FISH –BASED DIET- CORRELATIONS BETWEEN METABOLISM OF CARBOHYDRATES, LIPIDIS AND PROTEINS. STUDY CASE POPULATION OF THE SULINA TOWN, DANUBE DELTA

Georgiana ENE[1], Cristian Lucian PETCU[2], Magda-Ioana NENCIU[3], Natalia ROȘOIU[4,5]

[1]Ovidius University, Doctoral School of Applied Sciences, Constanta, Romania, 124 Mamaia, Blvd., RO-900527, Tel: (+40)241.606.408, (+40)763.357.610, *E-mail: enegeorgiana.biologie@yahoo.com*

[2]Ovidius University, Faculty of Dental Medicine, Department of Biophysics, Constanta, Romania, 1 University Alley, Building B, Campus, RO-900145, Tel: (+40)241.545.697, (+40)726.251.948, *E-mail: crilucpet@yahoo.com*

[3]National Institute for Marine Research and Development Grigore Antipa, 300 Mamaia Blvd., RO-900581, Constanta, Romania, Tel: (+40)766.734.030, *E-mail: magdalena.nenciu@gmail.com*

[4]Ovidius University, Faculty of Medicine, Department of Biochemistry, Constanta, Romania, 1 University Alley, Building B, Campus, RO-90014, Tel: (+40)241.545.697, (+40)722.737.516, *E-mail: natalia_rosoiu@yahoo.com*

[5]Full Member of the Academy of Romanian Scientists, 54 Splaiul Independentei 050094, Bucharest, Romania, (+40)722.737.516

Abstract

According to literature data, the normal values of biochemical parameters in blood vary by sex, age, geographical region, and type of diet. The aim of this study was to analyze the benefits of a fish-based diet among the population of Sulina, in the Danube Delta (3,663 individuals), by performing a comparative hepatic evaluation, lipid profile, serum glucose levels and total protein profile of these patients. Fish is an important source of protein with high biological value, containing all essential aminoacids and low lipid levels. The novelty of the research is represented by the analyzed geographical area. The Danube Delta had no medical analysis laboratory until 2010, when the RoutineMed Sulina laboratory was opened. Patients had a set of biochemical tests in the RoutineMed Sulina laboratory and declared they eat fish or fish-based products at least once a week. Tests were performed on 200 patients for the evaluation of the liver of these patients: Aspartate Amino Transferase, alanine amino transferase, de Ritis ratio, High Density Lipoprotein, Low Density Lipoprotein, total lipids, total cholesterol, triglycerides and 200 tests for the evaluation of the serum glucose levels and total protein. Both women and men were involved in the research and patients were grouped into age ranges: 20-40 years, 40-60 years, > 60 years. The values

obtained were statistically analyzed using the SPSS v. 20 software and then compared to the ranges considered normal for these parameters. The results obtained showed that patients with a fish-based diet seem to be healthier than those with a diet in which fish meat is scarce, as their blood biochemical parameters values are closer to normal, which leads to the conclusion that including fish and fish products in people's regular diet is beneficial in preventing lipid, protein and carbohydrate metabolism disorders and preserving the overall health of the body.

1 INTRODUCTION

Sulina town is surrounded by the waters of the Danube Delta and the Black Sea. Through its position and climate, Sulina is located in a specific natural environment. It is the eastern most point of Romania. (Bondar et al., 2010). The rivers ecosystem is dominated by fish (about 160 species, of which 75 are freshwater) forming local and migratory fish fauna. The stagnant water ecosystem is made up of lakes, streams, and former branches whose average water volume is about 2.6 km^3 (Bondar et al., 2010). Fishing is one of the oldest occupations of the Danube Delta population and still represents an important economic activity engaging more than half of the active Delta population (http://www.ddbra.ro/). Thus, fish represents a major source of food for local inhabitants. According to the 2011 census, 82.82% of the population were romanians, 11.43% lipovans, 1.8% greeks, 1.29% ukrainians and 2.3% of other or undeclared ethnicity. Industrial activities decreased due to the closure of canned fish factory and shipyard restriction. Tourism activity is growing among residents of Sulina. Sulina as a free port has a multitude of recipes in terms of cooking fish such as those related to traditions and customs and those relating to the daily diet and the celebration. Although each ethnic group has its own recipes fish dishes are similar (Bondar et al., 2010).

Based on literature data, normal values of biochemical parameters vary by gender, age, diet and geographical region, and there are indications that a fish-based diet really helps people maintain normal levels of lipids and other analytes at the biochemical level (Cleeman, 2001). The nutrition and endocrine diseases or the screening of healthy people are made through sets of laboratory analyses such as: glucose, triglycerides, lipoproteins, ionogram, total calcium, cholesterol, transaminases, proteins, phosphorus, magnesium, hemoglobin, iron (Roșoiu, 2010). The liver is the most

important seat of degradation and synthesis of lipids metabolism but the main role of liver is in glucose metabolism (Roşoiu, 2008).

2 STUDY AREA

From the geographical point of view, Sulina is located between N 45^0 08' 33''- N 45^0 09' 45'' parallels and E 29^0 36' 27''- E 29^0 46' 21''meridians (Figure 1).

The natural resources of Sulina, Danube Delta and the Black Sea that people benefit from are: soil resources, fishery resources, plant resources, forestry, landscape resources, renewable natural energy resources (wind, solar, wave Black Sea), marine hydrocarbon resources.
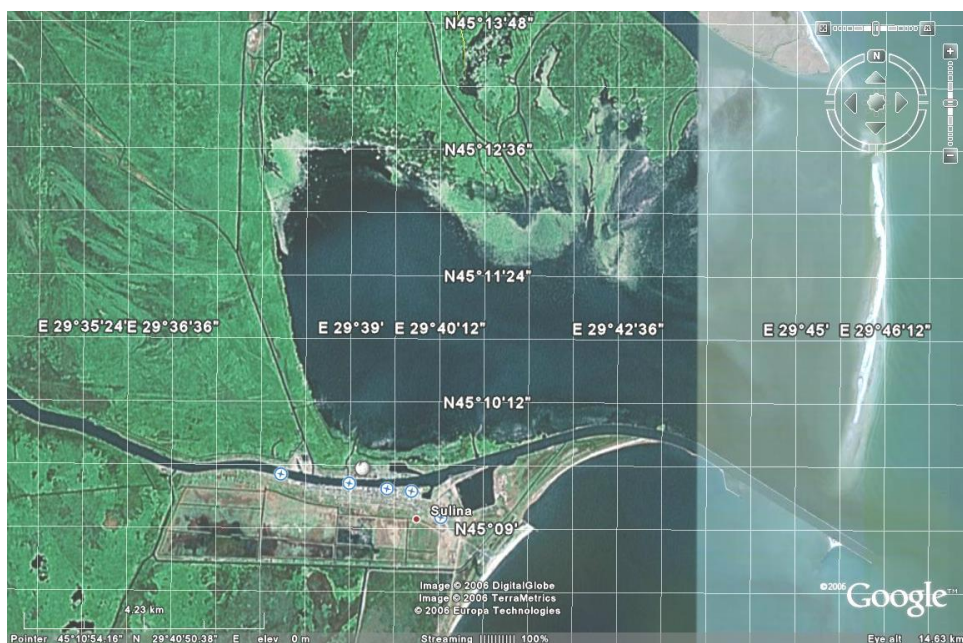

Figure 1. Location of the studied area ,Sulina, 2006 (after Bondar et al., 2010).

3 OBJECTIVES

Although average productions annual catches of fish species have declined in recent decades, the fish remained the staple food of the population of Sulina, Danube Delta (Table 1).

Although studies on fish consumption have been conducted in Romania, Sulina, Danube Delta, has not yet been researched and the main

objective of this research was to determine whether the fish-based diet really help people maintain normal levels of lipids and other analytes at the biochemical level.

The consumption of fish and fish products in Romania is estimated at 6.3 kg/inhabitant/year (Eurostat, 2016). However, there are indications that the consumption is increasing, mainly due to the health benefits associated with the consumption of fish.

Table 1. Average annual productions (tons) per year of fish species caught in the Danube Delta (after Bondar et al., 2010)

Category	1895-1930	1931-1964	1965-1970	1971-1984	1985-1998	1999-2004
Carp	1305	915.6	508.2	244.6	63.1	142
Bream	90.4	178.8	348	857.1	1123	830
Crucian carp	313.2	374.1	665.8	101	0.07	-
Tench	3.33	58.6	620.7	127.1	8.5	-
Carrassius	14.6	176.2	10.7	2709	1886	1020
Rudd	1784	1649	3789	1324	1057	-
Phytophagus	30.6	21.5	0.17	14.7	72.3	-
Pike	304.6	761.9	1858	508.3	49.9	63
Catfish	55.4	186.8	1066	411.4	80.5	96
Roach	1327	1232	696.3	349.4	232.5	248
Zander	465.3	638.7	615.8	494.4	205.5	86
Sturgeon	467.9	257.4	199.2	106.5	17.8	19.3
Pontic shad	349.1	407.6	799.2	1290	534.9	180
Others	543.6	-	-	66.5	10.9	53.3
Total	7054	6858	11177	8604	5342	2738

4 METHODS

Patients were subjected to a set of biochemical tests in the RoutineMed Sulina laboratory. They all declared they eat fish or fish-based products at least once a week. Tests were performed on 200 patients for evaluation of the liver: AST, ALT, AST/ALT, total lipids, total cholesterol, HDL cholesterol, LDL cholesterol, LDL/HDL, total cholesterol/HDL, triglycerides and 200 tests for the evaluation of the serum glucose levels and total protein. Both women and men were involved in the research and patients were grouped into age ranges: 20-40 years, 40-60 years, > 60 years. Blood was collected and analyzed during September 2013 - January 2016.

For serum biochemistry the following measuring instruments were used: Rotofix 32 A centrifuges, SAPPHIRE 350 automatic biochemistry unit and Audit Diagnostics reagents.

5 RESULTS

The study included 200 patients, 97 males (48.5%) and 103 females (51.5%) (Figure 2A); the distribution by age groups is: 53 are in the age range (20-40) years (26.5%), 79 are in the age range (40-60) years (39.5%) and 68 are in the age range (60 -...) years (34.0%) (Figure 2B). The mean of **total lipids** in the two groups of patients with a reference value $L_{sup} = 800$ mg/dL indicates that the results for both women and men are within normal limits (Figure 2C). When the means of **total cholesterol** in the two groups of patients are compared with a reference value $L_{sup}=200mg/dL$: for the group of male patients the mean total cholesterol - $M_M = 179.89$ mg/dL and for the group of female patients the mean total cholesterol - $M_F = 190.01$ mg /dL (Figure 2D). Comparing the mean **HDL cholesterol** in both groups of patients with a reference value $L_{sup} = 100$ mg/dL: for the group of male patients the mean HDL cholesterol - $M_M = 49.54$ mg/dL; for the group of female patients the mean HDL cholesterol - $M_F = 54.82$ mg / dL (Figure 2E). Comparing **LDL Cholesterol** mean values of the two groups of patients with a reference value $L_{sup} = 180$ mg / dL: for the group of male patients the mean LDL cholesterol - $M_M = 110.81$ mg/dL; for the group of female patients the mean LDL cholesterol - $M_F = 123.07$ mg / dL (Figure 2F). When the means of **triglycerides** are compared in the two groups of patients with a reference value $L_{sup} = 200$ mg/dL: for the group of male patients average amount of triglycerides - $M_M = 123.65$ mg/dL; for the group of female patients average amount of triglycerides - $M_F = 100.40$ mg/dL (Figure 3A). **LDL cholesterol/HDL-cholesterol** has a value of 2.9 in women and 3.3 in men: for male patients, the mean LDL cholesterol/HDL cholesterol is $M_M = 2.27$; for female patients, the mean LDL cholesterol/HDL cholesterol is $M_F = 2.29$ (Figure 3B). **Total cholesterol/HDL cholesterol**: for male patients, the mean total cholesterol / HDL cholesterol is $M_M = 3.70$; for female patients (N = 103), the mean total cholesterol/HDL cholesterol is $M_F = 3.55$ (Figure 3C). The mean of **glucose** in the two groups of patients with a reference value 70-115 mg/dl indicates that both women and men results are within normal limits (Figure 3D). The mean of **total protein** in the two groups of patients with a reference value 6.4-8.3 g/dL indicates again that both women and men results are within normal limits (Figure 3E). **Ritis ratio (RDR), representing the ratio of**

AST and ALT: for male patients, the average value of AST/ALT is $M_M = 1.00$; for female patients, the mean value of AST/ALT is $M_F = 1.09$ (Figure 3F).

6 DISCUSSIONS

Under normal physiological conditions, the LDL cholesterol/HDL-cholesterol ratio has a value of 2.9 in women and 3.3 in men. The risk of coronary artery disease increases significantly if the value ratio exceeds 3.5 in women and 3.8 in men (Atanasiu, 2004). The determination of cholesterol and lipid metabolic disorders assesses their status, the risk of atherosclerosis, coronary stenosis and myocardial infarction. HDL cholesterol is a group synthesized and secreted by lipoprotein hepatocytes. HDL cholesterol is recommended to assess accurately the risk of developing atherosclerosis. HDL has an important role in cholesterol metabolism, participating in its transportation from extrahepatic tissues to the liver for excretion and catabolic (Gordon et al., 1989). Ritis ratio (RDR), representing the ratio of AST and ALT, whose normal values are between 0.7 and 1.4 (Roșoiu, 2005). ALT, is found primarily in the liver (the liver cells being located mainly in the cytosol) and in descending order of concentration in the kidneys, myocardium, skeletal muscles and pancreas. ALT is an indicator of cytolysis most frequently explored in the opinion of the majority of the best placed to detect even minimal liver damage. ALT is more specific to liver disease than AST (Fischbach, 2009).

Fish is a very important part of a healthy diet. Fish and other seafood are the major sources of healthful long-chain omega-3 fats and are also rich in other nutrients such as vitamin D and selenium, high in protein, and low in saturated fat. There is strong evidence that eating fish or taking fish oil is good for the heart and blood vessels. An analysis of 20 studies involving hundreds of thousands of participants indicates that eating approximately one to three servings of fatty fish a week reduces the risk of dying from heart disease by 36 percent (Mozaffarian&Rimm, 2006). Eating fish once or twice a week may also reduce the risk of stroke, depression, Alzheimer's disease, and other chronic conditions (Raji et al., 2014).

The results obtained in the 200 patients from Sulina who declared they eat fish products at least once a week confirm the literature data and prove that people who consume fish regularly tend to be healthier.
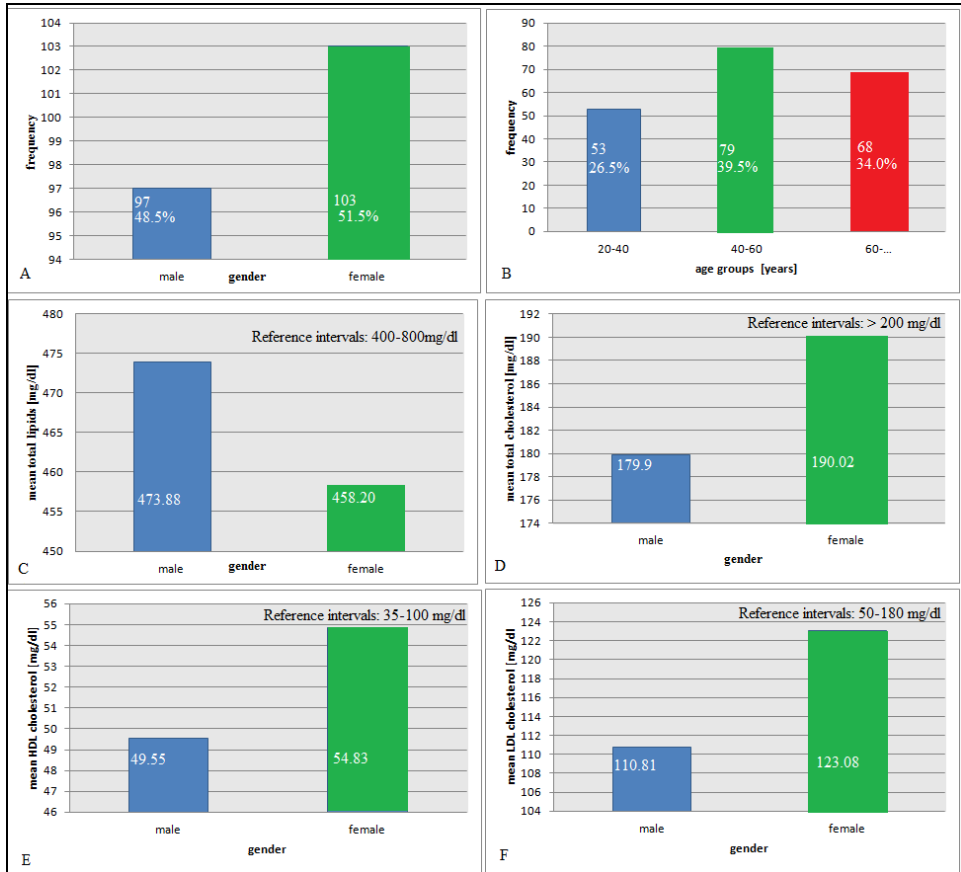
Figure 2. A, B: Patients distribution by gender and age groups; C: Mean total lipids in males and females; D: Mean total cholesterol in males and females; E: Mean HDL cholesterol in males and females; F: Mean LDL cholesterol in males and females
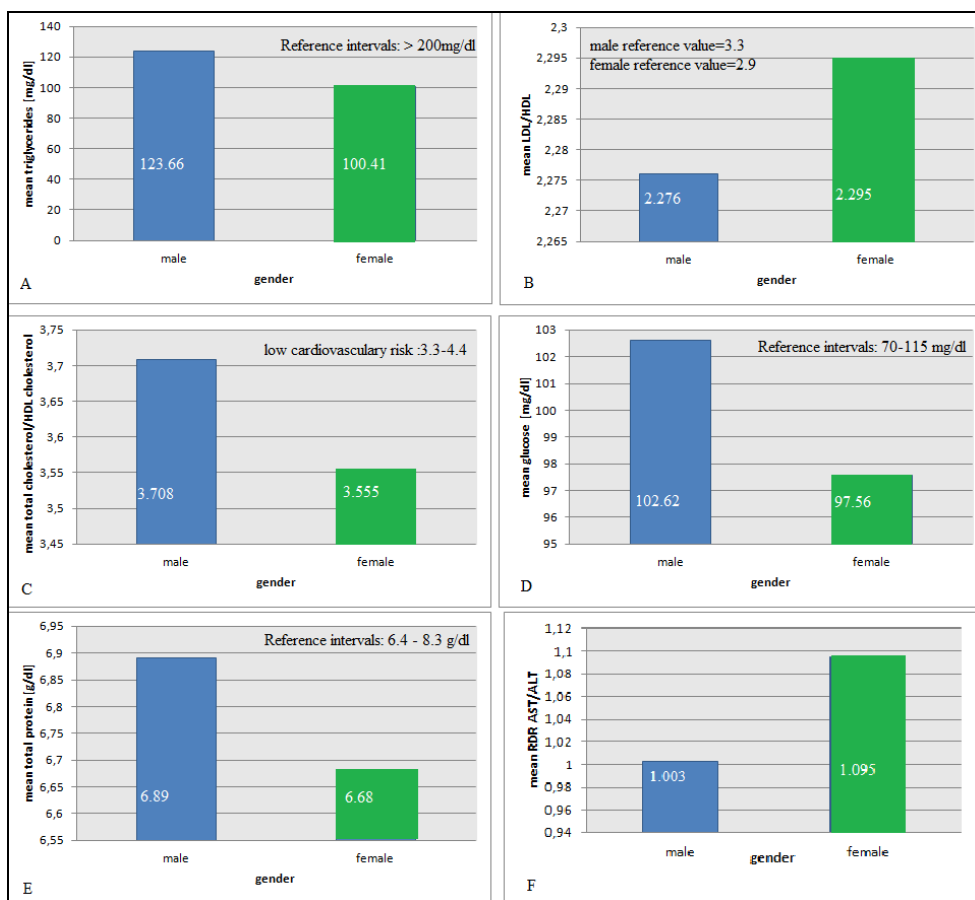
Figure 3. A: Mean triglycerides in males and females; B: LDL cholesterol/HDL-cholesterol ratio; C: Total cholesterol/HDL cholesterol; D: Mean glucose in males and females; E: Mean total protein in males and females; F: Mean RDR AST/ALT in males and females.

7 CONCLUSIONS

Because of the geographical area they live in, the inhabitants of Sulina, Danube Delta, are isolated and are constrained to eat more fish than other types of meat, Sulina being surrounded by water. People are more active than those from inland, they move around only with boats and they maximize the energy provided by total lipids, the final result being an overall good health. It is recommended that 50% of daily calories should come from carbohydrates, the recommended protein requirement is 0.8 to 1-1.25 g/kg/body/day, and 200 grams of fish provide almost 25% of the daily

protein requirement for an adult. Fish is an important source of protein with high biological value, containing all essential amino acids.

From the statistical analysis performed on the results of this research (200 patients: AST, ALT, AST/ALT, total lipids, total cholesterol, HDL cholesterol, LDL cholesterol, LDL/HDL, total cholesterol/HDL, triglycerides, serum glucose levels and total protein) it resulted that fish and fish products really help people maintain the normal level of total lipids and the liver metabolism. The consequences of inadequate lipids level are an increased risk of atherosclerotic cardiovascular disease; favoring excess weight/obesity. However, including fish/fish products in the regular diet (at least once a week) resulted in an overall good health state of the investigated patients.

ACKNOWLEDGEMENT

REFERENCES

Atanasiu, V. 2004, Lipid compounds. In *Medical Biochemistry*, 223p Edit. Niculescu, Bucharest, 223 p

Bondar, C., Dima, V., James, E. & Lungu, E. 2010, *Sulina Monograph*, Volume 1, Publisher Rawex Coms SRL, Bucharest, 496p

Bondar, C., Dima, V., James, E. & Lungu, E. 2010, *Sulina Monograph*, Volume 2, Publisher Rawex Coms SRL, Bucharest, 451p

Cleeman, J.I. 2001, Executive Summary of the Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation and Treatment of High Blood Cholesterol in Adults, *JAMA*, **285**: 2486-2497

Danube Delta Biosphere Reserve Administration, http://www.ddbra.ro/, accessed April 2016

Eurostat, http://ec.europa.eu/eurostat, accessed April 2016

Fischbach, F. 2009, *Chemistry Studies, A Manual of Laboratory and Diagnostic Tests*, 7th edition, Lippincott Williams & Wilkins Publishers, Philadelphia, 755p

Gordon, D.J., & Rifkind, B.M. 1989, High Density Lipoprotein-the Clinical Implications of recent studies, *N Engl J Med*, **321**(*19*): 1311-1316

Mozaffarian, D. & Rimm, E.B. 2006, Fish intake, contaminants, and human health: evaluating the risks and the benefits, *JAMA*, **296**: 1885-1899

Raji, C.A., Erikson, K.I., Lopez, O.L., Kuller, L.H., Gach, H.M., Thompson, P.M., Riverol, M., & Becker, J.T. 2014, Regular fish consumption and age-related brain gray matter loss, *Am J. of Prev Med.*, **47**(*4*): 444-451

Roşoiu, N. 2010, *Metode şi tehnici de laborator in biochimie [Methods and Laboratory Techniques in Biochemistry Volume 2: Clinical Biochemistry: diagnostic value]* publisher Ex Ponto, Constanta; 760p

Roşoiu, N. 2008, *Biochimie clinică [Clinical Biochemistry],* Vol. 1, Publisher Muntenia. Constanța, 347p

Roşoiu, N. 2005, *Biochimie medicală [Medical Biochemistry]*, Vol. 2, Publisher Muntenia, Constanța, 321p