EUROPEAN Journal of Medicine

Has been issued since 2013.
E-ISSN 2310-3434
2018. 6(1). Issued 2 times a year

Journal is indexed by: **CAS Source Index** (USA), **CiteFactor** (Canada), **CrossRef** (UK), **EBSCOhost Electronic Jornals Service** (USA), **Electronic scientific library** (Russia), **Open Academic Journals Index** (Russia), **ResearchBib** (Japan), **Sherpa Romeo** (Spain).

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Website: http://ejournal5.com/
E-mail: ejm2013@mail.ru

Founder and Editor: Academic Publishing House Researcher s.r.o.

Release date 16.03.18.
Format 21 × 29,7/4.

Headset Georgia.

Order № 17.

European Journal of Medicine

2018

Is. 1

C O N T E N T S

Articles and Statements

Published in the Slovak Republic
European Journal of Medicine
Has been issued since 2013.
E-ISSN: 2310-3434
2018, 6(1): 3-12

DOI: 10.13187/ejm.2018.1.3
www.ejournal5.com

Articles and Statements

Water Electrolysis-Processes in Catholyte and Anolyte Results with Differential Non-Equilibrium Water Spectrum

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Abstract

A two stage model of the physicochemical processes at the electrolysis of pure water is proposed. The presence of nascent hydrogen in the catholyte and nascent oxygen in the anolyte during the first stage explains the antioxidant properties of the catholyte and the strong biocidal action of the anolyte. In the second stage the nascent hydrogen and oxygen are combined into hydrogen and oxygen molecules, respectively. The comparison between their average energies with the average energy of the control sample of water shows an increase in the average energy of the catholyte and decrease in the average energy of the anolyte. This indicates that some changes in the structure of the activated water have occurred.

There are results with method of Differential Non-equilibrium Water Spectrum (DNES) for alteration of energy of hydrogen bonds in catholyte and anolyte according control sample of water.

Keywords: electrochemically activated water (ECAW), catholyte, anolyte, nascent hydrogen, nascent oxygen, water spectrum DNES, energy of hydrogen bonds.

1. Introduction

Water is the main factor of the life on our planet. Even more, according to one of the co-authors of this paper the life has started in the warm thermal springs on the earth (Ignatov, Mosin, 2010). Water regulates the vital processes in the living things actively participating in the metabolism and their adaptation to the environment. That's why every change of its composition and structure influences the live matter either aiding its development and stability or on contrary provoking its destruction. It seems strange that despite such enormous importance scientists from different countries have seriously started paying attention and investigating the unusual properties of the water put to different kind of influence only in the last decades. In this direction a great attention was paid to its electrochemical activation (Bakhir, 1999; Kloss, 1988; Petrushenko, Lobyshev, 2004; Prilutsky, Bakhir, 1997; Zenin, 1999). Notwithstanding the easy description of the chemical processes the explanation of the obtained properties of the alkaline (catholyte) and acidic (anolyte) water solutions are still not fully convincing (Ball, 2008). Even more, in the majority of the existing descriptions and experiments NaCl has been used which gives a satisfactory

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explanation of the biocidal properties of the anolyte but does not work in case of pure water which obtains same properties. The explanation probably relates not only to the chemical changes in the water composition but also to the changes of its structure.

The main questions that researchers have to answer are as follows.

1. What physical and chemical changes occur in the water during its electrolysis?

This question should be investigated in two aspects:

a) pure water is processed;

b) NaCl or other minerals are added to the water solution.

2. What parameters of the activated water play a major role for the explanation of its unusual properties?

3. What the mechanism of the action of the activated water on the living things is?

In this paper attention is paid to the question 1a) i.e. the activation of water without additional mineralization, free of ions of other elements. A model of the corresponding physicochemical processes is proposed.

Every model has to describe satisfactory the observed effects of the modeled process. In our case the known effects of the activated water (catholyte and anolyte) reported by different investigators concern especially its influence on the living things. An explanation for the described effects could be the increased presence of both molecular hydrogen (H_2) in the catholyte solution and molecular oxygen (O_2) in the anolyte solution. But such an increase can be achieved through saturation of the water solution with these gases as well. It is true that in this case the water properties will be changed but without same effects. It is reasonable to assume that the increased concentration of OH^-ions in the catholyte as well as H^+ ions in the anolyte cause such an action. Therefore, one may conclude **that the action of the direct current on distilled or de-ionized water leads to changes in its chemical composition and structure different for the catholyte and anolyte.**

Electrolysis of pure water.

According to the modern notion the following reaction takes place at the cathode :

$$2H^+ + 2e^- \rightarrow H_2\ \hat{I} \quad (1)$$

The analogous reaction at the anode is as follows:

$$2O^{2-} - 2e^- \rightarrow O_2\ \hat{I} \quad (2)$$

Actually, the above reactions run in two stages.

At the cathode:

First stage $H^+ + e^- \rightarrow H^*$ (3)

Second stage $H^* + H^* \rightarrow H_2$ (4)

At the anode:

First stage $OH \rightarrow O^* + 2e^- + H^+$ (5)

Second stage $O^* + O^* \rightarrow O_2$ (6)

Therefore, according to formula (3) $\mathbf{H^*}$ – atomic hydrogen is produced at the cathode during the first stage. It is called also nascent hydrogen and possesses high reactivity. Atomic oxygen $\mathbf{O^*}$ called also nascent oxygen is produced at the anode, and it is highly reactive as well. These atoms can react between them according to formulae (4) and (6), and the obtained hydrogen and oxygen molecules can be either separated from the solution and go to the air or remain dissolved in the water. If there are molecules of hydrogen and oxygen at the cathode and the anode obtained according to formulate

(4) and (6) they can react with the nascent hydrogen and oxygen respectively as follows:

$$4H^* + O_2 \rightarrow 2H_2O \quad (7)$$

$$O^* + H_2 \rightarrow H_2O \quad (8)$$

and water molecules will be produced.

2. Materials and Methods

2.1. Electrolysis apparatus for catholyte and anolyte

The experiments were conducted with the diaphragm electrolysis apparatus “Wasserionisierer Hybrid PWI 2100”, equipped with four titanium electrodes coated with platinum. The voltage of the electric power supply – 220 V, the frequency of the electric current – 50 Hz, the power of the electric current 0.2–0.7 A; the time of electro processing – 30–40 min; the volume of the electroactivated water: anolyte – 0.3 l; catholyte – 0.9 l; power consumption – 70 Watts.

The electrolysis cell was formed by two electrodes – a positively charged anode and a negatively charged cathode connected to different poles to a DC source. Interelectrode space was filled with water, which is an electrolyte capable of conducting the electrical current.

The anolyte had pH = 3.2 and ORP = +1070 mV;

The catholyte had pH = 9.0 and ORP = -300...-500 mV);

2.2. NES and DNES Spectral Analysis

The device for DNES spectral analysis was made by A. Antonov on an optical principle. For this was used a hermetic camera for evaporation of water drops under stable temperature (+22–24 °C) conditions. The water drops were placed on a water-proof transparent pad, which consists of thin maylar folio and a glass plate. The light was monochromatic with filter for yellow color with wavelength at λ = 580±7 nm. The device measures the angle of evaporation of water drops from 72.3° to 0°. The DNES-spectrum was measured in the range of -0.08– -0.1387 eV or λ = 8.9–13.8 μm using a specially designed computer program. The main estimation criterion in these studies was the average energy ($\Delta E_{H...O}$) of hydrogen O...H-bonds among H_2O molecules in water samples and human blood serum.

3. Results and Discussion

3.1. Nascent hydrogen in catholyte and nascent oxygen in anolyte

It is supposed in the proposed model that nascent hydrogen and nascent oxygen are produced in the catholyte and anolyte respectively. Their presence is substantial for the processes that may occur in the above solutions. The most probable assumption is that they will take part in reactions (4) and (6) or for example (7) and (8). Another possibility for them is to be stabilized in small quantity. It has to be underlined that the stabilization possibility depends on the probability to encounter corresponding re-agents from the above mentioned reactions. If this probability is small there is a possibility other molecules that can not react with them to play the role of stabilizers. For example, the water molecules due to their dipole character allowing for the formation of dimmers, thrimmers and larger structures could play such a role. In that case stabilized atoms of nascent hydrogen will remain in the catholyte, and stabilized atoms of nascent oxygen will remain in the anolyte. Since the nascent hydrogen is an active reducer it will attack admixtures that could be reduced when the catholyte is blended with another medium. This could explain the catholyte action as a strong antioxidant (Hanaoka, 2001; Kokichi et al., 2004). Probably, this could explain its healthy influence in case of different stress-related or due to the action of free radicals deceases. Such influence has been observed by many researchers (Hayashi, Kawamura, 2002; Komatsu, 2001; Lee et al., 2006; Sanetaka Shirahata et al., 2012; Yahagi et al., 2000; Ye, et al., 2008; Ye et al., 2004).

In a similar way the anolyte put in another medium will demonstrate its strong oxidative property and will oxidize all present organic admixtures and will destroy microorganisms, bacteria and viruses (Gluhchev et al., 2015; Karadzov et al., 2014; Kirkpatrick, 2009; Kumar et al., 1999; Miroshnikov, 2002; Suzuki et al., 2002; Tanaka et al., 1996; Zinkevich et al., 2000), which makes it an excellent biocidal, disinfection and harmless for the people and the environment mean.

3.2. Differential Non-equilibrium spectrum NES and DNES

A convenient method for studying of liquids is non-equilibrium differential spectrum. It was established experimentally that the process of evaporation of water drops, the wetting angle θ decreases discreetly to zero, and the diameter of the water drop basis is only slightly altered, that is a new physical effect (Antonov, 1995; Antonov, Yuskesselieva, 1983). Based on this effect, by means of the measurement of the wetting angle within equal intervals of time is determined the function of distribution of H_2O molecules according to the value of f(θ). The distribution function is denoted as the energy spectrum of the water state. The theoretical research established the dependence between the surface tension of water and the energy of hydrogen bonds among individual H_2O-molecules (Antonov, 1995).

For calculation of the function f(E) represented the energy spectrum of water, the experimental dependence between the wetting angle (θ) and the energy of hydrogen bonds (E) is established:

$$f(E) = \frac{14{,}33 f(\theta)}{[1-(1+bE)^2]^2} \qquad (9)$$

where $b = 14.33$ eV^{-1}

The relation between the wetting angle (θ) and the energy (E) of the hydrogen bonds between H_2O molecules is calculated by the formula:

$$\theta = arcos\ (-1 - 14.33E) \qquad (10)$$

The energy spectrum of water is characterized by a non-equilibrium process of water droplets evaporation, therefore, the term non-equilibrium spectrum (NES) of water is used.

The difference *Δf(E) = f (Esamples of water) – f (Econtrol sample of water)* – is called the "differential non-equilibrium energy spectrum of water" (DNES).

Thus, the DNES spectrum is an indicator of structural changes in water, because the energy of hydrogen bonds in water samples differ due to the different number of hydrogen bonds in water samples, which may result from the fact that different waters have different structures and composition and various intermolecular interactions – various associative elements etc (Ignatov et al., 2014; Ignatov et al., 2015). The redistribution of H_2O molecules in water samples according to the energy is a statistical process of dynamics.

Figure 1 shows the average NES-spectrum of deionised water. On the X-axis are depicted three scales. The energies of hydrogen bonds among H_2O molecules are calculated in eV. On the Y-axis is depicted the function of distribution of H_2O molecules according to energies f(E), measured in reciprocal unit eV^{-1}. Arrow A designates the energy of hydrogen bonds among H_2O molecules, which is accepted as most reliable in spectroscopy. Arrow B designates the energy of hydrogen bonds among H_2O molecules the value of which is calculated as:

$$\bar{E} = -\ 0.1067 \pm 0.0011\ \text{eV} \qquad (11)$$

Arrow C designates the energy at which the thermal radiation of the human body, considered like an absolute black body (ABB) with a temperature +36.6 °C, is at its maximum.

A horizontal arrow designates the window of transparency of the Earth atmosphere for the electromagnetic radiation in the middle infrared range of the Sun toward the Earth and from the Earth toward the surrounding space. It can be seen that the atmosphere window of transparency almost covers the NES-spectrum of water.

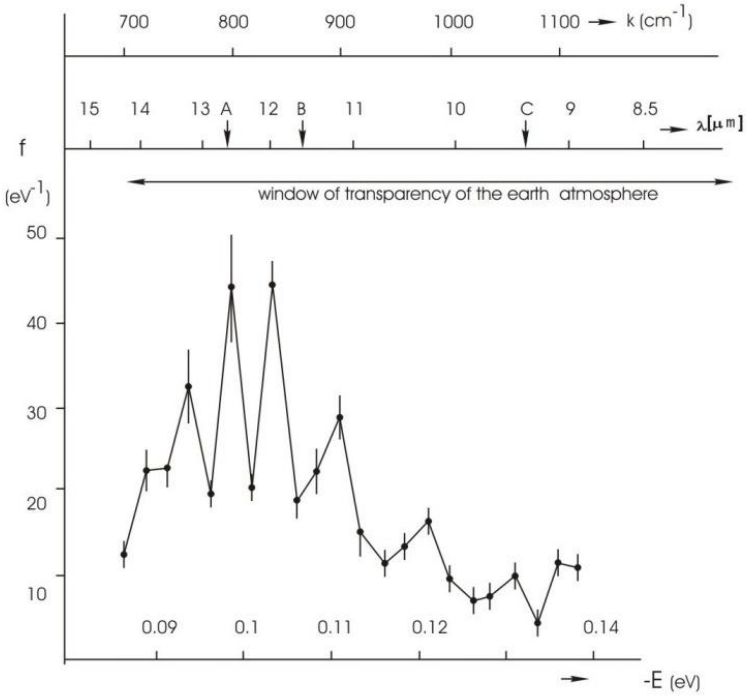

Fig. 1. The NES-spectrum of deionized water (chemical purity – 99.99 %; pH – 6,5–7,5; total mineralization – 200 mg/l; electric conductivity – 10 μS/cm): the horizontal axis shows the energy of the H...O hydrogen bonds in the associates – E (eV); the vertical axis – the energy distribution function – f (eV^{-1}); *k* – the vibration frequency of the H–O–H atoms (cm^{-1}); λ – wavelength (μm)

The presence of nascent hydrogen and nascent oxygen in the hydrolyzed water could produce changes in its state. Indeed, using the method NES (Antonov, 1995; Ignatov, Mosin, 2014) as a measure of the energy spectrum of the water stage a useful information could be obtained about the structural changes in water and the average energy of hydrogen bonds among individual H_2O molecules in samples. It was experimentally established (Ignatov, Mosin, 2014) that the surface pressure was increased in the catholyte and more molecules were included in a unit volume. The average energy E of the hydrogen bonds H...O between water molecules H_2O, measured for the catholyte and anolyte, accordingly when a de-ionized water is used is E = -0.1293 eV for the catholyte and E = -1221 eV for the anolyte. The difference $\Delta E_{H...O}$ between the average energy of the control sample of water E = -0.1251 eV, evaluated by the method DNES (Ignatov, Mosin, 2014) and the average energy of the catholyte and anolyte is respectively ΔE= -0.0042 ± 0.0011 eV and ΔE= 0.003 ± 0.0011 eV.

4. Discussion and Conclusions

A two-stage model describing physicochemical processes stemming from the electrolysis of pure water is suggested in the paper. The production of nascent hydrogen and nascent oxygen during the first stage is used as a basic assumption. The enriched concentration of these components in the solutions explains the antioxidant action of the catholyte the strong biocidal effect of the anolyte. The observed difference in the average energy and hydrogen bonds between catholyte and anolyte is an indication of structural changes that have taken place in the activated water. The development of more general model describing the physicochemical processes and analysis of the content and structure of activated water in case of electrolysis of weak water mineralization is a subject of future work.

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Published in the Slovak Republic
European Journal of Medicine
Has been issued since 2013.
E-ISSN: 2310-3434
2018, 6(1): 13-19

DOI: 10.13187/ejm.2018.1.13
www.ejournal5.com

Correlation between Lead Serum Level and Total Immunoglobulin E(Ige) Level in School-Aged Children

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Abstract

Correlation between lead serum level and total Immunoglobulin E (IgE) level in school-aged children. Gusmira Y.H., Irsa, L., Lubis, B., Evalina, R., Lubis, M. Lead poisoning may cause various health problems including immune system. Research objective is to determine the correlation between serum lead levels with total IgE level in school-aged children. A cross sectional study had been conducted in Juli 2015 to children aged 9-12 years old in an elementary school industrial area with lead exposure, Medan, Sumatera Utara. Children with atopy and worm infection were excluded. Lead serum level and total immunoglobulin E level were obtained. Data analysis to see correlation between blood lead level and total IgE level was used Pearson correlation test (r) if normal data distribution and Spearman test if abnormal data distribution with significance level P <0.05. From 42 subjects, 27 were boys and 15 girls, the mean age was 10.3 year (SD 1.09), 18 subjects with medium risk of allergic. The mean serum lead level was 2.57 µg/dL (SD 0.58, 95 %CI 2.42-2.74), the mean IgE total level was 1155 IU/L (SD 3340, 95%CI114.43-2196.22). From the Spearman test we obtained r= 0.023, p=0.887. It can be concluded that here was an increase in total IgE levels and blood lead levels but statistically no correlation between lead serum level and total IgE level among school aged children.

Keywords: lead serum level, total immunoglobulin E, children.

1. Introduction

Plumbum or better known as lead is one of heavy metals that can cause health problems including disturbances of hematology, nervous system, cardiovascular, kidney, endocrine, and immunology (Lubis et al., 2003; Farias et al., 2014). The toxic effects in children can arise from the lowest level of lead concentration at 5 µg/dL to the lethal concentration at about 150 µg/dL. According to the Center for Disease Control and Prevention (CDC) the toxic effect of lead can be observed at the serum level of 5 µg/dL (Farias et al., 2014; Mener et al., 2014).

One of the effects of lead exposure is impairment of immune system. The immune system is particularly sensitive to lead poisoning. Lead is immunotoxic even at a very low level (Mener et al., 2014). Lead will shift T-helper 1 (Th1) dependent immunity toward T-helper 2 (Th2) dependent immunity resulting in increasing allergic sensitization through increasing production of interleukins, cytokines, and immunoglobulin E (IgE) (Mener et al., 2014). An increase in IgE level will stimulate allergic reactions. This may be related to increased prevalence of allergies in recent

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years. Data from the World Allergy Organization (WAO) 2013 stated that the prevalence of allergy is increasing worldwide by the rate of 10-40 %. Epidemiological studies show that indoor and outdoor pollution affect respiratory health, including an increased prevalence of asthma and allergic diseases (Pawankar et al., 2013). The impact of allergies is the decreased quality of life, increased medical costs, impaired concentration, growth and development (Sudewi et al., 2009). Previous research conducted in Medan, North Sumatera, reported that air lead levels in industrial areas in Medan had exceeded the threshold value from the government which was 2µg/m (Mener et al., 2014; Pawankar et al., 2013; Sudewi et al., 2009; Girsang, 2008). The objective of this study is to determine the correlation between blood lead levels and total immunoglobulin E level in primary school children.

2. Materials and methods

This study was a cross sectional study determining the correlation between blood lead level and total IgE level in elementary school age children in Medan. The research was conducted at Al Washliyah Elementary School Timbang Deli Village, Medan Amplas district, North Sumatra in June 2015. Sample size was calculated using sample size formula for correlation study. Sample were obtained consecutive sampling method and consisted of 4th to 6th grader elementary school students. The inclusion criteria were children aged 9-12 years old and exclusion criteria are children with atopy or allergies and infected with a worm. The samples were given questionnaires for baseline characteristic and risk of atopy and examination of lead level, total IgE level, skin prick test (SPT) and stool examination. Risk of atopy was determined using Indonesian Pediatric Allergy Immunology Working Grup trace card scoring system. Blood is taken using a vacutainer as much as ± 6cc in venous area of cubiti. Then divided into 3cc inserted into tubes that already contain EDTA (preservative Ethylene Diamine Tetraacetic Acid) for examination of lead and 3cc are inserted into empty tubes for examination of total IgE. The study profile can be seen in Fig. 1.

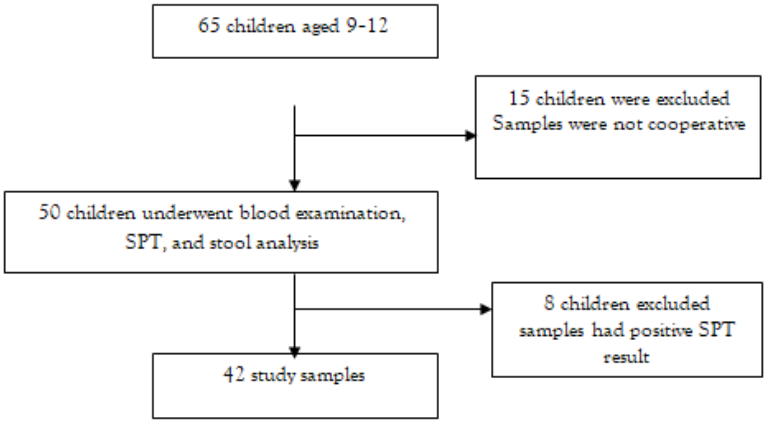

Fig. 1. Study Profile

The serum lead concentration was evaluated with Atomic Absorption Spectrophotometer (AAS) in µg/dL units, with equation (1).

$$\frac{1000}{\text{sample weight}} \text{x} \frac{50}{1000} \text{x AAS concentration (mg/L)} \quad (1)$$

Total IgE level was examined using Mini Vidas tool with ELFA (Enzyme Linked Fluorescence Immuno Assay) method. The normal value of total IgE level is <150 IU/ml.

Data was analyzed using Statistical Package for the Social Sciences software (SPSS). Bivariate analysis was performed using chi-square. Data analysis to see correlation between blood lead level and total IgE level was used Pearson correlation test (r) if normal data distribution and Spearman test if abnormal data distribution with significance level P <0.05.

3. Results

We obtained of 42 elementary school children aged 9-12 years who met the criteria of inclusion and exclusion. The baseline characteristics are shown in Table 1.

Table 1. Baseline characteristics

Characteristics	N
Sex, n(%)	
Female	15 (36)
Male	27 (64)
Nutritional status, n(%) Good nutritional status Malnourished	 29 (69) 13 (31)
Atopy risk, n (%)	
Low risk	24 (57)
Medium risk	18 (43)
IgE Level, n (%) High (≥150 IU/ml) Normal (<150 IU/ml)	26 (62) 16 (38)
Lead level, mean (SD), µg/dl ≥2,57 µg/dl < 2.57 µg/dl	2,57 (0,58) 23 (55) 19 (45)
Age, year, mean (SD)	10.3 (1.09)
Body weight, kg, mean (SD)	27.79 (7.57)
Body height, cm, mean (SD)	132.33 (8.89)
BW/BH, %, mean (SD)	95.91 (15.04)

Of 42 samples obtained, 64 % were male with an average age of 10.3 years. The mean height and weight were 27.79 kg and 132.33 cm, respectively. Most samples had good nutritional status with average of weight for height of 95.91 %. More than half of the samples (57 %) had a low atopy risk and 62 % of the samples had total immunoglobulin E levels of >150 IU/ml. Fifty five percent of samples in this study had serum lead level of 2.58 µg/dl. Table 2 showed the factors affecting lead exposure in this study where painted house had a significant relationship with lead exposure in this study (p <0.01).

Table 2. Factors affecting lead esposure

	Lead level		P
	<2.57 µg/dl (n=19)	≥2.57 µg/dl (n=23)	
Sex Male Female	 14 5	 13 10	 0.25[a]
Age 9 years 10 years 11 years 12 years	 7 7 4 1	 7 6 7 3	 0.34[b]
Nutritional Status Malnutrition Good nutritional status	 7 12	 6 17	 0.45[a]
Total IgE level >150 IU/ml <150 IU/ml	 12 7	 14 9	 0.88[a]

House location			
Near main road / very crowded	6	3	0.06[b]
Big crowded road	6	5	
Less crowded road	7	15	
Painted house			
Yes	17	19	0.43[a]
No	2	4	
Painted part of the house			
Only the insides	7	1	0.01[a]
Both inside and outside	10	19	
Chipped paint			
Yes	11	9	0.23[a]
No	6	11	

[a] Chi square
[b] Mann witney

Table 3 showed that 33 % of children used tap water for drinking, but there was no significant relationship between drinking water sources and lead (p = 0.95). In terms of subject habits, we also did not find significant relationship with lead level. Table 4 showed that there was no correlation between lead level and total immunoglobulin E level with r = 0.023 (p = 0.087).

Table 3. The sources of lead exposure in terms of subject habits

	Lead levels		**p**
	≥2.57 µg/dl (n=23)	<2.57 µg/dl (n=19)	
Daily water source			
Tap water	14	12	0.95[a]
Well water	7	4	
Bottled water	2	3	
Canned food / drink consumption by family members			
Yes	4	1	0.24[b]
No	19	18	
Street foods consumption			
Yes	5	3	0.47[b]
No	18	16	
Nail biting, sucking fingers, biting pencil / pen habit			
Yes	7	6	0.94[c]
No	16	13	
Handwashing habit before/after meal			
Yes	14	9	0.26[a]
No	3	1	
Sometimes	6	9	

[a] Mann-Whitney
[b] Fisher Exact
[c] Chi Square

Table 4. The correlation of lead levels with total immunoglobulin E levels

Parameter	Mean	SD	r*	95%CI	P
Pb, µg/dl	2.57	0.58	0.023	2.42-2.74	0.887
IgE, IU/ml	1155.33	3340.27		114.43-2196.23	

*Spearman test

4. Discussion

We found no correlation between serum lead level and total IgE level in elementary school children in this study (p = 0.887, r = 0.023). This result is different from previous studies. Earlier experimental and epidemiological studies have shown that lead exposure is involved in the changing of humoral and cellular mediated immunity and developing allergies by increasing IgE, eosinophils and bronchial responses (Mishara, 2009; Sun et al., 2003; Wells et al., 2014; Min et al., 2008). A study in Cairo in 2011-2012 enrolling 200 children to determine the association between lead exposure and bronchial asthma, there was an association between lead level with total IgE in children aged 5-14 years, and the relationship between lead level and asthma severity was found (Mohammed et al., 2015).

A study conducted in the USA on 1430 children aged 2-12 years showed an association between blood lead level and total IgE level and reported that elevation of lead level at 1 µg/dl will increase IgE level by 11.1 % (Wells et al., 2014). A study on 279 children in Missouri found that blood lead levels was ranging from 1-45 µg/dL, and found an association between elevated blood lead level and elevated total IgE level (Lutz et al., 1999). Study in Kairo found a positive association between the serum lead and total IgE levels was statistically significant in subjects with *D. farinae* sensitization, which indicated that the immunologic effects of lead exposure may be greater in people with allergic sensitization (Kim et al., 2016). The difference from the results of this study may be due different geographic areas, lifestyles, dietary habits, genetic variation, exposure rates, duration, concentrations, and laboratory methods.

The mechanism of the relationship between lead exposure and total IgE elevation remains unclear. However, lead exposure will cause in IgE-mediated type1 hypersensitivity. Lead will increase Th2 immune response and inhibit Th1 response and also increase Th2 / Th1 cytokine ratio. The effect is dependent on the enhanced production of cytokines and interleukins (IL), thereby promoting isotype switching to IgE. Exposure to lead seems to be associated with atopic sensitization and modulation of several cytokines (e.g. IL-12, IL-10, interferon (IFN)-ɣ, and IL-4 production) and with T-cell dysregulation (Wells et al., 2014; Yang et al., 2014; Wang et al., 2017). Lead exposure during allergic sensitization may induced increased tracheal responsiveness, eosinophil count, and IL-4 concentration. Lead exposure has also result in a decrease in lymphocyte count, IFN-ɣ concentration and IFN-ɣ/IL-4 ratio (Wang et al., 2017).

Sixty two percent of samples in this study had total IgE levels above 150 IU/ml. Study in Taiwan found lead level were positively linked with serum IgE (β=0.26 kU/l per In-unit increase lead concentration (Wang et al., 2017). In accordance with the results of this study can be concluded that any increase in lead levels have positive correlation with increased levels of IgE. But in our study did not examine other factors that might cause elevated IgE levels.

In this study we found that school-aged children in Medan Amplas area had an average lead level of 2.57 µg/dl, with the lowest value of 1.48 µg/dl and the highest value was 3.99 µg/dl. There was no safe lead level observed in these children (*Centers for Disease Control and Prevention*, 2014). The lead level in children in elementary school was still within the threshold limit, which according to the Center for Disease Control and Prevention (CDC) is of 5 µg/dL (*Centers for Disease Control and Prevention*, 2014). We must to prevent further elevation of lead level in the serum of children in elementary school, where normally no lead levels are detected in the serum more attention is needed to reduce the effects of lead poisoning.

The location of this study was an industrial area where it had bus terminals and some large factories producing molding and building material components, liquor, fodder, snacks, and others. According to a 2008 study, Medan Amplas had the highest airborne lead content with the concentration of 32.67 µg/m^3 (Pawankar et al., 2013). This becomes one of the factors of lead exposure in children living in the area. Potential sources of lead includes vehicle exhaust fumes with leaded gasoline, industrial pollution, lead dust that attached to roadside foods or beverages, as well as exposure from the workplaces of parents which are brought home such as smelting or metal recycling, welding, and printing. This is the same with the study in Cairo study, where the average children who have lead levels >10µg/dl are living in industrial environments, urban areas, and slums (Moawad et al., 2016).

Other lead sources from environment are from city plumbing pipes, lead dust on floors, cosmetics, and food or beverages in canned package (Suherni, 2010). WHO establishes a lead limit

in water of 0.1 mg/L. In contaminated the water source, almost all lead is present in the sediment, and partly dissolved in water. Indonesia also has a lead threshold limit for clean and drinking water based on Permenkes RI number 416 dated 1990 that is equal to 0.05 mg/L (Naria, 2005).

A research in Canada in 2012, involving 306 children aged 1 to 5 years, indicated that there was an association between increased blood lead level of 1.78 mg/dL and lead contaminated water, which reached 3.3 ug/L (Levallois et al., 2014). This study found that twenty-seven children used tap water as the source of water for daily necessities and found no association between lead level and drinking water sources, but they did not examine the concentration of lead in the water source for daily needs in the location.

Most children in our study always washed their hands before meal, did not eat street foods, were not non-food eater, had no nail or pencil biting habit. This reduced the lead exposure. The main route of lead exposure in children is through the digestive tract. The curiosity and hand-to-mouth habit of children give easier access for lead to enter the digestive tract thus increasing the lead exposure risk. In addition, the absorption of lead in the gastrointestinal tract of children is three times greater than adults (Levallois et al., 2014).

In the study, the numbers of male samples were more than female and there was no relationship between lead and gender. In the study of NHANES III (National Health and Nutrition Examination Surveys) between 1999-2004 in the United States, males had a slightly higher incidence of lead poisoning than females. This may be related to outdoor activities, where males play more frequently in the outside than females (Jones et al., 2009). A meta-analysis study in China on children aged 0-18 years also found that males had slightly higher risk than females in the lead poisoning (Li et al., 2015).

The limitations of this study are environmental risk factors, the study relied on reporting by parents; the specific products and environments were not directly tested for presence of lead and we did not examine other factors affecting the increase in IgE. For the future, further research or cohort is needed to determine whether there is an increase in lead levels in school-aged children and possible risk factors that lead to elevated levels.

5. Conclusion

There is no correlation between serum lead level and total IgE level in elementary school-aged children. But it should be of concern to us that all children in this study has been exposed to lead, where there should be no lead in the blood and most of the children has high total IgE level. This may be due to several factors which are not investigated in this study. Attention is needed to present further increase in the serum lead level and to minimize the effects caused by lead poisoning.

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Published in the Slovak Republic
European Journal of Medicine
Has been issued since 2013.
E-ISSN: 2310-3434
2018, 6(1): 20-42

DOI: 10.13187/ejm.2018.1.20
www.ejournal5.com

VITA Intense – Product with Negative Oxidation-reduction Potential (ORP) as Important Quality for Antioxidant and Inhibition Growth of Tumor Cells Effects. Anti Aging Effects

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Abstract

We studied the mathematical model of interaction with water VITA intense of LavaVitae company (Austria). In this report are submitted data about the interaction of VITA intense with water, obtained by non-equilibrium (NES) and differential-equilibrium energy spectrum (DNES) of water. The average energy ($\Delta E_{H...O}$) of hydrogen H...O-bonds among individual molecules H_2O after treatment of VITA intense with water measured by NES- and DNES-methods is ΔE=-0.0136±0.0011 eV for VITA intense. This result suggests the restructuring of $\Delta E_{H...O}$ values among H_2O molecules with a statistically reliable increase of local extremums in DNES-spectra. The research is performed for VITA intense, with study of pH and oxidative reduction potential (ORP). There is review of the effects of the chemical composition of VITA intense – anti-inflammatory, antioxidant etc. With methods NES we show the following effects – relaxing effect of nervous system, anti-inflammatory and inhibition of tumor cells. As results of these effects VITA intense has anti aging influence. The base of this influence is anti-inflammatory effect. This article deals with the review of the basic biophysical-biochemical and biological processes underlying the VITA intense. The author is studying their physical-chemical properties and biophysical and biological effects on human organism. Additionally, by using IR, NES, and DNES methods are investigated various samples of water from Bulgarian water springs: the melt water from Glacier Rosenlaui, Swiss Alps, as well as the human blood serum of people with excellent health and cancer patients between 50 and 70 years old. Other experiments were performed on a 1 % (v/v) solution of VITA intense in deionized water. As an estimation factor in NES and DNES was measured the values of the average energy of hydrogen bonds ($\Delta E_{H...O}$) among H_2O molecules in water samples, as well as a local extremums in the NES and DNES-spectra of various samples of water and the human blood serum at E = -0.1387 eV and λ = 8.95 μm. For a group of people in critical condition of life and patients with malignant tumors the greatest values of local extremums in IR-, DNES-spectra were shifted to lower energies relative to the control healthy group. Further we applied this method for calculation of percent distribution of H_2O molecules in all studied water samples according to energies of hydrogen bonds ranged from (-0.08 to -0.1387 eV). It was shown that mountain water is among the most important factors for human longevity and human health. The variety of ions (K^+, Na^+, Ca^{2+}, Mg^{2+}, Mn^{2+}, Fe^{2+}, Fe^{3+}, Zn^{2+}, SO_4^{2-}, Cl^-, HCO_3^-, CO_3^{2-}), the chemical-physical parameters (pH, electroconductivity) and the decreased content of deuterium in studied water samples renders beneficial effects of these types of water on human health. We are applying the conclusions for the effects of mountain water on human health and longevity as base

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for the conclusion on the effects of VITA intense. The base is also the results with methods NES and DNES on human serum and expected effects from VITA intense.

VITA intense has negative oxidative reduction potential (ORP) and this is special property of this product for antioxidant effects again free radicals.

Keywords: VITA intense, anti-inflammatory, antioxidant, anti-aging, mathematical model, NES, DNES, negative oxidative reduction potential (ORP).

1. Introduction

Water is the main substance of life. The human body of an adult person is composed from 50 to 55 % of water. With aging, the percentage of water in the human body decreases. Hence, the factor of water quality and its amount in organism is an essential factor for the research (Pocock et al., 1981; Howard, Hopps, 1986). Water is present in the composition of the physiological fluids in the body and plays an important role as an inner environment in which the vital biochemical processes involving enzymes and nutrients take place. Water also is the main factor for metabolic processes and aging (Ignatov, 2012). Earlier studies conducted by us have demonstrated the role of water, its structure, the isotopic composition and physical-chemical properties (pH, temperature) on the growth and proliferation of prokaryotes and eukaryotes in water with different isotopic content (Ignatov, Mosin, 2012; Ignatov, Mosin, 2013). These factors, the structure and composition of water are of great importance in many biophysical studies. The peculiarities of the chemical structure of the H_2O molecule and weak bonds caused by electrostatic forces and donor-acceptor interaction between hydrogen and oxygen atoms in H_2O molecules create favorable conditions for formation of directed intermolecular hydrogen bonds (O–H…O) with neighboring H_2O molecules, binding them into complex intermolecular associates which composition represented by general formula $(H_2O)_n$, where n can vary from 3 to 50 (Keutsch & Saykally, 2011). The hydrogen bond is a form of association between the electronegative O-atom and a H-atom, covalently bound to another electronegative O-atom, is of vital importance in the chemistry of intermolecular interactions, based on weak electrostatic forces and donor-acceptor interactions with charge-transfer (Pauling, 1960). It results from interaction between electron-deficient H-atom of one H_2O molecule (hydrogen donor) and unshared electron pair of an electronegative O-atom (hydrogen acceptor) on the neighboring H_2O molecule.

The product of LavaVitae VITA intense is combining of herbs from Alps, Aloe Vera, Green tea, Edelweiss, Aronia, Vitamins and Magnesium, Selenium. The research is with methods NES and DNES. There is research of ORP and pH and there are executing the conclusions from electrochemically activated waters – anolyte and catholyte for anti-inflammatory effects (Ignatov et al., 2014).

The aim of this research is to show the usefully of VITA intense on the base of the following results and conclusions. The research are from various samples of water from Bulgarian water springs: the melt water from Glacier Rosenlaui, Swiss Alps, as well as human blood serum of people with excellent health and cancer patients between 50 and 70 years old. In frames of this research on the water quality were investigated 415 people living in the municipalities of Teteven, Yablanitza. Ugarchin, Lukovit, Lovech district; Dolni Dabnik, Pleven district, Kuklen, Plovdiv district (Bulgaria), where is lived the most of long lived people and their siblings, were studied.

The authors also performed the research of 1% (v/v) solution of VITA intense on the distribution of H_2O molecules according to the energies of hydrogen bonds, as well as studies of the NES and DNES spectrum and the biophysical effect of this type of water on human body.

2. Materials and Methods

2.1. NES and DNES Spectral Analysis

The device for DNES spectral analysis was made by A. Antonov on an optical principle. For this was used a hermetic camera for evaporation of water drops under stable temperature (+22–24 ^oC) conditions. The water drops were placed on a water-proof transparent pad, which consists of thin maylar folio and a glass plate. The light was monochromatic with filter for yellow color with wavelength at λ = 580±7 nm. The device measures the angle of evaporation of water drops from 72.3^o to 0^o. The DNES-spectrum was measured in the range of -0.08– -0.1387 eV or λ = 8.9–13.8 μm using a specially designed computer program. The main estimation criterion in these studies was the average energy ($\Delta E_{H...O}$) of hydrogen O…H-bonds among H_2O molecules in water

samples and human blood serum.

2.2. Product of LavaVitae – VITA intense

The product LavaVitae VITA intense of 1 liter is including – Natural and high quality plant extracts from the Swiss Alps, Aronia, Edelweiss, Aloe Vera, Vitamin C (180.0 mg), Vitamin E (15.0 mg), Vitamin A (17.0 mg), Vitamin A (17.0 mg), Vitamin B_1 (1.5 mg), Vitamin B_2 (1.7 mg), Vitamin B_3 (17.0 mg), Vitamins B_5 (10.0 mg), Vitamin B_6 (4.2 mg), Vitamins B_3 (17.0 mg), Water-soluble B Vitamin (17.0 mg) Vitamin B_9, Vitamin M, Vitamin B_{11}, (0.4 mg), Vitamin B_9, (7.4 μg), Vitamin D_3, (5.0 μg), Magnesium (62.5 mg), Selenium (25 μg).

2.3. Studying the Bulgarian Long Living People and Centenarians

Interviews have been conducted with 415 Bulgarian centenarians and long living people and their siblings. Their heredity, body weight, health status, tobacco consumption, physical activity, attitude towards life has been analyzed. With using DNES method was performed a spectral analysis of 15 mountain water springs located in municipalities Teteven and Kuklen (Bulgaria). The composition of water samples was studied in the laboratory of "Eurotest Control" (Bulgaria). Statistics methods were attributed to the National Statistical Institute of Bulgaria.

2.4. Studying the ***Human B***lood Serum

1 % (v/v) solution of human blood serum was studied with the methods of IR-spectroscopy, non-equilibrium (NES) and differential non-equilibrium (DNES) spectral analysis. The specimens were provided by Kalinka Naneva (Municipal Hospital, Bulgaria). Two groups of people between the ages of 50 to 70 were tested. The first group (control group) consisted of people in good clinical health. The second group included people in critical health or suffering from malignant diseases.

2.5. IR-spectroscopy

IR-spectra were registered on Brucker Vertex ("Brucker", Germany) IR spectrometer (a spectral range: average IR – 370–7800 cm^{-1}; visible – 2500–8000 cm^{-1}; the permission – 0,5 cm^{-1}; the accuracy of wave number – 0,1 cm^{-1} on 2000 cm^{-1}) and on Thermo Nicolet Avatar 360 Fourier-transform IR.

2.6. Statistical Processing of ***Experimental D***ata

Statistical processing of experimental data was performed using the statistical package STATISTISA 6.0 using the Student's *t*- criterion (at $p < 0.05$).

3. Results and Discussions

3.1. Applications of VITA intense for Human Health. The information is from the company LavaVitae

Product features: LavaVitae Company –VITA intense product

– Two bottles (2x500 ml) covers up the need for a monthly (~ 33 ml per a day);

– A synergy of essential vitamins and minerals with natural high-quality plant extracts and aromas from the Swiss Alps;

– Highest bioavailability in liquid form;

– Without GMO. Free of animal ingredients and allergens according to regulation (EU) No. 1169/2011;

– Supports a healthy diet and an active lifestyle.

The Table 1 shows the chemical composition of VITA intense

Table 1. The chemical composition of VITA intense

Nutritional Information	Quantity in g per Serving (33 ml)	Quantity in g per 100 ml
Calorific value	18,53 kcal / 77,66 kJ	56,14 kcal / 235,33 kJ
Fat	< 0,1	< 0,1
saturated fatty acids	<0,05	<0,1
Carbohydrates	4,5	13,67
of which sugar	4,43	13,43
Protein	< 0,05	< 0,1
Balla substances	0,3	0,9
Salt	< 0,05	< 0,1

Nutrient	Quantity in mg per Serving (33 ml)
(Vitamins and Minerals)	
Vitamin A	0,750
Vitamin B1	1,5
Vitamin B2	1,7
Niacin	17,0
Pantothenic acid	10,0
Vitamin B_6	4,2
Biotin	0,150
Folic acid	0,400
Vitamin B_{12}	0,0075
Vitamin C	180,0
Vitamin D_3	0,005
Vitamin E	15,0
Magnesium	62,5
Selenium	0.0025

Another Nutrients	Quantity in mg per Serving (33 ml)	Quantity in mg per 100 ml
Organic Aronia juice	1,980	6.000
Organic Aloe Vera extract	17,5	53,0
Edelweiss extract	17,5	53,0
Green tea extract	6,9	21,0

Vitamins and Minerals:

Vitamin A

Improve eyesight and the immune system. Contributes to the maintenance of normal skin.

Vitamin B Complex

It strengthens nerves and ensures continuous supply of power. In the bustle of everyday life and times of increased willingness to perform the essential water-soluble vitamins support the metabolism and increase the stress tolerance. Thus counteract against shiftlessness and fatigue.

Vitamin B_1

Contributes to the normal function of the nervous system, psyche and heart.

Vitamin B_2

Contributes to the normal production of red blood cells and metabolism of iron.

Protect cells against oxidative stress.

Vitamin B_3 or Niacin

Helps maintaining normal skin.

Vitamin B_5 or Pantothenic acid

Contributes to the normal synthesis and metabolism of steroid hormones, Vitamin D and some neurotransmitters.

Vitamin B_6

Contributes to the normal metabolism of proteins and glycogen, to the normal production of red blood cells and regulating hormone balance.

Water-soluble B Vitamin other names: Vitamin H, Coenzyme R, Biotin

Helps maintaining normal skin, hair.

Vitamin B_9, Vitamin M, Vitamin B_{11}

Contributes to the normal synthesis of amino acid, formation of blood and plays an important role in cell division. Contributes to the normal growth of maternal tissues during pregnancy.

Vitamin B_{12}

Contributes to the normal formation of red blood cells and plays an important role in cell division.

Vitamin C

Anti oxidant. Supports formation of collagen for skin, blood vessels and bone and aids in wound healing. Protects cells from oxidative stress.Supports the immune system. Increases the excretion of heavy metals via the kidneys and improves iron absorption.

Vitamin B_3

Contributes to the normal function of the muscles and maintaining normal bone and teeth. Contributes to the normal utilization of calcium and phosphorus and contributes to the normal calcium level in blood.

Vitamin E

Protect cells against oxidative stress and contributes to a young, tight and healthy skin. Increases the amount of collagen in the skin. Supports wound healing through the support of cell division.

Magnesium

Contributes to the normal muscle function and maintaining to normal bone and teeth. Supporting protein synthesis, regulates electrolyte balance and plays an important role in cell division.

Selenium

Contributes to the normal function of the immune system and the thyroid. Protect cells against oxidative stress.

Helps maintaining normal hair and nails as well as for normal formation of sperm. Natural and high quality plant extracts from the Swiss Alps. Extracts from plants provide an effective and long lasting protection against free radicals. For this reason Vita Intense contains a number of effective and health promoting extracts which are carefully matched to achieve an optimum synergistic effect with maximum bioavailability.

Aronia (*Aronia melanocarpa*)

The Aronia (Fig. 1) or "Black chokeberry" originates from North America is now increasingly cultivated in Middle Europe, including Switzerland.

Fig. 1. Aronia

The shell of the Aronia is naturally rich in vitamins (such as Vitamin A, C, E, K and the entire group of the B Vitamins), minerals and trace elements (such as calcium, magnesium, potassium, zinc and iron). The Aronia shows record antioxidants levels for protecting cells against oxidative stress (Zheng, Wang, 2003). But the most interesting active ingredient is hidden in the group of polyphenols, whose main representatives are the flavonoids and anthocyanins. The Aronia contains an unsurpassed amount of flavonoids and the highest proportion of anthocyanins compared to other berries (Table 2) (Zheng, Wang 2003).

Table 2. Composition of different berries (Zheng, Wang 2003)

ORAC, Anthocyan- und Phenolgehalt der Beeren im Vergleich (nach Zheng und Wang 2003)

Beerensorte	ORAC (!mol Trolox Equivalent/g FG	Anthocyane (mg Cyanidin-3-Glukoside/g FG)	Phenole (mg Gallussäureequivalent/gFG)
Blaubeere	28,9	1,20	4,12
Cranberries	18,5	0,32	3,15
Preiselbeere	38,1	0,45	6,52
Apfelbeere	160,2	4,28	25,56

Figure 2 shows the results of anti oxidative parameter of Aronia (mmol/l) for TEAC.

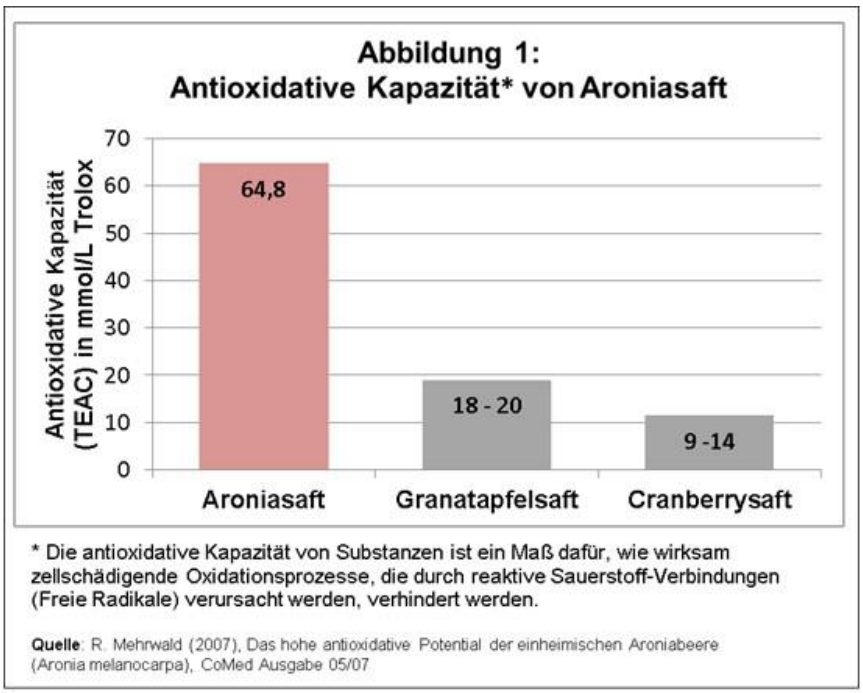

Fig. 2. Oxidative parameter of Aronia (mmol/l) for TEAC

The Aronia has antioxidant (Olas et al., 2008), anti-inflammatory (Ohgami et al. 2005; Borisova et al., 1994), anti oxidative stress (Valcheva-Kuzmanova et al., 2005) antibacterial and antiviral effect (Kokotkiewicz et al., 2010). Further these antioxidants show positive effects on blood sugar (Ryszawa et al., 2005) and of blood lipid levels (Valcheva-Kuzmanova et al., 2006). They protect the heritage ~antimutagenic effect: (Gasiorowski et al., 1997; Dimova et al., 1997), the cardiovascular system (Ryszawa et al., 2006), the gastrointestinal tract (Valcheva-Kuzmanova et al., 2005; Matsumoto et al., 2004) and the liver (Borisova et al., 2004). While flavonoids improve blood circulation and the cardiovascular system (via relaxation of the vessel walls), anthocyanins reduce platelet formation (Ryszawa et al., 2006). Both flavonoids show unsurpassed antiviral, antimicrobial, antiallergic and anticarcinogenic effects (Bermudez-Soto et al., 2006; Lala, Marlik, 2006; Zhao et al., 2004; Marlik, 2003, Zhao et al., 2003).

Green tea *(Camelia sinensis)*

Green tea (Fig. 3) has been shown to preventative against many diseases. Green tea has been proven for everyday primary care, prevention of mild illnesses or weakened constitution, improving fitness and metabolism or for setting an ideal weight. (Lambert, Yang, 2003).

Fig. 3. Green tea

The extract of Green tea (*Camelia sinensisis)* naturally rich in many vitamins, minerals and trace elements (calcium, iron, fluoride, potassium, manganese and magnesium), tea polyphenols and catechins (especially EGCG = Epigallocatechingallate). The combination of these powerful ingredients provides an antioxidative (Chan et al., 1997), anti-inflammatory, cardiotonic (Hollman et al., 1997) and cancer preventive effects (Lambert et al., 2003). Green tea contributes to the dental health, reduces deposits in the blood vessels and protects against atherosclerosis (Chacko et

al., 2010). While the antioxidant properties are generally ascribed to the high content of tea polyphenols (Balentine et al., 1997), the positive influence on the body weight is attributable to the caffeine and catechins. The caffeine/catechins combination stimulates metabolism, increases energy expenditure, fat digestion and resorption in the stomach and the intestine (Bartista et al., 2009). Furthermore, the German Medical Journal reported in April 2010 that the catechins contained in green tea (EGCG) harmless "toxic" plaques in Alzheimer's.

Aloe Vera

Sumerian and Egyptian records occupied that the "plant of immortality" or the "blood of the gods" has been used medicinally 5,000 years ago. Scientists were able to detect more than 200 active ingredients till today. The Aloe Vera (Fig. 4) is naturally rich in polysaccharides, glycoproteins, vitamins, amino acids, enzymes and phytochemicals (such as essential oils, saponins, tannins, salicylic acid, sterols and aloin) in an unique pharmacological combination (Choi et al., 2001).

Fig. 4. Aloe Vera

Esua and Rauwald reported 2005 of a novel bioactive glucan, which attributed to the significant anti-inflammatory effect. The Aloe further harmonized the metabolism in all organs by regulation of the basic substances and the acid-base balance. In folk medicine the “wonderful desert lily” shows an excellent effect in inflammation (especially in the gastrointestinal tract), skin diseases (especially in Eczema, Dermatitis and Psorias), sunburns and wound raptures (Vogler, Ernst, 1999).

Edelweiss (*Leontopodii alpinum*) – Pure ingrediens, real effect! The most famous and most symbolic flower of the Alps is naturally rich in antioxidants, terpenes, caffee acid derivatives (Leontopodic acid) and Leoligin. So it is very popular in the cosmetic- (especially in UV-protective- and anti-aging products), food- and dietary supplement industries. In the Alpine region it is particularly known Edelweiss (Fig. 5) as "Bellyache flower" (such as abdominal pain, indigestion, diarrhea or intestinal colic)

Fig. 5. Edelweiss

The secondary plant substances of the “Edelweiss” support in the prevention and treatment of gastrointestinal complaints (Dobner et al., 2003) and in respiratory-, neurologic-, muscular- and cardiovascular diseases.

In addition to the outstanding antiinflammatory, antibacterial (Dobner et al. 2004) – even with infections (Stuppner, 2000) – and analgesic effects (Speroni et al., 2006) the “Edelweiss” shows a superior property as a scavenger and antioxidant (Schwalger et al., 2005) and preserves so cells from damage (also by molds; (Costa et al., 2009). While Leoligin (Fig. 6) proved highly effective against thickening of the inner wall of blood vessels (Reisinger et al., 2009), an unknown terpene shows a significant cancer hostile activity against human leukemia cells (Wang et al. 2007). Moreover (Hornick et al., 2008) can showed after taking an extract from the whole root an improvement in the capacity of memory and kick off a discussion for taking this extract in the prevention and treatment of dementia.

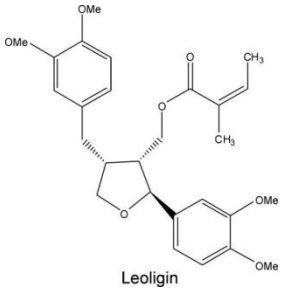

Fig. 6. The structure of Leoligin in Edelweiss

3.2. *Comparative analysis between longevity of long living people, centenarians and their siblings and the quality of water*

In frames of the research on the water quality 121 long living people from Bulgaria over 90 years of age have been studied together with their 294 siblings. The average lifespan of long lived people and centenarians in mountain areas is 94.1 years. For the average lifespan of long lived people in plain areas the result is 90.6 years. The most adult person from mountain areas is 104 years old and for plain areas is 97 years old. For the brothers and sisters of long live people from mountain areas the average lifespan is 88.5 years. For the brothers and sisters of long live people from plain areas the average lifespan is 86.4 years. The difference in life expectancy of the two groups of people is reliable and it corresponds to the Student's *t*-criteria at $p < 0.05$ with a confidence level of $t = 2.36$. There are distances of no more than 50–70 km between these places and the only difference is the mountain water and air.

There have been 21519 residents in Teteven and 142 of them were born before 1924. Figure 7 demonstrates the interrelation between the year of birth (1912–1924) of long living people (age) and their number (Teteven municipality, Bulgaria).

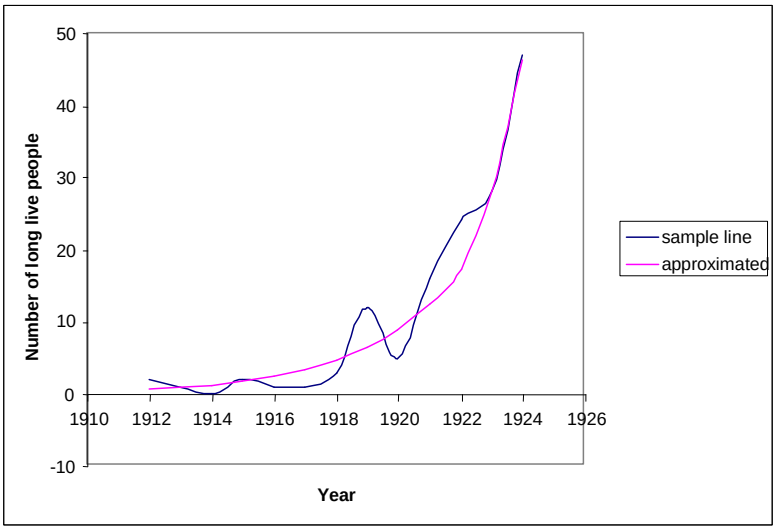

Fig. 7. Interrelation between the year of birth of long living people (age) and their number in Teteven municipality, Bulgaria

From the standpoint of genetics, the process of aging is associated with disruption of the genetic program of the organism and gradual accumulation of errors during the process of DNA replication. Aging may be associated with the accumulation of somatic mutations in the genome and be influenced by free radicals (mainly oxygen and primary products of oxidative metabolism) and ionizing radiation on DNA molecules as well (Woodhead, 1984; Adelman et al., 1988). Such mutations can reduce the ability of cells to the normal growth and division and be a cause of a large number of various cell responses: inhibition of replication and transcription, impaired cell cycle division, transcriptional mutagenesis, cell aging that finally result in cell death. Cells taken

from the elderly people show a reduction in transcription when transferring information from DNA to RNA.

From the standpoint of dynamics, aging is a non-linear biological process, which increases over time. Accordingly, the rate of aging increases with time. The accumulation of errors in the human genome increases exponentially with time and reaches a certain stationary maximum at the end of life. L. Orgel shows that, for this reason, the probability of cancer occurrence increases with age (Orgel, 1963). Figure 8 shows L. Orgel's results on the interrelation between age and the number of cancer cases. The accumulation of errors in synthesis of abnormal proteins increases exponentially over time with age. Cells taken from elderly people show the reduced levels of transcription or transmission of information from DNA to RNA. Therefore, the probability of cancer increases with age. The interrelation between the number of Bulgarian centenarians in the mountainous municipality of Teteven and their age is close to exponential.

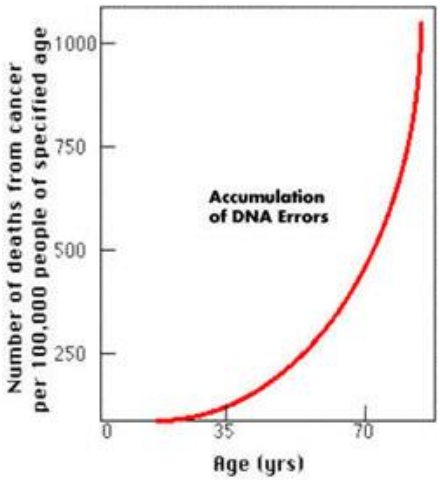

Fig. 8. Interrelation between age and the number of cancer patients (Orgel, 1963)

Here are submitted the data for Bulgaria:

1) Varna district – 44 centenarians per 1 million of inhabitants, plain and sea regions;

2) Pleven district – 78 centenarians per 1 million of inhabitants, plain regions;

3) Teteven district – 279 centenarians per 1 million of inhabitants, hills and mountainous regions;

4) Bulgaria – 47 centenarians per 1 million of inhabitants.

3.3. Clinical studies with human blood serum testing

A convenient method for studying of liquids is non-equilibrium differential spectrum. It was established experimentally that the process of evaporation of water drops, the wetting angle θ decreases discreetly to zero, and the diameter of the water drop basis is only slightly altered, that is a new physical effect (Antonov, 1995; Antonov, Yuskesselieva, 1983). Based on this effect, by means of the measurement of the wetting angle within equal intervals of time is determined the function of distribution of H_2O molecules according to the value of f(θ). The distribution function is denoted as the energy spectrum of the water state. The theoretical research established the dependence between the surface tension of water and the energy of hydrogen bonds among individual H_2O-molecules (Antonov, 1995).

For calculation of the function f(E) represented the energy spectrum of water, the experimental dependence between the wetting angle (θ) and the energy of hydrogen bonds (E) is established:

$$f(E) = \frac{14{,}33 f(\theta)}{[1-(1+bE)^2]^2} \qquad (1)$$

where b = 14.33 eV^{-1}

The relation between the wetting angle (θ) and the energy (E) of the hydrogen bonds between H_2O molecules is calculated by the formula:

$$\theta = arcos\ (-1 - 14.33E) \qquad (2)$$

The energy spectrum of water is characterized by a non-equilibrium process of water droplets evaporation, therefore, the term non-equilibrium spectrum (NES) of water is used.

The difference $\Delta f(E) = f\ (E\ samples\ of\ water) - f\ (E\ control\ sample\ of\ water)$ – is called the "differential non-equilibrium energy spectrum of water" (DNES).

Thus, the DNES spectrum is an indicator of structural changes in water, because the energy of hydrogen bonds in water samples differ due to the different number of hydrogen bonds in water samples, which may result from the fact that different waters have different structures and composition and various intermolecular interactions – various associative elements etc. (Ignatov et al., 2014; Ignatov et al., 2015). The redistribution of H_2O molecules in water samples according to the energy is a statistical process of dynamics.

Figure 9 shows the average NES-spectrum of deionised water. On the X-axis are depicted three scales. The energies of hydrogen bonds among H_2O molecules are calculated in eV. On the Y-axis is depicted the function of distribution of H_2O molecules according to energies f(E), measured in reciprocal unit eV^{-1}.

Arrow A designates the energy of hydrogen bonds among H_2O molecules, which is accepted as most reliable in spectroscopy.

Arrow B designates the energy of hydrogen bonds among H_2O molecules the value of which is calculated as:

$$\bar{E} = -\ 0.1067 \pm 0.0011\ eV \qquad (3)$$

Arrow C designates the energy at which the thermal radiation of the human body, considered like an absolute black body (ABB) with a temperature +36.6 °C, is at its maximum.

A horizontal arrow designates the window of transparency of the Earth atmosphere for the electromagnetic radiation in the middle infrared range of the Sun toward the Earth and from the Earth toward the surrounding space. It can be seen that the atmosphere window of transparency almost covers the NES-spectrum of water.

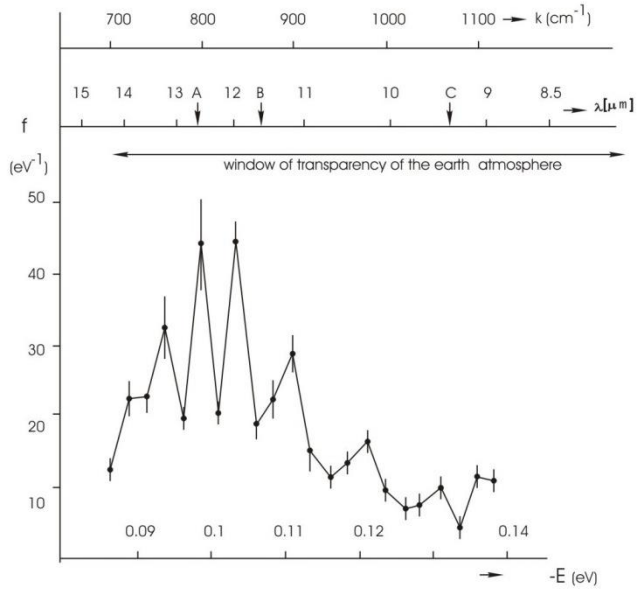

Fig. 9. The NES-spectrum of deionized water (chemical purity – 99.99 %; pH – 6,5–7,5; total mineralization – 200 mg/l; electric conductivity – 10 μS/cm): the horizontal axis shows the energy of the H...O hydrogen bonds in the associates – E (eV); the vertical axis – the energy distribution function – f (eV^{-1}); *k* – the vibration frequency of the H–O–H atoms (cm^{-1}); λ – wavelength (μm)

We have conducted studies of 1 % (v/v) solution of human blood serum taken from two groups of people between 50 and 70 years of age by IR, NES and DNES spectral analysis. The first group consisted of people in excellent health. The second group consisted of people in a critical state and patients with malignant tumors. The average energy of hydrogen bonds ($\Delta E_{H...O}$) between H_2O molecules in the blood serum was investigated as the main biophysical parameter. The result was registered as a difference between the NES-spectrum of 1 % solution of human blood serum and the NES-spectrum of deionized water control sample – DNES-spectrum, measured as the difference $\Delta f(E)$ = f (samples of water) – f (control sample of water). The DNES-spectrum obtained from the first group has a local extremum energy ($\Delta E_{H...O}$) at E = -9.1±1.1 meV and from the second group at E = -1.6±1.1 meV. The results between the two groups have a statistical difference in Student's criterion at $p < 0.05$. For the control group of healthy people the value of the largest local maximum in the DNES-spectrum was detected at E = -0.1387 eV, or at a wavelength λ = 8.95 μm. For the group of people in a critical health state and the patients with malignant tumors, the analogous values of the largest local maximums of the DNES-spectrum shifted to lower energies compared with the control group of people. For a group of people in critical health condition and patients with malignant tumors the greatest values of local extremum in the IR-spectrum are shifted to lower energies relative to the control group. In IR-spectrum of human blood serum are detected 8 local maxima at λ = 8.55, 8.58, 8.70, 8.77, 8.85, 9.10, 9.35 and 9.76 μm (Krasnov, Gordetsov, 2009). The resulting peak at λ = 8.95 μm in the IR-spectrum (Ignatov, 2012) approaching the peak at λ = 8.85 μm was monitored by Russian researchers. In the control group of healthy people the average value of the energy distribution function f(E) at λ = 8.95 μm compiles E = 75.3 eV, and in a group of people in critical condition – E = 24.1 eV. The norm has statistically reliable result for human blood serum for the control group of people having cancer at the local extremum of f(E) ~24.1 eV^{-1}. The level of reliability of the results is $p< 0.05$ according to the Student's t-test. In 1995 were performed DNES-experiments with an impact on tumor mice cells in water solutions containing Ca^{2+} (Antonov, 1995). There was a decrease in the DNES-spectrum compared with the control sample of cells from a healthy mouse. The decrease was also observed in the DNES-spectrum of human blood serum of terminally ill people relative to that of healthy people. With increasing of age of long-living blood relatives, the function of distribution of H_2O molecules according to energies at -0.1387 eV decreases. In this group of tested people the result was obtained by the DNES-method at E = -5.5±1.1 meV; the difference in age was of 20–25 years in relation to the control group. It should be noted that many of Bulgarian centenarians inhabit the Rhodopes Mountains areas. Among to the DNES-spectrum of mountain waters the similar to the DNES-spectrum of blood serum of healthy people at λ = 8.95 μm, was the DNES-spectrum of water in the Rhodopes. The mountain water from Teteven, Boyana and other Bulgarian provinces has similar parameters. Tables 1, 2 and 3 show the composition of mountain water springs in Teteven and Kuklen (Bulgaria) and local extremums in NES-spectra of water samples. The local extremums is water samples were detected at E = -0.11 eV and E = -0.1387 eV. The value measured at E = -0.11 eV is characteristic for the presence of Ca^{2+} in water. The value measured at E = -0.1387 eV is characteristic for inhibiting the growth of cancer cells. Experiments conducted by A. Antonov with cancer cells of mice in water with Ca^{2+} demonstrated a reduction of this local extremum to a negative value in spectra. Analysis by the DNES-method of aqueous solutions of natural mineral sorbents – shungite (carbonaceous mineral from *Zazhoginskoe deposit* in Karelia, Russia) and zeolite (microporous crystalline aluminosilicate mineral from Most village, Bulgaria) showed the presence of a local extremum at E = -0.1387 eV for shungite and E = -0.11 eV for zeolite (Mosin, Ignatov, 2013, Ignatov, Mosin, 2014). It should be noted that owing to the unique porous structures both the natural minerals shungite and zeolite are ideal natural water adsorbers effectively removing from water organochlorine compounds, phenols, dioxins, heavy metals, radionuclides, and color, and gives the water a good organoleptic qualities, additionally saturating water with micro-and macro-elements until the physiological levels (Mosin & Ignatov, 2013). It is worth to note that in Bulgaria the main mineral deposits of Bulgarian zeolites are located in the Rhodope Mountains, whereat has lived the greatest number of Bulgarian centenarians. It is believed that water in these areas is cleared out in a natural way by mineral zeolite. Company LavaVitae has medicine product with high quality ZEOLITH detox.

3.4. *Results of 1 % (v/v) solution in deionized water of VITA intense*

The research with the NES method of water drops is received with 1 % solution VITA intense, and deionized water as control sample. The mathematical models of 1 % (v/v) solution VITA intense gives the valuable information for the possible number of hydrogen bonds as percent of H_2O molecules with different values of distribution of energies (Table 2 and Fig. 10). These distributions are basically connected with the restructuring of H_2O molecules having the same energies.

Table 2. The distribution (%, $(-E_{value})/(-E_{total\ value})$ of H_2O molecules in 1 % water solution of VITA intense (product of LavaVitae, Austria) and control deionized water

-E(eV) x-axis	1 % water solution VITA intense (LavaVitae) y-axis (%((-E_{value}) */ (-$E_{total\ value}$)**	Control Sample Deionized water y-axis (%((-E_{value})*/ (-$E_{total\ value}$)**	-E(eV) x-axis	1 % water solution VITA intense (LavaVitae) y-axis (%((-E_{value}) */ (-$E_{total\ value}$)**	Control Sample Deionized water y-axis (%((-E_{value})*/ (-$E_{total\ value}$)**
0.0937	0	0	0.1187	0	0
0.0962	0	14.3	0.1212	**17.7**[2]	0
0.0987	0	0	0.1237	0	0
0.1012	0	14.3	0.1262	11.7	3.6
0.1037	0	0	0.1287	0	3.6
0.1062	11.7	14.3	0.1312	5.7	7.1
0.1087	0	0	0.1337	11.7	0
0.1112	**5.7**[1]	**14.3**	0.1362	5.7	7.1
0.1137	0	0	0.1387	**30.1**[3]	7.1
0.1162	0	14.3	–	–	–

E= -0.1112 eV is the local extremum for relaxing effect on nervous system

E=-0.1212 eV is the local extremum for anti-inflammatory effect

E= -0.1387 eV is the local extremum for inhibition of development of tumor cells of molecular level

Notes:

* The result (-E_{value}) is the result of hydrogen bonds energy for one parameter of (-E)

** The result (-$E_{total\ value}$) is the total result of hydrogen bonds energy

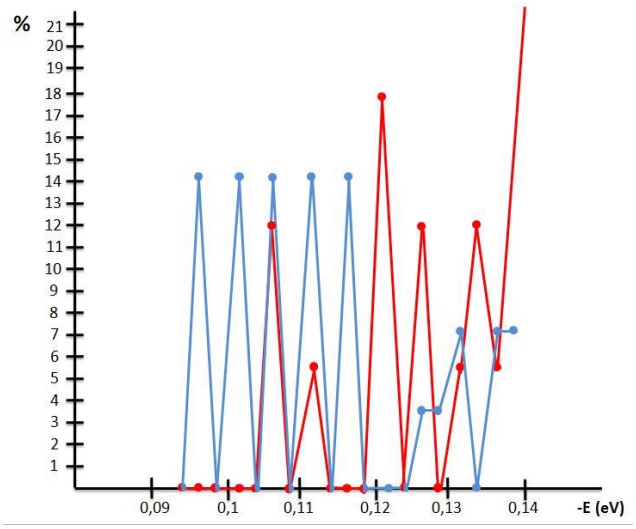

Fig. 10. Mathematical model (Ignatov, Mosin, 2013) of 1 % water solution of VITA intense (product of LavaVitae, Austria).

Figure 10 shows the distribution (%, $(-E_{value})/(-E_{total\ value})$ of H_2O molecules in and 1 % (v/v) of water solution of VITA intense (product of LavaVitae, Austria) (red line) and control sample deionized water (blue line).

Notes:

E= -0.1112 eV is the local extremum for relaxing effect on nervous system

E=-0.1212 eV is the local extremum for anti-inflammatory effect

E= -0.1387 eV is the local extremum for inhibition of development of tumor cells of molecular level

The experimental data obtained testified the following conclusions from the mathematical model of in 1 % (v/v) water solution of VITA intense (product of LavaVitae, Austria) and control deionized water. The distribution (%, $(-E_{value})/(-E_{total\ value})$ of water molecules in mathematical model of in 1 % (v/v) water solution of VITA intense (product of LavaVitae, Austria) and control deionized water. The distribution (%, $(-E_{value})/(-E_{total\ value})$ of water molecules in VITA intense (product of Lava Vitae, Austria) according control sample is different. However, for the value E = -0.1387 eV or λ = 8.95 μm there is the biggest local extremum (30.1 (%, $(-E_{value})/(-E_{total\ value})$) corresponding to the re-structuring of hydrogen bonds among H_2O molecules for inhabitation of development of tumor cells of molecular level. This difference may indicate on the different number of hydrogen bonds in water samples, as well as their physical parameters **(**pH, ORP**),** resulting in different distribution of H_2O molecules and different values of H_2O molecules with ratios of $(-E_{value})/(-E_{total\ value})$. Particularly it was observed the statistical re-structuring of H_2O molecules in water samples according to the energies. The experimental data may prove that stipulates the restructuring of H_2O molecules on molecular level and may be used for the prophylaxis of inhibition of development of tumor cells. For the value E = -0.1112 eV or λ = 11.15 μm there is the local extremum with sight minus (5.7 (%, $(-E_{value})/(-E_{total\ value})$) according control sample (14.3 (%, $(-E_{value})/(-E_{total\ value})$) corresponding to the re-structuring of hydrogen bonds among H_2O molecules. The experimental data may prove that influence stipulates the restructuring of H_2O molecules on molecular level and has biophysical effect for relaxing effect on nervous system. For the value E = -0.1212 eV or λ = 10.23 μm there is bigger local extremum (17.7 (%, $(-E_{value})/(-E_{total\ value})$) corresponding to the re-structuring of hydrogen bonds among H_2O molecules for anti-inflammatory effect. The experimental data for Lava Intense may prove that stipulates the restructuring of H_2O molecules on molecular level and the biophysical effects are:

E= -0.1112 eV is the local extremum for relaxing effect on nervous system

E=-0.1212 eV is the local extremum for anti inflammatory effect

E= -0.1387 eV is the local extremum for inhibition of development of tumor cells of molecular level.

As a result of different energies of hydrogen bonds, the surface tension of 1% (v/v) solutions of water samples with VITA Intense is increasing. The increasing of surface tension is regarding the control samples. This effect is connected with preservation of the energy in human body as result of biochemical process among water molecules and bio molecules.

3.5. VITA intense (product of the company LavaVitae)

The average energy ($E_{H...O}$) of hydrogen H...O-bonds among individual H_2O molecules in 1 % (v/v) solution of VITA Intense is measured at E=-0.1261 eV. The result for the control sample (deionized water) is E=-0.1125 eV. The results obtained with the NES method are recalculated with the DNES method as a difference of the NES (1 % (v/v) solution of VITA intense) minus the NES (control sample with deionized water) equaled the DNES spectrum of 1 % solution of VITA Intense. Thus, the result for 1 % solution of VITA intense recalculated with the DNES method is ΔE=-0.0136±0.0011 eV. The result shows the increasing of the values of the energy of hydrogen bonds in 1 % (v/v) solution of VITA intense regarding the deionized water. This is effect of stimulation on human body. The results show restructuring of water molecules in configurations of clusters, which influence usefully on human health on molecular and cellular level. The effects are describing with mathematical model of 1 % solution of VITA intense.

3.6. Results with pH and ORP

There are valid the following results of pH as indicator for acid alkaline medium of the products of LavaVitae. There are the results also of ORP or Oxidation-reduction potential.

The results are for 1% (v/v) of solutions of products, which are made from deionized water. This research is performed with Georgi Gluhchev from Bulgarian Academy of Science. The results of pH of deionized water is 6.05 and of ORP is 119.7. Table 3 shows the results of pH and ORP.

Table 3. Results of products of company LavaVitae for pH and ORP

Product	pH	ORP (mV)	Coordinates Fig. 11
VITA intense	4.07±0.02	- 104.5	Point 1 (4,07; -104.5)
BOOST	3.60±0.02	+113.6	Point 2 (3,90;113.6)
ZEOLITH detox	8.01±0.02	+109.5	Point 3 (8,01;103.3)
Deionized water	6.05±0.02	+119.7	

Figure 11 shows the dependence between the acidity and basicity (pH) of electrochemically activated solutions and the oxidation-reduction potential (ORP). The pH value within the interval from 3 to 10 units and the ORP within the interval from -400 mV to +900 mV characterize the area of the biosphere of microorganisms. Outside these ranges of pH and ORP the microorganisms will hardly survive.

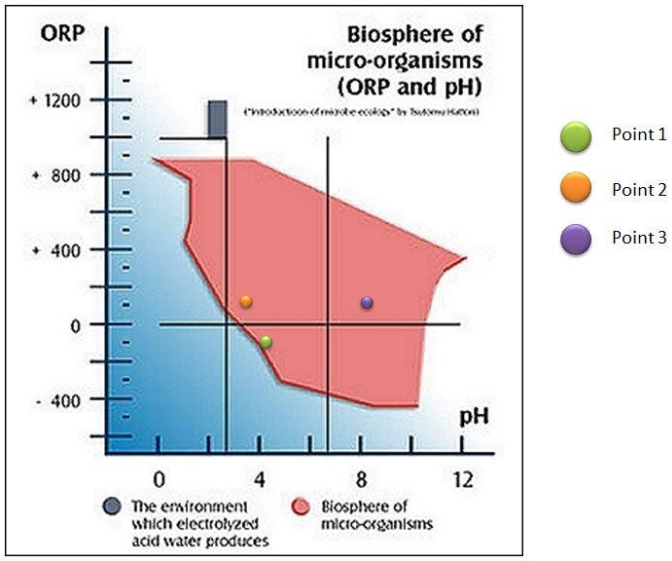

Fig. 11. The dependence between acidity and basicity (pH) of solutions and the ORP on the biosphere of micro-organisms (point 1; VITA intense), (point 2; BOOST), point 3; ZEOLITH detox)

The result of 1 % (v/v) solution of VITA intense is 4.07 or acidic medium. The result of ORP is (-104.5). The result of ORP with negative charge is connected with charge with negative value, which has antioxidant and permanent antioxidant activity. In the VITA intense there are the following antioxidants – Vitamins C, E, D. Figure 11 shows the dependence between acidity and basicity (pH) of solutions and the ORP on the biosphere of micro-organisms. The result of VITA intense with point 1 with coordinates (4,07; -104.5) is the biosphere of micro-organisms. VITA intense is useful for human health also with liquid form.

3.7. Effects of Magnesium and Selenium, Vitamins C and E in VITA intense again oxidative stress

VITA intense is product with negative Oxidation-reduction potential (ORP) or (-104.5) mV. This results shows that there are particles in VITA intense with negative charge again free radicals.

There are also active antioxidants as Vitamin C and Vitamin E. There are special role in the liquid product the ions of Magnesium – Mg^{2+}. These ions neutralize one of the most danger radicals – hydroxyl radical OH (Glamello et al., 1993). Magnesium deficiency increased cytotoxicity of the added oxyradicals. These results suggest that increased oxidative endothelial cell injury may contribute to vascular injury during Mg deficiency (Dickens et al., 1992). The magnesium deficiency and oxidative stress have both been identified as pathogenic factors in aging and in several age-related diseases. The link between these two factors is unclear in humans although, in experimental animals, severe Mg^{2+} deficiency has been shown to lead to the increased oxidative stress (Begona et al., 2000). A negative correlation between magnesium balance and oxidative stress was observed suggesting that the same etiological factor (chronic stress) initiate decreases in both free and total magnesium concentrations and simultaneously increase oxidative stress intensity. These findings support the need for magnesium supplementation with antioxidant vitamins for people living in conditions of chronic stress (Cernak et al., 2000).

Selenium has role for anti oxidative stress in the enzymes, which are connected with anti oxidant effects (Stewart et al., 1999).

Vitamin E, as a powerful antioxidant residing mainly in biomembranes, may provide effective protection against oxidative membrane damage and resultant age-related deterioration, especially in the elderly. We hypothesized that appropriate levels of vitamin E supplementation would protect erythrocyte membranes from oxidative stress and thus improve membrane fluidity in healthy middle-aged and elderly people (Sun et al., 2012).

The combination of antioxidative treatment with vitamins E and C decreases fetal malformation rate and diminishes oxygen radical-related tissue damage (Cederberget al., 2001).

3.8. Effects of Ca^{2+}, Mg^{2+}, $Zn^{2+}$and Mn^{2+} in water on biophysical and biochemical processes in the human body

The research of distribution of local extremums (eV^{-1}) in spectra of various water samples as a function of distribution of H_2O molecules according to energy f(E) at λ = 8.95 μm shows the analogue extremum at analogous values of f(E), E and λ, which was detected in water with $Zn^{2+}$and Mn^{2+} ions earlier demonstrated inhibiting the growth of cancer cells. Magnesium (Mg^{2+}), zinc (Zn^{2+}) and manganese (Mn^{2+}) ions dissolved in water have influence on enzymes, which are antioxidants (Ignatov, Mosin, 2015). The research of China team was categorized three groups of elements from the rice and drinking water according to their effect on longevity: Sr, Ca, Al, Mo, and Se, which were positively correlated with longevity: Fe, Mn, Zn, Cr, P, Mg, and K, which had a weak effect on local longevity, and Cu and Ba, which had a negative effect on longevity (Lv et al., 2011). There was a positive correlation between the eSOD activity and the age and a negative correlation between the eSOD activity and concentration of Zn^{2+} in plasma. An inverse correlation was also found between the content of Zn^{2+} ions in plasma relative to the age. The prevalence of Zn^{2+} deficiency is increased with age; with normal Zn^{2+} levels it is observed in about 80 % of adult people and only in 37 % of the non-agenarians. Aging is an inevitable biological process that is associated with gradual and spontaneous biochemical and physiological changes and the increased susceptibility to diseases. Because the nutritional factors are involved in improving the immune functions, metabolic balance, and antioxidant defense, some nutritional factors, such as Zn, may modify susceptibility to disease and promote healthy aging. *In vitro* (human lymphocytes exposed to endotoxins) and *in vivo* (old or young mice fed with low zinc dietary intake) studies revealed that zinc is important for immune efficiency (innate and adaptive), antioxidant activity (superoxide dismutase), and cell differentiation *via* clusterin/apolipoprotein J. The intracellular Zn homeostasis is regulated by metallothioneins (MT) *via* an ion release through the reduction of thiol groups in the MT molecule (Mocchegiani, 2007). Zinc in composition of water improves the antioxidative enzymes in red blood cells (Malhotra, Dhawan, 2008).

The antioxidants against free radical damage include tocopherol (vitamin E), ascorbic acid (vitamin C), β-carotene, glutathione, uric acid, bilirubin, and several metalloenzymes including glutathione peroxidase (Se), catalase (Fe), and superoxide dismutase (Cu, Zn, Mn) and proteins such as ceruloplasmin (Co). The extent of the tissue damage is the result of the balance between the free radicals generated and the antioxidant protective defense system (Machlin, Bendich, 1988). There was reported the antioxidant effects of water on rats (Abdullah, 2012). The norm in water for

Zn^{2+} and Mn^{2+} according to the World Health Organization (WHO) should be less than 20 μg. For the Na^{+} content the norm according to the WHO is less than 20 mg.

The interesting results on the concentration of Ca^{2+} in water were obtained in USA and Canada. According to the statistical information the most number of centenarians in Canada per 1 million of population is observed in Nova Scotia (210 of centenarians per 1 million). In the water from Nova Scotia the Ca^{2+} content makes up 6.8 mg/l. N. Druzhyak, Russia showed that in the places wherein live the most number of centenarians the Ca^{2+} content in water was 8–20 mg/l. The only risk factor regarding the increased Ca^{2+} content in water is cardiovascular diseases.

The following reactions occur in water if there are high concentrations of Ca^{2+} and Mg^{2+} ions: the reaction of limestone ($CaCO_3$) and gypsum ($CaSO_4 \cdot 2H_2O$) with water to separate the calcium (Ca^{2+}), carbonates (CO_3^{2-}) and sulfate (SO_4^{2-}) ions. By increasing the mineralization of water the content of Ca^{2+} ions decreases. During the concentration of the solutions Ca^{2+} ions are precipitated. With the increase of carbon dioxide (CO_2) in water and decreasing of the pH value the content of Ca^{2+} increases. The reaction of interaction of dolomite ($CaCO_3 \cdot MgCO_3$) with water makes the formation of Mg^{2+} ions. Hydrocarbonates (HCO_3^{-}) and carbonates (CO_3^{2-}) ions are formed by reaction of interaction of karst rocks, CO_2 and water. For example, in Zamzam water there is Ca^{2+} – 299.7 mg/l; Mg^{2+} – 18.9 mg/l; Zn^{2+} – 0.001 mg/l.

4. Conclusion

From the NES and DNES spectrum and mathematical model of 1% (v/v) solution of VITA Intense and deionized water as control sample are valid the following conclusions for biophysical effects for VITA Intense (LavaVitae company)

- relaxing effect on nervous system;
- anti inflammatory effect;
- inhibtion of development of tumor cells of molecular level;

In 1 % (v/v) solution of VITA intense there is restructuring of water molecules in configurations of clusters, which influence usefully on human health on molecular and cellular level.

The biophysical effects of VITA intense are connected also with antioxidant effects. The VITA intense is recommended as anti aging solution for prophylaxis (Ignatov et al., 2015). The scientific studies show that the inflammations are one of the basic reasons for aging. The recommendation is connected with additionally using or including in VITA intense of the additional mineral. The structuring of water clusters with highest energy of hydrogen bonds at 8.95 μm makes the water in human body more “active” as medium of biochemical and biophysical processes. This is similar like the human organism to be younger (Ignatov, Mosin, 2012). The quality of the water with which will be using is very important. There are types of water which will increase the effects. For these effects is recommended additional scientific project with pH is in progress.

As a result of different energies of hydrogen bonds, the surface tension of 1% (v/v) solutions of water samples with VITA intense is increasing. The increasing of surface tension is regarding the control sample. This effect is connected with preservation of the energy in human body as result of biochemical process among water molecules and bio molecules;

It worth to note that IR-spectrum of VITA intense is most similar to the IR-spectrum of blood serum of healthy group of people with a local extremum at λ=8.95 μm. The similar spectral characteristics possess mountain water from Teteven and other Bulgarian sources electrochemically activated water catholyte (Ignatov et al., 2014). The basic research is with electrolysis device (Ignatov et al., 2014) Studying the human blood serum by NES and DNES-methods show that by measuring the average energy of hydrogen bonds among H_2O molecules and the distribution function of H_2O molecules on energies it is possible to show a vital status of a person and associated life expectancy. Our data indicates that water in the human body has the IR-spectrum resembling the IR-spectrum of human blood serum. On the characteristics of the IR-spectrum of water also exerts an influence the presence of deuterium in water samples. In the research there is the optimal composition of mountain and melt water from areas where are lived the long live people and centenarians. The decreased content of deuterium in studied water samples with residual deuterium content of 60-100 ppm, the variety of ions (K^{+}, Na^{+}, Ca^{2+}, Mg^{2+}, Mn^{2+}, Fe^{2+}, Fe^{3+}, Zn^{2+}, SO_4^{2-}, Cl^{-}, HCO_3^{-}, CO_3^{2-}), and chemical-physical parameters (pH, electroconductivity) of studied water samples renders beneficial effects of this type of water on

human health. We have also obtained new proofs for biophysical and biochemical effects of Ca^{2+}, Mg^{2+}, Zn^{2+} and Mn^{2+} in composition of water on human organism and DNES-spectra of water. There are obtained new results of chemical composition of water from Glacier Rosenlaui, Swiss Alps.

VITA intense is an elixir from alpine herbs that strengthens the body powerfully. It is most useful during the autumn, winter and spring seasons, when it is the time of the viruses. It contains a complex of edelweiss, aronia, aloe vera and green tea. And most importantly is, that the antioxidant vitamins C and E, as well as Magnesium are together in a liquid that gets absorbed from the body perfectly. The magnesium neutralizes one of the most dangerous radicals, explains the scientist – the hydroxyl radical OH. This radical is one of the reason for Parkinson, Alzheimer and whole status of the brain and nervous system. VITA intense is one of the not so many liquid products from this type, which imports into the body negative antioxidant charges with effects again tumor cells. Inflammations are basic reason for aging process in human body. In VITA intense there are the following effects – anti aging, relaxing effect on the nervous system, antioxidant, inhibition of development of tumor cells, stimulating on the body. The people take one product with a lot of effects. VITA intense id in glass bottle and this is very important for the quality. People feel rush of energy, life vigour and a sense of happiness.

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Published in the Slovak Republic
European Journal of Medicine
Has been issued since 2013.
E-ISSN: 2310-3434
2018, 6(1): 43-51

DOI: 10.13187/ejm.2018.1.43
www.ejournal5.com

Aikido Game Activities as Stimulation Factor for a Proper Behavior in Conflict Situations

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Abstract

The conflicts between different personalities offer contradictions, division of opinions and clashes between people. In a field like sport activity the trainers should be able to identify the motives for the conflicts that burst out among the sportsmen and the methods to overcome them. The goal of the study is to prove the positive influence of the physical activity demonstrated in the aikido game over the ways in which one may react in case of conflict situation. The respondent group consists, generally, of 64 youngsters aged from 18 to 24, divided in to two groups: Control group of 24 young people who do not practice sport; and Experimental group of 40 young people that are actively practicing aikido in various clubs in this country. We applied the approved Ken Thomas 'Test (Thomas-Kilmann Conflict Mode Instrument) (Thomas, 1977). We used some given keys to determine the five instruments for reaction to conflict situations: 1) Competing; 2) Collaborating; 3) Compromising; 4) Avoiding; and 5) Accommodating. The analysis made is defining some generalizations: the young people practicing aikido have more harmonious structure of the tactics configuration to be used as a response to conflict situations in comparison to the group of non-involved in sport their peers. They are inclined to cooperation, adaptation and reasonable compromise in such environment. Competition and avoidance are not preferable techniques to resolve conflicts. In the same time non-involved in sport activities young people are governed by attitudes for competiveness; sometimes their behavior and motives are commanded by the will to make compromise and the desire to avoid conflicts to which it is difficult to adapt. The youngsters with higher physical aikido activities improve their will and tenacity; they are striving to achieve the goals and to assume definite responsibilities. The research of Ignatov with long living people and centenarians shows how physiological status is important for healthy life and adaptations after conflicts again stress (Ignatov et al., 2014). For that the sport is useful for young people.

Keywords: Conflict, Sport, Aikido game.

1. Introduction

The recent rapid development of society as a whole and particularly technologies modernization, especially the information systems, both drive to an evolutionary transformation of physical activities into mental. As a result of the enhanced knowledgeable activity, it is observed an increase of the psychical tension. The modern person demonstrates obvious limitation of his/her

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physical activities. The last EU reports have registered, in particularly, a notable decrease of the physical activity standards among the population. In these reports the Bulgarian citizens are ranked last and the statistics is rather alarming: „82 per cent of the Bulgarians have never practiced any kind of physical activities or sport“ (Eurobarometer, 2010), and ever since 2009 the motive inactivity of the population has been grown by 20 per cent. Probably, the negative trend will be preserved in future times if this problem is not focused seriously.

This considerable degradation of the physical activity in general, which is respectively connected with the motion limitation, is regarded as one of the fundamental, radical reasons for socially important and substantial diseases among the aged population such as ischemic heart disease (IHD), known also as coronary artery disease (CAD), hypertension (HTN or HT), known also as high blood pressure (HBP), neurological diseases such as stroke, diabetes and obesity. Physical activity in the various groups of children and grow-ups occurs to be a significant factor simulating growth and development processes. Hypodynamics is a stepping stone to the delay in child's physical and psychical development and to the advance of different acute and chronic diseases, aberrations and anomalies such as obesity, diabetes, acute and chronic diseases, postural distortions such as incorrect spinal position, scoliosis, kyphosis, fallen arch and flat feet (Markovska et al., 2010; Gencheva et al., 2010). In accordance with a NCPHA Report, there is a constant reduction of the physical efficiency in the time period 2008 - 2012, on the one hand, and on the other, the statistics have registered an increase of the postural disturbances. In reference to the spinal distortions in the group of 7-18 olds, Bulgarian children are ranked at fourth position (5.3 per mille), and the 2008 Annual Report records the two most widespread problems: overweight (average 15 per cent) and obesity (average 5 per cent); the tendency is to increase these situations and thus to create a serious precondition for another level, ever higher, of degradation of whatsoever kind of physical activity [ncpha.government.bg].

The pragmatic advantage of regular execution of some motions (physical exercises) is proved ever since Antiquity. The ancient Greek physician and scientist, Hippocrates of Kos, described different methods to practice physical exercises and massage in order to cure people suffering from spinal deformations. Gl. Tissot (1750-1826) pronounces his famous phrase 'so far as the effects of motion are concerned, it could substitute each medicine; however, all healing agents cannot replace the motive effects'; thus he reveals the great acceptation of the physical exercises. Movement occurs to be the most important factor influencing human adaptation to the changing environment. The considerable degradation of physical activities, which is bound up to the limitation of motions, opens the door for serious destructions of human active life and work. It has unfavorable influence on his/her psyche. Irritability, fatigue and anxiety are going up. The mutual determination of inner (psychic) and outer (physical) activity is broken.

On the other hand, the mutual determination of psychic and physical factors in the structure of human activity is obviously proven. While developing physical capacity there is also an improvement of the activity structure, itself, as well as an improvement of its psychic processes. (Kaykov, 1997). It is obvious that there is also a substantial decrease of the power of negative emotional experiences such as anxiety, fear, anger, aggression, etc. That is why, physical activity is among of the basic factors that are decisive for the health and life-style qualities of the modern person.

It is proved that the purposeful physical activity exerts favorable influence on the behavior modes needed for the resolution of various conflict situations (Gavrilov et al., 2016).

In modern times conflicts are among the most fundamental problems in the relationships between people. The conflict, per se, is a process in which one party involved accepts that its interests are set against the other party or are negatively affected by the other (Wall, Callister, 1995). Similar is the definition, given by K. Markov in 2013: at the root of each conflict there are subject-object oppositions; the reasons for them are underlying in the incompatible interests, needs and values which quite often occur to be salient in a real counter fight between both parties. Most often the emerged conflict is transformed into a real aggression, and in some cases such aggressive communications are converted into some forms of open violence. Science scrutinizes the various conflicts (military, religious, emotional, etc.) but those that are examined in depth are some that are most often observed in the human communication, such as the interpersonal.

Interpersonal conflicts are related to the contradictions between particular psychic processes, psychic statuses, distinctive features and attributes of the personality (Lee, 2008; Wall, Callister,

1995). We may add to them such as motivation conflicts, conflicts of the non-realized intention, role conflicts, adaptation conflicts as well as conflicts concerning non-adequate estimation. Basic structures of the inner personal world are motivations and experiences („I want "), values („ I am ought ", „I must/I should ") and self- assessment („I can" or „I cannot "). While a misbalance among these three structures occurs, interpersonal conflicts different in intensity, substance and duration emerge on the surface.

The conflicts between different personalities offer contradictions, division of opinions and clashes between people. These conflicts may be reduced to the level of persuasion conflicts; they are sources of the perception that different people apprehend one and the same situation in different ways and that each of the parties involved is an entirely complete believer in his/her personal individual understandings (Huseinagić, Hodžić,2010). These conflicts are studied in depth and are directly related to the objectivity of the human relations (Wall, Callister et al., 1995). Their specific features are as follows: 1) Availability of counter interests; 2) Awareness by each conflicting party of the existence of opposing interests; 3) Persuasion in both parties that the Other has harmed or will do harm the respective party's interests; 4) A fluent mutual interaction between the parties; and 5) Direct actions carried out by one or by both parties which are harmful for the interests of the other party (Andreeva, 2007). Basic attitudes and adjustments of the individual and the psychic status are strongly influencing on the conflicts and their specific development. Fundamental dipoles (bipolar attitudes) are attraction-repulsion, love-hate.

The various situational conflicts may impede efficiency and to avert one from the satisfaction to do the things s/he is doing. Each conflict may happen to be a threat for specific social relationships and for the stability of the social communications. Because of this, it is necessary to study the conflict causes and their effects on the personal emotions, cognitions, motivations and behaviors. The knowledge of the conflict and its evaluation in a holistic manner may be of help for the people involved in resolving them; and thus, to prevent their further escalation up to a destruction level. While exercising some activity and having in mind that nobody can be protected from such confusions, everyone may build up attitudes reactive towards such conflict situations, (Nauta, Kluver, 2004). Doing a special research, David Antonioni (1998) finds out that the people, who are thorough, attractive and open, may manage with skill and competence the conflict situation using constructive and acceptable behavioral models. The researcher makes reference to something more: by drawing attention to the fact that there is no perfect style of conflict management, he accents that every situation should be resolved individually and each one needs a special instrumentarium to be used for the purpose. According to Green, B., Leslie, A.D. and Marks, M. (2001), the management design of the group conflicts is influencing on effectiveness of the inner group relations and, so, occurs to influence on the conflict resolution processes, as a whole.

In a field like sport activity the trainers should be able to identify the motives for the conflicts that burst out among the sportsmen and the methods to overcome them. The study work of Laios A. and G. Tzetzis (2005) is based on a sample of 42 professional football, basketball and volleyball trainers inquired through questionnaires. The researchers have specified that the trainers use five conflict management styles and the most effective ones for team conflict resolution are: collaboration, compromise or avoid while the most non-effective are competition and accommodation. Huseinagić E. and A. Hodžić (2010) find out that majority of team trainers from the European championship think the conflicts should be avoided because they are unpleasant experiences in the everyday life and in sport as well. So, the conflicts may be solved administratively instead contractively and operationally.

Being a martial art, aikido offers a specific type of physical activity which is particularly influential on psychic and physical development of everybody who is practicing it; it contributes to the creation of a sense of readiness to find an individual conflict solution in each single personal value structure (Kaykov et al., 1998). Translated, the word aikido means method or way (do) to coordinate and harmonize (ai) the mental energy (ki). Ultimately, it is a doctrine for coordination of the body and mind, for their fusion, amalgamation of the individual physical and psychic capacities; as a result of these processes the individual practitioner becomes an all-round man (Drumev, Marinov, 2016). A. Nocque (1977) indicates that „aikido is a way practically to study, to apply to practice and to aware the laws concerning the unity of the body and spirit. This unity is implicitly necessary since the spirit expresses its power through the body".

The aikido game is ruled by strictly defined series of movements demonstrated in various techniques in order to neutralize the enemy. Each of them may turn aside and redirect, i.e. ward out, each attack, notwithstanding how mane attackers have launched it - one or several. This tactic is a method demonstrating how everyone may move from the basic consummative attitude to the self-defenceinstrumentarium toward a higher level. At such a higher stage the art itself is in a process of transformation into a system of teaching principles for coordination between the human Self and the environment in order to harmonize the oppositions; and clears up all doubts that it is an art aiming physical destruction. As B. Sosa and B. Robbins (1987) underlines, „aikido is occasionally termed a martial art without fight. The person who is training aikido should respect the body of his/her enemy. Despite the fact that many of its techniques are unhealthy and unsound, the pain is provisional and should never give birth to any lasting harms and damages".

Combined with the idea how to improve self-deference technical skills and the heightened physical activity, the entire aikido philosophy exercised some kind of ascendancy over us while discussing how to organize this study and how to define the working hypothesis. At the end of the day it is as follows:

We presuppose that the physical activity in the aikido game would friendly influence the process to form more harmonious structure of the whole set of techniques that are usually necessary to react to the conflicts.

The goal of the study is to prove the positive influence of the physical activity demonstrated in the aikido game over the ways in which one may react in case of conflict situation.

To achieve this goal and to confirm the working hypothesis we had to solve the following basic tasks:

1. To reveal the theoretical foundations of the problem focused on.
2. To reveal what is the real status of the tactics methods that are usually used by the respondents for reaction in conflict situations.
3. To analyze the results of the field work and to confirm or to reject the working hypothesis.

The respondent group consists, generally, of 64 youngsters aged from 18 to 24, divided into two groups: Control group of 24 young people who do not practice sport; and Experimental group of 40 young people that are actively practicing aikido in various clubs in this country.

2. Materials and Methods

During the research process we used complex methods, including literature resources study, observation and talk. In order to determine what are the typical ways to react to conflicts, we applied the approved Ken Thomas 'Test [Thomas-Kilmann Conflict Mode Instrument]. We used some given keys to determine the five instruments for reaction to conflict situations: 1) Competing; 2) Collaborating; 3) Compromising; 4) Avoiding; and 5) Accommodating. The information we obtained was processed via a variation analysis and a hypotheses test for independent samples (Student's t-criteria with guarantied probability P≥95,0 %).

3. Results and Discussion

After a detailed mathematic-statistic processing of the information collected during the field work we discovered the following regularities related to the ways of conflict reactions.

The young people from the Control group, who do not involve in sport activities, organize these reactive methods in the following configuration – see Figure 1.

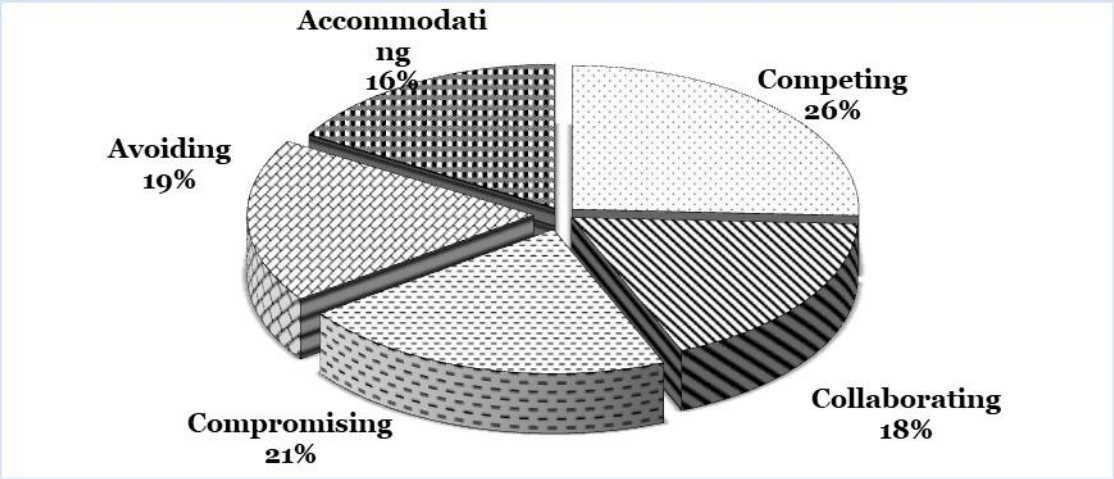

Fig. 1. Response tactics to conflicts in the group of sport non-involved young people

First rank, competing with average rating result of 7,42 units or 26 per cent of all conflict response methods. The respondents' preference for this tactics shapes a trend to manifest the personal Self of the individual actor. The respondents, who have answered in this manner, obviously prefer to manifest themselves in particular specific and tangible situations in their everyday life, includingly in conflict situations. Leading attitude here is to meet the requirements of their private exclusive interests and, so, disturbing the interests of the others. The choice of this conflict reaction mode almost always has a negative effect on their individual physics. The psychological tension increases and leads to anxiety, anger, hate and even abhorrence as experience. Naturally is has some negative reflections on the health of both conflicting parties and leads to health complications.

Second rank, compromising with average rating result of 6,17 units or 21 per cent in the Control group. This choice is an index that the non-involved with any sport activities young respondents have considered the motives and the arguments of the opposite party in the conflict and are striving to avoid the situation. Therefore, they take measures so as to not to meet the influence of its negative conflicting factors and to prevent the situation, respectively some event, from happening; more or less, the behavior is diplomatic and the reactions of the other party involved and, so, affected, do not increase the psychical tensions.

Third rank, avoiding with average rating result of 5,33 units or 19 per cent in the Control group. In this category the respondents try not to be directly involved in the arising conflict. They are in search for ways to avoid the conflict situation; however, they do not undertake any active action to resolve it. Using this instrument, they lose the capacity to achieve their own individual goals and give a chance to the other pretender, the opposite party in the game, to do so. This tactic is not quite favorable as far as the respondents demonstrate they are ready to get out of a situation which they should solve.

Fourth rank, collaborating with average rating result of 5,21 units and 18 per cent preference for it from all registered methods. It occurs to be the most positive way to react in such cases and is a search for finding all opportunities and possible capabilities to slow down the tension in the emerged conflict situation. In the majority of occurrences this predisposition helps them to manage the conflict situation and leads to the needed positive attitudes to play a hardball in the communication with the opposite game actor.

Fifth, last rank, accommodating with average rating result of 4,75 units and preferred by 16 per cent of the respondent group of sport non-involved young persons. Through the will registered for adaptation and adjustment to the situation, the non-involved in sport activities young people demonstrate that they are ready carefully to attend to the opponent stands, to debate the various arguments and to strive to understand their goal rationality. In most cases the individual personal skills, per se, to accommodate to the situation and to declare respect to the opposite party, involved in the conflict, create a priori a favorable climate for conflict resolution.

The so recognized and ranked pragmatic tactics for conflict reaction by the young people, who are not involved in sport activities, is of asymmetrical structure, non-harmonically

constructed, and may be considered as not quite perfect. In most of the cases the actions of the respondents may lead to plunging into the conflict situation, to extending and intensifying its negative effects which finally might lead to a stalemate without a chance to reveal any probable room for solution.

The configuration of the conflict reaction methods chosen by the Experimental group, consisting young people actively practicing aikido, is rather different – see Figure 2.

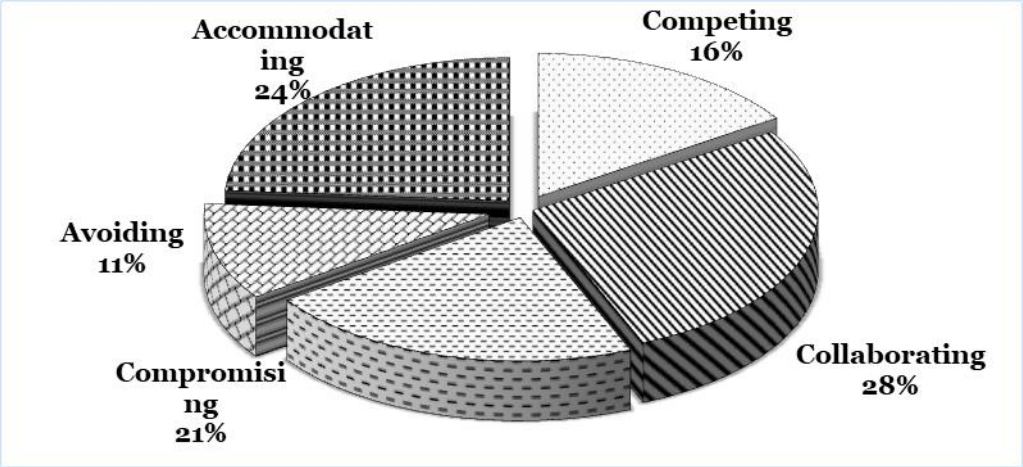

Fig. 2. Response tactics to conflicts in the group of actively involved in aikido game young people

First rank, collaborating with average 8,30 unites and 28 per cent preferred from all 5 methods in the Experimental group; thus this tactics is registered as the most preferred method for conflict reaction by the respondents who are involved in aikido training. The philosophy of aikido, grounded on the roads to harmony and lack of competing elements during the training practices, helps the people to build up a set of skills indicating unity, integrity and inner-structural arrangement and cohesion of collectivity, per se. At the end of the day this can be transferred in all activities in all other fields they are or maybe, in future, involved; and in case of any conflict situation this set of techniques subserves to obtain harmonious structure of the relationships with the other collectivities with whom they communicate. This is a positive drive which is built up on the idea to communicate, having in mind positive emotions and feelings such as understanding, good intensions, respect and demonstration of mutual good will; it has positive influences on the human health, as a whole. On the other hand, the lack of competition in aikido game eliminates the contest, per se, in pursuit of the dream „to be the first in the field" in any competition and, thus, the goal to be "victorious" at any rate is transformed into "let's do it together". There is a lack of dispute, not-agreement, and fight. There is a dominance of balance, harmony and solidarity, and unity.

Second rank, accommodating with average rating result of 7,08 units and 24 per cent preference in the experimental group of actively involved in aikido. The respondent arguments are associated with readiness to enter into the skin of the other party and neglecting one's individual interests. The respondents accept the opponent's viewpoint without confrontation. Keeping self-control is among the fundamental factors that are perceived, de facto, as guarantees for the effectivity in hard and tough situations – conflicts pertain to such situations. This sense of self-control may be created due to the determined strongly-willed aikido trainings and, so, it helps the practitioners to build up pragmatic skills allowing them to accept the conflict without any plunge into despair.

Third rank, compromising, with average rating result of 6,40 units and 21 per cent preference as a way to react in conflict situation. The rationally award compromise is among the tactical maneuvers in each conflict situations. The respondents are ready to make certain diplomatic moves back. In some cases, compromising is even necessary if the opponent manifest a correct behavior. The effect may even be better if both parties step back, at least partially, some of their goals in order to get a mutual agreement and to manage the conflict situation.

Fourth rank, competing, with average rating result of 4,68 units and 16 per cent preference. This tactics demonstrates one of the specific characteristics of the Self-concept; such an orientation is a manifestation that the goal of each party in the conflict is to verify identity validity of its

individual singular Self. However, predominantly its consequences are expressed by many different negative effects. In this case the respondents aim to make all possible efforts in order to obtain their goals and work hard on real fulfillment of this mental map, invest a lot of efforts, make attempts to demonstrate their individual rational thinking and advantages in front of the other.

Fifth rank, avoiding, with average rating result of 3,25 units and 11 per cent preference in the experimental group. This low rating permits us to think that aikido practitioners would undertake the activities, necessary to solve the conflict situation without seeking techniques how to avoid the conflict situation. This reaction tactics is typical for cases in which there is a need not to allow the conflict to extend and third parties to be involved in it. Thus, via avoiding, a chance is given to the opponent to take responsibility for the resolution of the emerged disputable problems.

Such a configuration of the methods used as a reaction to conflict situations possesses a harmonious structure. The activities of the young people practicing aikido are quite adequate to various conflict situations. Evidence we have found out in the significant distance between the data obtained in the Experimental group (EG) of the aikido training young people and the findings in the Control group (CG) of the non-involved in sport activities young people – Table 1 and Figure 3. After a variation analyses we have got the normal Gaussian division of the obtained values (the coefficients for As. and Ex. do not go above the critical values at the level of significance – $\alpha = 0.05$) (Brogley, Petkova, 1988). This has allowed us to compare the reactions of the respondents from both groups via each separate method for conflict reaction and to apply Student's t-criterion for independent samples.

Table 1. Response tactics to conflicts in both groups: of aikido non-involved (CG) and aikido practitioners (EG). Comparative data

№	Indices	CG		EG		D	d%	P%
	Reaction Tactics	$\overline{X}_1$	S	$\overline{X}_2$	s			
1	Competing	7,42	1,95	4,68	1,65	2,74	36.9	99,9
2	Collaborating	5,21	1,98	8,30	2,10	3,09	59.3	99,9
3	Compromising	6,17	1,76	6,40	1,68	0,23	3.7	40,1
4	Avoiding	5,33	2,06	3,25	1,06	2,08	39.0	99,9
5	Accommodating	4,75	1,51	7,08	1,86	2,33	49.1	99,9

First. Collaborating, as a result of systemic aikido training activities, occurs to be the preferred selected technique as a response to any conflict situations in the experimental group (EG). The average score in the respective respondent group is 8,30 units (S=2,10). The achieved result is with 3,09 units higher than the result achieved by the sport non-involved youngsters; the latter rate this criterion at third place with 5,21 units (S=1,98). This distinction is supported by a high guarantied probability (P=99,9 %). This shows 59,3 per cent higher probability the aikido practitioners' behavior to be more adequate to the emerged conflict situation and their capacity to track down painless path to going out of it. These young people would not permit themselves to get into contradiction and confrontation with the opponent. Their approach is directed to turning back to their own individual interests in return for obtaining communication harmony and preservation of good mutual relations.

Second. Accommodating: aikido practitioners' capability to adapt to the changing environment of the conflict situation is greater in comparison with the ability of non-involved in sport young people. The distinction in the accommodating index is backed by 2,33 units, i.e. P = 99,9 per cent. The first achievement – aftermath of effectivity of training burden during the various physical activities (including aikido), – is the capacity of the former respondents to quick accommodation of the separate systems and organs in the physical body. There is also adaptation

of the psychical and functional processes as a response to the high demands of the activity, per se. This shapes an adequate response signals for readiness and this readiness to react is carried over conflicting situations. The probability aikido training persons to adapt to conflict situation environment is of 49,1 per cent higher, which pragmatically means to be able to accept occurrences without being desperate and to act adequately to the opponent's behavior and actions.

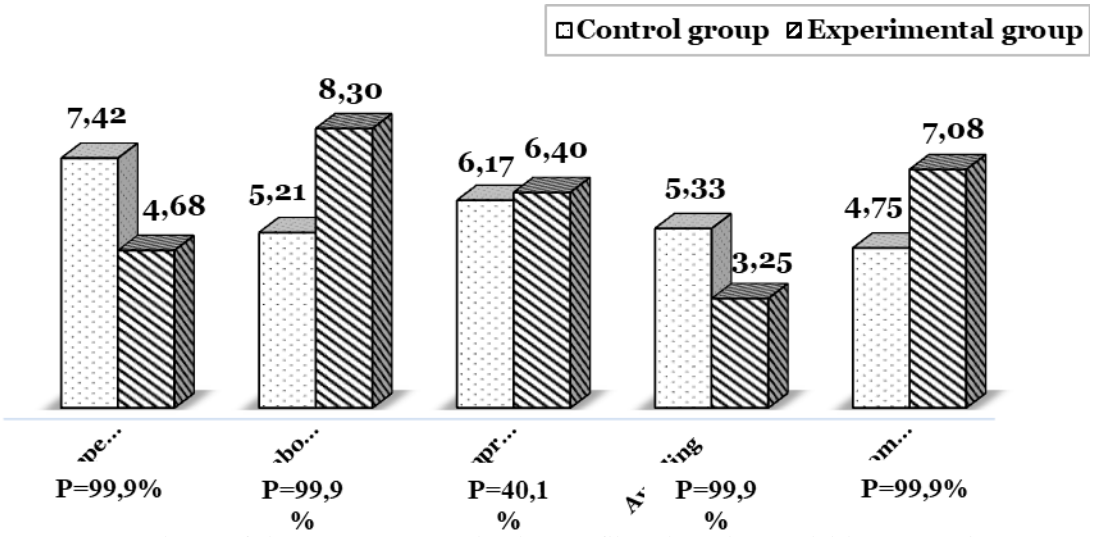

Fig. 3. Comparison of the response tactics in conflict situations within CG and EG

Third. Compromising: in some conflict situations rational compromise occurs to be a useful tool to exit the situation. There are some distinctions in average rating results registered in the data collected for this index; however, they are not reliable (P=40,1 per cent), and so, from this finding we may conclude that the attitudes towards this conflict reaction technique are of equal value.

Fourth. Competing is a response tactics in conflict situations preferred by the young people in the EG – $\overline{X}$ =4,68 (S=1,65) first for the CG – $\overline{X}$ =7,42 (S=1,95). The distinction is reliable (P=99,9 per cent). In this case non-involved young people accept conflict in a literal manner as a competing situation and enter into it with the will 'to win at any rate', irrespectively of the negative sequences. In the group of the aikido practitioners, the notion of competition is supported by high degree of will for collaboration and this is expressed by a respect to the opponent while communicating with him/her.

Fifth. Avoiding is characterized by lack of drive for solidarity, lack of trend to achieve one's personal aims and non-acceptance, even rejection, of responsibility; it appears as one of the unfavorable ways to react to the conflicts. In the control respondent group, the tendency to avoid is more clearly expressed than in the experimental respondent group. The distance between both groups as average rating results here is 2,08 units under P=99,9 per cent. The youngsters with higher physical aikido activities improve their will and tenacity; they are striving to achieve the goals and to assume definite responsibilities.

4. Conclusion

The analysis made is defining some generalizations: the young people practicing aikido have more harmonious structure of the tactics configuration to be used as a response to conflict situations in comparison to the group of non-involved in sport their peers. They are inclined to cooperation, adaptation and reasonable compromise in such environment. Competition and avoidance are not preferable techniques to resolve conflicts. In the same time non-involved in sport activities young people are governed by attitudes for competiveness; sometimes their behavior and motives are commanded by the will to make compromise and the desire to avoid conflicts to which it is difficult to adapt.

The research working hypothesis has been proved: physical activities involved in the aikido game favorably affect formation of more harmonious tactical configuration of the response in conflict situations. The challenge for us in future will be to study the methods for conflict resolution

in collective sports such as football, basketball, volleyball, etc., as well as whether there are gender distinctions registered. This will be an object of our further researches.

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Published in the Slovak Republic
European Journal of Medicine
Has been issued since 2013.
E-ISSN: 2310-3434
2018, 6(1): 52-58

DOI: 10.13187/ejm.2018.1.52
www.ejournal5.com

Polarimetry and Spectral Methods in Prediction of Children Disease after Adhesive Peritonitis

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Abstract

The aim of the study was to establish objective parameters of the field of laser and incoherent radiation of different spectral ranges (UV, visible, IR) as a non-invasive optical method of interaction with different samples of biological tissues and fluids of patients to determine the dynamics of peritonitis and choosing the best personal treatment. The objects of study were selected venous blood plasma of children and rats, histological sections of rat intestine in the postoperative period. As diagnostic methods have been used ultraviolet spectrometry samples of blood plasma in the liquid state, infrared spectroscopy middle range (2.5–25 microns) dry residue of plasma polarization and laser diagnostic technique of thin histological sections of biological tissues.

Keywords: peritonitis, biological tissues, blood plasma, UV spectroscopy, IR spectroscopy.

1. Introduction

The problem of adhesion formation, despite the long period of intensive study, and a large number of publications on this topic (Saravelos, 2007; Nogales, 2008), remains valid Postoperative adhesions break sharply quality of life for millions of people around the world, leading to complications with repeated operation accesses to the development of small bowel obstruction and chronic abdominal and pelvic pain. Currently, the basis of laboratory diagnosis of acute surgical diseases of the abdominal cavity is detecting changes of blood components as a quantitative values, and in a good state that accompany the development of inflammatory and destructive processes at both the local and at the system level. However, the accuracy of such a diagnosis is less than 70 %, as these criteria are obviously not characterized as inflammatory – destructive process as an individual response to it in each case. Therefore, the search for new, informative diagnostic parameter of acute surgical diseases of the abdominal cavity using non-invasive techniques such as spectral and polarization laser diagnostic techniques is becoming increasingly important (Nogales, 2008).

Also, equally important is to find simple and objective criteria that would provide an opportunity to assess the effectiveness of patient care in the postoperative period. In this paper examined the use for these purposes spectrometry method (Guminietsky, 1997), based on studies of UV absorption spectra of plasma venous blood as at the time of admission and in the postoperative period.

The aim of the study was to establish objective parameters of the field of laser and incoherent radiation of different spectral ranges (UV, visible, IR) as a non-invasive optical method of interaction with different samples of biological tissues and fluids of patients to determine the dynamics of peritonitis and choosing the best personal treatment. The objects of study were selected venous blood

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plasma of children and rats, histological sections of rat intestine in the postoperative period. As diagnostic methods have been used ultraviolet spectrometry samples of blood plasma in the liquid state, infrared spectroscopy middle range (2.5-25 microns) dry residue of plasma polarization and laser diagnostic technique of thin histological sections of biological tissues.

2. Materials and methods

In this section reviewed the results of spectrophotometric method, based on studies of UV absorption spectra of plasma venous blood as at the time of admission and in the postoperative period. In total, the study involved 91 patients, operated on various forms of peritonitis, appendiceal origin. Among them were 44 children with local forms of peritonitis, 25 with diffuse peritonitis, 22 with diffuse peritonitis. 30 patients constituted the comparison group (combined treatment by traditional methods); 61 – the main group, they performed traditional treatment combined with the use of ozonized saline solution (0.9 % NaCl). The control group consisted of 28 children operated on inguinal hernia. According to a retrospective analysis conducted by us, 318 patient records (2001-2010), the NCS in 73.75 % of cases observed at the age of 7-14 years, as a control, primary and comparison group included patients of appropriate age.

The main group is divided into two subgroups:

- subgroup l (n = 31) – children, operated on the HVZ abdomen, which besides standard treatment was carried total blood irrigation of the abdominal cavity ozonized saline (0.9 % NaCl);
- subgroup 2 (n = 30) – children, operated on the HVZ of the abdominal cavity, in which the standard treatment is supplemented with intraoperative lavage of the abdominal cavity ozonized saline and intravenous ozonized saline (0.9 % NaCl).

Ozonation of saline 0.9 % NaCl treated by ozonation ozonizer for UM-80 for 15 min. When washing the abdominal cavity a solution with a concentration of 5 mg/liter in volume of 2 liters was used. The solution was removed from the abdominal cavity after washing. Used for intravenous solution with a concentration of 3-5 mg/l. Speed intravenous is 7 ml/min.

The object of study during ultraviolet spectrometry (UV-spectrometry) was venous blood plasma of patients who acted in BSMU in City Children's Hospital with the clinical peritonitis, appendiceal origin. Since blood plasma is optically inhomogeneous medium, the spectrophotometric studies to determine its absorption to account for light scattering technique we used spherical photometer with a prefix to the spectrophotometer SF-4 and SF-5[15-16]. The spectral resolution setup was 1.5-2.0 nm.

We studied the spectra of transmittance τ in the spectral range λ = 220-320 nm every 5 nm, followed by calculation of the optical density D according to the formula D =-lgτ. The choice of spectral range was made due to the fact that the electronic absorption spectra of all organic constituents of blood plasma are in the X <320 nm (Angelsky, 2006; Angelsky, 2003; Angelsky, 2002). Blood plasma taken from a peripheral vein and diluted with distilled water at a ratio of 1:100, filled with quartz k'yuvetu thickness of 1 cm and placed it in the center of the spherical photometer. The relative error of the obtained values of D at these experimental conditions in the long-wave maximum D absorption at λ, = 280 nm not more than 0.5 %. All measurements were carried out on fresh samples (up to 3 days), which prior to studies were stored at low temperature.

3. Results and discussion

It is shown that the development of acute surgical diseases of the abdominal cavity growth occurs mainly optical density D plasma compared to control (for donors) in the λ = 280 nm. Next, the results of researches UV absorption spectra of blood plasma and their changes during development of peritonitis, as in the case of severe inflammatory diseases relevant to in vivo studies are not possible. Measured absorption spectra of blood plasma in the spectral range λ = 220-320 nm, but since the information is an area adjacent to λ = 280 nm, the paper analyzed the results in the range λ = 250-320 nm.

Thus, the study of the spectral dependence of the optical density of plasma venous blood from wavelength measurements showed that the wavelength λ = 280 nm is a maximum optical density. From our point of view the dynamics of change of optical density plasma of venous blood in the λ = 280 nm is associated with the activity of plasma globulin, which include fibrinogen, which promotes the formation of adhesions in the abdominal cavity. This makes it possible to predict the development of adhesions in the abdominal cavity. Therefore, this spectral maximum

can be selected for testing of differences in optical density plasma used in the diagnostic evaluation of treatment methods.

The basis of spectroscopic methods is the measurement of the intensity of absorption, emission or scattering of light by a substance from the light frequency (or wavelength) (Angelsky, 2001; Angelsky, 2002). In optical absorption spectroscopy are used in infrared and ultraviolet regions, as well as Raman spectroscopy and luminescence spectra. Each spectrum corresponds to a certain wavelength. In different areas of light absorption has a different nature. Absorption of energy within the optical spectrum varies the rotational, vibrational energy of molecules or excitation energy external valence electrons. To improve the rotational energy of the molecule is relatively small enough energy corresponding absorption lies in the far infrared (IR) region (the region of large wavelengths). To increase the vibrational energy of the molecule (to excite vibrations of atoms relative to each other) need large quantities of photons and the absorption is in the near infrared region. Even greater quanta of energy required to excite outer electrons of the molecule – absorption in the visible and ultraviolet region. The method of spectroscopic analysis was used to determine the amount of the substance. The Beer-Lambert law – one of the fundamental laws of spectroscopy, says, "The absorption of light is proportional to the number of molecules absorbing material on its way".

In the IR spectrum are four areas: I – visible, 2 – near, 3 – and 4 fundamental, far away. To study the organic matter is most important fundamental infrared region, which lies in the range from 5000 to 200 cm^{-1}. Lines in the 600-1300 cm^{-1} are excellent and even specific related molecules, so it is called the area "fingerprint" of the molecule. In this area fall into a single bond stretching vibrations of C=O, C=N, N=O. On the contrary, the absorption frequency of double bonds C=C, C=O, N=O was characterized, that is a little different for different molecules and lie in the 1500-1950 cm^{-1}. Absorption of triple bonds is even more short-wave region (for C=C at 2100-2250 cm^{-1}) IR spectrum is quite specific property of each chemical compound, as well as position isomers, geometric isomers and molecules containing protons have different spectra. In this regard, the IR spectrum of each substance has a distinct pattern.

IR spectra were used to identify the compounds and establish their degree of purity (quality) and they can be used for qualitative analysis of mixtures for controlling the progress of the reaction. However, the most common and important application of IR spectra is the clarification and confirmation of the intended structure of the compounds. The presence of almost any functional group in a molecule can be set through them. Also the possibility of using infrared spectroscopy and quantitative analysis is considered.

In our experimental setup spectrum recorded automatically and used to measure infrared spectrophotometer “Specord 80/851R” provided the photometric accuracy of ± 0,2 %. The control program embedded computer prevents incorrect and inconsistent parameter provides a linear correlation between basal line 10 wave numbers.

These factors ensure objectivity and high precision spectroscopic analyzes.

The current level of knowledge in the field of spectroscopy can determine the appearance of new ("pathological") and the disappearance of "normal" chemical substances (ATP, GTP, UTP, etc.) that are in various pathological conditions subject to rapid metabolism to form the corresponding hydrolysis products – monophosphate. Also it is possible to determine the concentration of protein molecules and phosphatides in the process of disintegration of cellular structures. In the studied area the infrared spectrum of fall and valence bonds free radicals.

Thus, infrared spectroscopy can be used to determine the level of several metabolites formed during pathological changes initiated by the inflammatory process and thus to assess the degree of metabolic processes.

In our opinion the most important is not to determine the amount of a substance (including a huge number of them), which is found in the blood of a sick child, and the correlation integral indicators infrared spectroscopic analysis of the severity of the inflammatory process and thus the level of metabolic disorders.

As an object of research using blood serum (healthy) children, prepared for spectral analysis (Angelsky, 2005; Angelsky, 2012; Guminestskij, 2006; Angelskaya, 2013). Serum was dried at room temperature for half – a petri dish. The dry residue was injected into the liquid paraffin and received a suspension, which is then subjected to IR-spectroscopy recording absorption spectra in the region 1200-1000cm^{-1}. Upon receipt of spectrogram peaks measured the height of the

absorption bands with maxima at 1170, 1 165, 1 150, 1140, 1130, 1100, 1070, 1025 cm^{-1} and calculated the average height of all peaks - C. Then determine the ratio of each peak prior to the subsequent: 1170/1165, 1165/1150, 1150/1140, 1140/1130, 1130/1100, 1100/1070, 1070/1025.

Informative values denote conditional symbols (M, m, D, c, R, x, S). Index M is the largest value obtained, and the index m - minimum. Also introduced an additional parameter – D, which is the difference between M and N. The mean of all values marked as indicator – p. Also calculated value of R – the ratio of peak height with a maximum at 1165 cm^{-1} to the height of the peak maximum at 1170 cm^{-1}, the value of x – the ratio of peak height with a maximum at 1130 cm^{-1} and the average value of the heights of peaks (C) and size S – the ratio of peak height with a maximum at 1100 cm^{-1} to the mean value of height (C).

In Fig. 1-4 is shown the infrared spectra of plasma venous blood of patients 1 and 2 on the 3rd and 7th day of treatment. Aligned spectra are characterized by absorption bands, among which we isolated two – first 1000-1800 cm^{-1} and the second 2800-3500 cm^{-1}. Characteristic was the difference in the behavior of the absorption spectrum in the region 1300-1700 cm^{-1} for patients of the 2nd subgroup 2 for different periods of treatment.

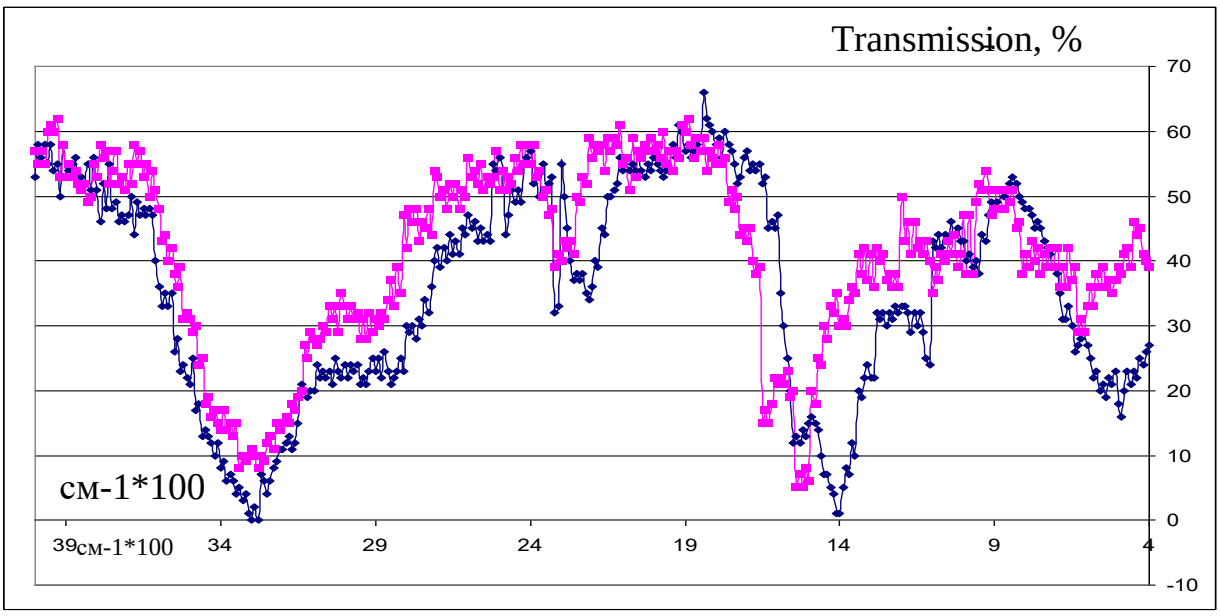

Fig. 1. IR transmission spectra of plasma venous blood of patients in group 2–1st subgroup on the 3rd and 7th day of treatment

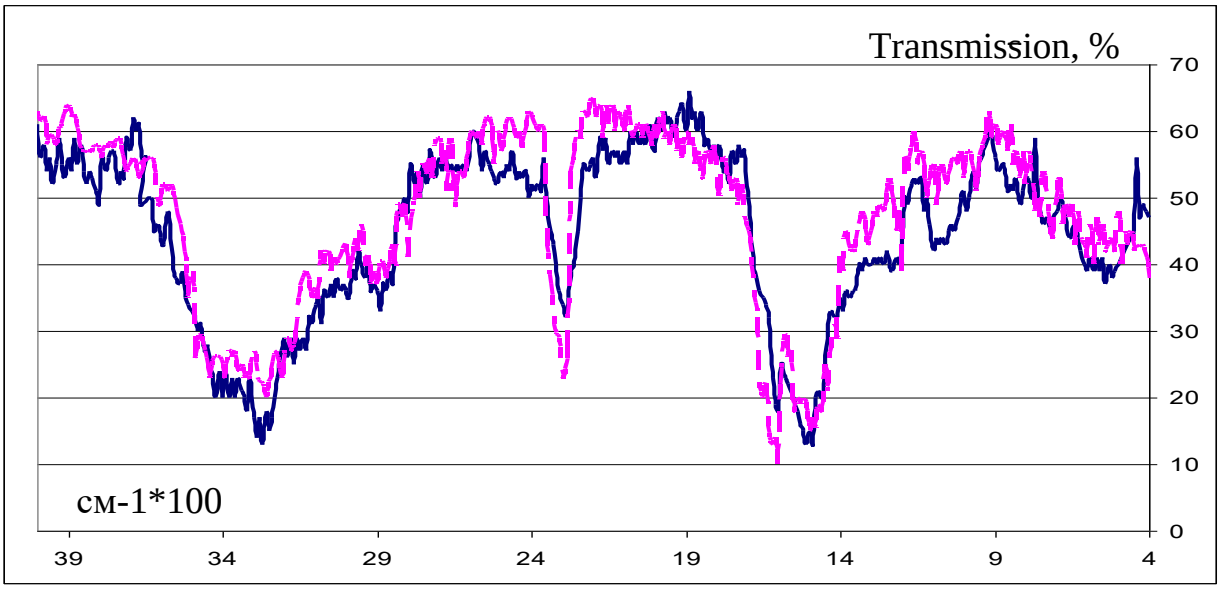

Fig. 2. IR transmission spectra of plasma venous blood of patients in group 2–2nd subgroup on the 3rd and 7th day of treatment

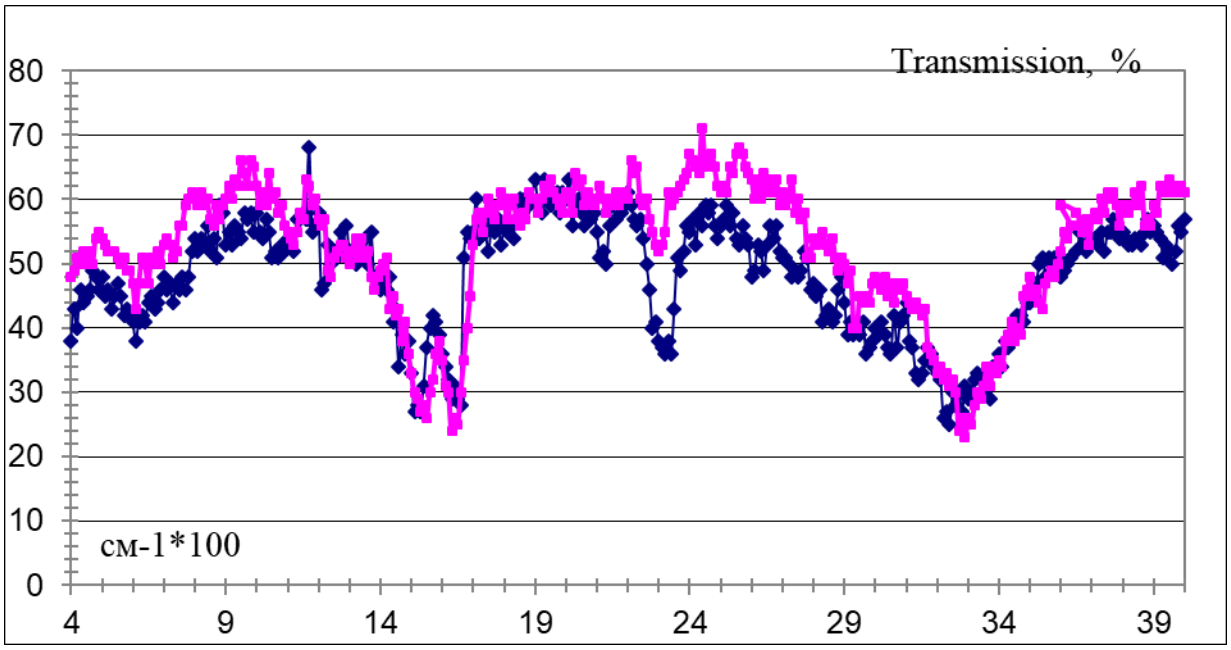

Fig. 3. IR transmission spectra of plasma venous blood of patients in group 1 on the 3rd and 7th day of treatment

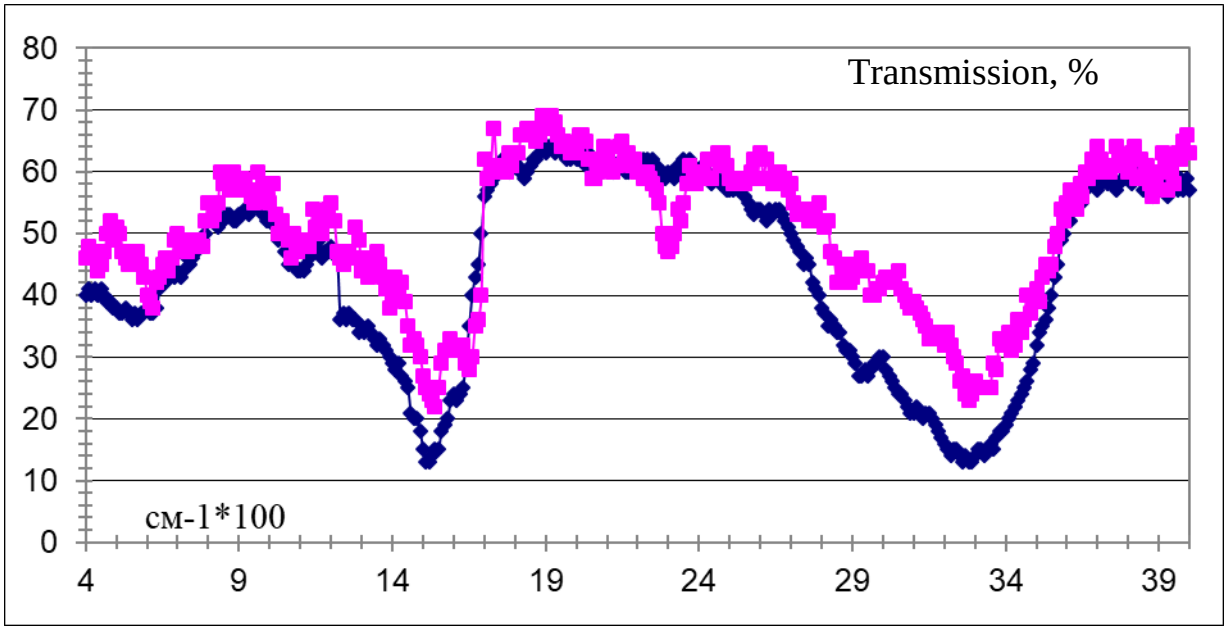

Fig. 4. IR transmission spectra of plasma venous blood of patients in the control group at 3 rd and 7 th day of treatment

In order to determine the values of IR spectroscopic parameters determined their range of children patients at the 3rd and 7th day of treatment. To do this, select the numerical values of each of the parameters in Table 1.

Table 1. Numerical values for IR- spectroscopy in children with peritonitis.

Values	IR spectroscopic parameters					
	D	M	*m*	R	S	*x*
Control group	0,55	1,23	0,30	0,43	0,50	0,45
1-st group	2,50	2,88	0,70	0,88	0,80	0,86
1st group, subgroup 1	0,75	3,23	0,10	0,4	0,52	0,5
2st group, subgroup 2	1,50	1,88	0,25	0,38	0,58	0,6

The results showed that using the method of IR spectroscopy in the frequency range 400-4000 cm^{-1} revealed the quantitative parameters of the degree of absorption of blood plasma of patients in different bands that will in the future conduct rapid analysis of the patient during his treatment at the time and technique to make choices treatment.

By using the method of IR spectroscopy were able to establish the effectiveness of the combined use of ozonized saline over the first 2 groups and 2 subgroups (washing + in/in the introduction of ozone) compared with the first subgroup (washing with ozone) 2 groups and nearly approach the targets in relation to prediction formation of adhesions in the abdominal cavity.

4. Conclusion

1. Thus, our study in this paper the spectral dependence of the optical density of plasma venous blood from wavelength measurements showed that the wavelength λ = 280 nm is a maximum optical density. From our point of view the dynamics of change of optical density plasma of venous blood in the λ = 280 nm is associated with the activity of plasma globulin, which include fibrinogen, which promotes the formation of adhesions in the abdominal cavity. This makes it possible to predict the development of adhesions in the abdominal cavity. Therefore, this spectral maximum can be selected for testing of differences in optical density plasma used in the diagnostic evaluation of treatment methods.

2. Using infrared spectroscopy in the frequency range 400-4000 cm^{-1} revealed the degree of absorption of quantitative parameters of blood plasma of patients in different bands that will in the future conduct rapid analysis of the patient during his treatment in time and make a choice method of treatment. By using the method of IR spectroscopy were able to establish the effectiveness of the combined use of ozonized saline over the first 2 groups and 2 subgroups (washing + in/in the introduction of ozone) compared with the first subgroup (washing with ozone) 2 groups and nearly approach the targets in relation to prediction formation of adhesions in the abdominal cavity.

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