

**"CAROL DAVILA" UNIVERSITY OF MEDICINE AND PHARMACY,
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DIABETES, NUTRITION AND METABOLIC DISEASES**

***Combating mercury and other toxic substances
from food***

PHD THESIS SUMMARY

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INTRODUCTION

Toxic substances, harmful to the environment but also to the human body, can have negative effects on our health. These substances can exist in the composition of some frequently used products or can be present in the air, water or soil [1]. Exposure to toxic substances is achieved by inhaling vapours, fumes, dust, by ingesting them or by absorption through the skin or mucous membranes. Depending on the toxicity degree, the time and method of exposure to the risk factors, but also on the reaction of each person to such substances - the way the immune system protects the body, the impact they can have on health can be determined [2].

The presence of heavy metals and other toxic substances in the human body is associated with brain degeneration, obesity, diabetes, cardiovascular diseases, neurological imbalances, and other health degradations [3]. Along with these, other factors such as the presence of an excessive amount of omega 6 polyunsaturated fatty acids and an unbalanced ratio between omega 6 and omega 3, as it happens in the contemporary Western diet, stimulate the pathogenesis of several diseases, including cardiovascular diseases, cancer, inflammatory and autoimmune diseases, while a higher level of omega 3 polyunsaturated fatty acids exerts mitigating effects [4].

Health isn't a trend, nutrition isn't a trend, detoxification isn't a trend, they only have applicability depending on the individual profile, health status, current ailments that require the adoption of other lifestyle changes.

This paper is structured in two distinct parts. The first part of this doctoral study - the general part - is dedicated to the presentation of theoretical notions, which highlight the current state of knowledge and the importance of deepening the chosen topic. The second part includes the original, personal contributions and the studies that were carried out are presented, with the main and secondary objectives, the results obtained, conclusions and perspectives for development. The results of this scientific work will contribute to a better understanding of the impact generated by the presence of heavy metals and other toxic substances in the human body, but especially to their management through a personalized protocol, related to each person, meaning a food plan and a dietary supplement protocol.

CURRENT STATE OF KNOWLEDGE

The general part refers to the current state of knowledge. It is structured in 9 chapters, as follows:

I.1. History of mercury and other toxic substances poisoning

Exposure to different sources of intoxication, repeatedly, without supporting a proper detoxification process and without identifying the sources of contamination so that they can be avoided, can be a trigger for the accumulation of toxic metals, which affects the proper functioning of the human body.

I.2. Sources of mercury poisoning (food, industry, soil, dental amalgam)

The main sources of mercury contamination are dental fillings with amalgam, food (fish, mollusks), food from agriculture where pesticides have been used, which contaminates the soil and water [5], plastic materials, neon light bulbs, light bulbs with low consumption energy. Inside the human body, the average half-life of inhaled mercury is about 60 days [6].

The clinical manifestations of chronic exposure to organic mercury have a gradual onset, the main area of action being the nervous system. Chronic exposure to mercury can cause symptoms such as fatigue [7], weakness, headache and reduced ability to concentrate [8]. In severe cases, chronic exposure leads to intellectual impairment and neurological abnormalities.

I.3. Sources of aluminum poisoning (food, cosmetic products)

Common symptoms of aluminum contamination are exhaustion, lack of concentration, long-term or short-term memory problems, calcium and phosphorus deficiency (risk of osteoporosis), muscle pain, anemia, digestive disorders, abdominal cramps, kidney failure, autoimmune diseases [8].

Possible sources of aluminum contamination are tap water (drinking and household water), kitchen utensils made of aluminum (cutlery, dishes, aluminum foil), aluminum packaging, food with synthetic additives, cosmetics (deodorants, antiperspirants, perfumes, toothpaste), vaccines, some medicines (antacid treatments) [9].

I.4. Sources of arsenic poisoning (food, drinking water)

Frequent sources of arsenic contamination (introduced into the body by inhalation, ingestion) are identified in soil and drinking water levels, food products resulting from repeated treatments with insecticides (cereals - wheat, rice; vegetables; marine products), electronic devices [10, 11]. Arsenic accumulation in rice is one of the most common [12] ingestion

sources, long-term and repeated consumption representing a major risk factor, both among adults and children [13].

Short-term exposure to arsenic affects blood vessels, leads to a change in erythrocytes, leukocytes, abnormal heartbeats, tingling sensations in the hands and feet, which induce a state of discomfort, up to the appearance of anxiety [14].

Long-term exposure to sources of arsenic contamination can cause skin lesions, peripheral vascular disease, neurological problems, diabetes, certain types of cancer, chronic fatigue syndrome [15].

Exposure and absorption of heavy metals in the body (mercury, aluminum, arsenic, lead) often impacts the health of the brain, the way it works, inducing various manifestations.

I.5. Sources of poisoning with different contaminants (food allergens, genetically modified organisms, pesticide residues, additives, residues of veterinary drugs)

Food must be associated with a daily investment in our health, avoiding eating anything, anytime, anywhere, our body not being a garbage can in which we throw everything that generates processes of fermentation or putrefaction and implicitly inflammatory processes. The role of each food in our daily menu helps us make rational, not instinctive choices, with the main objective being the quality of our life. Food allergens, additives, genetically modified organisms, pesticides can bring major benefits to certain food productions, but their negative impact on our health can be a decisive factor in our everyday choices.

I.6. Pathophysiology of poisoning with mercury, aluminum, arsenic and other food contaminants

The presence of heavy metals affects especially the brain, peripheral nerves, hematogenous marrow, digestive tract, cardiovascular system and kidneys. The affected organs and the severity of the manifestations depend on the amount of metals present in the body, the type of metals, the age of the patient, the chronicity and the period of exposure.

The most obvious toxic effects generated by the presence of food allergens, genetically modified organisms, pesticide residues, food additives, veterinary drug residues are manifested at the level of the skin, immune system, kidney function, but also at the level of the central nervous system.

I.7. Clinical manifestations in mercury, aluminum, arsenic, and other food contaminant poisonings (signs and symptoms related to neurological, renal, gastrointestinal toxicity, cancer)

In the context of inadequate nutrition, the individual potential of each of us cannot be manifested and, hence, the reduced ability to support detoxification, the blocking of the body's

regenerative function, the reduction of physical and intellectual performance. The brain functions on the basis of essential fatty acids, in an optimal ratio of omega 6: omega 3 and carbohydrates. Their sources are food and that is why it is very important that daily meals ensure the intake necessary for a good functioning [16].

According to the latest studies, the incidence of brain disorders is increasing, both in children and adults. One in eight seniors develops Alzheimer's disease and one in eight children is diagnosed with brain development imbalances, including autism, ADHD, and ADD [17-19]. Worldwide, there are an estimated 24 million cases of dementia, doubling in the past 20 years [20].

Anxiety, learning disabilities, lack of focus, depression, memory impairment, sleep imbalances are much more present in recent years, especially in people who do not provide daily food for the brain (omega 3), which is in a continuous degradation, manifested through different symptoms [21]. In the United States of America, the most common drugs recommended by specialists are antidepressants [22]. Indeed, there are also cases when depression is induced by emotional problems, but most often the health of the brain is affected.

There is scientific evidence that shows that the appearance of imbalances in the human body can be prevented by lifestyle changes, especially from a dietary point of view.

We face frequent scientific dilemmas regarding the maximum acceptable level of exposure, both for heavy metals and other toxic substances. At the same time, it is highlighted that the presence in the body of heavy metals and other food contaminants cannot be considered safe, being clinical manifestations even at low levels of them in the body.

Due to the high level of toxicity, metals such as arsenic, cadmium, lead, mercury play an important role in mental health, but not only because they have the ability to be located anywhere in the body, in the thyroid, prostate, heart, muscles, bones. The presence of metals influences the chemistry of the body, inducing major imbalances, especially due to the generated demineralization.

Recent studies place particular emphasis on chronic exposure to heavy metals and other toxic substances, even at extremely low levels, which produces disturbances in physiological processes of the body through an imbalance of the detoxification capacity and the oxidant/antioxidant balance. In this context, taking into account the oxidative stress induced by this accumulation of heavy metals or other toxic substances in the human body, aspects related to the impact of antioxidants in the human body must be considered.

I.8. Diagnostic methods in mercury, aluminium, arsenic and other food contaminants poisoning (blood, saliva, urine, gut microbiome, tissue mineral analysis, dried blood)

Blood, saliva, urine, intestinal microbiome analysis, tissue mineral analysis, as well as tests performed on dry blood allow a fast assessment of the risk of heavy metal accumulation in the human body, diagnosis being necessary in establishing a personalized protocol. The collection rules and the compliance of the patient with certain conditions before are important to avoid confusion in the heavy metal toxicity diagnosis.

I.9. Treatment of mercury, aluminium, arsenic and other food contaminants poisoning (principles of allopathic treatment, nutrition, food supplements)

Arsenic, mercury, lead and cadmium generate cellular chaos in the human body. These are the top four most toxic heavy metals according to the CDC's Priority List of Hazardous Substances. When toxic metals enter the body, a part is eliminated through some natural detoxification processes depending on the capacity of each individual body, but there are also amounts of toxic metals that accumulate at the cellular level. In order to remove toxic metals accumulated over time, it is important to establish a chelation procedure, which supports the detoxification of the body in a constant way, with chelators being administered at regular intervals. A decrease in the concentration of chelators or irregular administration of treatments can generate a retransmission of heavy metals at an organic level, especially at the level of the brain. The personalization of the protocol on an individual level allows an adaptation to the needs of each patient, to obtain the most beneficial results.

WORKING HYPOTHESIS AND GENERAL OBJECTIVES OF THE RESEARCH

II.1. The motivation of the study

This doctoral research aims to identify and implement an integrated concept of chelation of heavy metals and other toxic substances that have entered the human body.

A diet structured according to the antioxidant capacity of each food can improve the body's defense capacity against free radicals. Thus, in the current study, I proposed the development of an integrated concept which includes personalized nutrition programs based on foods with an antioxidant role and therapies with antioxidant supplements, which can antagonize the effects induced by the presence of heavy metals and other toxic substances in the human organism. Thus, through this holistic approach of each patient, I also aim to identify personalized solutions that can help specialists in their efforts to improve the life quality of patients intoxicated with various heavy metals or other toxic substances that enter the human body through food.

II.2. The objectives of the research study comprise the main objective and the secondary objectives as follows:

The main objective is:

- highlighting the correlation between intoxication with heavy metals or other toxic substances that have entered the human body and the antioxidant capacity of the body, especially through the concentration of omega 3, respectively the omega 6: omega 3 ratio

Secondary objectives are:

- the evaluation of the omega 3 supplements sold in the geographical area of Romania, through the comparative analysis of their composition, considering the concentrations of omega 3 (DHA, EPA)
- the development and use of an omega 3 supplement, with a higher concentration of DHA compared to EPA, to ensure the proper functioning of the brain, without heavy metals in the composition, with an optimal TOTOX level
- the evaluation of the dietary profile in a group of 77 patients, who had high omega 3 deficiencies and high values of omega 6: omega 3 ratios

- the identification of risk factors (sources of contamination) from the menu adopted by the 77 subjects: heavy metals, food additives, genetically modified organisms, pesticides, excess sugar, excess fat, antibiotic residues and hormones in food
- the development of a personalized nutrition plan for each subject for a period of 120 days, with monitoring of the evolution according to the symptoms mentioned during the anamnesis
- the development and implementation of a personalized detoxification protocol, without the involvement of allopathic treatments, based on foods with a high antioxidant capacity and dietary supplements with an antioxidant role, especially omega 3 and glutathione
- the correlation between certain foods consumption and exposure to heavy metals and other toxic substances (food additives, genetically modified organisms, pesticides, antibiotics, growth hormones)
- the correlation between nutritional status, antioxidant foods and severity of heavy metals exposure, food additives, pesticides, genetically modified organisms, antibiotics, growth hormones.

II.3 Work material

The research was carried out using a descriptive survey based on qualitative and quantitative methods in a population of patients intoxicated with heavy metals (mercury, aluminium, arsenic) whose diet was based on foods containing additives, pesticides, genetically modified organisms, antibiotics and growth hormones.

The subjects were selected from the patients who had presented at the Nutribalance nutrition consulting office, Bucharest.

The recruitment took place between October 2017 and July 2021, and participation in the study was conditioned by meeting the inclusion criteria:

- the presence of results that revealed heavy metal poisoning
- presence of results for omega 3 and omega 6 concentrations: omega 3
- the manifestation of symptoms specific to the respective contaminations
- the division into working groups was carried out periodically, depending on the manifested symptoms and the adopted dietary profile.
- the considered exclusion criteria were:
- lack of specific analyzes and interpretations for heavy metals

- the lack of specific analyzes and interpretations for omega 3, respectively omega 6: omega 3 concentrations
- the absence of manifestations associated with the presence of heavy metals
- neoplastic diseases

The data used was extracted from the patients' assessment sheets and from the Nutribalance database of the Nutribalance Nutrition Consulting Cabinet, Bucharest and the data collection type was longitudinal.

Each patient was explained the methodology of the study and the benefits of a customized, complex protocol developed by an interdisciplinary committee, starting from the individual results to the investigations performed, avoiding the use of standard treatment schemes. Each patient consented to be part of this study.

All procedures involving human subjects were performed in accordance with the guidelines set forth in the Declaration of Helsinki (October 2013).

Initial assessment included medical history, demographics, risk factors, lifestyle (focusing on dietary profile), results and interpretations of heavy metal analyses, omega 3 concentration, omega 6: omega 3 ratio value. Thus, the demographic parameters of the patients, clinical and biological characteristics were considered. Regarding demographics, the following were analysed: sex, age, rural or urban provenance (important in the development of the daily food plan), the field of activity in which they work, the average monthly income, the number of working hours per week.

II.4 Research methods

The study followed a retrospective interventional protocol, based on subjects enrolment at presentation in the Nutribalance nutrition consulting office, Bucharest, depending on the results of specific heavy metal poisoning analyzes. Also, one of the conditions for enrolling in the study was represented by the presence of results related to the analyzes for omega 3 fatty acids and for the omega 6: omega 3 ratio.

The dosage of omega 3, omega 6 fatty acids and heavy metals was carried out outside the office, the subjects presenting themselves with the results and their interpretation.

We studied the dietary profile of each study participant through the face-to-face interview method.

We assessed the dietary supplement plan for each study participant through the face-to-face interview method.

We studied by comparison the composition of omega 3 supplements from the geographical area of Romania mentioned by the study participants as being part of their supplement scheme.

Together with a team of researchers and doctors from Norway, we developed an omega 3 supplement, which was produced in a factory in Norway, imported and distributed in Romania, registered at the National Research Institute - Food Bioresource Development and at the State Office for Inventions and Trademarks. This omega 3 supplement stands out for the high concentration of DHA compared to EPA, for the optimal level of TOTOX, for the absence of heavy metals its composition, for the power of absorption, evaluated by analyzes carried out before and after use.

The study included a sample of 77 patients, of which 40 men and 37 women, aged between 13 and 69 years, with a monitoring period of 120 days, during which each subject in the study group followed a personalized program of food and food supplements, adapted to the analyzes performed (mercury, aluminium, arsenic, omega 3, omega 6: omega 3 ratio). 77 subjects were selected, whose results to the considered analyzes exceeded the upper limit of the reference range for mercury, aluminium or arsenic. The value of the omega 6: omega 3 ratio was much higher than the reference value, and they were deficient in omega 3.

In the anamnesis stage, we took into account the symptoms, the conditions specific to the presence of these metals in each patient, the omega 3 concentration, the omega 6: omega 3 ratio, possible sources of contamination with the identified metals, dietary behaviour (risk factors and foods with a therapeutic role), lifestyle.

At the end of the period we refer to, namely 120 days, we assume that there will be a difference in terms of existing symptomatology in subjects diagnosed with potentially toxic metals, as well as increases in omega 3, decreases in specific values for the omega ratio 6: omega 3 and reducing the amount of heavy metals in the blood in the first stage of diagnosis.

Specific symptoms are often the first indicators of contamination and help identify the contaminated person. Symptoms that occur because of toxic metal accumulation include reduced ability to concentrate, attention deficit, central nervous system disorders, kidney disease, liver disease, sleep disorders, emotional instability, depression, significant weight gain, cardiovascular disease, diabetes. Some common symptoms with those manifested in the context of an omega 3 deficiency and an increase in the specific values of the omega 6: omega 3 ratio are observed, a context in which we decided to study the correlation between the presence of metals with potential for intoxication (mercury, aluminium, arsenic), omega 3 deficiency and increased omega 6: omega 3 ratio.

The analysis of the food profile takes into account the frequency of the main food groups, mainly looking at the excessive consumption of farmed fish or possibly contaminated food, but also the exposure to repeated vaccinations [23, 24], the sources of drinking water, the cosmetic products used.

The re-evaluation of each participant was carried out at an interval of 2 weeks, taking into account the weight, the manifested symptoms, the impact of the eating plan on the health status, from a physical and intellectual point of view.

During this 120-day period new assessments were recorded for a complete heavy metal profile, for omega 3, for the ratio of omega 6: omega 3, but also taking into account the manifested symptoms.

II.5 Statistical analysis

The applied research method is statistical, with independent, quantitatively evaluated (laboratory analyses) and dependent (observable, physically characterized) variables. The obtained data were entered into specific databases, using Microsoft Office Excel 2019, and the statistical analysis was performed with the Python program. Thus, with the help of descriptive data, the obtained values could be interpreted, depending on the shape of the distribution. The libraries used were pandas, numpy, scipy (for data import, data processing, running statistical tests), but also matplotlib, seaborn (for graph generation). Also, arithmetic mean, standard deviation, minimum value and maximum value for quantitative (numerical) variables.

Qualitative variables were expressed in absolute and relative frequencies (expressed as percentages).

Student's t-test for dependent samples was used to compare data samples before and after the implementation of the diet plan and supplements according to the analyzed variables

Numerical results are presented as mean +/- standard deviation.

II.7 The RESULTS of this study highlight the beneficial impact of the personalized diet and the supplements used on the decrease in the omega 6: omega 3 ratio, the significant increase in the concentration of omega 3, which supports the detoxification process of heavy metals, associated with a significant improvement of the symptoms shown by each individual patient.

As a result of the anamnesis taken in the initial stage for the 77 subjects in the study group, the presence of the following was observed: attention deficit in 77 out of 77 subjects, reduced ability to concentrate in 77 out of 77 subjects, sleep disorders in 77 out of 77 of subjects,

cardiovascular disease in 21 of 77 subjects, development of nervous tics in 62 of 77 subjects, mood changes in 77 of 77 subjects, muscle weakness in 77 of 77 subjects, change in visual acuity in 77 of 77 subjects . All 77 subjects were identified as consuming farmed fish (raw or cooked), which implies exposure to methylmercury, even though they had different frequency in the weekly menu.

The presence of amalgam fillings is found among 69 subjects from the selected group who present symptoms specific to chronic fatigue syndrome.

Following the individual evaluations, I developed for each patient a personalized program of nutrition and food supplements, for a period of 120 days, referring to the lifestyle of each subject, to food preferences, to the staged evolution of each participant in the study.

The common element in the case of this batch of subjects was the dietary supplement protocol, which included:

- an omega 3 supplement, with an increased concentration of DHA – 0.15 ml/kg body weight/day

- the clay of Râciu, being used both in the internal cure - 2 spoons of clay were added to 500 ml of alkaline water during the night and the "clay milk" was drunk the next day, before breakfast, including the remaining undissolved powder), as well as in an external cure (foot baths made daily, in the evening, 2 hours after dinner, for 30 minutes, using 200 g of Râciu clay, 5 l of warm water, but also wraps with Râciu clay, in cures of 3 weekly sessions, throughout the duration of the study

- coriander essential oil -1 drop *3/day, sublingual
- selenium - 200 mcg/day
- probiotic -1 capsule *3/day
- ramp extract – 2 ml *3/day
- coriander tincture – 2 ml *3/day
- pomegranate juice – 150 ml/day – in the morning
- liquid chlorophyll - 30 ml/day - in the morning
- glutathione 600 mg/ 4 ml - 3 infusion treatments, administered weekly, throughout the duration of the study, but with the possibility of extending or reducing the frequency, depending on individual tolerance

- infrared sauna - exposure to the infrared sauna was carried out 3 times each week, throughout the study period

During the study period, the symptoms of each patient were monitored, a personalized plan of food supplements and nutrition was followed, avoiding the administration of additional treatment schemes.

The re-evaluation of each patient was carried out at an interval of 14 days, during which the evolution of the food profile was analyzed, defined as a favourable one in the management of the mentioned symptomatology.

At each reassessment, the food program was adapted according to the compatibility with each food or food group, the symptoms that appeared because of the introduction of new foods (for example: improving the ability to concentrate, reducing attention deficit, improving the quality of sleep, weight loss, reduction in frequency and intensity of nervous tics, reduction in muscle pain).

After the 120-day period of monitoring the evolution of each subject in the study group, we evaluated the symptoms mentioned at the anamnesis stage, the analyzes for mercury, aluminium, arsenic, omega 3, the omega 6: omega 3 ratio and we performed a comparative analysis between the initial evaluation and the final one.

Through the analysis of the 77 participants in the study group, after the 120-day monitoring period, we observed the presence of the following: predisposition to attention deficit in 6 out of 77 subjects (an improvement is observed, but elements specific to attention deficit are present on different types of activities), reduced ability to concentrate in 6 out of 77 subjects (an improvement compared to the initial stage is observed), sleep disorders in 0 out of 77 subjects, cardiovascular diseases in 0 out of 77, development of nervous tics in 1 out of 77 77 subjects, mood changes in 0 of 77 subjects, muscle weakness in 0 of 77 subjects, change in visual acuity in 0 of 77 subjects (Table 1). Also, the chronic fatigue symptoms of the 77 subjects improved after farmed fish was excluded from their diet.

Although fish is recognized for its significant omega 3 intake, the presence of mercury can negate the benefits and is recommended to be avoided. Individual tolerance is the one that has a decisive role in the frequency of certain food groups in the weekly menu, depending on the detoxification capacity, a function that can be limited, including at the DNA level.

Figure 1 shows the distribution of patients according to the results of specific omega 3 concentration analyses. The results show a significant decrease in the percentage of patients whose omega 3 concentrations were in the range [1-3] from 64.94% to 13%, and an important increase in the percentage of patients who had concentration values in the range [3.1-5].

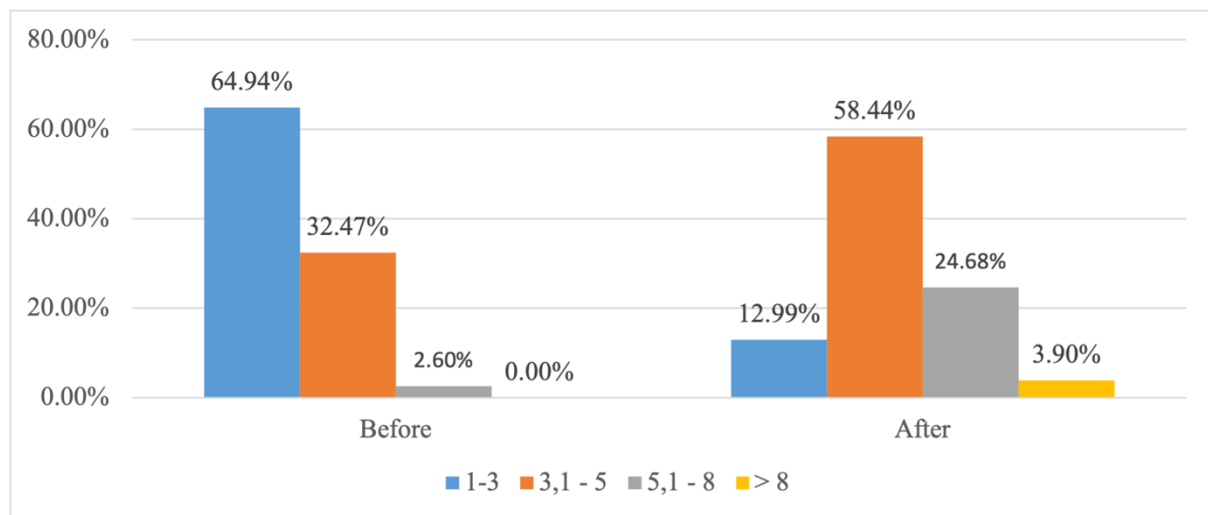


Figure 1. Distribution of patients according to the results of specific omega 3 concentration analyses

Figure 2 shows the distribution of patients according to the results of specific analyzes of the omega 6: omega 3 ratio. From this figure it can be seen that the highest percentage of patients had values greater than 50 of the omega 6: omega 3 ratio initially, and after the implementation period of the customized protocol, this percentage decreased 26 times.

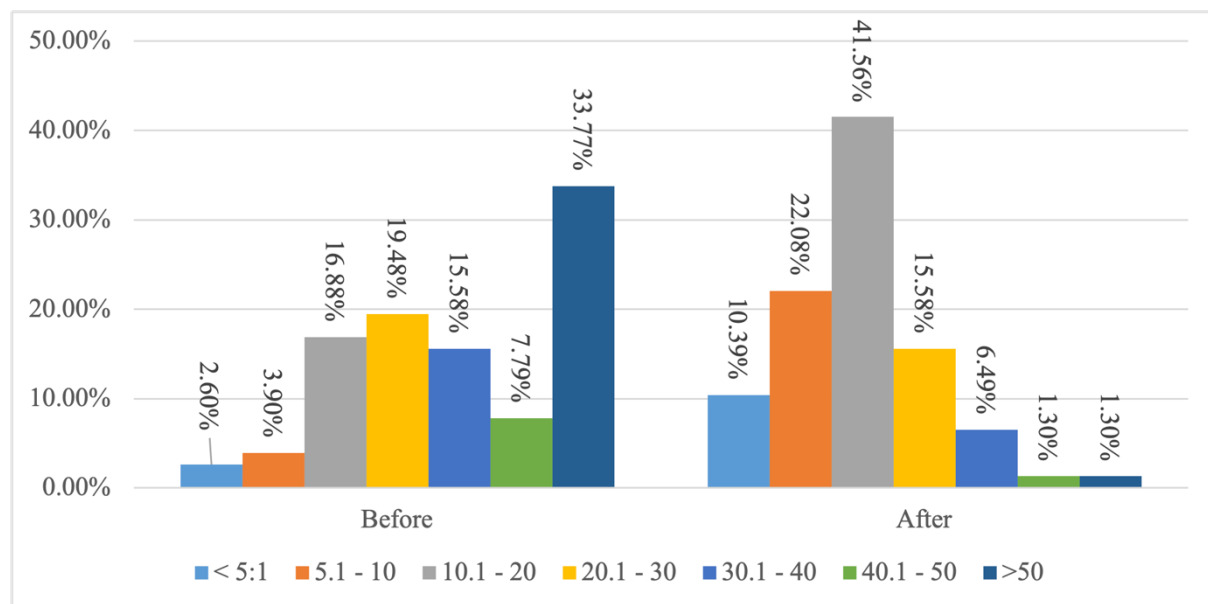


Figure 2. Distribution of patients according to the results of specific analyzes of the omega 6: omega 3 ratio

Initially, the study group had results for omega 3 in the range of 1.10 - 6.80, with an average of 2.71, in the context where the reference range is 8-12.

	Initial			
omega 3 concentration	1-3	3,1 – 5	5,1 - 8	> 8
number of participants	50	25	2	0
participants %	64,94%	32,47%	2,60%	0%

omega 6: omega 3 ratio	< 5:1	5.1-10	10.1-20	20.1-30	30.1-40	40.1-50	>50
number of participants	8	17	32	12	5	1	1
participants %	10,4%	22,08%	41,56%	15,58%	6,49%	1,30%	1,30%

Table 4. Distribution of patients based on omega 6: omega 3 ratio intervals - after

Based on the results presented in Figure 1 and Figure 2, the average concentration of omega 3 increased from 2.71 to 4.59, and the result of the ratio omega 6: omega 3 a decreased from 37.97 to 15.56, after implementing a customized protocol for 120 days.

The calculation for the omega 6: omega 3 ratio before and after following the customized protocol:

- Result: Statistics = 11.087, $p = 0.00000000000000001520$
- #Average initial Omega6: Omega3 result: 37,966
- #Average final Omega6: Omega3 result: 15,395

	Initial average	Final average
omega 3 concentration	2,71	4,59
omega 6: omega 3 ratio	37,97	15,56

Table 5. Averages of omega 3 concentration and omega 6: omega 3 ratio - before and after

Following the treatment, a reduction in the specific values for the omega 6: omega 3 ratio in the blood was observed (37,966 -> 15,395).

The value of $p < 0.05$ causes a rejection of the null hypothesis, which means that the mean change is statistically significant.

It is observed that for the 10 selected omega 3 products used by the study participants, the daily dose varies between 82.5 – 378 mg DHA, respectively 75 – 530 mg EPA, the average being 184.65 mg DHA and 295.8 mg EPA.

Manufacturer name	Supplement name	DHA concentration / daily dose	EPA concentration / daily dose
MY VITAMINS	OMEGA 3 PLUS	90 mg	530 mg
DOPPEL HERZ	Omega -3 extra 1000 mg	120 mg	180 mg
SOLGAR	OMEGA – 3	378 mg	504 mg
NOW	Molecularly Distilled Omega – 3	240 mg	360 mg
LABORATOIRE BIOCYTE	OMEGA 3 KRILL	82.5 mg	180 mg
SECOM	Algal Omega 3	150 mg	75 mg

SOLGAR	Omega 3-6-9	221 mg	344 mg
MOLLER*S	Omega 3	250 mg	310 mg
LYSI	OMEGA -3	95 mg	145 mg
OLIMP SPORT NUTRITION	OMEGA 3	220 mg	330 mg

Table 6. Concentration of DHA and EPA for daily doses of the products used by participants

The recommended daily dose for an adult weighing 60 kg is 2000 - 3000 mg of omega 3, of which the majority component must be DHA.

The product developed as part of the carried-out research:

- DHA: 1429 mg/ 10 ml product = daily dose

- EPA: 571 mg/ 10 ml product = daily dose

Manufacturer name	Supplement name	DHA concentration / daily dose	EPA concentration / daily dose
NUTRIBALANCE	OMEGA 3 OIL	1429 mg	571 mg

Table 7. Concentration of DHA and EPA for daily doses for Omega 3 Oil

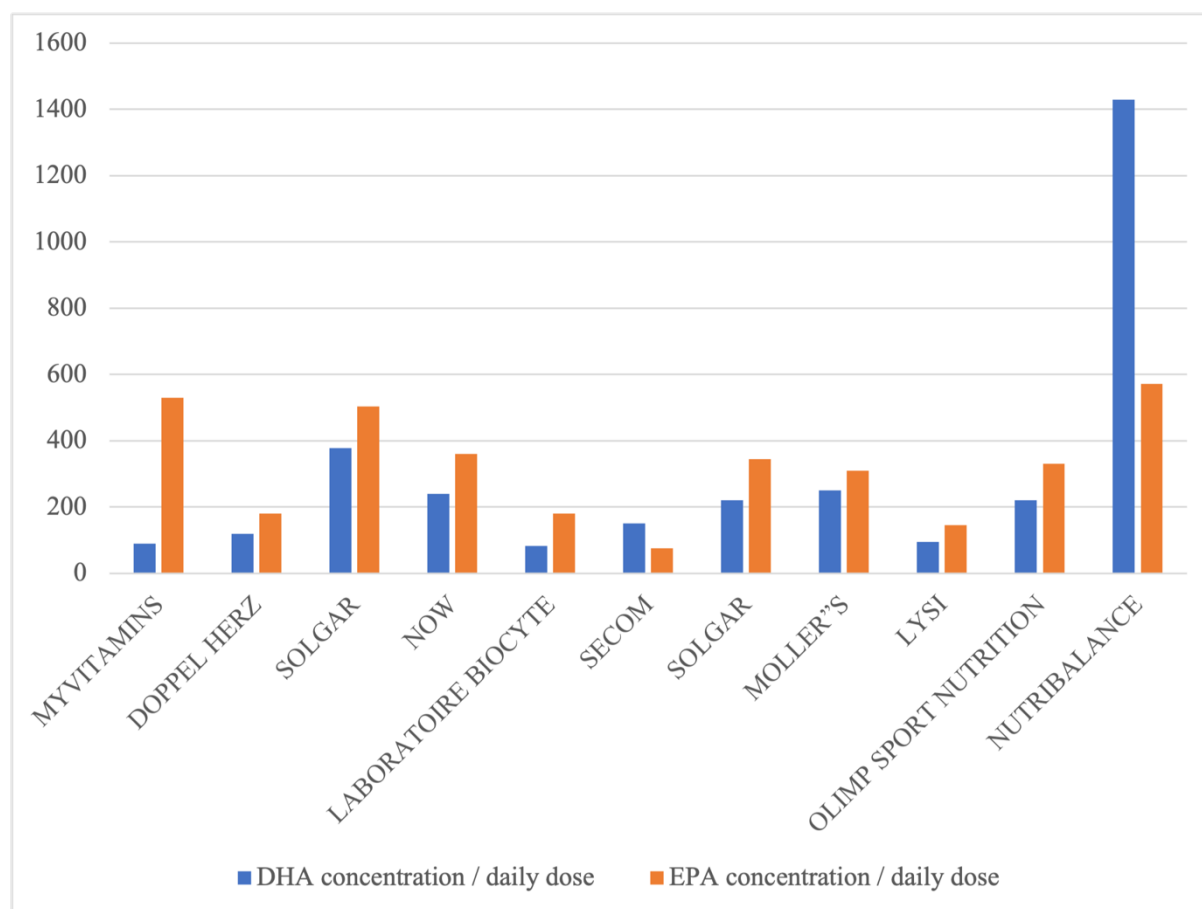


Figure 3. Concentration analysis of DHA and EPA for daily doses

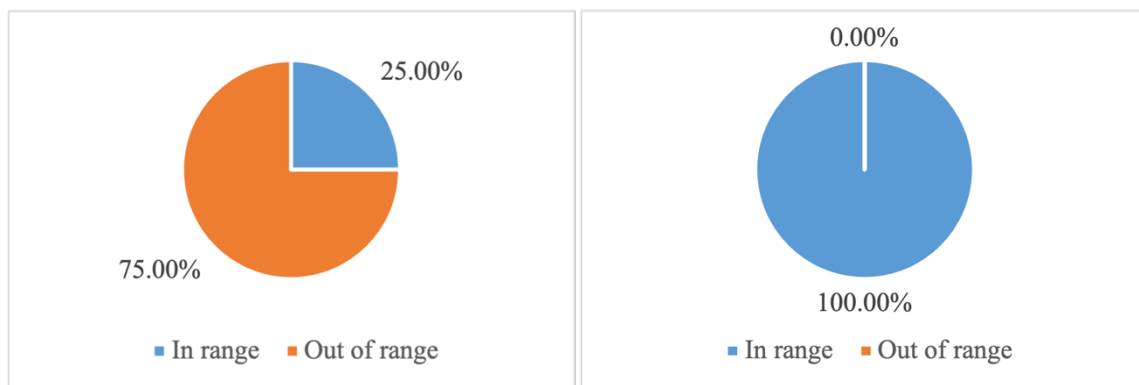


Figure 4. Distribution of participants in whom the presence of mercury was identified - initial and final

Calculation for **mercury** before and after following the custom protocol:

- Result: Statistics = 9.200, $p = 0.000000000000005531467$
- #Average Hg initial: **7,287**
- #Average Hg final: **3.638**

Following the implementation of the customized protocol, a reduction in blood mercury levels was observed.

The p value < 0.05 rejects the null hypothesis, which means that the mean change is statistically significant.

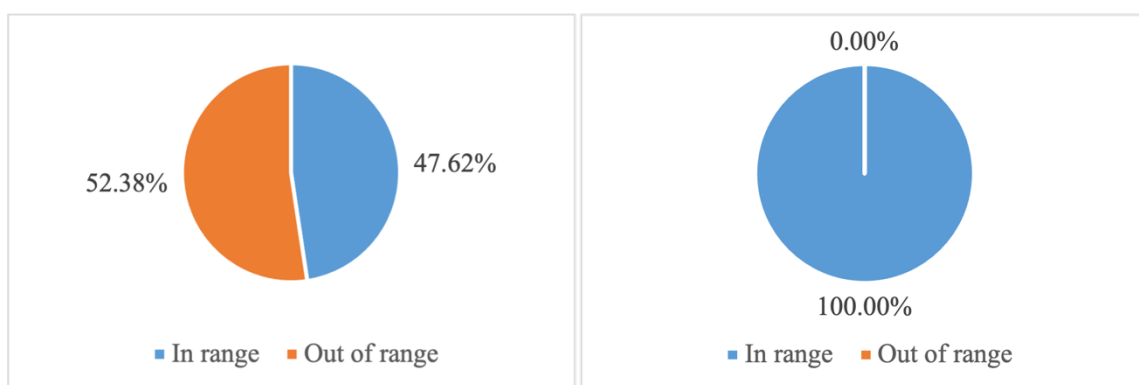


Figure 5. Distribution of participants in whom the presence of aluminium was identified - initial and final

Calculation for **aluminium** before and after following the custom protocol:

- Result: Statistics = 5.995, $p = 0.000000006390336422749$
- #Average Al initial: **10,625**
- #Average Al final: **5,586**

Following the implementation of the customized protocol, a reduction in blood aluminium levels was observed.

The value $p < 0.05$ rejects the null hypothesis, which means that the change in mean is statistically significant.

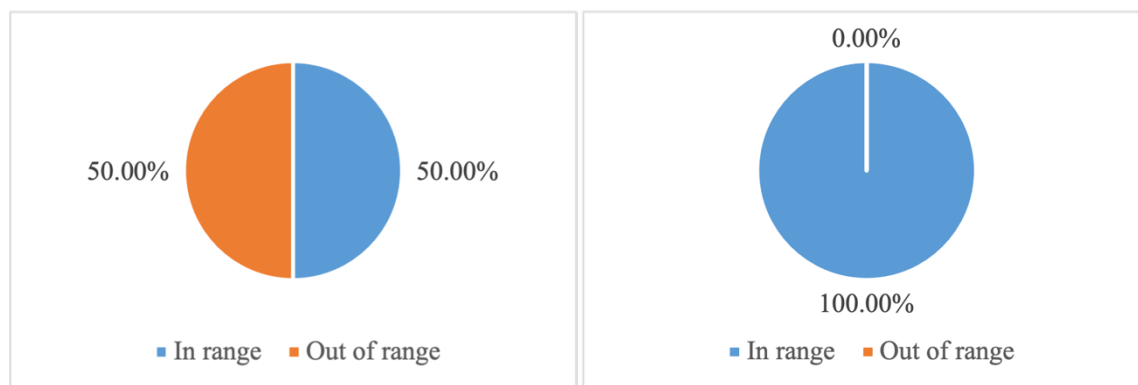


Figure 6. Distribution of participants in whom the presence of arsenic was identified – initial and final

Calculation of **arsenic** before and after following the custom protocol

Result: Statistics = 4.198, $p = 0.00007236442789135302$

#Average As initial: **2,580**

#Average As final: **1,690**

Following the implementation of the customized protocol, a reduction in blood arsenic levels was observed.

The p value < 0.05 rejects the null hypothesis, which means that the mean change is statistically significant.

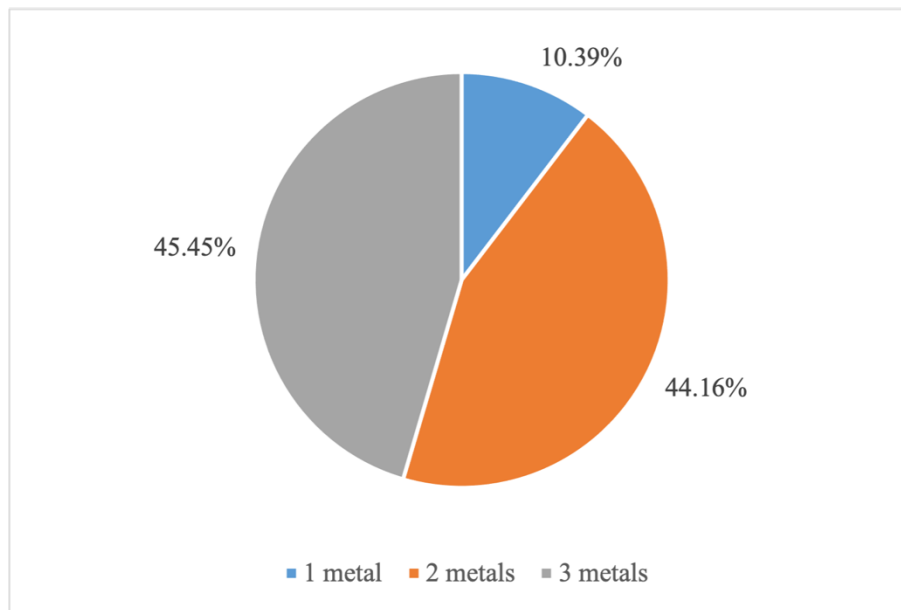


Figure 7. Distribution of participants according to the number of heavy metals present – before

The correlation matrix was also used to determine which parameters are correlated and what the direction of the correlation may be:

It is observed that there is a correlation between the consumption of foods with a high content of omega 3 and the concentration of omega 3 in the blood. It is a positive correlation, which means that the introduction of foods with a significant intake of omega 3 allows an increase in the concentration of omega 3 of the blood.

Also, the consumption of foods high in omega 6 is inversely related to the concentration of omega 3 in the blood, meaning that as the consumption of foods rich in omega 6 increases, the concentration of omega 3 in the blood decreases.

	initial omega 6 foods	final omega 6 foods	initial omega 6 omega 3 result	final omega 6 omega 3 result	initial omega 3 foods initial	final omega 3 foods	initial omega 3 result	final omega 3 result	initial antioxidant foods	final antioxidant foods	initial metal number
initial omega 6 foods	100%	15%	-5%	6%	-40%	-37%	-8%	-3%	-49%	-31%	-4%
final omega 6 foods	15%	100%	-28%	-12%	9%	-21%	-3%	-4%	7%	-19%	-14%
initial omega 6 omega 3 result	-5%	-28%	100%	59%	-26%	-8%	-60%	-47%	-18%	-15%	-5%
final omega 6 omega 3 result	6%	-12%	59%	100%	-17%	1%	-41%	-62%	-14%	-11%	-1%
initial omega 3 foods initial	-40%	9%	-26%	-17%	100%	10%	26%	26%	85%	16%	9%
final omega 3 foods	-37%	-21%	-8%	1%	10%	100%	-6%	0%	16%	85%	6%
initial omega 3 result	-8%	-3%	-60%	-41%	26%	-6%	100%	51%	19%	6%	13%
final omega 3 result	-3%	-4%	-47%	-62%	26%	0%	51%	100%	21%	14%	12%
initial antioxidant foods	-49%	7%	-18%	-14%	85%	16%	19%	21%	100%	14%	-2%
final antioxidant foods	-31%	-19%	-15%	-11%	16%	85%	6%	14%	14%	100%	14%
initial metal number	-4%	-14%	-5%	-1%	9%	6%	13%	12%	-2%	14%	100%

Figure 8. Corelation matrix

	omega 3 result before	omega 3 result after	omega 6 omega 3 result before	omega 6 omega 3 result after	AI before	AI after	As before	As after	Hg before	Hg after	predisposition to attention deficit after	reduced ability to focus after	cardiovascular diseases before	development of nervous tics before	development of nervous tics after	change in visual acuity before
omega 3 result before	100%															
omega 3 result after	51%	100%														
omega 6 omega 3 result before	-60%	-47%	100%													
omega 6 omega 3 result after	-41%	-62%	59%	100%												
AI before	27%	18%	-22%	-4%	100%											
AI after	29%	12%	-15%	6%	86%	100%										
As before	25%	8%	-19%	-10%	19%	13%	100%									
As after	25%	7%	-16%	-10%	17%	11%	98%	100%								
Hg before	-32%	-29%	44%	56%	-9%	7%	-5%	-5%	100%							
Hg after	-31%	-26%	42%	52%	-8%	9%	-2%	-2%	99%	100%						
predisposition to attention deficit after	-11%	-8%	21%	-5%	-6%	1%	-12%	-10%	21%	18%	100%					
reduced ability to focus after	-11%	-8%	21%	-5%	-6%	1%	-12%	-10%	21%	18%	100%	100%				
cardiovascular diseases before	17%	11%	-10%	5%	29%	30%	79%	75%	10%	14%	-18%	-18%	100%			
development of nervous tics before	-26%	-14%	36%	18%	-31%	-15%	-15%	-14%	27%	25%	14%	14%	-14%	100%		
development of nervous tics after	3%	9%	-4%	-9%	2%	-1%	7%	8%	3%	2%	-3%	-3%	19%	6%	100%	
change in visual acuity before	-27%	-11%	34%	15%	-28%	-14%	-16%	-15%	27%	26%	15%	15%	-19%	96%	6%	100%

Figure 9. Correlation matrix

Analyzing the correlation coefficients between the different considered variables we can draw some extremely useful conclusions:

- the initial omega 3 result is strongly correlated with the initial omega 6 result, but also with the final omega 6 result, the correlations being negative (-60%, respectively -41%). This tells us that a person who has a high omega 3 concentration will have a low omega 6 concentration.
- another negative correlation is found between the omega 3 result and mercury, which tells us that people with a high concentration of omega 3 tend to have a low concentration of mercury
- we find an extremely strong, positive correlation between the concentration of mercury and the result of the omega 6: omega 3 ratio, both in the initial and in the final stage, which suggests an increase in the concentration of mercury in people who have high values for the omega 6: omega 3 ratio
- one of the strongest positive correlations is found at the level of two variables: arsenic and cardiovascular diseases, which indicates the close connection and impact of arsenic on visual ability. A high blood concentration of arsenic strongly impacts visual acuity.
- at the same time, there is a strong and dependent correlation between the visual acuity variable and the development of nervous tics; practically they evolve in the same direction.
- another important correlation is that between the specific values for the omega 6: omega 3 ratio and the development of nervous tics. The two variables are positively correlated, which means that for an increase in the specific values for the omega 6: omega 3 ratio, it is very likely for the patient to develop nervous tics.

CONCLUSIONS

This study highlights that the chelation of heavy metals process of is a complex one, which implies the presence of a complementarity between food therapy and that of nutritional supplements, avoiding a single direction approach, because results can be obtained for a short period of time or insignificant.

- the presence of heavy metals (mercury, aluminium, arsenic) can be associated with an unbalanced eating behaviour, with the tendency to eat foods with a high sugar content, refined carbohydrates, unhealthy fats, thus easily inducing increases in acidity omega 6 fats, implicitly increases in specific values for the omega 6: omega 3 ratio
- the analyzed subjects do not tend to constantly introduce foods with a beneficial role, they are accepted only if they are imposed, in the first stage
- increased omega 6: omega 3 ratio and high omega 3 deficiencies (identified in the initial testing stage) reflect nutritional imbalances
- the decrease in the specific values for the omega 6: omega 3 ratio and the significant increases in omega 3 (identified in the final stage of testing) reflect the beneficial impact of nutrition on health, the reduction of the risks associated with the initial values
- the increase in the concentration of omega 3, the reduction of the specific values for the omega 6: omega 3 ratio can support the chelation process
- the heavy metal chelation process is a complex one, which requires the presence of a complementarity between the therapy with food supplements and the nutritional one, avoiding a single direction approach, because results can be obtained for a short period of time or insignificant
- the maintenance stage is important: avoiding foods with intoxication potential, transforming this 120-day period into a lifestyle

DISCUSSIONS

Both in Romania and worldwide, we have not acknowledged a research of this type (on the correlation between the presence of metals with the potential for intoxication, the lack of omega 3, the increase in specific values for the omega 6: omega 3 ratio and their associated symptoms) to have been carried out to date.

The international researches present in the specialized literature at this moment have been carried out on topics collateral to the theme of our study and also demonstrate, as we assumed at the beginning of the study, that there is a correlation between the presence of potentially toxic metals, omega deficiency 3 and their associated symptoms.

A study addressing the correlation between low levels of omega 3 fatty acids and neuropsychiatric disorders is the review by Thomas Larrieu and Sophie Layé (2018) [21]. In this bibliographic study the topic analyzed was related to the effects of food on mood and the relevance of omega 3 fatty acids in depression and anxiety. The reviewed papers indicated that low intake of omega-3 fatty acids may predispose certain individuals to depression and anxiety and that the introduction of dietary supplements based on long-chain omega-3 fatty acids represent an interesting strategy for the prevention or treatment of depression and anxiety disorders in certain individuals. This review reinforces the idea of the utility of omega 3 fatty acids in the daily diet as a beneficial tool for designing and testing new non-pharmacological strategies in the treatment of neuropsychiatric disorders such as mood disorders.

Regarding combating heavy metal poisoning with omega 3 fatty acids, a rat study [25] supports the claim that omega 3 fatty acids could mitigate the adverse effects of heavy metal poisoning by improving intestinal gland cell line survival rates (IEC 6). The results also suggest that omega 3 fatty acids protected IEC 6 cells from heavy metal damage, leading to the conclusion that dietary omega 3 fatty acid supplementation is a promising therapeutic practice to mitigate heavy metal damage. Also, in another study on cadmium neurotoxicity [26] it was shown that omega 3 fatty acids act as an antioxidant with neuroprotective impact and treatment against cadmium toxicity, and polyunsaturated fatty acid supplementation is more useful in co-treatment than in pre-treatment.

Other collateral studies on the effect of omega 3 fatty acids on health status are reviewed in the bibliographic study carried out by Peter Van Dael [27]. It details aspects of expert recommendations regarding dietary supplementation with omega 3 fatty acid

supplements that may beneficially impact certain health issues. For example, expert recommendations generally support a beneficial effect of omega 3 fatty acids on cardiovascular health and recommend a daily intake of 500 mg of DHA and EPA or 1-2 servings of fish per week. Other expert recommendations for treating depression include 200–300 mg/day; up to 1–2 g omega 3 fatty acids/day for major depressive disorder. Other studies in this review support a beneficial role for omega 3 fatty acids in reducing the risk of preterm birth, with a daily intake of 600-800 mg of DHA during pregnancy. In conclusion, the available scientific studies support that dietary recommendations for omega 3 fatty acids should be established for the general population and for subjects with specific physiological conditions.

DEVELOPMENT PERSPECTIVES, CONTINUATION OF THE RESEARCH STUDY

Collaborative partnerships July – December 2022

- Palazzo Fiuggi – under the coordination of Prof. Dr. David Della Morte (July – September 2022, with the possibility of extension)
- Viva Mayr – under the coordination of Prof. Dr. Maximilian Schubert (September – October 2022)
- Life Co – starting in November 2022

Shaping an education on the value of food in physical, emotional and intellectual health (online programs, nutrition courses in schools, kindergartens, corporations, writing and publishing information documents – articles, brochures, books)

Information campaign, awareness of the concept of medical nutrition, which isn't becoming a measure of the amount of food and extra pounds but is related to the individual needs of the modern man, determining the state of health.

ORIGINAL CONTRIBUTIONS

- The most used omega 3 supplements by the study participants, sold in the geographical area of Romania, were evaluated.
- An omega 3 supplement with a higher concentration of DHA compared to EPA (1429 mg DHA, 571 mg EPA/ 10 ml product) was developed and implemented in the individual protocol for the subjects of the study group, following the absorption at an individual level after 120 days of use.
- The dietary profile was evaluated and the risk factors (sources of contamination) in the usual diet were identified for a group of 77 subjects, who presented significant omega 3 deficiencies and major increases in specific values of the omega 6: omega 3 ratio.
- Nutrition and chelation plans were developed and implemented for each subject for a period of 120 days, laying the foundations of an integrated, personalized concept, related to the individual needs of each study participant.
- The correlation between consumption of certain foods and exposure to heavy metals as well as other toxic substances has been realized.
- The correlation between nutritional status, antioxidant foods and the severity of exposure to heavy metals, food additives, pesticides, genetically modified organisms, antibiotics, growth hormones was realized.

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A. Anuțoiu (Stoica), D. M. Cheța, « Detoxification therapy and diet plan for mercury, aluminium, arsenic poisoning », Romanian Medical Journal, vol. LXVIII, No 3, 2021.

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A. Anuțoiu (Stoica), D. M. Cheța, « Digestive disorders induced by the accumulation of arsenic in the human body », Romanian Journal of Medical Practice, vol. XVI, No 3 (80), 2021.

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A. Anuțoiu (Stoica), V. Anăstăsoaie, D. M. Cheța, « Correlation between the presence of metals with potential for intoxication, omega 3 deficiency, increased omega 6: omega 3 ratio and their associated symptoms », Romanian Medical Journal, vol. 69, No 2, 2022.

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