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## OMEGA-UNSATURATED FATTY ACIDS – THEIR PRESENCE IN THE NUTRITIONAL REGIME FOR THE PREVENTION OF CERTAIN SOCIALLY SIGNIFICANT DISEASES

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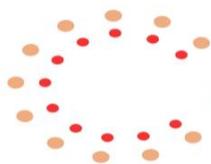
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### Abstract

The essential fatty acids provide normal biological functions of the human body and are important parts of the dietary intake in the prophylaxis and prevention of various diseases – cardiovascular diseases, hypertension, arthritis, asthma, and autoimmune disorders.

In this study, we present the results of a survey on the intake of dietary supplements containing omega-unsaturated fatty acids in addition to the diet in order to prevent some socially significant diseases. As a result of the survey, in order to satisfy the respondents' desire regarding the quality of the food supplements offered, we have developed a gas chromatographic method.

The results of the study highlight the benefits of taking omega-3 unsaturated fatty acids among different age groups for a better quality of life. We also offer a fast and reliable analytical method to verify the quality and safety of the food supplements on the market in Bulgaria.



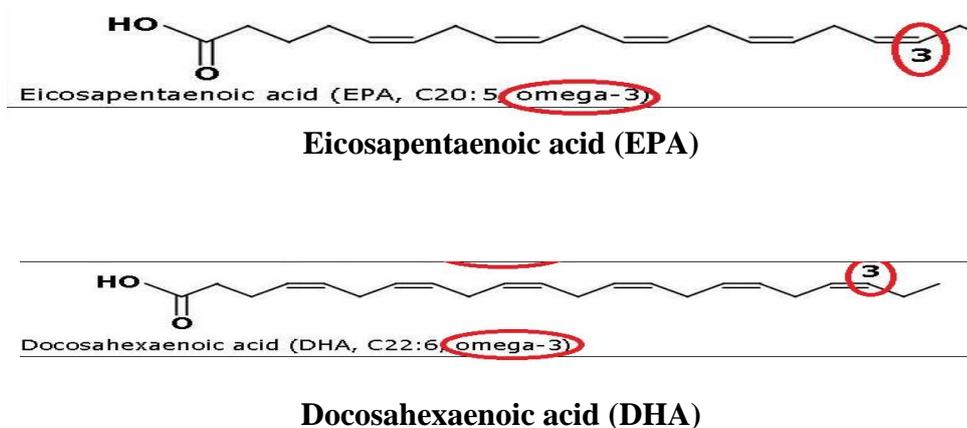
**Keywords:** omega fatty acids, diseases, nutritional diet, chromatographic method.

## Introduction

The most important and nutritionally applicable omega-3 unsaturated (essential) fatty acids are the *Eicosapentaenoic acid (EPA)* and the *Docosahexaenoic acid (DHA)*.

The polyunsaturated fatty acids (PUFA) contain two or more double bonds separated by methyl group. They differ not only in the number of the double bonds but also in their position regarding the beginning of the chain (designated with  $\Delta$  „delta”) or the last atom (designated with the letter  $\omega$  “omega”). Depending on the position of the double bond regarding the last atom, they are classified as polyunsaturated omega-9, omega-6 and omega-3 fatty acids (15).

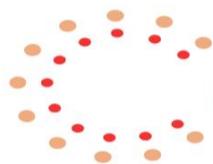
**Figure 1.**



*Figure 1. Polyunsaturated fatty acids - structure*

## Materials and methods

When preparing this article, we conducted a survey in the MEDLINE data base (accessible via PubMed) and Google Scholar. We used the terms: “omega unsaturated fatty acids” in correlation with “diseases” and “nutritional diet”. The statistical method with a social element – a survey. Analytical method – a developed gas chromatographic method.



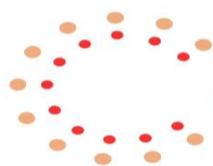
## Results and discussion

Fatty acids are contained in phospholipids and triacylglycerols. The presence of polyunsaturated fatty acids determines the biological activity of the phospholipids, the properties of the biological membranes, the interaction of the phospholipids and their transport and receptor activity. It has been established that the long-chain (C22, C24) polyunsaturated fatty acids are involved in the memorization mechanism and in the behavioral reactions as well (4).

Up until twenty years ago, the omega-3 fatty acids were mainly regarded as cardiovascular protectors, while today they are also highly recognized for the role they play in the immune regulation (including the protection they provide from inflammatory, autoimmune and tumor processes), their anti-rheumatic effect as well as their indispensable role in the structuring of the child's brain and the maintenance of the cognitive functions and the good mood of adults (1).

The concept of cardiac protection by supplementing the diet with omega-3 unsaturated fatty acids (UFA) definitely underlies the rule of medical knowledge and clinical thinking contained in the handbooks of the American Heart Association, which recommend taking omega-3 UFA (7, 10).

The guidelines of the World Health Organization (WHO) regarding dietary nutrition as well as the Food references recommend a total intake of fats between 20 and 35% of the total number of calories (3).

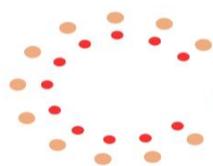


The most popular recommendations regarding the intake of omega UFA are those published by the AHA, the UK Committee of Medical Aspects of Food and Nutrition Policy, the World Health Organization and the European Food Safety Authority (EFSA) (3, 5). The American Heart Association (AHA) recommends a daily intake of about 1g EPA + DHA per day to patients with a proven heart disease. People who need to lower the level of the triglycerides are recommended to take 2-4g EPA + DHA per day under a doctor's supervision. The AHA also recommend that the patients without a documented coronary heart disease should also take omega-3 fatty acids by consuming different types of fish (preferably fatty fish) at least twice a week and include in their diet oils rich in alpha-Linoleic acid (flax seeds, rape and soya oil; flax seeds and walnuts). The Scientific Advisory Committee of Nutrition (SACN) of England recommends a daily intake of up to 450g of DHA + EPA for the different age groups. **Table 1** (6).

In the prophylaxis of cardiovascular diseases, the 4/1 proportion of omega-6/ omega-3 is related to the decrease of general death rate to 70%. The same proportion seems to be the optimal proportion for brain mediated functions (12, 13).

**Table-1: Adequate intake of omega-3 for different age groups**

Age group	Years	Omega – 3 UFA	Men (g / day)	Women (g / day)
<i>Babies</i>	0 – 6 months	ALA, EPA, DHA	0,5	0,5
<i>Babies</i>	7 – 12 months	ALA, EPA, DHA	0,5	0,5
<i>Children</i>	1 – 3 years	ALA	0,7	0,7



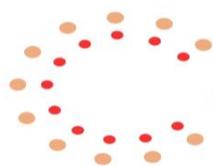
<i>Children</i>	4 – 8 years	ALA	0,9	0,9
<i>Children</i>	9 – 13 years	ALA	1,2	1,0
<i>Adolescents</i>	14 – 18 years	ALA	1,6	1,1
<i>Adults</i>	19 years of age and adults	ALA	1,6	1,1
<i>Pregnant women</i>	all ages	ALA		1,4
<i>Nursing mothers</i>	all ages	ALA		1,3

The main source of polyunsaturated higher fatty acids in the nutritional diet of the Bulgarians is the vegetable oil (mainly the sunflower oil) which is rich in omega-6. The transformation of ALA into EPA has been limited within the range of 10-20% (11).

Therefore, it is important to maintain the proper omega-3: omega-6 balance. The 1:4 proportion of omega-3 and omega-6 is recommended for prophylaxis of cardiovascular diseases (CVD), while the proportion of omega-3 and omega-6 in most nutritional ranges between 1:15 и 1:16, 7 (9)

The sources of omega-3 fatty acids, which are predominant in the diet, are different oils and fish. Fish is the main source of EPA and DHA, while the vegetable oils are the main source of ALA (8).

Some of the general sources of vegetable oils containing omega-3 ALA fatty acids are eatable walnuts, salvia – seed oil, chia – seed oil, flax seed oil, seaweed oil, hemp seed oil and some of the sources of animal omega 3 EPA and DHA fatty acids include – fish oil, calamari oil and krill oil. (2.14). **Table 2. Table 3.**

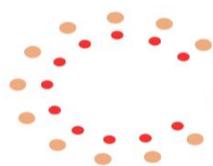
**Table - 2.  $\alpha$ -linoleic acid (ala), as percentage content in food.**

Name	Botanical name (L.)	% ALA
Flax seed	<i>Linum usitatissimum</i> L.	18,1
Hemp seed	<i>Cannabis sativa</i> L.	8,7
Walnuts	<i>Juglans regia</i> L.	0,6
Hazelnuts	<i>Corylus avellana</i> L.	0,1

**Table - 3.  $\alpha$ -linoleic acid as percentage content in oil.**

Name	Botanical name (L.)	% ALA
Kiwi seeds	<i>Actinidia deliciosa</i>	63
Chia seeds	<i>Salvia hispanica</i>	58
Flax seeds	<i>Linum usitatissimum</i>	53 – 59
Cranberry	<i>Vaccinium vitis-idaea</i>	49
Common purslane	<i>Portulaca oleracea</i>	35
Black raspberry	<i>Rubus occidentalis</i>	33
Hemp seed	<i>Cannabis sativa</i>	19
Rapeseed (Kanola)	<i>Brassica napus</i>	9 - 11

The supplementation of the nutritional diet with vitamins, mineral salts, amino acids, omega-3, 6, 9 unsaturated fatty acids, medicinal plants extracts, products intended for weight loss and memory improvement, anti-stress teas, fitness supplements and others – that have been the interests of most of the people visiting pharmacies, drug stores, MI.M and online trade websites in Bulgaria over the last years.



Through this survey we aim at establishing to what extent the consumers are able to assess the main things regarding: the therapeutic use of the Omega unsaturated fatty acids; information to be obtained prior to the admission of the product; product features; willingness to use the product; economic capacity to purchase and inclination to use the product.

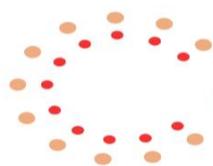
The survey has been made to collect information about: the socio-demographic features of the subjects (age, gender, education, profession); Maximum 42 question on the 10 factors; The participants assess each parameter based on a numerological scale of 1 to 4 (1 indicates an opinion “not important at all”, 2 – “unimportant”, 3 – “average importance”, 4 – “very important”).

The results have been processed using the statistical programme. A factor analysis with a variable rotation has been used in the original Food Choice Questionnaire (FCQ), as it has also been used to group the questions by factors.

The respondents taking part in this survey are 200 participants aged 16-75. The average age of the participants is 38.8 years for the respondents from the city of Sofia and 32.7 years for the respondents from the city of Plovdiv. The education and the profession of all the participants are quite varied (medical representatives were not included in this survey).

**Table - 4. Comparison of the respondents in terms of gender.**

Factor	Women		Men		Sig p-value
	mean	SD	mean	SD	
Health	2,92	0,67	2,48	0,7	0,003
Mood	2,35	0,73	1,92	0,74	0,004



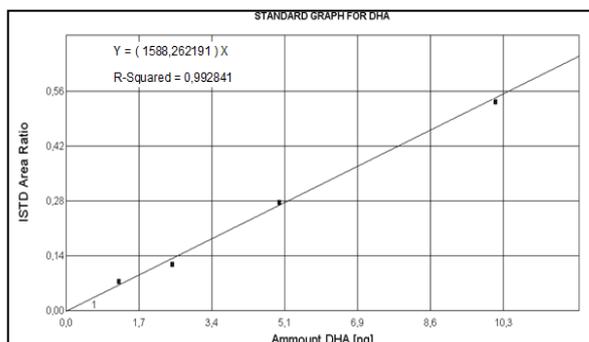
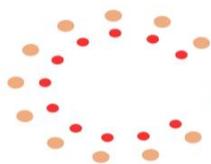
Amenities	2,71	0,71	2,48	0,72	0,041
Organoleptic features	3,21	0,56	2,6	0,62	0,000
Content	2,41	0,88	2,2	0,69	
Price	2,66	0,78	2,41	0,71	
Weight control	2,61	0,88	2,05	0,74	0,001
Knowledge of the product	2,18	0,65	2,03	0,70	
Ethical aspects	1,91	0,59	1,83	0,66	

The table shows the proportion between the two sexes of the respondents included in the extract in terms of different factors influencing their choice of food supplements containing Omega unsaturated fatty acids. A significant difference has been registered in five of the nine factors: health, mood, amenities, organoleptic features and weight control, respectively.

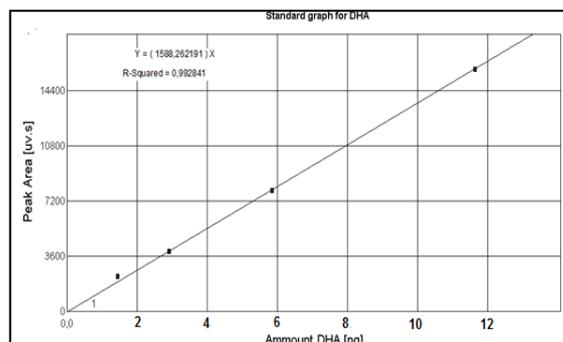
Regardless of the fact that no statistically significant differences have been found in the answers given by the respondents, both sexes have shown a substantial interest in the content of the food supplements (Factor 5 of the survey), as a result of which we have developed a prompt and reliable analytical method.

The gas chromatographic method has been validated in terms of the analytical parameters: linearity; sensitivity; reproducibility.

Linearity – the dependence of the area of the chromatographic peak of DHA on the quantity entered in the chromatograph was examined by obtaining chromatograms of a number of methylated standard solutions of DHA and nonadecanoic acid used as an internal reference.



**Figure 2.** A standard DHA graph with an internal reference



**Figure 3.** A standard DHA graph based on a method involving an external reference

The standard graph shows that for quantities of up to 400 ng entered into the chromatograph, the dependence of the proportion of the areas of mutilated DHA and the methylnonadecanoic acid (the internal reference) of the quantity of DHA is linear.

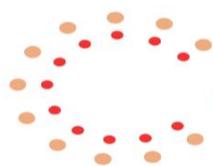
Reproducibility.

The repeatability of the measurement of the area of the DHA peak (without using an internal reference) and the proportion of the areas of the chromatographic DHA peaks and the internal reference has been determined by means of a threefold performance of the analytical procedure in the derivatization and extraction using hexane when adding 400 µl of a standard DHA solution with a concentration of 500 µg/ml in the reaction container.

The developed gas chromatographic method has shown positive values of the analyzed omega unsaturated fatty acids, which is in conformity with the requirements for satisfactory qualitative and quantitative indicators of the tested objects that are applicable on the Bulgarian market.

## Conclusion

The conducted survey on the attitude of the consumers towards the Omega unsaturated fatty acids in the cities of Sofia and Plovdiv has shown that there is a statistically significant



difference regarding their use. The development of optimal and easily executable pharmacological analytical methods for analysis regarding the content of Omega-3 unsaturated fatty acids is also very important for the consumers.

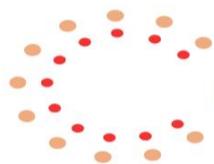
The developed analytical method allows an analysis of samples containing Omega unsaturated fatty acids.

Qualitative analysis – the specified options allow the identification of the peaks of the methylated fatty acids by the times of retention through their comparison with the available comparative substances.

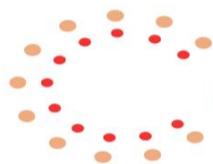
Quantitative analysis: comparison of the areas of the chromatographic peaks of the components of the analyzed sample with comparative substances; assessment of the relative values of the areas of the chromatographic peaks of the analytes with reference to the sum total of the areas of all components of the analyzed object; analysis of the fatty acids in objects consisting entirely of lipids.

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