured fish but present in post-monsoon cultured and both season wild fish (Table 2). Mean Docosatrienoic acid in wild *A. testudineus*was 0.444±0.003% and 0.621±0.002%, collected in pre-monsoon and post-monsoon season whereas in cultured fish the amount was 0.257±0.002 % in post-monsoon. Amount of Adrinic acid was 0.344±0.002 % and 0.335±0.006% in pre-monsoon and post-monsoon wild fish. Post-monsoon cultured koi samle contains 0.371±0.000 % Adrinic acid (Table 2). Inwild fish result ofDocosatrienoic acid represented that it differed significantly ($p<0.05$) between wild fish depending on season. Significant difference ($p< 0.05$) also observed when compare between wild fish of both season and cultured fish of post-monsoon (Appendix D-Table 13).

Result of Adrinic acidshowed that no significant difference ($p> 0.05$) is observed between wild fish of both seasons. Again Significant difference ($p < 0.05$) can be described when compare among wild fish (both pre-monsoon and post-monsoon) and post-monsoon cultured fish (Table 2).

Docosapentanoic acid(DPA) (C22:5)was not detected in cultured fish in post-monsoon but present in pre-monsoon cultured fish. In wild fish the amount of DPA were0.756±0.004%

and 0.582±0.006% in pre-monsoon and in post-monsoon respectively (Table 2). Cultured fish contains 0.014±0.000% DPA in pre-monsoon. Result narrated that Significant difference ($p < 0.05$) is found between wild fish of both season (pre-monsoon and post-monsoon) (Appendix D-Table 13). Moreover significant difference ($p< 0.05$) can be described when compare among wild fish (both pre-monsoon and post-monsoon) and pre-monsoon cultured fish.

Eicosadienoic acid (C20:2)was not found in cultured fishbut present in wild koi fishes (Table 2). Wild koi contained0.321±0.002%Eicosadienoic acid in pre-monsoon and 0.466±0.000% in post monsoon.Result also expressed that significant difference ($p< 0.05$)exists between pre-monsoon wild and post-monsoon wild koi fish(Table 2).

In pre-monsoon, ratio of n-3/n-6were 0.83and 0.7 for wild and cultured koi fish respectively whereas in post-monsoon these ratio were 0.82 and 0.67 respectively for wild and cultured fishes.

4 DISCUSSION

Fish as a food is expected to provide relief from malnutrition, especially in developing countries (Ashraf *et al.*, 2011). It provides superior quality protein to that of meat, milk and eggs and well balanced essential amino acid profile, necessary minerals and fatty acids (Hossain, 1996). Fish may be the sole accessible and affordable source of animal protein for poor households in urban or semi-urban areas.Fish contains the most important nutritional components and serves as a source of energy for human beings. Foran*et al.* (2005) concluded that, fish is a highly portentous food consumed by a larger percentage of population because of its availability and palatability.Koi is a popular fish for Bangladeshis. It is found in open water as well as cultured in close water body.

Cultured fish is provided with nutrient rich foods in addition to natural productivity in the pond. On the other hand captured fish has to depend totally on natural food for its sustenance. The nature and quality of nutrients in most animals depend largely on their food type. These variations have direct relationship on body composition, health status and growth of fish. Moreover Stansby (1954) has established that information on the proximate composition of fish in respect to the nutritive value is important to compare with other source of animal protein foods such as meat and poultry products. Islam and Joadder (2005) reported that proximate composition of fish depends on season, sex and reproductive cycle.

A comparative study was undertaken to determine the moisture, ash, protein, lipid, amino acid and fatty acid profile in wild and cultured koi fishes of Jessore district, Bangladesh. Depending on the habitat and season, results showed a wide variation in proximate composition amino acid and fatty acid profile of wild cultured koi fish.

4.1 Length-Weight Relationship

Length-Weight relationships can be used to predict weight from length measurements made in the yield assessment (Pauly, 1993). Like any other morphometric characters, the length-weight relationship can be used as a character of differentiation of taxonomic units and the relationship changes with the various development events in life such as metamorphosis, growth and onset of maturity (Thomas *et al.,* 2003).

Fish can attain either isometric growth, negative allometric growth or positive allometric growth. An organism shows isometric growth when no change is associated with body shape as grows. Negative allometric growth implies the fish becomes more slender as it increase in weight while positive allometric growth implies the fish becomes relatively stouter or deeper-bodied as it increases in length (Riedel *et al*., 2007). In this study, it was found that there were a positive association between length and weight for the both wild and cultured *Anabas testudineus*. These findings were more or less similar to the results of Bala*et al*. (2009), Imam *et al.* (2010),Haruna (2006), Sangun*et al., (*2007) and Naeem*et al.* (2010).

4.2 Nutritional Composition

In fishes nutritional composition means the composition of the fish flesh. It is the most common method to evaluate nutritional quality. Proximate composition generally comprises the estimation of moisture, protein, fat and ash contents of the fresh fish body. According to FAO (1991) normally fish contains 72% water, 19% protein, 8% fat, 0.5% calcium, 0.25% phosphorus and 0.1% vitamin A, D, B etc. Knowledge of the proximate composition of fishes is essential to estimate their nutritional value. FAO (1991) also reported that the measurement of some proximate profiles such as protein contents, carbohydrates, lipids, moisture contents and ashpercentage is often necessary to ensure that they meet the requirements of food regulations and commercial specifications.

Again the percentage of proximate composition showed a wide variation in wild and cultured fish. Alemu*et al.* (2013) showed that both exogenous and endogenous factors control fish body composition.Proximate composition of fish may vary widely from species wise as well assame species. Khuda, M.Q. (1964) studied the proximate composition of different species and different individual of same species. Both times he found difference. But the difference is significant in different species as compared with same species.

4.2.1 Moisture Content

Moisture content is expressed as the amount of water as a percentage (%) and the remaining portion is dry matter content. The major component of fish muscle is moisture. In general the range fall between 60-80% in many fish (Murra and Burt, 2001).Moisture content may vary depending on age, fat content, habitat and feeding etc.

In our present study seasonal variation is observed in moisture content of wild and cultured fish. The moisture content varied from (73.06-80.62)%. Seasonally the highest value was found in pre-monsoon wild koi fish 79.62±0.29 % and the lowest value is in cultured fish of post-monsoon season 73.91±0.16%.

Hossain*et al.* (2015) analyzed proximate composition of *Anabas testudineus.*They found 72.60±0.17% moisturein koi fish.Stansby (1954) andRubbi*et al.* (1987) described that moisture content varied from 72.2-83.6% with an average 77.64%. According to Brogstorm (1961) fish contains 66-84% moisture. Moisture content was 70% in *Mystusvitatas*.

Nutritional composition of mola (*AmblyPharyngodonmola*), dhela (*osteobramacotio*), Chapila (*Gudusiachapra*), Punti (*Puntius stigma*) tengra (*Mystustengra*) and kankila (*Xenentodoncancila*) was estimated by Nurullah*et al.* (2003). They showed that moisture content ranged from (72.97-76.36)%.

However, moisture content varied seasonally and also varied from species to species. Again moisture content varies depending on environment where they survive. All these findings coincide well with the present findings of moisture content.

4.2.2 Ash Content

Ash content was found to vary from (2.37-3.71)%. Inpre-monsoon wild and cultured fish illustrated that they contain 3.62±0.02% and 2.50±0.03% ash respectively on the contrary in post monsoon the amount was 3.02±0.01% and 2.90±0.03%.

Body composition is true reflector of its feeding habits and type of food availability (Ashraf *et al.,* 2011). Comparison of nutrient values of wild and cultured *Heterobranchusbidorsalis*and and *Clariasgariepinus*werestudiedbyOnyia*et al.* (2013). They obtained that there remainssignificant differences ($p<0.05$) in crude fiber and ash.

Hossain*et al.* (2015)described that *Anabas testudineus* contains 3.02±1.12% ash. However Begum and Hoque (1986) found 2.06% ash in shrimp. Both scaly and non-scaly fresh water fish contains 0.85-5.11% ash (Rubbi*et al.,* 1987).Matto*et al.* (1991) estimated 1.2%, 1.3%, 1.3% and 1.1% ash in *sardinapulchardus, Thunnusthynnus, Mugilcephalas*and*Atherinahepsetus*respectively. These values are more or less supported our present findings.Alemu*et al.* (2013) worked on composition of Nile Tilapia and described that ash and gross energy decreased from 15.8 to 13.3, 1.2 to 1.0% and 65 to 55.9 kcal/100 g respectively with increase in age.

But the result is contradictory with Venkatarman and Chari (1951). This might be due to habitat, season, age, geographical location etc. He also observed that the proximate composition in fish depend upon some factors, e.g. size, age, sex, seasonal change and habitat.

4.2.3 Crude ProteinContent

Crude protein contents in both cultured and wild fish varied based on season (Pre-monsoon and post-monsoon). Findings described that crude protein contents of wild koi fishes in pre-monsoon and post-monsoon were respectively 12.13±0.01% and 14.74±0.02%. On the other hand cultured koi fish contains 16.04±0.02% in pre monsoon and 17.92±0.02 % in post-monsoon. The highest value was recorded in post-monsoon cultured fish (17.92±0.02%) and the lowest value in wild fish (12.13±0.01%) of pre-monsoon. Seasonally protein content may vary between (12.09-17.96)%.

Amount of protein in fish muscle is between 15-20 per cent (Murraand Burt, 2001). Protein content may vary considering age, fat content, spawning and starvation etc.Hossain*et al.,*(2015) described that Anabas *testudineus* contains 16.18±0.13% protein.Our present investigation coincides with the work.

Nurullah*et al.,* (2003) worked on Nutritional composition of mola, dhela, Chapila, Punti, tengra and kankila. They found that protein content varied between (14.08-21.70)%.Irianto and Irianto (1997) studied on proximate composition of Nile Tilapia in Indonesia. The report showed that amount of Protein was 20.1%. *Puntiusticto* contains 18.62% protein (Banu*et al.,* 1985).

Nargis (2006) worked on Protein, carbohydrate, fat, ash and moisture contents in the body muscle of *Anabas testudineus*. The compositions varied seasonally in relationto reproductive cycle of the fish. Significant correlation existed between moisture andcarbohydrate, moisture and fat, moisture and protein, moisture and ash, protein andash, fat and carbohydrate, fat and ash, protein and carbohydrate. The protein content found to be higher in medium sized fishes and gradually decreased with the increase of age. Fat content was higher in large-sized male than that of the females. Carbohydrate content was slightly higher in male than the female. Our present study has similarity with this variation.

Rubbi*et al.* (1987) found 18.40±0.25% protein in prawn whereas Aykoryed*et al.,* (1951) described 19.1% protein in shrimp. Begum and Hoque (1986) reported 17.35% protein in shrimp and Kamal *et al.* (2000) found 18.64% protein in *Macrobrachiumrosenbergii.*Onyia*et al.* (2013) worked on comparison of nutrient values of wild and cultured *Heterobranchusbidorsalis*and and *Clariasgariepinus.* They obtained that there remains significant difference (p<0.05) in crude protein content of wild and cultured *H. bidorsalis*(55.03±6.21%, 54.00±4.90%), and *C. gariepinus*(50.20±0.38%, 48.90±2.70%).

Chandrasharkar and Deosthale (1993) worked on proximate composition, amino acid, mineral and trace element content of the edible muscle of 20 Indian fish species. They showed that a wide variation exist between species in protein content (marine 8%-21%, fresh water 13.5% - 17.3%).

Govindan (1985) showed that both marine and fresh water fish contained 9%-25% protein,where (16-19) % is regular limit. Matto*et al.* (1991) estimated amount of protein in *sardinapulchardus, Thunnusthynnus, Mugilcephalas*and*Atherinahepsetus* and it was 18.6%, 18.3%, 19.6% and 18.7% respectively. Islam and Joadder (1990) estimated protein content of six fresh water fish and exposed that these fishes contains 11-16.75% protein. The above discussion also has similarity with our result.

4.2.4 Lipid Content

Lipid content narrated that lipid content may vary between (3.56-5.61)% in pre-monsoon and post-monsoon. Findings described that in pre-monsoon lipid content was 3.82±0.02% and 5.14±0.01% in wild and cultured fish respectively on the contrary in post monsoon the amount was 3.61±0.02% and 5.56±0.01%. Among these result the highest value of lipid content was marked in cultured fish of post monsoon season (5.56±0.01) % as the lowest was in wild fish of pre monsoon (3.61±0.02) %.

Hossain*et al.* (2015) who worked on proximate composition of *Anabas testudineus fo*und that amount of lipid was 5.31±0.37%. Our findings have similarity with the result of Hossain*et al.* (2015)

Nargis (2006) showed significant correlation between moisture, fat and ash in Koi fish. Fat content was higher in large-sized male than that of the females.

Seasonal Variation in the biochemical composition of mackerel was studied by Venkatarman and Chari (1951). They found that fat was varied seasonally.Alemu*et al.* (2013) made their experiment on Nile Tilapia *(Oreochromisniloticus)* fillet. They concluded that moisture and fat contents increased from 79.5 to 80.8 and 0.4 to 0.7 % respectively, with increase in age of fish.

Kamal *et al.* (2000) found 1.8% lipid in *Macrobrachiumrosenbergii.*Chandrasharkar and Deosthale (1993) showed that a wide variation exists between species in lipid content and the range was 0.65-1.3%.He also reported that positive correlation remains between protein and moisture whereas inverse relationship is maintained between fat and moisture.

Rubbi*et al.* (1987) described that lipid content varied from 0.8-1.55% in 27 fresh water species both scaly and non-scaly. Lipid content was 1.0% in *Heteropneustes fossils* and *Clariusbatrachus.* Stansby (1954) reported that certain fresh water fish contained 5.0% lipid. The above discussion supported our present findings.

4.3 Amino Acid

Amino acids are the building blocks of proteins and serve as body builders. They are utilized to form various cell structures, of which they are key components and they serve as source of energy (Babsky*et al*. 1989).

Fish protein comprises of all the ten essential amino acid in desirable quantity for human consumption. Fish protein is very rich in such amino acid as methionine, lysine and low in tryptophan compared to mammalian protein (Nowsad, 2007).

Our present findings described that amount of methionine in wild koi fish were 0.313±0.017% and 0.350±0.012% in pre-monsoon and post-monsoon whereas in cultured fish the amount were 0.457±0.013% and 0.490±0.017% respectively. Result expressed that amount of methionine is higher in post-monsoon cultured fish and lower in pre-monsoon wild fish.

Amount of lysine in pre-monsoon wild and cultured *Anabas* were0.300±0.015% and 0.367±0.007% respectively and in post monsoon the amount was 0.370±0.012% and

0.440±0.017%. Among these result the highest value was marked in cultured fish of post monsoon season aswell as the lowest value was in wild fish of pre monsoon and they showed significant difference (p< 0.05).

Findings described that in pre-monsoon amount of glutamic Acid was 0.990±0.015% and 1.147±0.0150 % in wild and cultured fish respectively on the other hand in post monsoon the amount was 1.040±0.023% and 1.260±0.017%. Among these result the highest value was marked in cultured fish in post-monsoon as well as the lowest was in wild fish of pre-monsoon.Nurullah*et al.* (2003) reported that among all the amino acids analyzed, glutamic acid content was found to be quite high whereas the other amino acids were slightly low.Iwasaki and Harada (1985) similarly reported that the main amino acids in fish muscle were aspartic acid, glutamic acid and lysine.

In pre monsoon wild and cultured fish amount of aspartic acid is 0.737±0.007% and 0.890±0.01 % whereas in post-monsoon the amount was0.760±0.017% and 0.930±0.006% respectively.Result described that amount of aspartic acid is higher in cultured fish than in wild. Wesselinova(2000) also reported that the amounts and types of amino acids in fish muscle were affected by catching time and habitat. Our present findings have similarity with both of these works.

In the present study, the ratio of EA/NEA amino acids were observed 0.23 and 0.27 in wild koi fish collected in two different period as pre-monsoon and post-monsoon. Moreover the ratio was 0.32 in pre-monsoon cultured fish and 0.33 in post-monsoon cultured fish(Table 2).

The results obtained from this study showed that cultured koi fish have well-balanced and high-quality protein source in the respect of EA/NEA ratio in post-monsoon. Again the ratio was higher in post-monsoon cultured fish.

4.4 Fatty Acid

The analysis of the fatty acid content indicates that wild fish are superior in terms of thenutritional value. More specifically, PUFAs are dominated in wild koi specimens mono unsaturated fatty acids (MUFAs) weredominant in farmed koi fish.

The most abundant fatty acids in the four fish species were palmitic and stearic acids for saturated fatty acids, palmitoleic and oleic acids for monounsaturated fatty acid, linoleic and docosahexaenoic acids for polyunsaturated fatty acid. Similar studies performed on tropical.

Stansby(1982) also reported that 20–50% of total fatty acid in some fish lipids was palmitic acid. In this study, Palmitic acid (C16:0) was the primary saturated fatty acid (SFA), contributing approximately 70% to the total SFA content of the lipids for both cultured and wild koi fish. Result showed that wild koi fish contains 29.377±0.002% and 26.138±0.001% palmitic acid in pre monsoon and post-monsoon as in cultured koi fish the amount was 30.277±0.147 % and 27.91±0.004%. This work coincides with our present findings.

Orban*et al.* (2002) similarly reported that palmitic acid (16:0), oleic acid (18:1), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) fatty acids constituted the basiccomponents of fatty acids of sea bass fillets.This result also partially supports our findings.

The level of the monounsaturated fatty acid is highly dependent on the level of oleic acid, since the primary constituent of this group is oleic acid.In this study, oleic acid was identified as the dominantmonounsaturated fatty acid among monounsaturatedfatty acids. Shchenikova*et al.* (1987) reported that the level forthe 16:1 of the monounsaturated fatty acids capturedis the Far East basin (Russia) was 10.5%.

The reason for the remarkable difference between the data of Shchenikova*et al*. (1987) may be for different species and for different geographical region and habitat.Sinanoglou and Miniadis-Meimaroglou (1998) described that amount of different fatty acid in fish lipid have direct relation to the geographical region where fishing wasconducted.

The high levels of oleic, palmitoleic, and arachidonic acids had been reported as a characteristic property of freshwater fish oils (Andrade*et al.,* 1995 *Osman et al.,* 2001). These results more or less similar to our findings. The reason may be that both are fresh water species.

Hassan *et al.* (2010) worked on total lipids and fatty acid profile in the liver of wild and farmed *catlacatla*fish. Proportions of saturated fatty acids (C14:0, C16:0, C18:0, C20:0) varied irregularly in lipids of the liver from both wild and farmed *Catlacatla.* MUFA (C18:1) was found in considerable amounts in the liver of both and PUFA such as C18:3 (n-6) and C20: 2 (n-6) were detected in the liver of the wild fish.Our present findings are similar with this result. In our experimental fish amount of PUFA is higher in wild than in cultured.

Piggott and Tucker (1990) suggested that the n-3/n-6 ratio is a better index in comparing the relative nutritional value of fish oils of different species.An n-3/n-6 ratio of 1:1is considered to be optimal for nutritional purposes (Simopoulos, 1989), particularly whenn-3 FAs consist mainly of EPA and DHA.

A dietary intake of fish with a high ratio of n-3/n-6 would therefore be beneficial. In pre-monsoon wild and cultured fish amount of n-3/n-6 ratio is 0.83and 0.7 whereas in post-monsoon the amount was 0.82 and 0.67 respectively.

Yeganeh*et al.* (2012) made a thorough study on seasonal variations in chemical composition and fatty acid profile of farmed and wild common carp *(cyprinuscarpio).* They concluded that in the farmed carp fillet, the major SFA and MUFA were similar to those in the wild one but linoleic acid was the major PUFA in all seasons n -3/n-6 PUFA ratios in the wild carp fillet were higher than in the farmed counterparts. This work also has similarity with our findings. In our present work the ratio (n -3/n-6) is highest in wild koi of pre-monsoon and lowest in farmed koi of post-monsoon.

5 CONCLUSION

Koi fish (*Anabas testudineus)* has high cultural value and consumer preference in Bangladesh. This is an excellent candidate for culture in derelict water bodies as it is able to live in poorly oxygenated water. It is commercially important due to its high demand for protein.

Findings of the study showed that seasonal variation remains in proximate, fatty acid and amino acid profile of wild and cultured *A. testudineus*. Amount of moistureand ash is higher in wild fish than cultured. On the other hand cultured koi fish contains higher amount of protein and lipid.

Wild fish totally depend upon natural food. Moreover cultured fish is provided with nutrient rich foods and the food is mainly rich in animal protein. As a result cultured fish contain more protein than wild. This amount is also varying based on season. In post-monsoon amount of protein is higher in both wild and cultured fish. The reason may be that change in water quality and other parameter of the environment where they inhibit. But more research is needed to draw a conclusion regarding these findings.

Protein is usually the nitrogen containing component. Protein consists of chains of hundreds or thousands of amino acids. In amino acid profile result indicates that EA/NEA ratio is the highest in post-monsoon cultured koi fish. It is well known that high quality protein have higher EA/NEA ratio. So it may be approved that cultured koi contains well balanced and high-quality protein.

Fish lipids have been recognized as being beneficial for human health. Total amount of lipid is higher in cultured fish. But fatty acid profile indicates that wild fish contains higher level of PUFA mainly EPA and DHA. Many studies have been carried out on the metabolism and function of polyunsaturated fatty acids (PUFAs) in general and on the levels and ratios of n-3 and n-6 fatty acids in particular. Long chain polyunsaturated fatty acids (PUFAs) like linoleic acid, arachidonic acids, α-linolenic acid, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are essential fatty acids (EFA's). Human body cannot synthesize these EFAs.It is well known that n-3 fatty acids, or a balanced n–3/n-6 ratio in the diet, are essential for normal growth and development and may play an important role in the prevention and treatment of coronary artery disease, diabetes, hypertension and cancer. It is thus important, for human health, to increase the consumption of fish or fish products, which are rich in polyunsaturated fatty acids of the n-3 family and poor in polyunsaturated fatty acids of the n-6 family.Farmed fish exhibited lower nutritional value in terms of FAs, especially in terms of high linolic and linolenic acid content and low n-3/n-6 ratio, which is important for human's health. So it may concluded that wild *Anabas* is an excellent source of PUFA of n-3 family.

Koi fish species do not provide the same nutrient profile depending on the seasons. Generally, this fish is an excellent source of protein, because of the amino acid composition and degree of digestibility. It is found that fatty acid composition and n-3/n-6 is quite high in wild koi fish in post-monsoon and it is preferable for our diet than cultured fish.Therefore, a comprehensive study on cultured and wild fishes is required for the further confirmation of the result of present study.

LITERATURE CITED

AHMED, A.T.A., RUBBI, S.F., MUSTAFA, G., MUSLEMUDDIN, M. and AHMED, N. 1981.The proximate composition of flesh and food of *Tilapia nilotica* and *T. mossambica.Dhaka University Studies***29**(2): 33-40.

ALEMU, L.A., MELECE, A.Y. and GULELAT, D.H. 2013. Effect of endogenous factors on proximate composition of Nile tilapia (*Oreochromisniloticus* L. fillet from Lake Zeway).*Am. J. Res.* **1**: 405-410.

ANDRADE, A.D., RUBIRA, A.F., MATSUSHIA, M. and SOUZA, N.E. 1995.Omega3 fatty acids in freshwater fish from South Brazil. J. *Am. Oil Chem. Soc.***72**(10):1207–1210.

ANONYMOUS, 1993.Amino acid analysis sytem instruction manual.Shimadzu HPLC amino acid analysis system. Shimadzu corporation, Analytical Instruments Division, Kyoto, Japan, pp. 63 – 65.

AOAC. 1995. Official Methods of Analysis of the Association of Official Analytical Chemists. 16th ed. Edited by Patricia Cunniff. Arlington, VA, USA.

ARNETT, J.J. 2000. Emerging adulthood: A theory of development from the late teens through the twenties. *Am. Psychol.***55**(5): 469

ARTS, M.T., ACKMAN, R.G. and HOLUB, B.J. 2001. “Essential fatty acids" in aquatic ecosystems: a crucial link between diet and human health and evolution. *Can. J. Fish.Aquat. Sci.***58**(1): 122-137

ASHRAF, M., ZAFAR, A., RAUF, A., MEHBOOB, S. and QURESHI, N.A. 2011. Nutritional values of wild and cultivated silver carp (*Hypophthalmichthysmolitrix*) and grass carp *(Ctenopharyngodonidella).Int. J. Agric. Biol.***13**: 210-214.

AURSAND, M., BLEVIK, B., RAINUZZO, J.R., JORGENSEN, L. and MOHR, V. 1994. Lipid distribution and composition of commercially farmed Atlantic Salmon (*Salmosalar*). *J. Sci. Food Agric.***64**(2): 239-248.

AUSTRENG, E. and REFSTIE, T. 1979.Effect of varying dietary protein level in different families of rainbow trout.*Aquaculture***18**(2): 145-156.

AYKROYD, W. R., PATWARDHAN, V. N. and RANGANATHAN, S. 1951. Nutritive value of of Indian foods and the planning of satisfactory diets (Health bulletin no. 23), Manager of publications, Govt. of India, New Delhi, India.

BABSKY, E.B., KHODOROV, B.I., KOSITSKY, G.I. and ZUBKOV, A.A. 1989.*Human Physiology*, Mir Publishers, Moscow.

BADIANI, A., ANFOSSI, P., FIORENTINI, L., GATTA, P P., MANFREDINI, M., NANNI, N., STIPA, S. and TOLOMELLI, B. 1996.Nutritional composition of cultured sturgeon (*Acipenserspp*).*J. Food Comp. Anal.***9**(2): 171-190.

BALA, U., LAWAL, I., BOLORUNDURO, P. I., ONIYE, S. J., ABDULLAHI, S. A. and BICHI, A. H. 2009. Study of ichtyofauna of Daberam reservoir in Katsina State.*Bayero J. Pure.and App. Sci.***2**:172-174.

BANU, C. P., NAHAR, B., BEGUM, M., and AHMED, K. 1985.Studies on the Protein, riboflavin and iron content of some fresh water fishes and prawns of Bangladesh.*Bangladesh J. Zool.***13**(1): 25-28.

BEGUM,S.A. and HOQUE, M. S.1986. Effects of temperature on the composition, color, texture and reconstitution of dehydrated shrimps.*Bangladesh J. Agri. Sci.***13**(1):1-4

BELTON, G. S., REINE, W. F., HUISMAN, J. M., DRAISMA, S. G., GURGEL, D. and FREDERICO, C. 2014. Resolving phenotypic plasticity and species designation in the morphologically challenging Caulerparacemosa–peltata complex (Chlorophyta, Caulerpaceae).*J.phycol.***50**(1):32-54.

BRAHNEY, C. P., ARIA, A. A., KOVAL, M. H. and NAJJAR, T. A. 1981.Angiolipoma of the tongue: report of case and review of literature. *J. oral.Surg.***39**(6):451.

BRAMMER, H. 2004. Can Bangladesh be protected from floods*?* Hugh Brammer.University Press.

CHANDRASHEKAR, K. and DEOSTHALE, Y.G., 1993. Proximate composition, amino acid, mineral and trace element content of the edible muscle of 20 Indian fish species. *J. food comp.Anal.***6**(2): 195-200.

DEMPSON, J. B., SCHWARZ, C. J., SHEARS, M. and FUREY, G. 2004. Comparative proximate body composition of Atlantic salmon with emphasis on parr from fluvial and lacustrine habitats.*J. Fish Biol.***64**(5): 1257-1271.

DOF., 2015. National fish week 2015 Compendium (in Bangla).Department of Fisheries, Ministry of Fisheries and Livestock, Bangladesh.pp.148.

DUNSTAN, G. A., VOLKMAN, J. K., BARRETT, S. M. and GARLAND, C. D. 1993. Changes in the lipid composition and maximisation of the polyunsaturated fatty acid content of three microalgae grown in mass culture. *J. Appl.Phycol.* **5**(1): 71-83.

EXLER, J., KINSELLA, J.E. and WATT, B.K. 1975. Lipids and fatty acids of important finfish: new data for nutrient tables. . *J.Am. Oil Chem. Soc.***52**(5): 154-159.

FAO.1991.Wholesomeness of irradiate food. Report of a joint FAO/IAEA/WHO experts committee.WHO.Tech. Report Series.**659**:7-34

FLOWER, 1925.Contributions to our knowledge of the duration of life in vertebrate animals.*I. Proc. Zool Society, London.***1**: 247-267.

FOLCH, J. B. and STANLY, G. H. 1957.A sample method for the isolation and purification total lipids from animal tissue.*J. Biol. Chem*.**226**:697-709

FORAN, J. A., CARPENTER, D. O., HAMILTON, M. C., KNUTH, B. A. and SCHWAGER, S. J. 2005. Risk-based consumption advice for farmed Atlantic and wild Pacific salmon contaminated with dioxins and dioxin-like compounds. *Environ.healthperspec.***33**:552-556.

FRSS, 2015. Fisheries statistical Report of Bangladesh. Fisheries Resources Survey System (FRSS), Department of Fisheries, Bangladesh. Volume **31**: 57

GOVINDAN, T. K. 1985. Fish processing technology. Oxford IBH publishing Co. Pvt. Ltd. New Delhi, India. pp. 21-76.

HARUNA, M. A 2006.length-weight relationship of four fish species chichlidae from magaga lake, kano, *Nigeria. Best Journal*. **3**:109-111.

HASSAN, M., SHAHID CHATHA, S.A., TAHIRA, I. and HUSSAIN, B. 2010. Total lipids and fatty acid profile in the liver of wild and farmed *catlacatla* fish. *Grasas Y Aceites*.**61**(1): 52-57.

HOSSAIN, M.A. 1996. Proximate and amino acid composition of some potential Bangladesh fish feed ingredients. *Bangladesh J. Zool*.**24**: 163-168.

HOSSAIN, M.N., AFROZ, H., HAQUE, M.Z. and BEGUM, M. (2015). Evaluation of nutritional properties of some small indigenous fishes species in Bangladesh. *Bangladesh J. Zool.* **6**(6): 102-109.

HUGHES, G.M. and SINGH, B.N. 1970. Respiration in an air breathing fish, the climbing perch, *Anabas testudineus* (Bloch) I .Oxygen uptake and carbon dioxide release in to air and water.*J. Expt. Biology.* **53**:265-280.

HUNTER, B.J. and ROBERTS, D.C. 2000. Potential impact of the fat composition of farmed fish on human health.*Nutr. Res.***20**(7): 1047-1058.

HUSS, H.H. 1995. Quality and quality changes in fresh fish. *FAO fisheries technical paper*.Pp. 348.

IMAM, T.S., BALA, U., BALARABE, M.L. and OYEYI, T.I 2010. Length-weight relationship and condition factor of four fish species from Wasai Reservoir in Kano, *Nigeria. African Studies.***6**:125-130.

IRIANTO, G. and IRIANTO, H.E. 1997. Post-harvest technology of Nile tilapia in Indonesia: a review. *FAO Fisheries Report***563**: 71-83.

ISLAM, M.N. andJOADDER, M.A.R. 2005.Seasonal variation of the proximate composition of freshwater Gobi, Glossogobiusgiuris (Hamilton) from the River Padma.*Pak. J. Bio. Sci.***8**(4):532-536.

IWASAKI, M. and HARADA, R. 1985. Proximate and amino acid composition of the roe and muscle of selected marine species.*J. food sci.* **50**(6):1585-1587.

KAMAL, M., RAHMAN, M. M., YASMIN, L., ISLAM, M. N., NURULLAH, M. and MAZID, M. A. 2000. Studies on the post mortem changes in shrimp and prawn during storage: II. Biochemical aspects of quality changes.*Bangladesh J. fish. Res.***4**(1):91-96.

KAMALUDDIN, A., MALEK, M. A. and SANAULLAH, M. 1977. Deshiokhaddeerpustiman.*Ins. Nutr. andFd. SCI. Dhaka University*. pp 1-20.

KHUDA, M.Q. 1964. Evaluation of mechanism of fish spoilage by measurement of fish spoilage by measurement of tyrocine values.*Pak. J. sci.Ind. Res***3**: 187-190

KINSELLA, J.E. 1987. Sea Foods and Fish Oils in Human Health and Disease. Marcel Dekker, Inc. New York: 231-236.

KRIS-ETHERTON, P. M., HARRIS, W. S. and APPEL, L. J. 2002.Fish consumption, fish oil, omega-3 fatty acids, and cardiovascular disease.circulation, spoilage by measurement of tyrocinevalues.*Pak. J. sci.Ind. Res* **3**: 187-190

LAURITZEN, L., HANSEN, H.S., JORGENSEN, M.H.and MICHAELSEN, K.F. 2001. The essentiality of long chain n-3 fatty acids in relation to development and function of the brain and retina.*Prog. Lipid Res.***40**(1): 1-94.

LOVE, R.M. 1970. The Chemical Biology of Fishes.Academic Press, Inc. London, U.K., 547 pp. *Lutjanusjohnii*(hira).*Pak. J. Pharm. Sci*., **4**(2): 91-101.

MATTO, M-DI, BOSCHI, V., MUSSO, S. S.,GRASSO, G. and MATTBQ, M.1990.Chemical composition and protein quality of some Mediterranean Revista-della-societa-Italian-di-Scienza-dell Alimentazione.**19**:1-2.23.

MAZUMDER, M.S.A., RAHMAN, M.M., AHMED, A.T.A., BEGUM, M. and HOSSAIN, M. A. 2008. Proximate composition of some small indigenous fish species (SIS) in Bangladesh.*Int. J. sust.Crop Pro.***3**(4): 18-23.

MINIADIS-MEIMAROGLOU, S., KORA, L. and SINANOGLOU, V. J. 1998.Isolation and identification of phospholipid molecular species in α wild marine shrimp Penaeuskerathurus muscle and cephalothorax.*Chem. Phys.Lipids.***152**(2):104-112.

MURRA, J. and BURT, J. R. 2001.The Composition of Fish.Ministry of Technology, Torry Research Station (No. 38).Torry advisory note.

NAEEM M., SALAM A. GILLANI Q. and ISHTIAQ, A. 2010**.**Length-weight relationships of *Notopterusnotopterus*and introduced *Oreochromisniloticus*from the Indus River, southern Punjab, *Pakistan. J. Appl. Ichthyol.* **26**: 620.

NARGIS, R. 2006. Seasonal variation in the chemical composition of body flesh of koi fish *Anabas testudineus* (Block) (Anabantidae, Perciformes). *Bangladesh J. Sci. Ind. Res.***41**: 219-226.

NICHOLS, D. S., NICHOLS, P. D.and MCMEEKIN, T. A. 1993. Polyunsaturated fatty acids in Antarctic bacteria.*Antarct. Sci.***5**(02): 149-160.

NOWSAD, A. K. M. 2007. In Participatory Training of Trainers: A new approach applied in Fish Processing, Bangladesh Fisheries Research Forum.

NURULLAH, M., KAMAL, M., WAHAB, M. A., ISLAM, M. N., AHASAN, C. T. and THILSTED, S. 2003. Nutritional quality of some small indigenous fish species of Bangladesh. In BAU-ENRECA/DANIDA Workshop on Potentials of Small Indigenous Species of Fish (SIS) in Aquaculture & Rice-field Stocking for Improved Food & Nutrition Security in Bangladesh, Small indigenous species of fish in Bangladesh (pp. 151-158).

ONYIA, L. U., MICHAEL, K. S., MANU, J. M. and SABO, M. 2013. Comparison of Nutrient Values of Wild and Cultured *Heterobranchusbidorsalis*and *Clariasgariepinus. Nigerian J. Fish. Aquacult.***1**(1): 7 – 12.

ORBAN, E., LENA, G. DI., NEVIGATO, T., CASINI, I., MARZETTI, A. and CAPRONI, R. (2002). Seasonal changes in meat content, condition index and chemical composition of mussels (*Mytilusgalloprovincialis*) cultured in two different italian sites. *Food chem.* **77**(1):57–65.

OSMAN, H., SURIAH, A. R. and LAW, E. C. 2001. Fatty acid composition and cholesterol content of selected marine fish in Malaysian waters. *Food chem.***73**(1):55-60.

SAOUD, I.P., BATAL, M., GHANAWI, J. and LEBBOS, N. 2008.Seasonal evaluation of nutritional benefits of two fish species in the eastern Mediterranean Sea.*Int.J.Food Sci. Technol.***43**(3): 538-542.

PAULY, D. 1993.Linear regressions in fisheries research.*J. Fish. Res. Board Canada.***30**:409-434.

PIGGOTT, G. M. and TUCKER, B. W. 1990. Sea Foods: Effects of Technology on Nutrition. New York: CRC Press, Marcel Dekker. pp. 155–170.

PUWASTIEN, P., RAROENGWICHIT, M., SUNGPUAG, P., and JUDPRASONG, K. 1999.Thai food composition tables.*Institute of Nutrition Mahidol University (INMU),* Thailand.

RAFFLENBEUL, W. 2001.Fish for a healthy heart.*European J. lipid Sci.Tech.***103**(5): 315-317.

RIEDEL, R., CASKEY, L.M. and HURLBERT, S.H. 2007. Length-weight relations and growth rates of dominant fishes of the Salton Sea: implications for predation by fish-eating birds. *Lake and Res.Manag*.**23**:528-535.

RUBBI, S. F. MUJIBAR, M., A. R. JAHAN, S. S. and MAJEDA, B.1987.Proximate composition and quality of some commercial species of fresh water fish.*J. sci. Reg*.**5**(1):1-20

SANGUN, L., AKAMCA, E. and AKAR M. (2007).Weight-Length Relationships for 39 Fish Species from the North-Eastern Mediterranean Coast of Turkey.*Turkish J. Fisheries Aqua. Sci.* **7**: 37-40.

SATO, M., YOSHINAKA, R., NISHINAKA, Y., MORIMOTO, H., KOJIMA, T., YAMAMOTO, Y. and IKEDA, S. 1986. Comparison of nutritive components in meat of wild and cultured bastard halibut Paralichthysolivaceus.*Bulletin of the Japanese Society of Scientific Fisheries*.

SHCHENIKOVA, N. V., PAVLYCHEVA, E. V., DAVYDOVA, S. A., ISA, S. V. and SOKOLOVA, L. I. (1987). Fatty acid composition of the lipids of the mantle and liver of cephalopod mollusks.*Aquaculture***6**:61–64.

SHEARER, K. D. 1994.Factors affecting the proximate composition of cultured fishes with emphasis on salmonids.*Aquaculture***119**(1): 63-88.

SIGURGISLADÓTTIR, S. and PÁLMADÓTTIR, H. 1993.Fatty acid composition of thirty-five Icelandic fish species.. J. *Am. Oil Chem. Soc.***70**(11): 1081-1087.

SIMOPOULOS, A. P. 1989. Summary of NATO advanced research workshop on dietary N-3 and N-6 fatty acids: Biological effects and nutritional essentiality. *J. Nutr.***199**: 512–528.

SINGH, M. B., SAROJNALINI, C. and VISHWANATH, W. 1990. Nutritive values of sun-dried *Esomusdanricus* and smoked *Lepidocephalusguntea*. *Food chem.***36**(2):89-96.

SPSS (1999).Computer program, MS. for Windows, version 10.01, SPSS Inc. USA.

STANSBY, M. E. (1982) Properties of fish oils and their application to handling of fish and to nutritional and industrial use. In: Martin RE, Flick GJ, Hebard CE, Ward DR (eds) Chemistry and biochemistry of marine food products. Avi Publishing Wesport, Connecticut, pp 75–92

STANSBY, M. E. 1954. Composition of certain species of fresh-water.*J. Food Sci*, **19**(1): 231-234.

SUSHCHIK, N.N., GLADYSHEVL, M.I. and KALACHOVA, G.S. 2007. Seasonal dynamics of fatty acid content of a common food fish from the Yenisei river, Siberian grayling, Thymallusarcticus. *Food Chem*.**104:** 1353–1358.

TADROS, A. B., HAMAIDA, H. and SAID, T. O. 2005. Chemical characteristics of El-Mex fish farm ponds. *Egyptian J. Aqu Res.***1**:191-212.

THOMAS, J.S., VENUS and B. M. KURUP 2003.Length-weight relationship of some deep-sea fish inhabiting continental slope beyond 250 m depth along West coast of India *Naga, world Fish Center Quart.,* **26:** 17-21

THURSTON, C. E. 1958. Variation in composition of Southern Alaska Pink Salmon.*Food Res*, **23**: 313-325.

VENKATARAMA, N. and CHARI, A.P. 1951. The elementary chemical composition of marine organism. Yale Univ. Press, New Haven, pp. 463-566

WATKINS, B. A., LI, Y., LIPPMAN, H. E. and FENG, S. 2003. Modulatory effect of omega-3 polyunsaturated fatty acids on osteoblast function and bone metabolism. *Bangladesh J. Sci. Ind. Res.***68**(6): 387-398.

WESSELINOVA, D. 2000. Amino acid composition of fish meat after different frozen storage periods.*J. Sci. Ind. Res***9**(4):41-48.

YEGANEH, S., SHABANPOUR, B., HOSSEINI, H., IMANPOUR, M. R. and SHABANI, A. 2012. Comparison of farmed and wild common carp (*Cyprinuscarpio*): seasonal variations in chemical composition and fatty acid profile. *Czech J. of Food Sci.***30**(6): 503-511.

YEGANEH, S., SHABANPOUR, B., HOSSEINI, H., IMANPOUR, M. R. and SHABANI, A. 2012. Comparison of farmed and wild common carp (*Cyprinuscarpio*): seasonal variations in chemical composition and fatty acid profile. *Czech J. of Food Sci.***30**(6): 503-511.

APPENDICES

Appendix A

Table 1: Physical characteristics of *Anabas testudineus* collected in two different seasons

Season	Wild			Cultured		
	Weight (g)	Length (cm)		Weight (g)	Length (cm)	
		Total length	Standard length		Total length	Standard length
Pre monsoon	26.58	9.3	7.8	40.62	10.2	8.8
	28.61	10.0	8.5	35.87	8.5	6.7
	24.92	8.5	7.2	39.93	9.8	7.9
	22.85	7.9	6.2	33.81	7.8	6.1
	23.75	8.0	6.4	34.52	8.5	6.5
	20.31	7.1	5.8	41.56	10.3	8.8
	21.96	7.5	6.0	37.76	9.0	7.5
	28.84	10.3	8.6	39.52	9.2	7.2
	27.45	9.5	7.6	42.76	10.6	8.7
	25.82	9.1	7.5	36.45	8.6	6.5
Post monsoon	31.79	13.0	10.9	52.25	13.5	11.2
	27.61	11.0	9.2	49.69	12	10.9
	26.82	9.8	7.5	42.78	10.5	9.1
	28.52	11.2	9.4	51.22	13.1	11.5
	30.67	12.2	10.5	50.48	12.2	11.4
	32.54	13.5	11.3	43.81	10.7	8.8
	31.86	13.3	11.0	42.52	10.5	8.5
	27.94	10.5	8.5	41.56	10.6	8.6
	29.67	11.6	9.7	52.84	13.3	12.0
	32.61	13.8	11.5	51.83	13.5	11.5

Table 2: Mean length and weight of wild and cultured fish of both seasons. Result showed Mean value ± SEM

	Wild		Cultured	
	Pre-monsoon (March- May)	Post-monsoon (September-November)	Pre-monsoon (March -May)	Post-monsoon (September-November)
Mean Total length (cm)	8.72±1.08 (7.10 -10.30)	11.99±1.38 (9.80-13.80)	9.25±0.93 (7.80 - 10.60)	10.49±1.91 (7.10 to 13.80)
Mean Total weight (g)	25.11±2.87 (20.31-28.84)	30.00±2.18 (26.82- 32.61)	38.21± 3.06 (33.81 -42.76)	47.90± 4.62 (41.56 to 52.84)

Appendix B

Table 3:Seasonal variation in the percentage of Nutritional composition of wild and cultured koi fish. Data are expressed as mean ± 1 SEM.Averages followed by Different letters within a row denote significant differences ($p < 0.05$) and the same letter denotes no significance ($p>0.05$).

Proximate composition(%)	Wild		Cultured	
	Pre-monsoon	Post-monsoon	Pre-monsoon	Post-monsoon
Moisture	79.62±0.29[a]	75.27±0.05[b]	74.04±0.18[cd]	73.91±0.16[d]
Ash	3.62±0.02[a]	3.02±0.01[b]	2.50±0.03[c]	2.90±0.03[d]
Protein	12.13±0.01[a]	14.74±0.02[b]	16.04±0.02[c]	17.92±0.02[d]
Lipid	3.82±0.02[a]	3.61±0.02[b]	5.14±0.01[c]	5.56±0.01[d]

Table 4: Mean±1 SEM for proximate compostion of wild and cultured koi fish of both seasons

				Moisture	Ash	Protein	Lipid
Group	Premonsoon_Wild	1		80.21	3.56	12.09	3.74
		2		80.62	3.71	12.12	3.81
		3		78.02	3.59	12.18	3.91
		4		80.25	3.64	12.15	3.77
		5		79.77	3.57	12.11	3.75
		6		78.78	3.62	12.09	3.91
		7		80.23	3.57	12.19	3.90
		8		78.35	3.67	12.17	3.79
		9		79.62	3.65	12.09	3.77
		10		80.35	3.62	12.13	3.82
		Total	Mean	79.6200	3.6200	12.1320	3.8170
			Std. Error of Mean	.28965	.01542	.01218	.02103
	Premonsoon_Cultured	1		73.76	2.60	16.09	5.17
		2		74.92	2.48	15.95	5.11
		3		73.53	2.42	16.08	5.14
		4		74.54	2.62	15.94	5.12
		5		73.23	2.37	16.05	5.11
		6		74.78	2.58	16.08	5.16
		7		73.52	2.54	16.12	5.17
		8		74.04	2.38	15.98	5.17
		9		73.86	2.54	15.99	5.13
		10		74.22	2.50	16.12	5.13
		Total	Mean	74.0400	2.5030	16.0400	5.1410

		Std. Error of Mean	.17956	.02829	.02181	.00781
Postmonsoon_Wild	1		75.24	3.07	14.76	3.56
	2		75.31	2.98	14.81	3.58
	3		75.25	3.01	14.65	3.69
	4		75.27	3.07	14.67	3.57
	5		75.24	2.99	14.75	3.56
	6		75.34	3.03	14.73	3.69
	7		75.45	2.97	14.79	3.69
	8		74.99	3.04	14.75	3.63
	9		75.53	3.06	14.81	3.57
	10		75.12	2.97	14.71	3.59
	Total	Mean	75.2740	3.0190	14.7430	3.6130
		Std. Error of Mean	.04824	.01278	.01726	.01795
Postmonsoon_Cultured	1		74.46	2.85	17.96	5.59
	2		74.22	3.01	17.85	5.56
	3		73.06	2.84	17.95	5.53
	4		73.87	2.85	17.81	5.56
	5		74.07	2.85	17.96	5.53
	6		73.54	2.79	17.93	5.54
	7		73.65	3.05	17.92	5.56
	8		73.39	3.03	17.89	5.61
	9		74.59	2.98	17.96	5.54
	10		74.25	2.75	17.96	5.55
	Total	Mean	73.9100	2.9000	17.9190	5.5570
		Std. Error of Mean	.15639	.03393	.01676	.00817
Total	Mean		75.7110	3.0105	15.2085	4.5320
	Std. Error of Mean		.38225	.06517	.33713	.13362

a. Limited to first 100 cases.

Table 5: One way ANOVA to analysis the significance variation in nutritional composition of Wild and Cultured *Anabas testudineus,* collected in two different seasons.

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Moisture	Between Groups	215.071	3	71.690	200.635	.000
	Within Groups	12.863	36	.357		
	Total	227.934	39			
Ash	Between Groups	6.413	3	2.138	363.531	.000

	Within Groups	.212	36	.006		
	Total	6.625	39			
Protein	Between Groups	177.197	3	59.066	19641.321	.000
	Within Groups	.108	36	.003		
	Total	177.306	39			
Lipid	Between Groups	27.773	3	9.258	4149.341	.000
	Within Groups	.080	36	.002		
	Total	27.853	39			

Table 6: Tukey HSDfor Proximate composition

Homogeneous Subsets

Moisture

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
Postmonsoon_Cultured	10	73.9100		
Premonsoon_Cultured	10	74.0400		
Postmonsoon_Wild	10		75.2740	
Premonsoon_Wild	10			79.6200
Sig.		.962	1.000	1.000

Means for groups in homogeneous subsets are displayed.
Uses Harmonic Mean Sample Size = 10.000.

Ash

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
Premonsoon_Cultured	10	2.5030			
Postmonsoon_Cultured	10		2.9000		
Postmonsoon_Wild	10			3.0190	
Premonsoon_Wild	10				3.6200
Sig.		1.000	1.000	1.000	1.000

Protein

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
Premonsoon_Wild	10	12.1320			
Postmonsoon_Wild	10		14.7430		
Premonsoon_Cultured	10			16.0400	
Postmonsoon_Cultured	10				17.9190
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

Lipid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
Postmonsoon_Wild	10	3.6130			
Premonsoon_Wild	10		3.8170		
Premonsoon_Cultured	10			5.1410	
Postmonsoon_Cultured	10				5.5570
Sig.		1.000	1.000	1.000	1.000

Appendix C

Table 7: Amino Acid composition (%) of wild and cultured koi fish in pre-monsoon and post-monsoon

Protein and Amino Acids	Wild (%)		Cultured (%)	
	Pre-monsoon	Post-monsoon	Pre-monsoon	Post-monsoon
Protein	12.13±0.01[a]	14.74±0.02[b]	16.04±0.02[c]	17.92±0.02[d]
Threonone[EA]	0.277±0.003[a]	0.320±0.012[a]	0.437±0.003[b]	0.460±0.023[b]
Valine[EA]	0.090±0.012[a]	0.100±0.006[a]	0.110±0.01[a]	0.140±0.017[a]
Methionine[EA]	0.313±0.017[a]	0.350±0.012[a]	0.457±0.013[b]	0.490±0.017[b]
Isoleucine[EA]	0.527±0.019[a]	0.620±0.023[a]	0.8433±0.029[b]	0.907±0.020[b]
Leucine[EA]	0.133±0.012[a]	0.170±0.012[ab]	0.170±0.01[ab]	0.210±0.006[b]
Lysine[EA]	0.300±0.015[a]	0.370±0.012[b]	0.367±0.007[b]	0.440±0.017[c]
Aspartic Acid[NEA]	0.737±0.007[a]	0.760±0.017[a]	0.890±0.01[b]	0.930±0.006[b]
Serine[NEA]	0.323±0.009[a]	0.360±0.023[a]	0.463±0.003[b]	0.490±0.006[b]
Glutamic Acid[NEA]	0.990±0.015[a]	1.040±0.023[a]	1.147±0.015[b]	1.260±0.017[c]
Glycine[NEA]	1.290±0.015[a]	1.380±0.012[b]	1.450±0.012[c]	1.580±0.017[d]
Alanine[NEA]	0.397±0.012[a]	0.420±0.017[ab]	0.490±0.01[bc]	0.550±0.029[c]
Histidine[NEA]	2.023±0.048[a]	2.240±0.023[a]	1.823±0.015[b]	1.870±0.006[c]
Tyrosine[NEA]	0.383±0.009[a]	0.440±0.012[ab]	0.473±0.007[b]	0.550±0.023[c]
Arginine[NEA]	0.407±0.009[a]	0.47±0.006[ab]	0.52±0.015[bc]	0.58±0.023[c]
EA/NEA	0.23	0.27	0.32	0.33

Data are expressed as mean ± standard error (n = 3).

Different letters within a row denote significant differences ($p < 0.05$) and the same letter denotes no significances ($p>0.05$)

[EA] Essential amino acids, [NEA]Non-essential amino acids.

Table 8: Values shows in Mean±1 SEM for Amino acids of Wild and cultured of both seasons

				Aspartic_Acid	Threonine	Serine	Glutamic_Acid	Glycine	Alanine	Valine	Methionine	Isoleucine	Leucine	Tyrosine	Histidine	Lysine	Arginine
Group	Premonsoon_Wild	1		.73	.28	.32	.98	1.27	.38	.09	.28	.55	.14	.38	1.98	.28	.41
		2		.75	.28	.34	.97	1.28	.39	.11	.33	.54	.15	.37	2.12	.29	.39
		3		.73	.27	.31	1.02	1.32	.42	.07	.33	.49	.11	.40	1.97	.33	.42
		Total	Mean	.7367	.2767	.3233	.9900	1.290	.3967	.0900	.3133	.5267	.1333	.3833	2.0233	.3000	.4067
			Std. Error of Mean	.00667	.00333	.0088	.01528	.0152	.0120	.0115	.01667	.01856	.0120	.0088	.04842	.0152	.0088
	Postmonsoon_Wild	1		.76	.32	.36	1.04	1.38	.42	.10	.35	.62	.17	.44	2.24	.37	.47
		2		.79	.30	.40	1.08	1.40	.45	.11	.37	.66	.19	.46	2.20	.35	.46
		3		.73	.34	.32	1.00	1.36	.39	.09	.33	.58	.15	.42	2.28	.39	.48
		Total	Mean	.7600	.3200	.3600	1.0400	1.380	.4200	.1000	.3500	.6200	.1700	.4400	2.2400	.3700	.4700
			Std. Error of Mean	.01732	.01155	.0230	.02309	.0115	.0173	.0057	.01155	.02309	.0115	.0115	.02309	.0115	.0057
	Premonsoon_Cultured	1		.91	.43	.47	1.15	1.45	.51	.12	.43	.89	.18	.48	1.80	.38	.53
		2		.88	.44	.46	1.12	1.43	.48	.12	.47	.85	.18	.48	1.85	.36	.54
		3		.88	.44	.46	1.17	1.47	.48	.09	.47	.79	.15	.46	1.82	.36	.49
		Total	Mean	.8900	.4367	.4633	1.1467	1.450	.4900	.1100	.4567	.8433	.1700	.4733	1.8233	.3667	.5200
			Std. Error of Mean	.01000	.00333	.0033	.01453	.0115	.0100	.0100	.01333	.02906	.0100	.0066	.01453	.0066	.0152
	Postmonsoon_Cultured	1		.93	.46	.49	1.26	1.58	.55	.14	.49	.91	.21	.55	1.87	.44	.58
		2		.94	.42	.48	1.23	1.55	.50	.17	.52	.94	.20	.51	1.86	.41	.54
		3		.92	.50	.50	1.29	1.61	.60	.11	.46	.87	.22	.59	1.88	.47	.62
		Total	Mean	.9300	.4600	.4900	1.2600	1.580	.5500	.1400	.4900	.9067	.2100	.5500	1.8700	.4400	.5800
			Std. Error of Mean	.00577	.02309	.0057	.01732	.0173	.0288	.0173	.01732	.02028	.0057	.0230	.00577	.0173	.0230
	Total	Mean		.8292	.3733	.4092	1.1092	1.425	.4642	.1100	.4025	.7242	.1708	.4617	1.9892	.3692	.4942
		Std. Error of Mean		.02530	.02388	.0216	.03223	.0325	.0198	.0075	.02290	.04806	.0092	.0191	.05046	.0159	.0202

a. Limited to first 100 cases.

Table 9: One way ANOVA to analysis the significance variations in amino acid content of wild and cultured fish of both seasons

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Aspartic_Acid	Between Groups	.082	3	.027	75.930	.000
	Within Groups	.003	8	.000		
	Total	.084	11			
Threonine	Between Groups	.071	3	.024	45.892	.000
	Within Groups	.004	8	.001		
	Total	.075	11			
Serine	Between Groups	.058	3	.019	39.158	.000
	Within Groups	.004	8	.000		
	Total	.062	11			
Glutamic_Acid	Between Groups	.129	3	.043	45.017	.000
	Within Groups	.008	8	.001		
	Total	.137	11			
Glycine	Between Groups	.135	3	.045	74.833	.000
	Within Groups	.005	8	.001		
	Total	.140	11			
Alanine	Between Groups	.044	3	.015	14.073	.001
	Within Groups	.008	8	.001		
	Total	.052	11			
Valine	Between Groups	.004	3	.001	3.294	.079
	Within Groups	.003	8	.000		
	Total	.008	11			
Methionine	Between Groups	.064	3	.021	31.946	.000
	Within Groups	.005	8	.001		
	Total	.069	11			
Isoleucine	Between Groups	.292	3	.097	60.852	.000
	Within Groups	.013	8	.002		
	Total	.305	11			
Leucine	Between Groups	.009	3	.003	9.541	.005
	Within Groups	.002	8	.000		
	Total	.011	11			
Tyrosine	Between Groups	.044	3	.015	24.582	.000
	Within Groups	.005	8	.001		
	Total	.048	11			
Histidine	Between Groups	.317	3	.106	45.176	.000
	Within Groups	.019	8	.002		
	Total	.336	11			
Lysine	Between Groups	.029	3	.010	18.391	.001
	Within Groups	.004	8	.001		
	Total	.034	11			
Arginine	Between Groups	.049	3	.016	24.722	.000
	Within Groups	.005	8	.001		
	Total	.054	11			

Table 10: Turkey HSD for significance test within and between groups of variables

Serine
Tukey HSD

Group	N	Subset for alpha = 0.05	
		1	2
Premonsoon_Wild	3	.3233	
Postmonsoon_Wild	3	.3600	
Premonsoon_Cultured	3		.4633
Postmonsoon_Cultured	3		.4900
Sig.		.256	.494

Means for groups in homogeneous subsets are displayed.

Glutamic_Acid
Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
Premonsoon_Wild	3	.9900		
Postmonsoon_Wild	3	1.0400		
Premonsoon_Cultured	3		1.1467	
Postmonsoon_Cultured	3			1.2600
Sig.		.272	1.000	1.000

Glycine
Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
Premonsoon_Wild	3	1.2900			
Postmonsoon_Wild	3		1.3800		
Premonsoon_Cultured	3			1.4500	
Postmonsoon_Cultured	3				1.5800
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.
Uses Harmonic Mean Sample Size = 3.000.

Alanine

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
Premonsoon_Wild	3	.3967		
Postmonsoon_Wild	3	.4200	.4200	
Premonsoon_Cultured	3		.4900	.4900
Postmonsoon_Cultured	3			.5500
Sig.		.811	.106	.181

Valine

Tukey HSD

Group	N	Subset for alpha = 0.05
		1
Premonsoon_Wild	3	.0900
Postmonsoon_Wild	3	.1000
Premonsoon_Cultured	3	.1100
Postmonsoon_Cultured	3	.1400
Sig.		.069

Methionine

Tukey HSD

Group	N	Subset for alpha = 0.05	
		1	2
Premonsoon_Wild	3	.3133	
Postmonsoon_Wild	3	.3500	
Premonsoon_Cultured	3		.4567
Postmonsoon_Cultured	3		.4900
Sig.		.366	.439

Means for groups in homogeneous subsets are displayed.
Uses Harmonic Mean Sample Size = 3.000.

Isoleucine

Group	N	Subset for alpha = 0.05	
		1	2
Premonsoon_Wild	3	.5267	
Postmonsoon_Wild	3	.6200	
Premonsoon_Cultured	3		.8433
Postmonsoon_Cultured	3		.9067
Sig.		.081	.285

Means for groups in homogeneous subsets are displayed.
Uses Harmonic Mean Sample Size = 3.000.

Leucine

Tukey HSD

Group	N	Subset for alpha = 0.05	
		1	2
Premonsoon_Wild	3	.1333	
Postmonsoon_Wild	3	.1700	.1700
Premonsoon_Cultured	3	.1700	.1700
Postmonsoon_Cultured	3		.2100
Sig.		.124	.089

Means for groups in homogeneous subsets are displayed.
Uses Harmonic Mean Sample Size = 3.000.

Tyrosine

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
Premonsoon_Wild	3	.3833		
Postmonsoon_Wild	3	.4400	.4400	
Premonsoon_Cultured	3		.4733	
Postmonsoon_Cultured	3			.5500
Sig.		.082	.393	1.000

Means for groups in homogeneous subsets are displayed.

Histidine

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
Premonsoon_Cultured	3	1.8233		
Postmonsoon_Cultured	3	1.8700		
Premonsoon_Wild	3		2.0233	
Postmonsoon_Wild	3			2.2400
Sig.		.654	1.000	1.000

Lysine

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
Premonsoon_Wild	3	.3000		
Premonsoon_Cultured	3		.3667	
Postmonsoon_Wild	3		.3700	
Postmonsoon_Cultured	3			.4400
Sig.		1.000	.998	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Arginine

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
Premonsoon_Wild	3	.4067		
Postmonsoon_Wild	3	.4700	.4700	
Premonsoon_Cultured	3		.5200	.5200
Postmonsoon_Cultured	3			.5800
Sig.		.064	.157	.081

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Appendix D

Table 11: Descriptive statistics for fatty acid of wild and cultured fish of both seasons

		N	Mean	Std. Deviation	Std. Error	Min	Max
Caproic_Acid	pre-monsoon wild	3	.077000	.0020000	.0011547	.0750	.0790
	post-monsoon wild	3	.673000	.0030000	.0017321	.6700	.6760
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.181333	.0015275	.0008819	.1800	.1830
	Total	12	.232833	.2738138	.0790432	.0000	.6760
Enanthic_Acid	pre-monsoon wild	3	.075000	.0040000	.0023094	.0710	.0790
	post-monsoon wild	3	.611000	.0030000	.0017321	.6080	.6140
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.345333	.0011547	.0006667	.3440	.3460
	Total	12	.257833	.2517068	.0726615	.0000	.6140
Caprylic_Acid	pre-monsoon wild	3	.058000	.0010000	.0005774	.0570	.0590
	post-monsoon wild	3	.440000	.0400000	.0230940	.4000	.4800
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.227700	.0090150	.0052048	.2190	.2370
	Total	12	.181425	.1796001	.0518461	.0000	.4800
Capric_Acid	pre-monsoon wild	3	.018000	.0020000	.0011547	.0160	.0200
	post-monsoon wild	3	.192000	.0040000	.0023094	.1880	.1960
	pre-monsoon cultured	3	.050667	.0011547	.0006667	.0500	.0520
	post-monsoon cultured	3	.132600	.0074081	.0042771	.1250	.1398
	Total	12	.098317	.0714601	.0206288	.0160	.1960
Lauric_Acid	pre-monsoon wild	3	.054000	.0030000	.0017321	.0510	.0570
	post-monsoon wild	3	.176000	.0010000	.0005774	.1750	.1770
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.057500	.0750884	.0216762	.0000	.1770
Myristic_Acid	pre-monsoon wild	3	1.630000	.0200000	.0115470	1.6100	1.6500
	post-monsoon wild	3	1.595000	.0040000	.0023094	1.5910	1.5990
	pre-monsoon cultured	3	1.524000	.0055678	.0032146	1.5180	1.5290
	post-monsoon cultured	3	.854000	.0050000	.0028868	.8490	.8590

	Total	12	1.400750	.3322360	.0959083	.8490	1.6500
Pentadecyclic_Acid	pre-monsoon wild	3	.082000	.0010000	.0005774	.0810	.0830
	post-monsoon wild	3	.252000	.0010000	.0005774	.2510	.2530
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.083500	.1074587	.0310206	.0000	.2530
Palmitic_Acid	pre-monsoon wild	3	29.377000	.0030000	.0017321	29.3740	29.3800
	post-monsoon wild	3	26.138000	.0020000	.0011547	27.1360	27.1400
	pre-monsoon cultured	3	30.276667	.2550163	.1472338	30.0200	30.5300
	post-monsoon cultured	3	27.91000	.0070000	.0040415	27.3540	27.3670
	Total	12	28.537667	1.3935227	.4022754	27.1360	30.5300
Margaric_Acid	pre-monsoon wild	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon wild	3	.128000	.0010000	.0005774	.1270	.1290
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.032000	.0578917	.0167119	.0000	.1290
Stearic_Acid	pre-monsoon wild	3	8.550000	.0200000	.0115470	8.5300	8.5700
	post-monsoon wild	3	8.245000	.0010000	.0005774	8.2440	8.2460
	pre-monsoon cultured	3	8.012667	.0145029	.0083732	7.9980	8.0270
	post-monsoon cultured	3	9.072000	.0062450	.0036056	8.4660	8.4780
	Total	12	8.320167	.2195996	.0633929	7.9980	8.5700
Arachidic_Acid	pre-monsoon wild	3	.198000	.0010000	.0005774	.1970	.1990
	post-monsoon wild	3	.226000	.0030000	.0017321	.2230	.2290
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.106000	.1112033	.0321016	.0000	.2290
Behenic_Acid	pre-monsoon wild	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon wild	3	.072000	.0010000	.0005774	.0710	.0730
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000

	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.018000	.0325660	.0094010	.0000	.0730
Palmitolic_Acid	pre-monsoon wild	3	4.782000	.0010000	.0005774	4.7810	4.7830
	post-monsoon wild	3	4.616000	.0070000	.0040415	4.6080	4.6210
	pre-monsoon cultured	3	2.707000	.0085440	.0049329	2.6980	2.7150
	post-monsoon cultured	3	1.608667	.0520801	.0300684	1.5490	1.6450
	Total	12	3.428417	1.3892147	.4010317	1.5490	4.7830
Oleic_Acid	pre-monsoon wild	3	43.107000	.0020000	.0011547	44.0650	44.0690
	post-monsoon wild	3	33.864000	.1798527	.1038380	33.6910	34.0500
	pre-monsoon cultured	3	42.351333	.0077675	.0044845	42.3450	42.3600
	post-monsoon cultured	3	44.994333	.0065064	.0037565	44.9880	45.0010
	Total	12	41.319167	4.6040679	1.3290799	33.6910	45.0010
Vaccenic_Acid	pre-monsoon wild	3	.356667	.0073711	.0042557	.3510	.3650
	post-monsoon wild	3	.145000	.0010000	.0005774	.1440	.1460
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.125417	.1525740	.0440443	.0000	.3650
Eicosenoic_Acid	pre-monsoon wild	3	.747333	.0032146	.0018559	.7450	.7510
	post-monsoon wild	3	.856667	.0037859	.0021858	.8540	.8610
	pre-monsoon cultured	3	.281000	.0020000	.0011547	.2790	.2830
	post-monsoon cultured	3	.435000	.0034641	.0020000	.4310	.4370
	Total	12	.580000	.2421483	.0699022	.2790	.8610
Hexadecatrienoic_Acid	pre-monsoon wild	3	.133333	.0005774	.0003333	.1330	.1340
	post-monsoon wild	3	.743667	.0032146	.0018559	.7400	.7460
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.219250	.3213082	.0927537	.0000	.7460
Hexadecadienoic_Acid	pre-monsoon wild	3	.103333	.0057735	.0033333	.1000	.1100
	post-monsoon wild	3	.503333	.0057735	.0033333	.5000	.5100
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000

	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.151667	.2166200	.0625328	.0000	.5100
Linolenic_Acid	pre-monsoon wild	3	1.613333	.0085049	.0049103	1.6070	1.6230
	post-monsoon wild	3	3.566667	.0152753	.0088192	3.5500	3.5800
	pre-monsoon cultured	3	.825333	.0060277	.0034801	.8190	.8310
	post-monsoon cultured	3	1.383000	.0030000	.0017321	1.3800	1.3860
	Total	12	1.847083	1.0792910	.3115645	.8190	3.5800
Linoleic_Acid	pre-monsoon wild	3	3.384333	.0050332	.0029059	4.3790	4.3890
	post-monsoon wild	3	10.41467	.0110604	.0063857	10.6920	10.7140
	pre-monsoon cultured	3	11.923333	.0040415	.0023333	11.9190	11.9270
	post-monsoon cultured	3	10.028333	.0028868	.0016667	10.0250	10.0300
	Total	12	9.259917	3.0244501	.8730835	4.3790	11.9270
Eicosadienoic_Acid	pre-monsoon wild	3	.321000	.0026458	.0015275	.3190	.3240
	post-monsoon wild	3	.465667	.0005774	.0003333	.4650	.4660
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.196667	.2122479	.0612707	.0000	.4660
Eicosatrienoic_Acid	pre-monsoon wild	3	.321667	.0005774	.0003333	.3210	.3220
	post-monsoon wild	3	.526667	.0030551	.0017638	.5240	.5300
	pre-monsoon cultured	3	.396333	.0070238	.0040552	.3890	.4030
	post-monsoon cultured	3	.583333	.0251661	.0145297	.5600	.6100
	Total	12	.457000	.1086328	.0313596	.3210	.6100
Arachidonic_Acid	pre-monsoon wild	3	1.238333	.0015275	.0008819	1.2370	1.2400
	post-monsoon wild	3	2.172667	.0596769	.0344545	2.1040	2.2120
	pre-monsoon cultured	3	.645667	.0011547	.0006667	.6450	.6470
	post-monsoon cultured	3	1.195333	.0552932	.0319235	1.1320	1.2340
	Total	12	1.313000	.5740279	.1657076	.6450	2.2120

Eicosapentanoic_Acid	pre-monsoon wild	3	.061667	.0005774	.0003333	.0610	.0620
	post-monsoon wild	3	.162000	.0010000	.0005774	.1610	.1630
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.328667	.0020817	.0012019	.3270	.3310
	Total	12	.138083	.1298296	.0374786	.0000	.3310
Docosatrienoic_Acid	pre-monsoon wild	3	.444000	.0055678	.0032146	.4380	.4490
	post-monsoon wild	3	.621000	.0026458	.0015275	.6190	.6240
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.256667	.0035119	.0020276	.2530	.2600
	Total	12	.330417	.2404471	.0694111	.0000	.6240
Adrenic_Acid	pre-monsoon wild	3	.343667	.0032146	.0018559	.3400	.3460
	post-monsoon wild	3	.334667	.0110604	.0063857	.3230	.3450
	pre-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	post-monsoon cultured	3	.370833	.0002887	.0001667	.3705	.3710
	Total	12	.262292	.1588539	.0458572	.0000	.3710
Docosapentanoic_Acid	pre-monsoon wild	3	.756000	.0070000	.0040415	.7510	.7640
	post-monsoon wild	3	.582000	.0105357	.0060828	.5710	.5920
	pre-monsoon cultured	3	.014333	.0005774	.0003333	.0140	.0150
	post-monsoon cultured	3	.000000	0E-7	0E-7	.0000	.0000
	Total	12	.338083	.3516342	.1015081	.0000	.7640
Docosahexanoic_Acid	pre-monsoon wild	3	.970667	.0055076	.0031798	.9650	.9760
	post-monsoon wild	3	1.408333	.0176163	.0101708	1.3920	1.4270
	pre-monsoon cultured	3	.971333	.0005774	.0003333	.9710	.9720
	post-monsoon cultured	3	.137833	.0002887	.0001667	.1375	.1380
	Total	12	.872042	.4804781	.1387021	.1375	1.4270

Table 12: One way ANOVA to test the analysis of variance for fatty acid content of wild and cultured fish of both seasons

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Caproic_Acid	Between Groups	.825	3	.275	71711.565	.000
	Within Groups	.000	8	.000		
	Total	.825	11			
Enanthic_Acid	Between Groups	.697	3	.232	35284.405	.000
	Within Groups	.000	8	.000		
	Total	.697	11			
Caprylic_Acid	Between Groups	.351	3	.117	278.555	.000
	Within Groups	.003	8	.000		

	Total	.355	11			
Capric_Acid	Between Groups	.056	3	.019	980.049	.000
	Within Groups	.000	8	.000		
	Total	.056	11			
Lauric_Acid	Between Groups	.062	3	.021	8266.800	.000
	Within Groups	.000	8	.000		
	Total	.062	11			
Myristic_Acid	Between Groups	1.213	3	.404	3427.244	.000
	Within Groups	.001	8	.000		
	Total	1.214	11			
Pentadecyclic_Acid	Between Groups	.127	3	.042	84678.000	.000
	Within Groups	.000	8	.000		
	Total	.127	11			
Palmitic_Acid	Between Groups	21.231	3	7.077	434.865	.000
	Within Groups	.130	8	.016		
	Total	21.361	11			
Margaric_Acid	Between Groups	.037	3	.012	49152.000	.000
	Within Groups	.000	8	.000		
	Total	.037	11			
Stearic_Acid	Between Groups	.529	3	.176	1084.906	.000
	Within Groups	.001	8	.000		
	Total	.530	11			
Arachidic_Acid	Between Groups	.136	3	.045	18134.400	.000
	Within Groups	.000	8	.000		
	Total	.136	11			
Behenic_Acid	Between Groups	.012	3	.004	15552.000	.000
	Within Groups	.000	8	.000		
	Total	.012	11			
Palmitolic_Acid	Between Groups	21.223	3	7.074	9980.448	.000
	Within Groups	.006	8	.001		
	Total	21.229	11			
Oleic_Acid	Between Groups	233.107	3	77.702	9577.015	.000
	Within Groups	.065	8	.008		
	Total	233.172	11			
Vaccenic_Acid	Between Groups	.256	3	.085	6167.620	.000
	Within Groups	.000	8	.000		
	Total	.256	11			
Eicosenoic_Acid	Between Groups	.645	3	.215	21144.678	.000
	Within Groups	.000	8	.000		

	Total	.645	11			
Hexadecatrienoic_Acid	Between Groups	1.136	3	.379	141950.865	.000
	Within Groups	.000	8	.000		
	Total	1.136	11			
Hexadecadienoic_Acid	Between Groups	.516	3	.172	10320.667	.000
	Within Groups	.000	8	.000		
	Total	.516	11			
Linolenic_Acid	Between Groups	12.813	3	4.271	48671.821	.000
	Within Groups	.001	8	.000		
	Total	12.814	11			
Linoleic_Acid	Between Groups	100.620	3	33.540	778490.818	.000
	Within Groups	.000	8	.000		
	Total	100.620	11			
Eicosadienoic_Acid	Between Groups	.496	3	.165	90095.636	.000
	Within Groups	.000	8	.000		
	Total	.496	11			
Eicosatrienoic_Acid	Between Groups	.128	3	.043	247.332	.000
	Within Groups	.001	8	.000		
	Total	.130	11			
Arachidonic_Acid	Between Groups	3.611	3	1.204	727.104	.000
	Within Groups	.013	8	.002		
	Total	3.625	11			
Eicosapentanoic_Acid	Between Groups	.185	3	.062	43623.902	.000
	Within Groups	.000	8	.000		
	Total	.185	11			
Docosatrienoic_Acid	Between Groups	.636	3	.212	16844.033	.000
	Within Groups	.000	8	.000		
	Total	.636	11			
Adrenic_Acid	Between Groups	.277	3	.092	2785.333	.000
	Within Groups	.000	8	.000		
	Total	.278	11			
Docosapentanoic_Acid	Between Groups	1.360	3	.453	11308.044	.000
	Within Groups	.000	8	.000		
	Total	1.360	11			
Docosahexanoic_Acid	Between Groups	2.539	3	.846	9924.335	.000
	Within Groups	.001	8	.000		
	Total	2.539	11			

Table 13: Tukey HSD to test the significance variations between and within groups of variables

Capric_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon wild	3	.018000			
pre-monsoon cultured	3		.050667		
post-monsoon cultured	3			.132600	
post-monsoon wild	3				.192000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Enanthic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon cultured	3	.000000			
pre-monsoon wild	3		.075000		
post-monsoon cultured	3			.345333	
post-monsoon wild	3				.611000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Caprylic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon cultured	3	.000000			
pre-monsoon wild	3		.058000		
post-monsoon cultured	3			.227700	
post-monsoon wild	3				.440000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Lauric_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
pre-monsoon cultured	3	.000000		
post-monsoon cultured	3	.000000		
pre-monsoon wild	3		.054000	
post-monsoon wild	3			.176000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Myristic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
post-monsoon cultured	3	.854000			
pre-monsoon cultured	3		1.524000		
post-monsoon wild	3			1.595000	
pre-monsoon wild	3				1.630000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Pentadecyclic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
pre-monsoon cultured	3	.000000		
post-monsoon cultured	3	.000000		
pre-monsoon wild	3		.082000	
post-monsoon wild	3			.252000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Palmitic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
post-monsoon wild	3	27.138000		
post-monsoon cultured	3	27.359000		
pre-monsoon wild	3		29.377000	
pre-monsoon cultured	3			30.276667
Sig.		.225	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Margaric_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05	
		1	2
pre-monsoon wild	3	.000000	
pre-monsoon cultured	3	.000000	
post-monsoon cultured	3	.000000	
post-monsoon wild	3		.128000
Sig.		1.000	1.000

Stearic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon cultured	3	8.012667			
post-monsoon wild	3		8.245000		
post-monsoon cultured	3			8.473000	
pre-monsoon wild	3				8.550000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

Aspartic Acids

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
pre-monsoon cultured	3	.000000		
post-monsoon cultured	3	.000000		
pre-monsoon wild	3		.198000	
post-monsoon wild	3			.226000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Behenic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05	
		1	2
pre-monsoon wild	3	.000000	
pre-monsoon cultured	3	.000000	
post-monsoon cultured	3	.000000	
post-monsoon wild	3		.072000
Sig.		1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Palmitolic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
post-monsoon cultured	3	1.608667			
pre-monsoon cultured	3		2.707000		
post-monsoon wild	3			4.616000	
pre-monsoon wild	3				4.782000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

Oleic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
post-monsoon wild	3	33.864000			
pre-monsoon cultured	3		42.351333		
pre-monsoon wild	3			44.067000	
post-monsoon cultured	3				44.994333
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

b.

Vaccenic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
pre-monsoon cultured	3	.000000		
post-monsoon cultured	3	.000000		
post-monsoon wild	3		.145000	
pre-monsoon wild	3			.356667
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Eicosenoic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon cultured	3	.281000			
post-monsoon cultured	3		.435000		
pre-monsoon wild	3			.747333	
post-monsoon wild	3				.856667
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Hexadecatrienoic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
pre-monsoon cultured	3	.000000		
post-monsoon cultured	3	.000000		
pre-monsoon wild	3		.133333	
post-monsoon wild	3			.743667
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Hexadecadienoic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
pre-monsoon cultured	3	.000000		
post-monsoon cultured	3	.000000		
pre-monsoon wild	3		.103333	
post-monsoon wild	3			.503333
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Linolenic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon cultured	3	.825333			
post-monsoon cultured	3		1.383000		
pre-monsoon wild	3			1.613333	
post-monsoon wild	3				3.566667
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Linoleic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon wild	3	4.384333			
post-monsoon cultured	3		10.028333		
post-monsoon wild	3			10.703667	
pre-monsoon cultured	3				11.923333
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Eicosadienoic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
pre-monsoon cultured	3	.000000		
post-monsoon cultured	3	.000000		
pre-monsoon wild	3		.321000	
post-monsoon wild	3			.465667
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Eicosatrienoic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon wild	3	.321667			
pre-monsoon cultured	3		.396333		
post-monsoon wild	3			.526667	
post-monsoon cultured	3				.583333
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Arachidonic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
pre-monsoon cultured	3	.645667		
post-monsoon cultured	3		1.195333	
pre-monsoon wild	3		1.238333	
post-monsoon wild	3			2.172667
Sig.		1.000	.591	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

b.

Eicosapentanoic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon cultured	3	.000000			
pre-monsoon wild	3		.061667		
post-monsoon wild	3			.162000	
post-monsoon cultured	3				.328667
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

b.

Docosatrienoic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05			
		1	2	3	4
pre-monsoon cultured	3	.000000			
post-monsoon cultured	3		.256667		
pre-monsoon wild	3			.444000	
post-monsoon wild	3				.621000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Adrenic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
pre-monsoon cultured	3	.000000		
post-monsoon wild	3		.334667	
pre-monsoon wild	3		.343667	
post-monsoon cultured	3			.370833
Sig.		1.000	.295	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Docosapentanoic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
post-monsoon cultured	3	.000000		
pre-monsoon cultured	3	.014333		
post-monsoon wild	3		.582000	
pre-monsoon wild	3			.756000
Sig.		.092	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Docosahexanoic_Acid

Tukey HSD

Group	N	Subset for alpha = 0.05		
		1	2	3
post-monsoon cultured	3	.137833		
pre-monsoon wild	3		.970667	
pre-monsoon cultured	3		.971333	
post-monsoon wild	3			1.408333
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.