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403: FISH BIOLOGY AND AQUACULTURE

Chemical composition of fish

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INTRODUCTION

Today there is an ever-increasing awareness about healthy food and fish is finding more acceptance because of its special nutritional qualities. In this context a proper understanding about the biochemical constituents of fish has become a primary requirement for the nutritionists and dieticians.

Fish and fishery products are used in animal feeds. In this case also, proper data on the biochemical composition is essential for formulating such products. Another vital area where accurate information on biochemical composition is a must is processing and preservation of fish and fishery products.

Fish is an easily perishable commodity and deterioration in quality is due to the changes taking place to the various constituents like proteins, lipids etc. Information on the biochemical constituents will help a processing technologist to define the optimum processing and storage conditions, so that the quality is preserved to the maximum extent.

PROXIMATE COMPOSITION

- The four major constituents in the edible portion of fish are water, protein, lipid (fat or oil) and ash (minerals). The analysis of these four basic constituents of fish muscle is often referred to as ‘proximate analysis’. Even though data on proximate composition are critical for many applications and investigations on these lines had been carried out from as early as the 1880s, reliable data on proximate composition of most of the species of fish are difficult to obtain.
- Fishes are a very heterogeneous and highly specialized group evolved through biochemical adaptation and evolution, The habitat and food intake of these species are equally diverse. Some species are exclusively marine while some are confined to freshwater habitats. Some survive in marine as well as freshwater environments. Some marine species migrate to fresh water for spawning whereas many freshwater species enter the sea for spawning. These widely different environmental conditions of temperature, salinity, pressure, availability of food etc. have profound influence on the biochemical composition. There may be group-specific or even species-specific differences in the biochemical composition.

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- The percentage composition of the four major constituents of fish viz. water, protein, lipid and ash (minerals) is referred to as proximate composition (it may be noted that the term does not indicate any degree of inaccuracy in the analysis). These four components account for about 96-98% of total tissue constituents in most cases. The range of values for these constituents in the edible portion of common fish species from Indian coastal waters are given below:
- Carbohydrates, vitamins, nucleotides, other non-protein nitrogenous compounds etc. are also present in small quantities. Though quantitatively minor components, these play vital roles in maintaining the system and thus are essential for growth and development of the organisms.

water	65-90 %
Protein	10-22 %
Fat	1-20 %
Mineral	0.5-5 %

WATER IN FISH TISSUE

- Water is essential for all living systems. Body fluids act as medium of transport of nutrients, metabolites etc. and water is the major component in these fluids. It is required for the normal functioning of many biological molecules. Proteins, for example, can maintain its native form and normal functions only in presence of water. The proportion of water in the flesh varies widely, though in a majority of cases the variation is much narrower, between 70-80%. One of the examples of very high water content is Bombay duck (*Harpodon nehereus*) a species found abundantly along the north-west coast of India, in which case the muscle tissue contains about 90% water. Water is present in two forms in the tissues, bound to the proteins and in the free form. These forms have well defined biological roles. Water is lost from the tissue in many ways during processing and this may affect the quality, especially the texture of the processed products.
- There exists an inverse relationship between the water content and lipid content of fish, such that the sum of the percentages of the two approximates 80 percent. The summation of oil and water, however, is not necessarily constant and frequently spans a range of 78 to 85 percent.

LIPIDS

- Lipids include a wide heterogeneous group of compounds. Lipids are defined as the fraction of any biological material extractable by solvents of low polarity. As can be seen, the definition itself is not a precise one, but that is thought to be the best to include all compounds belonging to this group. Any material extracted with 'fat solvents' like ethyl alcohol, ether, chloroform, hexane, petroleum ether etc. is classified as a lipid. The important type of compounds included in this group are fatty acids, glycerides, phosphoglycerides, sphingolipids, aliphatic alcohols and waxes, steroids and combination of the above type of compounds with proteins, peptides carbohydrates etc. In the case of fish tissues, the major components of lipids are triacylglycerol and phosphoglycerides, both containing long chain fatty acids. Smaller proportions of other components are also present.
- Variations in the lipid content are much wider than that in protein. Fish with fat content as low as 0.5% and as high as 16-18% are of common occurrence. In many species, there is a build up of lipids during the feeding season and decrease during spawning. In fatty fish like oil sardine, mackerel, herring etc. the main site of storage of lipids is the muscle. The lipid content such fish show wide variations with season and sexual maturity. The lipid content of the muscle of oil sardine (*Sardinella longiceps*) is about 3-4% in June-July, which increases to about 18% by November-December

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- In animals energy is stored mainly as fat. When excess energy is available from food, it is stored as fat and it is utilized during periods of low energy availability. This is true in the case of fish also. The storage sites for fat (fat depot) are different for different species. In some cases it is the liver and in some other cases it is the adipose tissue. In a great majority of cases, the depot fat is mainly triacylglycerol.
- Phospholipids, another important constituents of lipids are essential components of cell membranes. It is the lipid-globular protein mosaic structure that determines important functions like permeability of cell membranes, transport of various substances into and outside the cell. Various types of phospholipids are essential for the proper functioning of the cell. Unlike in the case of depot fat, the proportions of phospholipids do not show wide variation. Normally it is in the range of 0.5 to 1% of tissue.
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CHANGES TO LIPIDS DURING PROCESSING/STORAGE

- Two types changes take place to lipids during processing and preservation of fish, hydrolysis and oxidation. Lipid hydrolysis results in release of free fatty acids and these free fatty acids cause protein denaturation. Denatured protein looses its characteristic properties and this leads to loss of quality, especially the texture. Lipid hydrolysis in the tissue is mainly due to the action of lipases and the activity of these enzymes will be low at low temperatures. Thus lipid hydrolysis and consequent deterioration of quality will be minimum in products kept at low temperatures (about –18 to –20°C).
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- The process is characterized by an induction period, during which the oxidation is slow, followed by an accelerating rate of oxygen absorption with concurrent development of hydroperoxides, which are the primary products of oxidation. The hydroperoxides undergo decomposition to various products like aldehydes, ketones, alcohols, carboxylic acids etc. some of which are volatile and some, non-volatile. The number and nature of these decomposition products depend on the position of the double bond being oxidized and the conditions under which the hydroperoxides are decomposed. The rancid flavour is the net result of these changes. Further oxidation of highly unsaturated fatty acids will lead to formation of polymerized products and under these conditions fish or oil will become totally unacceptable.
- Presence of air, elevated temperature, catalysts etc. are the main agents which promote oxidation. Avoiding these conditions can minimize the risk of oxidation. Another effective method of preventing oxidative degradation is the use of antioxidants. Antioxidants prevent or retard the propagation of free radical chain reaction and thus minimize the damage caused by oxidation. Several naturally occurring and synthetic antioxidants like tocopherols, butylated hydroxyanisol (BHA), butylated hydroxytoluene (BHT), propyl gallate etc. are used (usually 0.1 to 0.2%) during the processing of fish oils.

FATTY ACID COMPOSITION OF FISH LIPIDS

- The major chemical entity in most lipid molecules like glycerides, phospholipids, wax esters is fatty acid. The nature of the fatty acids present in fish lipids is very complex. Fatty acids with carbon chain varying from 10 to 22 and unsaturation varying from 0-6 double bonds are of common occurrence. A great majority of the fatty acids, whether saturated or unsaturated, have an even number of carbon atoms in the molecules.
- Odd numbered acids are present, but quantitatively, they are very insignificant. Another important characteristic is that in the unsaturated acids which have more than one double bond (polyunsaturated), the double bonds are separated by a methylene group and have cis-configuration. The proportion of trans isomers is usually very negligible. High degree of unsaturation, with 5 or 6 double bonds per molecule is very common and abundant in fish, which is seldom observed in the lipids of other animals or plants of terrestrial origin. These features make the fatty acids of fish unique.

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- As in the case of lipid content, the fatty acid composition of fish lipids also shows wide variations. Proportion of individual fatty acids may vary from species to species. Even within the same species this composition may vary depending factors like feed intake, spawning migration etc. Commercially produced fish oils made from the same species of fish often vary quite widely in fatty acid composition and there is, at times, quite significant variation in fatty acid composition of the same species from year to year. The fatty acid profile of depot lipids is different from that of other tissue lipids. Depot lipids generally are richer in saturated acids when compared with lipids muscle tissue.
- The number of fatty acids present in the lipids of any species is quite high. About fifty different acids (including isomeric forms) have been identified in some species. However, a comparatively small number of acids account for about 85-90% of the total fatty acids. Myristic, palmitic and stearic acids are the important saturated acids present in fish from Indian waters. Among the monounsaturated group, palmitoleic and oleic acids are the important members and in the polyunsaturated group, arachidonic acid, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are the major components.

ENZYMES

- Enzymes are ubiquitous in living systems and are the agents that make chemical reactions possible in a diversity of life forms. Enzymes of fish and other marine organisms are comparatively less studied group of enzymes. Fish are poikilothermal animals and their enzymes are accordingly cold adapted.
- Autolysis or endogenous biochemical reactions catalyzed by enzymes are responsible for the loss of prime quality in seafood. Accordingly the development of methods to preserve the quality of fish must evolve from an understanding of seafood enzymes and aim to target key enzyme catalyzed reactions that contribute to loss of appearance, flavour, texture, nutrition and functional properties.

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- Numerous enzymes influence the physical properties of seafood. Among the most important enzymes that can influence the seafood texture are those involved with energy metabolism and the onset of rigor mortis, endogenous and exogenous proteolytic enzymes and trans glutaminases. Among all the enzymes, the nucleotide degrading enzymes are comparatively well studied as they directly affect the quality of seafood during post mortem. After death ATP levels are quickly depleted as the muscle enters rigor mortis. Enzymatic catabolism of ATP and related compounds, determines the ultimate eating quality of fish. In post mortem muscle, the conversion of ATP to ADP, ADP to AMP, AMP to IMP takes place within 24 h or less. These changes are thought to be totally enzymatic. The enzymes, involved in the post mortem degradation are ATPase, AMP deaminase, 5 nucleotidase, Inosine nucleosidase and xanthine oxidase.

- Another enzyme which is important in determining the quality of fish is myosin ATPase which favours protein denaturation. Inactivation of myosin ATPase in fish muscle to prevent or retard denaturation may be a method in the future to facilitate effective utilization of fish and shellfish.

PROTEIN

- Quantitatively, protein is the second major component in muscle tissues of fish and is generally present in the range of 16-18%. Protein content of fish is considered low if it is below 15%. The extent of variations in protein level is comparatively low. Feeding habits, spawning cycle etc. affect the level of protein in the tissues.
- The amount of protein in fish muscle is usually somewhere between 15 and 20 per cent, but values lower than 15 per cent or as high as 28 per cent are occasionally met with in some species.
- All proteins, including those from fish, are chains of chemical units linked together to make one long molecule. These units, of which there are about twenty types, are called amino acids, and certain of them are essential in the human diet for the maintenance of good health. Furthermore, if a diet is to be fully and economically utilized, amino acids must not only be present but must also occur in the correct proportions. Two essential amino acids called lysine and methionine are generally found in high concentrations in fish proteins, in contrast to cereal proteins for example. Thus fish and cereal protein can supplement each other in the diet. Fish protein provides a good combination of amino acids which is highly suited to man's nutritional requirements and compares favourably with that provided by meat, milk and eggs.
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THE MINOR COMPONENTS OF FISH MUSCLE

Carbohydrates:

- The amount of carbohydrate in white fish muscle is generally too small to be of any significance in the diet; hence no values are given in the tables. In white fish the amount is usually less than 1 per cent, but in the dark muscle of some fatty species it may occasionally be up to 2 per cent. Some molluscs, however, contain up to 5 per cent of the carbohydrate glycogen.

Minerals and Vitamins:

- These include a range of substances widely different in character that must be present in the diet, even if only in minute quantities, not only to promote good health but also to maintain life itself.
- Although fish is very unlikely to be the only source of an essential mineral in the diet, fish does provide a well balanced supply of minerals in a readily usable form. The table of mineral constituents of fish muscle gives values averaged from a large number of species and is intended to serve only as a rough guide. It would be impracticable in this short note, and of limited value, to give a detailed analysis for individual species.

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- Composition tables for fish often include a value for total ash. Since ash consists largely of a number of different minerals, and the total rarely exceeds 1-2 per cent of the edible portion, this figure has also been omitted, except from the table of fish products.
- 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 necessary for good health in humans and domestic animals are present to some extent in fish, but the amounts vary widely from species to species, and throughout the year.
- The vitamin content of individual fish of the same species, and even of different parts of the same fish, can also vary considerably. Often the parts of a fish not normally eaten, such as the liver and the gut, contain much greater quantities of oil-soluble vitamins than the flesh; the livers of cod and halibut for example contain almost all of the vitamins A and D present in those species. In contrast, the same two vitamins in eels, for example, are present mainly in the flesh.
- Water-soluble vitamins in fish, although present in the skin, the liver and gut, are more uniformly distributed, and the flesh usually contains more than half the total amount present in the fish. The roe, when present, is also a good source of these vitamins.

EXTRACTIVES

- These substances are so called because they can easily be extracted from fish flesh by water or water-based solutions. Unlike the proteins, substances in this group have comparatively small molecules; the most important extractives in fish include sugars, free amino acids, that is free in the sense that they are not bound in the protein structure, and nitrogenous bases, which are substances chemically related to ammonia. While many of these extractives contribute generally to the flavour of fish, some of them, known as volatiles, contribute directly to the flavours and odours characteristic of particular species; as the name suggests, volatiles are given off from the fish as vapours. Most of the extractives are present at very low concentrations but, because of their marked flavour or odour, are nonetheless important to the consumer. Detailed analyses of these substances have not been given because of the large variation existing both between and within species. An additional complication is the way in which the concentrations of these compounds change during storage and spoilage.
- When fish is stored after capture, the amount of some of the extractives present will change with time; thus measurement of the amount can often indicate the storage time and hence indirectly the quality. Extractive compounds whose concentration in fish varies directly with time of storage have long been studied since they may provide indicators of the quality of fish.

FACTORS AFFECTING THE COMPOSITION OF FISH

- The composition of a particular species often appears to vary from one fishing ground to another, and from season to season, but the basic causes of change in composition are usually variation in the amount and quality of food that the fish eats and the amount of movement it makes. For example, fish usually stop feeding before they spawn, and draw on their reserves of fat and protein. Again, when fish are overcrowded, there may not be enough food to go round; intake will be low and composition will change accordingly. Reduction in a basic food resource, plankton for example, can affect the whole food chain. An example of how abundance of food supply can markedly change the composition of a species is shown by the sheepshead, an American freshwater fish: when taken from certain small lakes that were overstocked, the sheepshead had an average fat content of 1 per cent, compared with 6-10 per cent for those taken from rivers or lakes where food was plentiful.

FISH FOR HUMAN CONSUMPTION

- Fish can form a very nutritious part of man's diet; it is rich in most of the vitamins he requires, it contains a good selection of minerals, and the proteins contain all the essential amino acids in the right proportions. Although the amount of protein in fish varies a little from species to species and, on occasions, within a species, the protein content for meat and for fish is roughly comparable.
- The extreme variability of composition of different species of fish accounts to some extent for the large variety of dishes than can be made from them; unfortunately fish are all too often lumped together in one category while pork, beef, lamb and mutton are invariably regarded as being quite distinct kinds of meat. In fact there is a much greater difference in composition, flavour and texture between, say, herring, haddock, halibut and salmon than there is between butcher meats, and this range is even wider when shellfish are included.

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Thank you