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First Report on Comparative Evaluation of Fat (Total Cholesterol and Fatty Acids) Profile in Farm Cultivated and River Water Fishes Community of *Labeo rohita*

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ABSTRACT

The present investigation deals with the study of, fat, cholesterol and major fatty acid content in river and farm cultivated *Labeo rohita* (Rohu). Triplicate samples of Rohu were obtained from the river flowing at Doiwala region of Dehradun (U.K) and from the cultivated farm. Fatty acids were determined by conventional biochemical methods, TLC and GC-MS. Fat content of farm cultivated Rohu ranged from 2.5-5 times that of river water samples. Monounsaturated fatty acids predominated in cultivated farm Rohu in comparison to Rohu in river water. Differences in fatty acid profile between river water and farm cultivated Rohu were greatest among Polyunsaturated Fatty Acids (PUFA), linoleic acid was substantially higher in all cultivated samples than in river water fish. Omega-3 fatty acid levels were also found to be highest in cultivated farm water fish in comparison to river water fish. The study thus indicates that, as per basis of nutritional screening in terms of omega-3 fatty acids, cultivated farm water Rohu are more appropriate for diet.

Key words: Omega fatty acids, Nutritional requirements, Farm cultivated, River water, Rohu

INTRODUCTION

Fish meat possesses high nutritional quality and is therefore a particularly recommended human dietary component. Information concerning the chemical and fatty acid composition of freshwater fishes is valuable to nutritionists who are interested in finding sources of low-fat, high protein foods, with desirable fatty acid compositions and acceptable amount of total cholesterol. Fish meat contains biologically active protein which is characterized by a very favourable composition of amino acids, a high omega-3 polyunsaturated fatty acid content such as Eicosapentaenoic acid (20:5 n-3, EPA) and Docosahexaenoic (22:6 n-3, DHA) and fat-soluble vitamins as well as it represents a good source of micro- and macro-elements. The shortage of α -Linolenic acid (18:3 n-3, ALA) is responsible for neurological disorders and poor growth [1]. DHA and EPA have been shown to have a positive effect in preventing hypertension and cardiovascular diseases [2]. Essential-polyunsaturated fatty acids such as ALA, EPA and DHA are not synthesized in the human body and effectively synthesized only by aquatic organisms; therefore, humans can receive these essential fatty acids by marine and freshwater fishes. The present study is in need to determine the important nutritional components present in fish, *Labeo rohita* (Rohu) as most of the population of India residing near the banks of the rivers and specific communities utilizes fish as their regular diet [3-6]. Recent studies were performed on *L. rohita* (Rohu) after exposure to phenolic compounds [7]. The study was performed to focus on the nutritional and pharmacological components of fish which makes the fish as an important nutritional and dietary supplement. The study was performed to describe the most important n-3 Poly Unsaturated Fatty Acids (PUFA) acids present in farm cultivated and river water *L. rohita* (Rohu) fishes communities.

MATERIALS AND METHODS

Collection of samples

Sampling of farm cultivated and river water Rohu fish species was performed for the comparison of fatty acid composition from Doiwala region of Dehradun (U.K).

Lipid extraction from meat

From fish muscle samples the total lipids were extracted [8]. Briefly mixture of chloroform-methanol solvent (2:1 v/v) was added to frozen samples in the ratio of 20:1 v/w of solvent-tissue. With solvent mixture the samples were homogenized three times for 10 min at 3000-4000 rpm.

After each homogenization step samples were cooled at 4°C for 1 h. For 1 g of tissue 4 ml of 0.034% of MgCl₂ was added. The extracts were incubated overnight at 4°C for complete partition of aqueous layer and organic (containing the extract of total lipids). The top aqueous layer was separated, and the bottom organic layer was dissolved with chloroform-methanol (2:1 v/v) and placed into a glass tube. The total lipid fraction was obtained by evaporating the lower phase. The total lipids from the feed samples were extracted by soxhlet for 4 h at condensation rate of 5-6 drops/s, dry extract for 30 min at 100°C cool. Total lipid contents were determined gravimetrically and stored at 4°C until analysis.

Preparation of fatty acid methyl esters and gas chromatography analysis

Preparation of Fatty Acid Methyl Esters (FAME)

FAME was prepared by standard IUPAC method which involved the esterification of FA by methanol. Briefly, 100 mg of fish lipid was taken in a 50 ml round bottom flask; 20 ml of chromatographic grade methanol was added followed by the addition of 0.5 ml of 1 N methanolic potassium hydroxide. The contents of the flask were refluxed for 30 min at 70°C until the droplets of fats were disappeared. On cooling, the reaction mixture was gently transferred to a separating funnel; 10 ml of n-hexane was added. Separating funnel was shaken gently. The upper hexane layer was recovered and mixed with distilled water. This hexane layer will be dried over anhydrous sodium sulfate and placed in sealed GC vials, kept at -20°C until GC analysis.

Fatty acid analysis

The FAMES were analyzed on a Perkin Elmer gas chromatograph model 8700 (Perkin-Elmer Ltd.) fitted with non-bonded biscynopropyl siloxane stationary-phase, polar capillary column Rt-2560 (100 × 0.25 mm) 0.2 µm film thickness (Supelco, Inc., Bellefonte, PA, USA) on FID. Oxygen-free nitrogen will be used as a carrier gas at a flow rate of 3.5 ml/min. Gas chromatographic conditions were as follows. The initial oven temperature was 150°C at rate of 4 min which was raised to 190°C at a rate of 2°C/min and further to 220°C held for 7 min. The injector and detector temperature were set at 260°C and 270°C respectively. A sample volume of 1.0 µl was injected. All of the quantification was done by a built-in data-handling program provided by the manufacturer of the gas chromatograph (PerkinElmer).

RESULTS AND DISCUSSION

The total lipids, cholesterol content, fatty acids profiles include minor amounts of odd-number, branched-chain, and even-number fatty acids as well as saturated components, the Mono Unsaturated Fatty Acids (MUFA) and Poly Unsaturated Fatty Acids (PUFA) were determined in farm cultivated and river water fish's community of *L. rohita*. The major saturated fatty acids (SFA) were C14:0 and C16:0. The C18:1 was the prominent MUFA predominant in farm cultivated fishes. The dominant PUFA are of the omega-6 series and are found chiefly in C18:2 fatty acids in farm cultivated fishes. The essential fatty acids compositions showed prominence in C18:3n-3 and C18:2n-6 in farm cultivated fishes. The branched chain fatty acids identified C15:0, C16:0, C17:0, C18:2 and C20:0 in farm cultivated fishes. The results are shown in Figures 1-3 and Table 1.



Figure 1: Preparation of fatty acid methyl esters of (a) farm cultivated and (b) river water communities of *Labeo rohita*

Table 1: Percent content of fat related parameters in farm cultivated and river water communities of *Labeo rohita*

S. No.	Fat content related parameters	Labeo rohita community	
		Farm cultivated	River water
1	Total lipid content (%)	2.56	1.89
2	Total cholesterol (%)	18	12
3	Fatty acids (saturated) (%)		
	C14:0 (Myristic acid)	56	34
	C16:0 (Palmitic acid)	58	36
4	C17:0 (Stearic acid)	54	45
	Fatty acids (Mono-unsaturated) (%)		
5	C18:1 (Oleic acid)	45	34
	Fatty acids (Poly-unsaturated) (%)		
	Omega -6 fatty acids	47	33
	C15:0	26	13
	C16:0	27	22
	C17:0	28	12
	C18:2	38	23
	C20:0	48	34

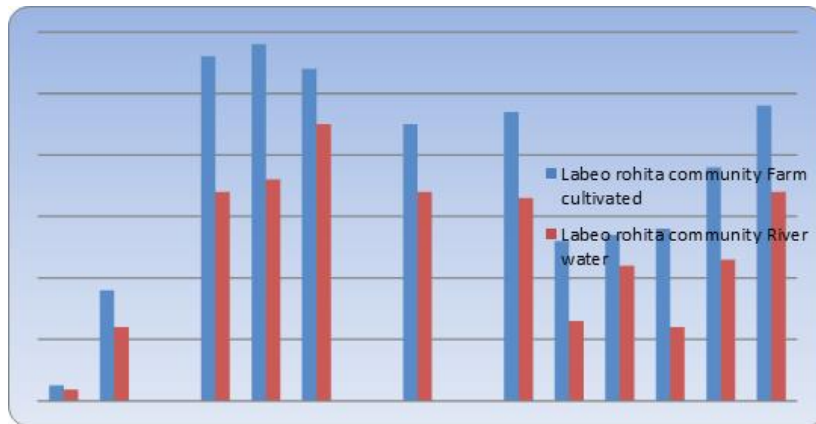


Figure 2: Percent content comparison of fat related parameters in farm cultivated and river water communities of *Labeo rohita*

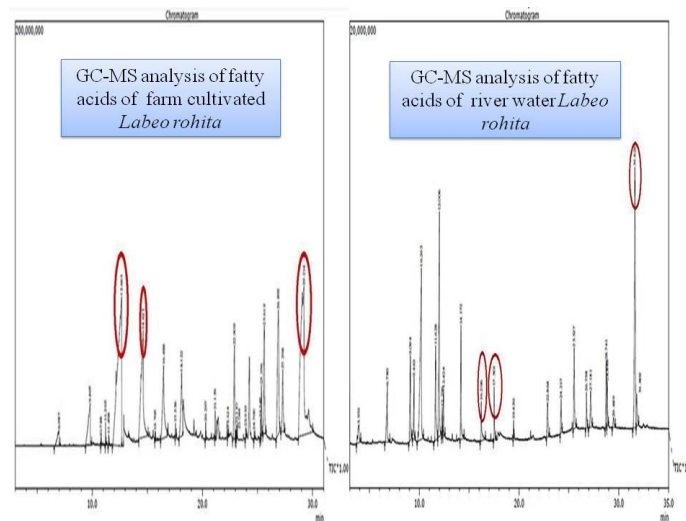


Figure 3: Peaks (fatty acids) variation as determined via GC-MS chromatogram of fatty acids extracted from farm cultivated and river water communities of *Labeo rohita*

CONCLUSION

Cultivated Rohu fish exceeded their wild counterparts in total fat, by as much as five times. Differences were found in total fat and most fatty acids between cultivated and river water fishes. Ratios of fatty acid classes in individual foods may not be an appropriate reflection of the nutritional contribution of various fatty acids. The present study thus suggests the importance of cultivation and rearing of edible fishes as is justified by the results. Proper feeding and nutrition enrichment leads to the good quality fishes which are safe as per nutrition point of view.

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