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Research Article

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Study on some Lipid Components in the muscle of some minor carps from "Dhubri- Town Markets"

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ABSTRACT

This quantitative study dealt with some lipid components related to human nutrition and health namely High Density Lipoprotein (HDL), Low Density Lipoprotein (LDL), Very Low Density Lipoprotein (VLDL), Triglyceride, important Omega-3-Fatty Acids namely α-Linolenic Acid (ALA), Eicosapentaenoic Acid (EPA), and Docosahexaenoic Acid (DHA) in the major consumable part the muscle in some minor carps found in the markets of Dhubri-Town; namely Mystus vittatus, Trichogaster fasciata, Glossogobius giuris, Nandus Meni and Macrognathus aral. Fishes were found to be rich source of High Density Lipoprotein and mentioned Omega-3-Fatty Acids. These were also found to be of lower contents of Low Density Lipoprotein, Very Low Density Lipoprotein and Triglyceride.

Key words: *Lipoprotein, Omega-3-Fatty Acid, minor carps.*

INTRODUCTION

Dhubri District is bounded both by inter-state and international border i.e. West Bengal and Bangladesh in the west, Goalpara and Bogaigoan district of Assam and Garo Hills district of Meghalaya in the east, Kokrajhar district in the north, Bangladesh and state of Meghalaya in the south. This district is located on the globe between 89.42 to 90.12 degree east longitude and 26.22 to 25.28 degree north latitude. The district is situated at 30 meters above the sea level on average. General topography of Dhubri district is plain with patches of small hillocks like Tokorabandha, Dudhnath, Chandardinga, Boukumari, Boropahar, Chakrasila etc. All these are situated in the north eastern part of the district. Mighty river Brahmaputra is flowing through this district from east to west with its tributaries like Champabati, Gourang, Gadadhar, Gangadhar, Tipkai, Sankosh,Silai, Jinjiram etc[1]. Most of tributaries are originates from the foot hills of Bhutan. The district is very rich in diversity of edible fishes for its wet-lands, The Brahmaputra and its tributaries. Right from Indian Major Carps to some Hill Stream Fishes and even some Brackish Water Fishes (seasonal attendants like *Tenualosa ilisha*) are available in the market till now.

Fishes are rich source of easily digestible fats and lipids including Cholesterols like High Density Lipoprotein (HDL), Low Density Lipoprotein (LDL), Very Low Density Lipoprotein (VLDL); Triglyceride, important Omega-3-Fatty Acids like α-Linolenic Acid (ALA), Eicosapentaenoic Acid (EPA), and Docosahexaenoic Acid (DHA) etc. in their muscles, brain and liver and are the main source of such nutrients for non-vegetarian people.

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Cholesterol, from the Ancient Greek *chole-* (bile) and *stereos* (solid) followed by the chemical suffix -*ol* for an alcohol, is an organic molecule. It is a sterol (or modified steroid),[2] a lipid molecule and is biosynthesized by all animal cells because it is an essential structural component of animal cell membranes that is required to maintain both membrane structural integrity and fluidity. Cholesterol enables animal cells to **(a)** not need a cell wall (like plants & bacteria) to protect membrane integrity/cell-viability and thus be able to **(b)** change shape and **(c)** move about (unlike bacteria and plant cells which are restricted by their cell walls).

In addition to its importance within cells, cholesterol also serves as a precursor for the biosynthesis of steroid hormones, bile acids, and vitamin D[3]. Cholesterol is the principal sterol synthesized by animals. All kinds of cells in animals can produce it. In vertebrates the hepatic cells typically produce greater amounts than other cells. It is almost completely absent among prokaryotes (bacteria and archaea), although there are some exceptions such as Mycoplasma, which require cholesterol for growth[4].

François Poulletier de la Salle first identified cholesterol in solid form in gallstones in 1769. However, it was not until 1815 that chemist Michel Eugène Chevreul named the compound "cholesterine"[5, 6].

High-density lipoprotein (HDL) is one of the five major groups of lipoproteins. Lipoproteins are complex particles composed of multiple proteins which transport all fat molecules (lipids) around the body within the water outside cells. They are typically composed of 80-100 proteins/particle (organized by one, two or three Apo-A; more as the particles enlarge picking up and carrying more fat molecules) and transporting none to hundreds fat molecules/particle. Unlike the larger lipoprotein particles which deliver fat molecules to cells, HDL particles remove fat molecules from cells which want to export fat molecules. The fats carried include cholesterol, phospholipids, and triglycerides; amounts of each quite variable.

Lipoproteins, in order of molecular size, largest to smallest, are chylomicrons, very low-density lipoprotein (VLDL), intermediate-density lipoprotein (IDL), low-density lipoprotein (LDL), and HDL. Lipoprotein molecules (all particles far smaller than human cells), enable the transportation of all lipids, such as cholesterol, phospholipids, and triglycerides, within the water around cells (extracellular fluid), including the bloodstream. HDL particles, unlike the larger particles, transfer fats away from cells, artery walls and tissues (around the body, body wide) through the bloodstream, back to both (a) LDL particles and (b) back to the liver for other disposition[7].

Increasing concentrations of HDL particles are strongly associated with decreasing accumulation of atherosclerosis within the walls of arteries[8] over weeks, years, decades. This is important because atherosclerosis, eventually, results in sudden plaque ruptures triggering clots within the artery opening, narrowing/closing the opening (s), i.e. cardiovascular disease, stroke (s) and other vascular disease complications body wide.

HDL particles (though vastly different from just cholesterol and other fat molecules *per-se*) are sometimes referred to as good cholesterol because they can transport fat molecules (including cholesterol, triglycerides, etc.) out of artery walls, reduce macrophage accumulation, and thus help prevent, even regress atherosclerosis over weeks, years, decades, thereby helping prevent cardiovascular disease, stroke (s) and other vascular disease complications body wide. In contrast, LDL particles (also far different from cholesterol *per-se*) are often called bad cholesterol or unhealthy cholesterol, because they deliver fat molecules to macrophages in the wall of arteries[9].

Omega-3 fatty acids - also called ω-3 fatty acids or *n*-3 fatty acids[10] - are polyunsaturated fatty acids (PUFAs) with a double bond (C=C) at the third carbon atom from the end of the carbon chain.[11] The fatty acids have two ends, the carboxylic acid (-COOH) end, which is considered the beginning of the chain, thus "alpha", and the methyl (CH_3) end, which is considered the "tail" of the chain, thus "omega." The way in which a fatty acid is named is determined by the location of the first double bond, counted from the methyl end, that is, the omega (ω-) or the n- end.

The three types of omega-3 fatty acids involved in human physiology are α-linolenic acid (ALA) (found in plant oils), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA) (both commonly found in marine oils). Marine algae and phytoplankton are primary sources of omega-3 fatty acids. Common sources of plant oils containing the omega-3 ALA fatty acid include walnut, edible seeds, clary sage seed

oil, algal oil, flaxseed oil, Sacha Inchi (*Plukenetia volubilis*) oil, *Echium* oil, and hemp oil, while sources of animal omega-3 EPA and DHA fatty acids include fish oils, egg oil, squid oils, and krill oil. Dietary supplementation with omega-3 fatty acids does not appear to affect the risk of death, cancer or heart disease[12,13]. Furthermore, fish oil supplement studies have failed to support claims of preventing heart attacks or strokes[14, 15, 16].

Omega-3 fatty acids are important for normal metabolism[8]. Mammals are unable to synthesize omega-3 fatty acids, but can obtain the shorter-chain omega-3 fatty acid ALA (18 carbons and 3 double bonds) through diet and use it to form the more important long-chain omega-3 fatty acids, EPA (20 carbons and 5 double bonds) and then from EPA, the most crucial, DHA (22 carbons and 6 double bonds)[17]. The ability to make the longer-chain omega-3 fatty acids from ALA may be impaired in aging.[18,19] In foods exposed to air, unsaturated fatty acids are vulnerable to oxidation and rancidity[20].

In this study the major lipid components related to human health and nutrition are quantitatively observed in the major consumable part of some commercially less important fishes (minor carps) namely *1) Mystus vittatus* (Order: Siluriformes family : Bagridae) locally known as "Tengra", 2) *Trichogaster fasciata* (Order: Perciformes Family: Osphronemidae) locally known as "kholisa", 3) *Glossogobius giuris*, (Order: Perciformes Family: Gobiidae) locally known as "Balia", 4) *Nandus Meni* (Order: Perciformes Family: Nandidae) locally known as "Meni", and 5) *Macrognathus aral* (Order: Synbranchiformes Family: Mastacembelidae) locally known as "Gota"; procuring from the two main fish markets of Dhubri Town the "New Market" and "Bou Bazar". 12-24 hours Ice-Preserves fishes are used for the study and the study was done in the months of March to June.

MATERIALS AND METHODS

Measured amount of fish-muscle are homogenized in fixed amount of methanol/chloroform (1:1) for 15 min using vortex mixing. Extracts were then centrifuged at 15000 xg to remove solids, and 1.0 mL of extract was added to 4 ml of isopropanol / water (3:2) and 1 ml of 5 M KOH and kept in separate labeled vials in deep freeze for assays. Total Cholesterol content in muscle homogenate is estimated by CHOD-PAP, End Point Assay Kit[21, 22]. Triglyceride content in body fluid is estimated by GPO-PAP, End Point Assay Kit[21, 22]. HDL Cholesterol content in body fluid is estimated by PEG- CHOD-PAP, End Point Assay with Lipid Cleaning Factor (LCF) Kit[21,22]. Low Density Lipoproteins (LDL), Very Low Density Lipoproteins (VLDL), Chylomicron Fats are precipitated by addition of Propylene Glycol 6000 (PEG). After centrifugation, High Density Lipoproteins (HDL) fraction remains in supernatant and is determined with CHOD-PAP method [21,22] (by the kit for Total Cholesterol). From the results of Total Cholesterol, Triglycerides and HDL fractionation of LDL Cholesterol and VLDL Cholesterol is done by using "Friedewald's Equation"[23]. The reagent kits for Total Cholesterol, Triglycerides and HDL assays were procured from Span Diagnostics (India) Ltd. All the photometric observations and biochemical assays were done in a semi automated biochemistry analyzer ("Lab Life Chem-Master" manufactured by Ranbaxy- Diagnova LTD). Necessary kit specifications and dilution factors were preprogrammed in the machine.

Important Omega-3-Fatty Acids related to human health and nutrition namely α-Linolenic Acid (ALA), Eicosapentaenoic Acid (EPA), and Docosahexaenoic Acid (DHA) are estimated fish-muscle-extract in reversed-phase HPLC workstation associated with mass sensitive charged aerosol detection system. [24, 25] The chromatographic devices and conditions in a gist are HPLC System: PerkinElmer-Flexar, Mobile Phase A: Water/formic acid/mobile phase B (900:3.6:100), Mobile Phase B: Acetone/acetonitrile/tetrahydrofuran/formic acid (675:225:100:4), Column: Acclaim™ C30, 250 × 3 mm, 3 μm (Dinorex), Column Temp.: 30 °C, Flow Rate of Eluent Gradient Pump: 1 mL/min, Flow Rate of Inverse Gradient Pump: 1 mL/min, Injection Volume: 2.00 μL, Detector: Corona ultra RS charged aerosol detector (Dinorex),, Corona Filter: High, Corona Nebulizer Temp.: 15 °C, Total Run Time: 30.1 min. α-Linolenic Acid (ALA), Eicosapentaenoic Acid (EPA), and Docosahexaenoic Acid (DHA) standards were procured from Sigma Aldrich (USA). The other regents and chemicals were procured from Ranbaxy-Ranchem LTD and E. Mark.

RESULTS

Results obtained so far were analyzed statistically[26] with the help of Microsoft Excel and presented in the following tables:

Table 1: - Shows amounts of different lipoproteins in muscles of different fishes (in µg/100mg of muscle)

Name of fishes ⟶	***Mystus vittatus***	***Trichogaster fasciata***	***Glossogobius giuris***	***Nandus Meni***	***Macrognathus aral***
Amount of Total Cholesterol (µg/100mg of muscle) ⟶	93.69 ±0.069642	82.472 ±0.173332 -11.9735% *	127.766 ±0.15387 36.37101% *	76.688 ±0.144582 -18.1471% *	106.664 ±1.447593 +13.8478% *
Amount of HDL Cholesterol (µg/100mg of muscle) ⟶	74.81 ±0.091269	63.788 ±4.042954 -14.7333% *	91.264 ±0.202919 +21.99439% *	62.514 ±0.207957 -16.4363% *	87.358 ±0.128117 +16.77316% *
Amount of LDL Cholesterol (µg/100mg of muscle) ⟶	15.3648 ±0.110805	11.7904 ±0.225436 -23.2636% *	32.1992 ±0.28923 +109.5647% *	11.6476 ±0.253976 -24.193% *	15.3764 ±1.467993 +0.075497% *
Amount of VLDL Cholesterol (µg/100mg of muscle) ⟶	3.5152 ±0.016848	2.8936 ±0.013422 -17.6832% *	4.3028 ±0.010911 +22.40555% *	2.5264 ±0.018999 -28.1293% *	3.9296 ±0.01256 +11.7888% *
Amount of Triglyceride (µg/100mg of muscle) ⟶	17.576 ±0.084238	14.468 ±0.067112 -17.6832% *	21.514 ±0.054553 +22.40555% *	12.632 ±0.094995 -28.1293% *	19.648 ±0.062801 +11.7888% *
"*" indicates Significant at p<0.05 and "-…%" indicate percent deviations.					

Table 2: - Shows percentage of High Density Lipoproteins (HDL) in Total Cholesterol

Name of fishes ⟶	***Mystus vittatus***	***Trichogaster fasciata***	***Glossogobius giuris***	***Nandus Meni***	***Macrognathus aral***
% of HDL in Total Cholesterol ⟶	79.85%	77.35%	71.43%	81.52%	81.90%

Table 3: - Shows amounts of important omega-3-fatty acids in muscles of different fishes (in µg/100mg of muscle)

Name of fishes ⟶	***Mystus vittatus***	***Trichogaster fasciata***	***Glossogobius giuris***	***Nandus Meni***	***Macrognathus aral***
Amount of α-Linolenic Acid (ALA) (µg/100mg of muscle) ⟶	345.894 ±1.305027	469.256 ±11.30982 +35.66468% *	373.986 ±2.111347 +8.121563% *	587.186 ±1.994664 +69.75894% *	304.056 ±2.326297 -12.0956% *
Amount of Eicosapentaenoic Acid (EPA) (µg/100mg of muscle) ⟶	643.276 ±9.750764	488.33 ±2.084665 -24.087% *	747.748 ±10.19043 +16.24062% *	559.26 ±9.417491 -13.0606% *	853.272 ±11.79223 +32.64477% *
Amount of Docosahexaenoic Acid (DHA) (µg/100mg of muscle) ⟶	446.094 ±10.01057	758.294 ±2.094814 +69.98525% *	560.162 ±7.98871 +25.5704% *	376.392 ±8.75014 -15.625% *	653.982 ±9.549912 +46.60184% *
"*" indicates Significant at $p<0.05$ and "-…%" indicate percent deviations.					

DISCUSSION

Mystus vittatus contained 93.69±0.069642 µg/100mg of (Total) Cholesterol in their muscle. *Trichogaster fasciata* contained 11.9735% less, *Glossogobius giuris* contained 36.37101% more, *Nandus Meni* contained 18.1471% less and *Macrognathus aral* contained 13.8478% more (Total) Cholesterol in their muscle than *Mystus vittatus*.

In *Mystus vittatus* content of HDL Cholesterol was 74.81±0.091269 µg/100mg in their muscle. *Trichogaster fasciata* contained 14.7333%less, *Glossogobius giuris* contained 21.99439% more, *Nandus Meni* contained16.4363%less and *Macrognathus aral* contained 16.77316% more of HDL Cholesterol in their muscle than *Mystus vittatus.*

Mystus vittatus contained15.3648±0.110805 µg /100mg Amount of LDL Cholesterol in their muscle. *Trichogaster fasciata* contained 23.2636%less, *Glossogobius giuris* contained 109.5647% more, *Nandus Meni* contained 24.193% less and *Macrognathus aral* contained 0.075497% more of LDL Cholesterol in their muscle than *Mystus vittatus.*

In *Mystus vittatus* content of LDL Cholesterol was 15.3648±0.110805 µg/100mg in their muscle. *Trichogaster fasciata* contained 23.2636%less, *Glossogobius giuris* contained109.5647% more, *Nandus Meni* contained 24.193% less and *Macrognathus aral* contained 0.075497% more of LDL Cholesterol in their muscle than *Mystus vittatus.*

Mystus vittatus contained17.576±0.084238 µg /100mg Amount of Triglyceride in their muscle. *Trichogaster fasciata* contained 17.6832% less, *Glossogobius giuris* contained 22.40555% more, *Nandus Meni* contained 28.1293%less and *Macrognathus aral* contained 11.7888% more of Triglyceride in their muscle than *Mystus vittatus*.

High Density Lipoproteins (HDL) in Total Cholesterol of *Mystus vittatus* was 79.85% in *Trichogaster fasciata* 77.35%, in *Glossogobius giuris* 71.43% in *Nandus Meni* 81.52% and in *Macrognathus aral* 81.90%.

α-Linolenic Acid (ALA) content of *Mystus vittatus* muscle was 345.894±1.305027 µg /100mg. *Trichogaster fasciata* contained 35.66468% more, *Glossogobius giuris* contained 8.121563% more,

Nandus Meni contained 69.75894% more and *Macrognathus aral* contained 12.0956% less of α-Linolenic Acid (ALA) in their muscle than *Mystus vittatus.*

Eicosapentaenoic Acid (EPA) content of *Mystus vittatus* muscle was 643.276±9.750764 µg/100mg. *Trichogaster fasciata* contained 24.087% less, *Glossogobius giuris* contained 16.24062% more, *Nandus Meni* contained 13.0606% less and *Macrognathus aral* contained 32.64477% more of α-Linolenic Acid (ALA) in their muscle than *Mystus vittatus.*

Docosahexaenoic Acid (DHA) content of *Mystus vittatus* muscle was 446.094±10.01057 µg/100mg. *Trichogaster fasciata* contained 69.98525% more, *Glossogobius giuris* contained 25.5704% more, *Nandus Meni* contained 15.625% less and *Macrognathus aral* contained 46.60184% more of α-Linolenic Acid (ALA) in their muscle than *Mystus vittatus*.

CONCLUSION

From this study it had been found that, though these fishes are less preferred for regular diet list or though they are commercially less important, yet they should be treated as important ingredient of healthy diet. As they provide high amounts of important omega-3-fatty acids and more HDL in comparison to LDL, VLDL and Triglyceride they may lead to wellbeing of lipid profile of human body, good for cardiac and hepatic functions etc. The problem of threat to the diversity of these species are: fishing in breeding season, mainly using dense mesh net (that used for making mosquito-nets) and habitat destruction including deforestation, mainly in foot hill areas which results in soil erosion and raising of river beds; dumping on wet-lands etc. So, for the good diversity and availability these species eco-restoration processes including public awareness programmes are wanting.

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