

Vortex Power Spring Water: Physical-Chemical Qualities of this Water compared to Mountain and Melt Water from Bulgaria, Russia and Glacier Rosenloui from Swiss Alps

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Abstract

In this research by using IR, NES, and DNES methods were investigated various samples of water from Bulgarian water springs: the melt water from Glacier Rosenloui, 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 Vortex Power Spring device (Vortex Power Company) for improvement of quality of tap water by saturation with oxygen. 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 extremum in the DNES-spectra of various samples of water and the human blood serum at $E = -0.1387$ eV and $\lambda = 8.95$ 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). In frames of the research was carried out the computer calculation of elemental polyhedral nanoclusters with a formula $(H_2O)_n$, where $n = 3-20$. Based on this data some important physical characteristics of water were obtained, e.g. the average energy of hydrogen bonding between H_2O molecules in the process of cluster formation was measured by the DNES method compiles -0.1067 ± 0.0011 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-}), chemical-physical parameters (pH, electroconductivity) and the decreased content of deuterium in studied water samples renders beneficial effects of this type of water on human health. Also our research has confirmed the efficiency of water treatment with the Vortex Power Spring apparatus for increasing the levels of oxygenation. Water subjected to the water treatment with the Vortex Power Spring apparatus has a higher permanent oxygen levels (1.26 ± 0.13 mg O_2/dm^3) relative to the control (0.9 ± 0.09 mg O_2/dm^3).

Keywords: deuterium, longevity, mountain water, IR, NES, DNES, Glacier Rosenloui, Vortex Power Spring.

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) in the growth and proliferation of prokaryotes and eukaryotes in water with different isotopic content (Mosin & Ignatov, 2012; Ignatov & Mosin, 2013; 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 H_2O molecule create the favorable conditions for formation of electrostatic intermolecular van der Waals, dipole-dipole forces and donor-acceptor interaction with transfer of electric charges between H-atoms and O-atom in H_2O molecules, binding them into water associates (clusters) with the general formula $(H_2O)_n$ where n varies from 3 to 50 units (Saykally, 2005; Ignatov, Mosin, 2013).

Other important indicator of water quality is its isotopic composition. The natural water consists on 99.7 mol.% of $H_2^{16}O$, which molecules are formed by 1H and ^{16}O atoms (Mosin & Ignatov, 2012). The remaining 0.3 mol.% is represented by isotope varieties (isotopomers) of water molecules, wherein deuterium forms 6 configurations of isotopomers – $HD^{16}O$, $HD^{17}O$, $HD^{18}O$, $D_2^{16}O$, $D_2^{17}O$, $D_2^{18}O$, while 3 configuration are formed by isotopomers of oxygen – $H_2^{16}O$, $H_2^{17}O$, $H_2^{18}O$.

The aim of this research was studying the various samples of water from Bulgarian water springs: the melt water from Glacier Rosenloui, Swiss Alps compared to Vortex Power Spring water (obtained after the water treatment by a Vortex Power Spring device), 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.

2. Material and Methods

2.1. Preparation of Water Samples with Varying Deuterium Content

For preparation of water samples with varying deuterium content we used D₂O (99.9 atom%) received from the Russian Research Centre "Isotope" (St. Petersburg, Russian Federation). Inorganic salts were preliminary crystallized in D₂O and dried in vacuum before using. D₂O distilled over KMnO₄ with the subsequent control of deuterium content in water by ¹H-NMR-spectroscopy on Bruker WM-250 device ("Bruker", Germany) (working frequency – 70 MHz, internal standard – Me₄Si) and on Bruker Vertex ("Bruker", Germany) IR spectrometer (a spectral range: average IR – 370–7800 cm⁻¹; visible – 2500–8000 cm⁻¹; the permission – 0,5 cm⁻¹; accuracy of wave number – 0,1 cm⁻¹ on 2000 cm⁻¹).

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 $\lambda = 580 \pm 7$ nm. The device measures the angle of evaporation of water drops from 72.3⁰ to 0⁰. The DNES-spectrum was measured in the range of -0.08– -0.1387 eV or $\lambda = 8.9–13.8$ 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 between H₂O molecules in water samples and human blood serum.

2.3. Vortex Power Spring Water Experiments

The experiments for improvement of quality of tap water by saturation with oxygen were performed on a Vortex Power Spring nuzzle device (Vortex Power Company). The experiment was performed with water samples subjected to rapid water treatment in the Vortex Power Spring nuzzles. The permanent oxidation was measured as quantity of oxygen needing for oxidation of organic compounds in a volume of the treated water (mg O₂/dm³).

2.4. Studying the Bulgarian Long Lived People and Centenarians

Interviews have been conducted with 415 Bulgarian centenarians and long lived 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.

2.5. Studying the Human Blood 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.

2.6. IR-spectroscopy

IR-spectra were registered on Bruker Vertex ("Bruker", 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 (K. Chakarova).

2.7. Statistical Processing of Experimental Data

Statistical processing of experimental data was performed using the statistical package STATISTISA 6 using the Student's *t*- criterion (at $p < 0.05$).

3. Results and Discussions

3.1. Comparative Analysis between Longevity of Long Lived People, Centenarians and Their Siblings

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 1 demonstrates the interrelation between the year of birth (1912–1924) of long living people (age) and their number (Teteven municipality, Bulgaria).

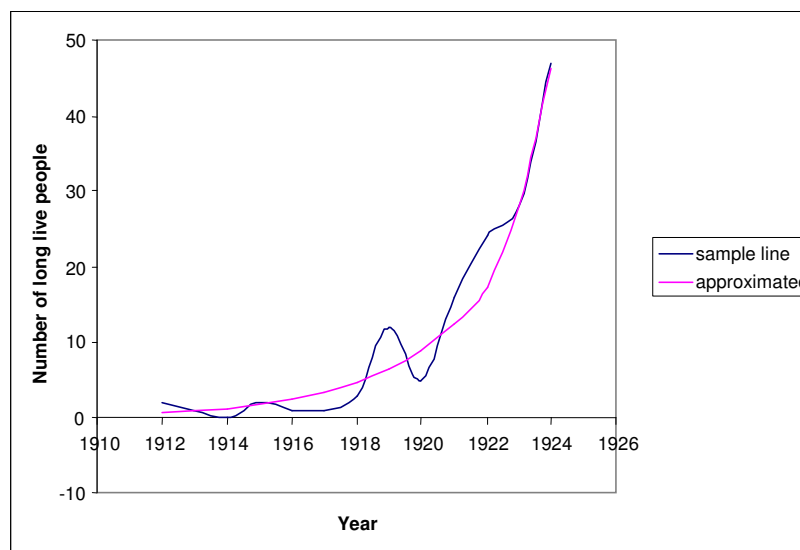


Figure 1: Interrelation between the year of birth of long living people (age) and their number in Teteven municipality, Bulgaria.

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 2 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.

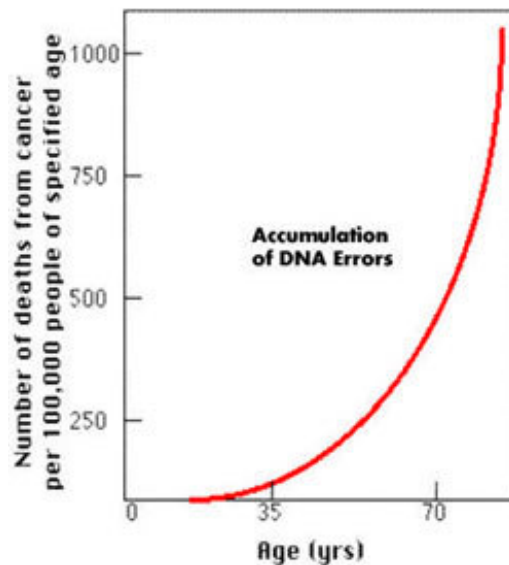


Figure 2: Interrelation between age and the number of cancer patients (Orgel, 1963).

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.

The analogous situation is observed in the Russian North. According to G. Berdishev (Berdishev, 1989), people inhabiting the Russian North – the Yakuts and the Altaians as well as the Buryats, drink the mountain water obtained after the melting of ice. Altai and Buryat as well as Kavkaz water sources are known as moderately warm, with temperatures of +8–10 °C; the water is generally ice-free in winter. This phenomenon is explained by the fact that the melt water contains a low percentage of deuterium compared with an ordinary tap water that is believed to have a positive effect on the tissue cells and metabolism. The melt water in Russia is considered to be a good folk remedy for increasing physical activity of the human body, enhancing the vitality of the organism and thought to have a beneficial effect on metabolism (Goncharuk *et al.*, 2013). In the world are also popular the water sources containing the melt water from Canada, Norway, Island and Alaska.

3.2. Clinical Evidence 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 0, and the diameter of water drop basis is only slightly altered, that is a new physical effect (Antonov, 1995; Antonov & Yuskesseliya, 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 H₂O molecules according to the value of $f(\theta)$. The distribution function is denoted as the energy spectrum of the water state. Theoretical research established the dependence between the surface tension of water and the energy of hydrogen bonds among individual H₂O-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:

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

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

The relation between the wetting angle (θ) and the energy (E) of the hydrogen bonds between H₂O molecules is calculated by the formula:

$$\theta = \arccos(-1 - 14.33E), \quad (2)$$

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 $\Delta f(E) = f(\text{samples of water}) - f(\text{control sample of water})$ – 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. The redistribution of H₂O molecules in water samples according to the energy is a statistical process of dynamics.

Figure 3 shows the average NES-spectrum of deionised water. On the X-axis are depicted three scales. The energies of hydrogen bonds among H₂O molecules are calculated in eV. On the Y-axis is depicted the function of distribution of H₂O molecules according to energies $f(E)$, measured in reciprocal unit eV^{-1} .

Arrow A designates the energy of hydrogen bonds among H₂O molecules, which is accepted as most reliable in spectroscopy.

Arrow B designates the energy of hydrogen bonds among H₂O molecules the value of which is calculated as:

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

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.

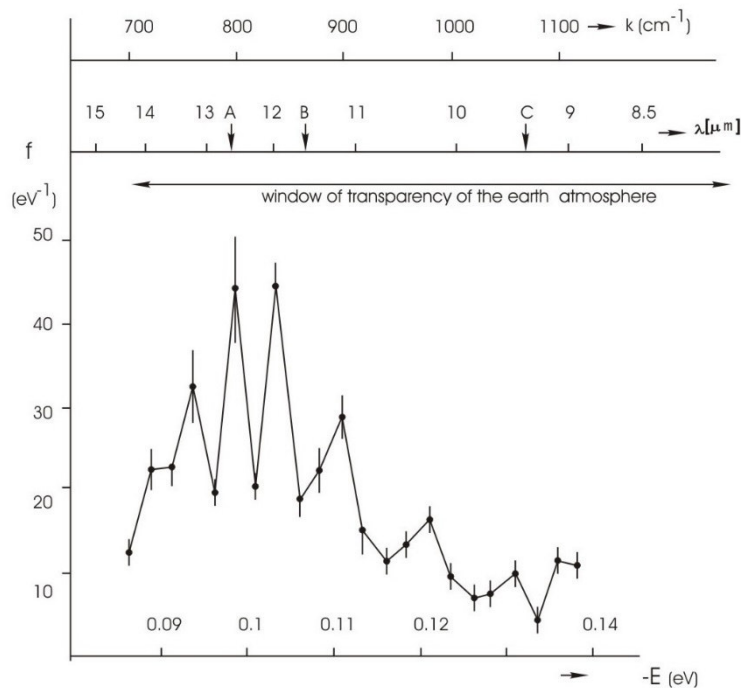


Figure 3: The NES-spectrum of deionized water (chemical purity – 99,99 %; pH – 6,5–7,5; total mineralization – 200 mg/l; electric conductivity – 10 μ S/cm): the horizontal axis shows the energy of the H...O hydrogen bonds in the associates – E (eV); the vertical axis – the energy distribution function – 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 H₂O 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 blood serum and NES-spectrum of deionized water control sample – DNES-spectrum, measured as the difference $\Delta f(E) = f(\text{samples of water}) - f(\text{control sample of water})$. The DNES-spectrum obtained from the first group has a local extremum energy ($\Delta E_{H...O}$) at $E = -9.1 \pm 1.1$ meV and from the second group at $E = -1.6 \pm 1.1$ 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 $\lambda = 8.95$ 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 $\lambda = 8.55, 8.58, 8.70, 8.77, 8.85, 9.10, 9.35$ and 9.76 μm (Krasnov, Gordetsov, 2009). The resulting peak at $\lambda = 8.95$ μm in IR-spectrum (Ignatov, 2012) approaching the peak at $\lambda = 8.85$ μm monitored by Russian researchers. In the control group of healthy people the average value of the energy distribution function $f(E)$ at $\lambda = 8.95$ μm compiles 75.3 eV, and in a group of people in critical condition – 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 $f(E) \sim 24.1$ eV⁻¹. The level of reliability of the results is $p < 0.05$ according to Student's t -test. In 1995 were performed DNES-experiments with impact on tumor mice cells in water containing Ca²⁺ (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 H₂O molecules according to energies at -0.1387 eV decreases. In this group of tested people the result was obtained by DNES-method at -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 $\lambda = 8.95$ 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 Ca²⁺ 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 Ca²⁺ 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 maximum 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, 2013c). 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 zeolite.

3.3. Composition of water in the mountain area in Teteven municipality, in Stara Planina Mountain, Kuklen municipality, Rhodopes Mountain (Bulgaria), and Glacier Rosenloui, Swiss Alps

The statistical data demonstrated that the difference between the age of long lived people in mountain and plain areas is reliable at the Student's t -criteria ($p < 0,05$) with a confidence level of $t = 2.36$. The analyses of water samples obtained from various water sources show the differences regarding the chemical composition, hardness, local extrema in NES-spectra of water eV⁻¹ at $(-0.1362$ – -0.1387 eV), and the isotopic shifts of D/H in water.

Tables 1, 2, 3 and 4 show the chemical composition of mountain water springs in Teteven, Kuklen (Bulgaria) and Glacier, Rosenloui, Swiss Alps and local extremums in NES-spectra of water samples. A new parameter is entered into Tables 1, 2 and 3 – a local extremum of energy at $(-0.1362$ – -0.1387 eV). This value was determined by the NES-spectrum as function of distribution of individual H₂O molecules according to energy $f(E)$. The function of distribution of H₂O molecules according to energy $f(E)$ for tap water in Teteven is 11.8 ± 0.6 eV⁻¹ (Table 1, 2, 3).

Table 1: The composition of mountain water springs in Zlatishko-Tetevenska Mountain (Teteven municipality, Bulgaria) and local extremums in NES-spectra of water

Indicators	Results of the research (mg/l)	Norm
Sodium (Na^+)	0.96	< 200
Calcium (Ca^{2+})	100.4	< 150
Magnesium (Mg^{2+})	12.65	< 80
Iron (Fe)	0.016	<0.2
Manganese (Mn^{2+})	0.0018	<0.2
Zinc (Zn^{2+})	0.18	<4.0
Sulfates (SO_4^{2-})	81.8	< 250
Chlorides (Cl^-)	3.96	< 250
Carbonates (CO_3^{2-})	< 2.0	-
Hydrocarbonates (HCO_3^-)	184.0	-
Other values	Results	
Active reaction (pH)	7.9 alkaline	6.5-9.5
Electroconductivity	536.8 $\mu\text{S}/\text{cm}$	< 2000
Hardness of water	16.5 dH hard	<33.7
Local extremum* eV^{-1} at (-0.1362--0.1387 eV)	36.9	>24.1

*Function of distribution of H_2O molecules according to energy $f(E)$.

Table 2: The composition of mountain water springs in Vasiliovska Mountain (Teteven municipality, Bulgaria) and local extremums in NES-spectra of water

Indicators	Results of the research (mg/l)	Norm
Sodium (Na^+)	4.5	< 200
Calcium (Ca^{2+})	55.5	< 150
Magnesium (Mg^{2+})	2.28	< 80
Iron (Fe)	0.0127	<0.2
Manganese (Mn^{2+})	0.0014	<0.2
Zinc (Zn^{2+})	0.006	<4.0
Sulfates (SO_4^{2-})	16.9	< 250
Chlorides (Cl^-)	3.4	< 250
Carbonates (CO_3^{2-})	< 2.0	-
Hydrocarbonates (HCO_3^-)	118.0	-
Other values	Results	
Active reaction (pH)	7.4 alkaline	6.5-9.5
Electroconductivity	285.0 $\mu\text{S}/\text{cm}$	< 2000
Hardness of water	7.9 dH slightly hard	<33.7
Local extremum* eV^{-1} at (-0.1362--0.1387 eV)	40.1	>24.1

*Function of distribution of H_2O molecules according to energy $f(E)$.

Table 3: The composition of mountain water spring Eco Hotel Zdravetz, Rhodopes Mountain (Kuklen municipality, Bulgaria) and local extremums in NES-spectra of water

Indicators	Results of the research (mg/l)	Norm
Sodium (Na ⁺)	7.6	< 200
Calcium (Ca ²⁺)	3.5	< 150
Magnesium (Mg ²⁺)	0.63	< 80
Iron (Fe)	0.007	<0.2
Manganese (Mn ²⁺)	0.002	<0.2
Zinc (Zn ²⁺)	0.007	<4.0
Sulfates (SO ₄ ²⁻)	26.8	< 250
Chlorides (Cl ⁻)	3.00	< 250
Carbonates (CO ₃ ²⁻)	< 2.0	-
Hydrocarbonates (HCO ₃ ⁻)	21.3	-
Other values	Results	
Active reaction (pH)	5.93 normal	6.5-9.5
Electroconductivity	536.8 μS/cm	< 2000
Hardness of water	1.4 dH soft	<33.7
Local extremum* eV ⁻¹ at (-0.1362--0.1387 eV)	59.3	>24.1

*Function of distribution of H₂O molecules according to energy f(E).

Table 4: The composition of mountain melt spring Glacier Rosenloui, Swiss Alps and local extremums in NES-spectra of water

Indicators	Results of the research (mg/l)	Norm
Sodium (Na ²⁺)	0.53	< 200
Calcium (Ca ²⁺)	8.7	< 150
Magnesium (Mg ²⁺)	0.6	< 80
Iron (Fe)	0.106	<0.2
Manganese (Mn ²⁺)	0.0023	<0.2
Zinc (Zn ²⁺)	0.009	<4.0
Sulfates (SO ₄ ²⁻)	8.4	< 250
Chlorides (Cl ⁻)	< 1.0	< 250
Carbonates (CO ₃ ²⁻)	< 2.0	-
Hydrocarbonates (HCO ₃ ⁻)	36.0	-
Other values	Results	
Active reaction (pH)	7.3 alkaline	6.5-9.5
Electroconductivity	82.3 μS/cm	< 2000
Hardness of water	<1.4 dH soft	<33.7
Local extremum* eV ⁻¹ at (-0.1362--0.1387 eV)	70.1	>24.1

*Function of distribution of H₂O molecules according to energy f(E).

Table 5 shows the results on water composition in field area of Dolni Dabnik. The maximum peak in NES-spectra of Dolni Dabnik water (eV⁻¹) is detected at (-0.1362--0.1387 eV), in water from Danubian Plain is detected at 23.2 eV⁻¹, and in water from Thracian Valley – at -21.3 eV⁻¹. In the water samples from Danubian Plain and Thracian Valley there are data for presence of nitrites (NO₂⁻), nitrates (NO₃⁻), ammonia (NH₄⁺),

phosphates (HPO_4^{2-}) more than the norm.

Table 5: The composition of artesian spring Sadovetz, Dolni Dabnik municipality and local extremums in NES-spectra of water

Indicators	Results of the research (mg/l)	Norm
Sodium (Na^+)	14.2	< 200
Calcium (Ca^{2+})	103.3	< 150
Sulfates (SO_4^{2-})	19.2	< 250
Magnesium (Mg^{2+})	64.0	< 80
Chlorides (Cl^-)	9.2	< 250
Carbonates (CO_3^{2-})	< 2.0	-
Hydrocarbonates (HCO_3^-)	184.4	-
Other values	Results	
Active reaction (pH)	7.3 alkaline	6.5-9.5
Hardness of water	29.1 dH Very hard	<33.7
Local extremum* eV^{-1} at (-0.1362--0.1387 eV)	23.2	>24.1

Table 6 shows the optimal chemical composition of water, hardness, the local extremum (eV^{-1}) at (-0.1362--0.1387 eV), and total mineralization of water as the middle result of different studies. The water samples were taken from areas between 600 and 1300 m attitude in Bulgaria and from Kavkaz, Russia. It is worth to note that these areas are populated by long living people.

Table 6: The optical chemical composition of water, hardness, local extremum eV^{-1} at (-0.1362--0.1387 eV) and total mineralization of water

Indicators	Results of melt and mountain water (Bulgaria) (mg/l)	Results of melt water (Russia) (mg/l)
Sodium(Na^+) + Potassium (K^+)	6.1	< 30
Calcium (Ca^{2+})	29.5	< 50
Magnesium (Mg^{2+})	1.5	< 10
Iron (Fe)	0.083	-
Manganese (Mn^{2+})	0.0017	-
Zinc (Zn^{2+})	0.007	-
Sulfates (SO_4^{2-})	21.9	< 100
Chlorides (Cl^-)	3.2	< 70
Carbonates (CO_3^{2-})	< 2.0	-
Hydrocarbonates (HCO_3^-)	69.7	< 100
Other values	Results	
Active reaction (pH)	6.7 normal	6.5-7.0
Electroconductivity	410.9 $\mu\text{S/cm}$	< 2000
Hardness of water	4.65 dH Moderately soft	<33.7
Total mineralization (g/l)	0.132	< 0.3
Local extremum* eV^{-1} at (-0.1362--0.1387 eV)	49.7	>24.1

3.4. 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 water samples as a function of distribution of H_2O molecules according to energy $f(E)$ at $\lambda = 8.95 \mu m$ shows the analogue peak at analogous values of $f(E)$, E and λ , which was detected in water with Ca^{2+} ions 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., J et al., 2011). There was a positive correlation between the eSOD activity and 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 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, E. 2007). Zinc in composition of water improves the antioxidative enzymes in red blood cells (Malhotra, A., Dhawan, D. K., 2008).

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, M. et al, 2000). 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, L. J., Bendich, A, 1988). There was reported the antioxidant effects of water on rats (Abdullah, A.M., 2012). The norm in water for Zn^{2+} and Mg^{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.

There are known the results obtained in USA and Canada on the concentration of Ca^{2+} in water. 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 $Ca^{2+} - 299.7$; $Mg^{2+} - 18.9$; $Zn^{2+} - 0.001$ mg/l.

3.5. Results on Vortex Power Spring water.

Resume from Vortex Power for type of Preparation

The main results of the research are demonstrated in Table 7 which presents the local extremums in NES spectra of water samples of different origin.

Table 7. The local extremums in NES spectra of water samples of different origin

Sources and Types of Waters	Local extremum* at (-0.1112...-0.1137 eV)	Local extremum** at (-0.1362...-0.1387 eV)	[%(-E _{value})/(-E _{total} value)] -0.1112 eV	[%(-E _{value})/(-E _{total} value)] -0.1137 eV	[%(-E _{value})/(-E _{total} value)] -0.1162 eV	[%(-E _{value})/(-E _{total} value)] -0.1187 eV
	eV ⁻¹ norm (>24.1)	eV ⁻¹ norm (>24.1)				
1.Zlatishko-Tetevenska Mountain (Bulgaria)	46.1±2.3	36.9±1.9	4.1	7.0	4.0	7.4
2.Vasiliovka Mountain (Bulgaria)	12.0±0.6	44.9±2.2	1	2	5	6.2
3.Rhodops Mountain (Eco hotel Zdravetz) (Bulgaria)	53.3±1.6	59.3±3.0	11.0	3.6	5.5	13.0
4. Deionized Water with Shungite (Bulgaria)	70.6±3.5	47.1±2.4	5.9	11.8	0	11.8
5. Deionized Water with Zeolite (Bulgaria)	0	50.1±2.0	0	0	0	12.5
6. Danubian Plain, Sadovetz (Bulgaria)	25±1.25	23.2±1.7	0	6.3	0	3.1
7. Tap Water Teteven (Bulgaria)	16.0±0.8	16.0±0.8	4.0	0	0	4.0
8. Glacier Rosenloui (Switzerland)	129.0±6.3	70.1±3.5	22.6	9.7	6.5	12.9
9. Vortex Power Water (Switzerland)	88.8±4.4	74.1±3.4	7.4	14.8	7.4	11.1
10. Deionized Water	38.7±1.9	32.3±1.6	3.2	6.4	4.8	3.2

Notes:

*The values (E= -0.1112...-0.1137 eV) correspond to the wavelengths ($\lambda=10.91-11.91\mu\text{m}$).

**The values (E= -0.1362...-0.1387 eV) correspond to the wavelengths ($\lambda=8.95- 9.10 \mu\text{m}$).

There are two conclusions from analyzing the NES spectra of water (Vortex Power Spring) obtained after the treatment by Vortex Power Spring apparatus. For the energy E= -0.1387 eV there was the local extremum with f(E) value measured at $74.1\pm 3.7 \text{ eV}^{-1}$. In 1992 A. Antonov demonstrated that in E= -0.1387 eV the aqueous suspension of tumor cells there was detected a decrease of local extremum corresponding to a statistical error. NES-spectra of aqueous solution containing Ca^{2+} (67 mg/l) had a local extremum of energy at -0,11 eV in NES-spectra. In 1992 it was established by A. Antonov that the differential spectra of karst water and water solutions of calcium carbonate (CaCO_3) with the same content of calcium (Ca^{2+}) ions were similar at statistical level to the Student *t*-criterion at $p < 0.05$. For Vortex Power Spring water the local extremum was detected at $88.8\pm 4.4 \text{ eV}^{-1}$. The result of tap water of Teteven was observed within a statistical error or $16.0\pm 0.8 \text{ eV}^{-1}$ for -0,11 eV and -0.1387 eV. The measurement of the DNES spectrum for Vortex Power Spring water was $72.8\pm 3.6 \text{ eV}^{-1}$ in the range (-0.1112– -0.1137 eV). The result in the range (-0.1362–-0.1387 eV) of DNES

spectrum was $58.1 \pm 2.9 \text{ eV}^{-1}$. These data may indicate that on the molecular level Vortex Power Spring water supposedly is more structurally organized than tap water and other analyzed water samples.

In 2012 the research of 1% solution of human blood serum was made by I. Ignatov and O. Mosin with the collaboration of K. Naneva from Municipality hospital, Teteven, Bulgaria. In the control group of healthy people, the function of distribution according to energy $f(E)$ at $\lambda = 8.95 \text{ }\mu\text{m}$ was detected at an average value of $75.3 \pm 3.8 \text{ eV}^{-1}$. In the group of people in critical condition this value was $24.1 \pm 1.2 \text{ eV}^{-1}$. The confidence level of the obtained results according to the Student t -criterion was at $p < 0.05$. The result of Vortex Power Spring water was $74.1 \pm 3.7 \text{ eV}^{-1}$. On the molecular level there was a possibility for decreasing the number of tumor cells in water samples. However, this depends on the health status and other factors – heredity, quality of water, medical prophylactics.

The distribution $[\%, (-E_{\text{value}})/(-E_{\text{total value}})]$ of H_2O molecules in Vortex Power Spring water according to energies of hydrogen bonds and local extremums in NES and DNES spectra of water solutions of natural minerals - shungite and zeolite are shown in Table in the ranges of $(-0.1112 \dots -0.1137 \text{ eV})$ and $(-0.1362 \dots -0.1387 \text{ eV})$. The average energy ($E_{\text{H}\dots\text{O}}$) of hydrogen $\text{H}\dots\text{O}$ -bonds among individual H_2O molecules in Vortex Power Spring water was measured at $E = -0.1195 \text{ eV}$. The result for the control sample (deionized water) was $E = -0.1102 \text{ eV}$. The results obtained with the NES method were recalculated with the DNES method as a difference of the NES (Vortex Power Spring) minus the NES (control sample) equalled the DNES spectrum of Vortex Power Spring water. Thus, the result for Vortex Power Spring water recalculated with the DNES method was $\Delta E = 0.0093 \pm 0.0011 \text{ eV}$. The results showed the increasing of the values of the energy of hydrogen bonds in Vortex Power Spring water regarding the deionized water. The result for the control sample (tap water from Teteven) was $E = -0.1115 \text{ eV}$. Thus, the result for Vortex Power Water measured with the DNES method was $\Delta E = -0.008 \pm 0.0011 \text{ eV}$.

Further it was carried out the research of melt water from Glacier Rosenloui, Alps. The local extremum at $(-0.1362 \dots -0.1387 \text{ eV})$ in Glacier Rosenloui water, Alps was detected at $f(E) 70.1 \pm 3.5$ and Vortex Power water at $74.1 \pm 3.7 \text{ eV}^{-1}$. For the healthy group of people 1% solution of human blood serum has $75.3 \pm 3.8 \text{ (eV}^{-1})$ and for people having tumor diseases – $24.1 \pm 1.2 \text{ (eV}^{-1})$. The results obtained with the NES method were recalculated with the DNES method as a difference of the NES (Glacier Rosenloui) minus the NES (the control sample deionized water) equalled the DNES spectrum of melt water from Glacier Rosenloui. Thus, the result for water from Glacier Rosenloui measured with the DNES method was $\Delta E = -0.0093 \pm 0.0011 \text{ eV}$.

Another important physical parameter was calculated with using the NES method – the average energy ($E_{\text{H}\dots\text{O}}$) of $\text{H}\dots\text{O}$ -bonds between H_2O compiled – $0,1067 \pm 0,0011 \text{ eV}$.

According to the analysis of various water samples by the NES and DNES methods can be drawn the main conclusions:

- The energy of hydrogen bonds of water in the samples was differed because of the different number of hydrogen bonds in the water samples, which may result from the fact that different waters have a different structure and various intermolecular interactions – various associative elements with different structure, clusters of formula $(\text{H}_2\text{O})_n$ with different n , connected into the molecular associates;
- As a result of different energies of hydrogen bonds, the surface tension of water samples was increasing or decreasing. For Vortex Power Spring water and melt water from Glacier Rosenloui there was the increasing of surface tension regarding the control samples;
- The redistribution of H_2O molecules in water samples according to the energy (statistical process of dynamics);
- The hydrogen bond network may be stabilized with metal cations and anions, contained in water. In ice the hydrogen bonds have the energy of 4.6 kcal/mol or 0.1995 eV . For liquid water the energy of hydrogen bonds makes up $1\text{--}3 \text{ kcal/mol}$ or $0.043\text{--}0.13 \text{ eV}$.

The further empirical results obtained under the project of “Water, Ecology and Longevity” on the research of the water quality, DNES-spectra of human blood serum and long living people in Bulgaria from 2012 till 2015, showed the difference in the age of the people who lived in mountainous and plain areas in Bulgaria that was statistically proven by the Student t -criterion at a confidence level $p < 0.05$. The most adult person in mountain was 104 and in plain areas 97 years old. The difference between mountainous and plain areas is 50–70 km. For the average lifespan of long living people in plain areas the result was 90.6 years. For the brothers and sisters of long living people from mountain areas the average lifespan is 88.5 years. The analyses show the difference in the water quality derived from different water springs and the differences in DNES-spectra of human blood serum. With increasing of age of long-living blood relatives, the function of distribution of H_2O molecules in DNES-spectra of human blood serum according to energies $f(E)$ at $-0,1387 \text{ eV}$ is decreasing. In this group of tested people the result obtained by the DNES method was – $\Delta E = -5,5 \pm 1,1 \text{ meV}$; the difference in age was of 20–25 years in relation to the control group.

3.6. Results on influence of Vortex Power device on Tap Water from Danubian Plain.

Permanent oxidation. Conception of Otto Warburg

The effects of turbulence (turbulent flows, vortex) on physical-chemical properties of water (pH, surface tension) have been widely discussed in scientific literature. The most profound effect exerts a combination of turbulence with the magnetic field. Other positive characteristics of this method of water treatment include reducing the surface tension of water; the pH, reducing the time of the water treatment, improving the palatability of the treated water, and longer-terms of water storage. As the most effective recognized structuring impacts is recognized the mixing of water with a magnetic stirrer to form vortex (within 30 seconds), and passing water through the funnel, providing its rotational movement.

These devices are additional elements mounted inside the water pipes by a threaded or flanged connection. The main element of the design is a spiral element that turns the flow of fluid from laminar into turbulent. Water being passed through a twisted counter-flow vortex generator forms turbulent flows (vortex) similar to river whirlpools. On this principle is based a number of effective water treatment devices such as Vortex Power Spring nozzle (Vortex Power Company). However, the nature of the physical processes in turbulent water flow is just beginning to explore. Therefore, reports on new alternative methods in water treatment and the effects upon the water structure are extremely rare.

The experiments for improvement of quality of tap water by saturation with oxygen via the water treatment were performed on a Vortex Power Spring device (Vortex Power Company) by the DNES-method. The experimental data demonstrated the increasing of the average energy of hydrogen bonds in a sample of tap water from Sadovetz, Dolni Dabnik, Danubian plain as the DNES difference before and after the water treatment with Vortex Power Spring apparatus. The result for the control sample (tap water from Sadovetz) was $E = -0.1133$ eV. The result after the treatment of the water sample by Vortex Power apparatus was measured as $E = -0.1246$ eV. Thus, the result for Vortex Power Water measured with the DNES method was $\Delta E = -0.0113 \pm 0.0011$ eV. The result for tap water as a control sample from the Danubian plain water with a local extremum at $(-0.1362 \text{--} -0.1387$ eV) was 12.5 eV^{-1} . The result for tap water after the treatment of the water sample from Danubian plain by Vortex Power apparatus was 75 eV^{-1} with a local extremum at $(-0.1362 \text{--} -0.1387$ eV).

The result before using of Vortex Power Spring apparatus on permanent oxidation of water was $0.9 \pm 0.09 \text{ mg O}_2/\text{dm}^3$. After the usage of Vortex Power Spring apparatus the result was $1.26 \pm 0.13 \text{ mg O}_2/\text{dm}^3$. Thus, there is statistically reliable result measured at $0.36 \pm 0.04 \text{ mg O}_2/\text{dm}^3$ on the influence of Vortex Power Spring apparatus on permanent water oxidation levels. The permanent oxidation was measured as quantity of oxygen needing for oxidation of organic compounds in a volume of the treated water ($\text{mg O}_2/\text{dm}^3$).

The conception of Otto Warburg is related to influence of oxygen on proliferating cancer cells and cellular respiration (Warburg, 1956). If cellular respiration is damaged then it is synthesized little amount of adenosine triphosphate (ATP) in damaged cells; it may be either that the oxygen consumption in cells has been decreased or that, with the undiminished oxygen consumption, the coupling between respiration and the formation of ATP has been metabolically broken. The authors suggest that the respiration of cancer cells never returns to normal. The important fact is that water subjected to the water treatment in the Vortex Power Spring apparatus has a higher permanent oxygen levels.

The results of these studies are demonstrated below:

The research of Tap Water from Danubian plain after the treatment by the Vortex Power Spring apparatus:

Local extremum at $(-0.1362 \text{--} -0.1387 \text{ eV}) - 75 \text{ eV}^{-1}$

The result of NES – (-0.1246 eV)

The research of Tap Water from Danubian plain:

Local extremum at $(-0.1362 \text{--} -0.1387 \text{ eV}) - 12.5 \text{ eV}^{-1}$

The result of NES – (-0.1133 eV)

3.7. Hydrogen Bonds in Liquids and Crystals

The peculiarities of chemical structure of 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 ($\text{O-H}\dots\text{O}$) with neighboring H_2O molecules, binding them into complex intermolecular associates which composition represented by general formula $(\text{H}_2\text{O})_n$, 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 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 structure of hydrogen bonding, therefore may be defined as

$O...H^{\delta+}-O^{\delta-}$. As the result, the electron of the H-atom due to its relatively weak bond with the proton easily shifts to the electronegative O-atom. The O-atom with increased electron density becomes partly negatively charged $-\delta^-$, while the H-atom on the opposite side of the molecule becomes positively charged $+\delta^+$ that leads to the polarization of $O^{\delta-}-H^{\delta+}$ covalent bond. In this process the proton becomes almost bared, and due to the electrostatic attraction forces are provided good conditions for convergence of $O...O$ or $O...H$ atoms, leading to the chemical exchange of a proton in the reaction $O-H...O \leftrightarrow O...H-O$. Although this interaction is essentially compensated by mutual repulsion of the molecules' nuclei and electrons, the effect of the electrostatic forces and donor-acceptor interactions for H_2O molecule compiles 5–10 kcal per 1 mole of substance. It is explained by a negligible small atomic radius of hydrogen and shortage of inner electron shells, which enables the neighboring H_2O molecule to approach the hydrogen atom of another molecule at very close distance without experiencing any strong electrostatic repulsion. The H_2O molecule has four sites of hydrogen bonding – two uncompensated positive charges at H-atoms and two negative charges at the O-atom. Their mutual disposition is characterized by direction from the centre of regular tetrahedron (nucleus of O-atom) towards its vertexes. This allows to one H_2O molecule in condensed state to form up to 4 classical hydrogen bonds, two of which are donor bonds and the other two – acceptor ones (taking into consideration the bifurcate (“two-forked”) hydrogen bond – 5) (Pasichnyk *et al.*, 2008).

A hydrogen bond according to Bernal–Fowler rules (Bernal & Fowler, 1933) is characterized by the following parameters:

- O-atom of each H_2O molecule is bound with four neighboring H-atoms: by covalent bonding with two own H-atoms, and by hydrogen bonding – with two neighboring H-atoms (as in the crystalline structure of ice); each H-atom in its turn is bound with O-atom of neighbouring H_2O molecule.
- On the line of O-atom – there can be disposed only one proton H^+ ;
- The proton, which takes part in hydrogen bonding situated between two O-atoms, therefore has two equilibrium positions: it can be located near its O-atom at approximate distance of 1 Å, and near the neighboring O-atom at the distance of 1.7 Å as well, hence both a usual dimmer $HO-H...OH_2$ and an ion pair $HO...H-OH_2$ may be formed during the hydrogen bonding, i.e. the hydrogen bond is partly electrostatic (~90%) and partly (~10%) covalent (Isaacs *et al.*, 2000). The state of “a proton near the neighboring oxygen” is typical for the interphase boundary, i.e. near water–solid body or water–gas surfaces.
- The hydrogen bonding of a triad $O-H...O$ possess direction of the shorter $O-H$ (\rightarrow) covalent bond; the donor hydrogen bond tends to point directly at the acceptor electron pair (this direction means that the H-atom being donated to the H-atom acceptor on another H_2O molecule).

The most remarkable peculiarity of hydrogen bond consists in its relatively low strength; it is 5–10 times weaker than chemical covalent bond (Pimentel & McClellan, 1960). In respect of energy the hydrogen bond has an intermediate position between covalent bonds and intermolecular van der Waals forces, based on dipole-dipole interactions, holding the neutral molecules together in gasses or liquefied or solidified gasses. Hydrogen bonding produces interatomic distances shorter than the sum of van der Waals radii, and usually involves a limited number of interaction partners. These characteristics become more substantial when acceptors bind H-atoms from more electronegative donors. Hydrogen bonds hold H_2O molecules on 15% closer than if water was a simple liquid with van der Waals interactions. The hydrogen bond energy compiles 5–10 kcal/mole, while the energy of $O-H$ covalent bonds in H_2O molecule – 109 kcal/mole (Arunan *et al.*, 2011). The values of the average energy ($\Delta E_{H...O}$) of hydrogen $H...O$ -bonds between H_2O molecules make up 0.1067 ± 0.0011 eV (Antonov & Galabova, 1992). With fluctuations of water temperature the average energy of hydrogen $H...O$ -bonds in of water molecule associates changes. That is why hydrogen bonds in liquid state are relatively weak and unstable: it is thought that they can easily form and disappear as the result of temperature fluctuations (Ignatov & Mosin, 2013).

Another key feature of hydrogen bond consists in its cooperatively coupling. Hydrogen bonding leads to the formation of the next hydrogen bond and redistribution of electrons, which in its turn promotes the formation of the following hydrogen bond, which length increasing with distance. Cooperative hydrogen bonding increases the $O-H$ bond length, at the same time causing a reduction in the $H...O$ and $O...O$ distances (Goryainov, 2012). The protons held by individual H_2O molecules may switch partners in an ordered manner within hydrogen networks (Bartha *et al.*, 2003). As the result, aqueous solutions may undergo autoprotolysis, i.e. the H^+ proton is released from H_2O molecule and then transferred and accepted by the neighboring H_2O molecule resulting in formation of hydronium ions as H_3O^+ , $H_5O_2^+$, $H_7O_3^+$, $H_9O_4^+$, etc. This leads to the fact, that water should be considered as associated liquid composed from a set of individual H_2O molecules, linked together by hydrogen bonds and weak intermolecular van der Waals forces (Liu *et al.*, 1996). The simplest example of such associate is a dimmer of water: $(H_2O)_2 = H_2O \cdots HOH$.

The energy of the hydrogen bonding in the water dimmer is 0.2 eV (~5 kcal/mol), which is larger than the energy of thermal motion of the molecules at the temperature of 300 K. Hydrogen bonds are easily

disintegrated and re-formed through an interval of time, which makes the water structure quite unstable and changeable (George, 1997). This process leads to structural inhomogeneity of water characterizing it as an associated heterogeneous two-phase liquid with short-range ordering, i.e. with regularity in mutual positioning of atoms and molecules, which reoccurs only at distances comparable to distances between initial atoms, i.e. the first H₂O layer. As it is known, a liquid in contrast to a solid body is a dynamic system: its atoms, ions or molecules, keeping short-range order in mutual disposition, participate in thermal motion, the character of which is much more complicated than that of crystals. For example H₂O molecules in liquid state under normal conditions (1 atm, +22 °C) are quiet mobile and can oscillate around their rotation axes, as well as to perform the random and directed shifts. This enabled for some individual molecules due to cooperative interactions to “jump up” from one place to another in an elementary volume of water. The random motion of molecules in liquids causes continuous changes in the distances between them. The statistical character of ordered arrangement of molecules in liquids results in fluctuations – continuously occurring deviations not only from average density, but from average orientation as well, because molecules in liquids are capable to form groups, in which a particular orientation prevails. Thus, the smaller these deviations are, the more frequently they occur in liquids.

In 2005 R. Saykally (USA) calculated the possible number of hydrogen bonds and the stability of water clusters depending on the number of H₂O molecules (Figure 4) (Saykally, 2005). It was also estimated the possible number of hydrogen bonds (100) depending on the number of H₂O molecules (250) in clusters (Sykes, 2007). O. Loboda and O.V. Goncharuk provided data about the existence of icosahedral water clusters consisting of 280 H₂O molecules with the average size up to 3 nm (Loboda & Goncharuk, 2010). The ordering of H₂O molecules into associates corresponds to a decrease in the entropy (randomness), or decrease in the overall Gibbs energy ($G = \Delta H - T\Delta S$). This means that the change in enthalpy ΔH minus the change in entropy ΔS (multiplied by the absolute temperature T) is a negative value. These results are consistent with our research of the DNES spectrum of water on which it may make conclusion about the number of H₂O molecules in elemental water clusters. The DNES spectrum of water has energy ranges from -0.08 to -0.14 eV. The spectral range lies in the middle infrared range from 8 to 14 μm (“window” of the atmosphere transparency to electromagnetic radiation). Under these conditions, the relative stability of water clusters depends on external factors, primarily on the temperature. We demonstrated that H₂O molecules change their position in clusters depending on the energy of intermolecular H...O hydrogen bonds. The values of the average energy ($E_{H...O}$) of hydrogen bonds between H₂O molecules in the formation of cluster associates with the formula (H₂O)_n compile -0.1067 ± 0.0011 eV. As the energy of hydrogen bonds between H₂O molecules increases up to -0.14 eV, the cluster formation of water becomes “restructuring”. In this case, the redistribution of energies among the individual H₂O molecules occurs.

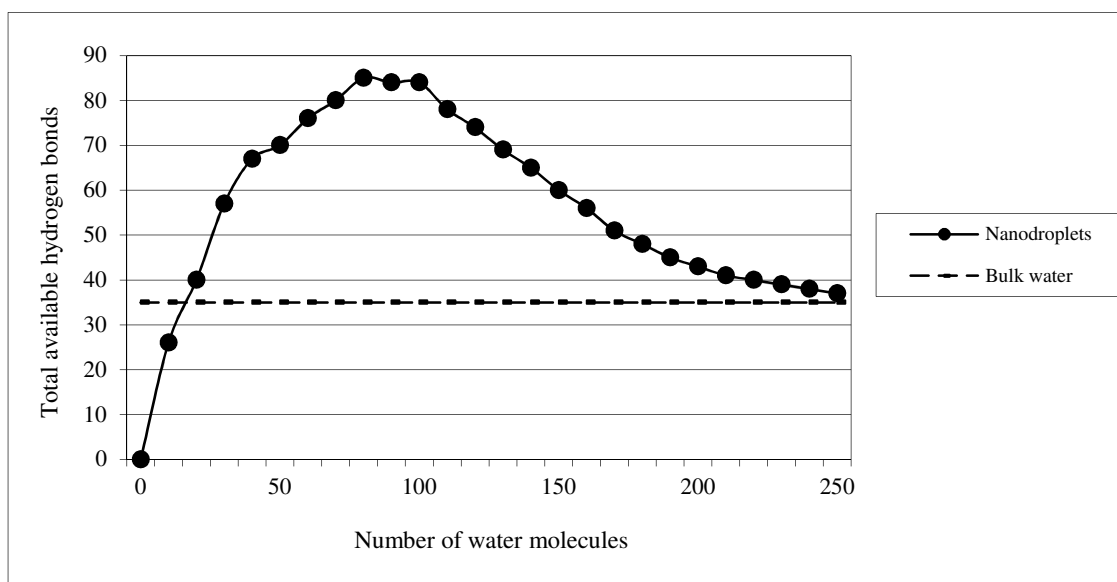


Figure 4: The total number of hydrogen bonds depending on the number of H₂O molecules in clusters.

All these experimental data including our data indicate that water is a complex associated non-equilibrium liquid consisting of associative groups containing according to the present data, from 3 to 20 individual H₂O molecules (Tokmachev *et al.*, 2010). Associates can be perceived as unstable groups (dimers, trimers, tetramers, pentamers, hexamers etc.) in which H₂O molecules are linked by van der Waals forces, dipole-dipole and other charge-transfer interactions, including hydrogen bonding. At room temperature, the degree of association of H₂O molecules may vary from 2 to 6. In 2000 it was deciphered the structure of the

water trimer, and in 2003 – tetramer, pentamer and the hexamer (Wang & Jordan, 2003). In 1993 K. Jordan (USA) (Tsai & Jordan, 1993) calculated the possible structural modifications of small water clusters consisting of six H₂O molecules (Figure 5a–c). Subsequently, it was shown that H₂O molecules capable of hydrogen bonding by forming the structures representing topological 1D rings and 2D chains composed from numerous H₂O molecules. Interpreting the experimental data, they are considered as pretty stable elements of the structure. According to computer simulations, elemental clusters are able to interact with each other through the exposed protons on the outer surfaces of hydrogen bonds to form more complicated clusters of more complex structure.

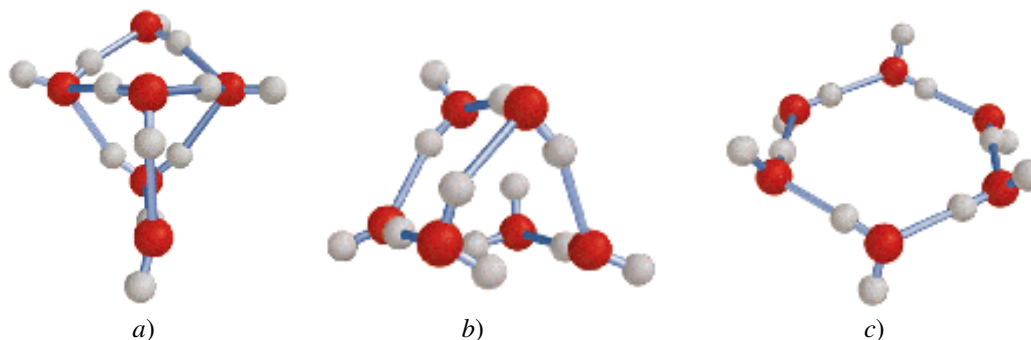


Figure 5: Computer calculations of small water cluster structures (a–c) with general formula (H₂O)_n, where n = 6 (Tsai & Jordan, 1993).

3.6. Melt water

The mountain water in springtime is the result of the melting of ice and snow accumulated in the mountains. Natural ice (ice I_h, hexagonal lattice) is usually much cleaner than water, because the solubility of all substances (except NH₄F) in ice is extremely low. The growing ice crystal is always striving to create a perfect crystal lattice and therefore displaces impurities. The melt water obtained after the thawing of ice has a certain “ice-like” structure, because it preserves the hydrogen bonding between water molecules; as a result it is formed complex intermolecular associates (clusters) – the analogues of ice structures, consisting of a different number of H₂O molecules. However, unlike the ice crystal, each associate has a very short time of existence as a result there occurs the constant processes of decay and formation of water associates having very complicated structure. The specificity of intermolecular interactions characteristic for the structure of ice, is kept in melt water, as it is estimated that in the melting of ice crystal is destroyed only 15% of all hydrogen bonds in the associates. Therefore, the inherent to ice connection of each H₂O molecule with four neighboring H₂O molecules is largely disturbed, although there is observed the substantially “blurring” of oxygen lattice framework. Processes of decay and formation of clusters occur with equal probability that is probably why physical properties of melt water are changed over time, e.g. dielectric permittivity comes to its equilibrium state after 15–20 min, viscosity – in 3–6 days (Mosin, 2012). The heating of fresh melt water above $t = +37^{\circ}\text{C}$ leads to a loss of the biological activity. The storage of melted water at $+22^{\circ}\text{C}$ is also accompanied by a gradual decrease in its biological activity; within ~16–18 hours it is reduced by 50%. The main difference between the structure of ice and water is more diffuse arrangement of the atoms in the lattice and disturbance of long-range order. The thermal oscillations (fluctuations) lead to the bending and breaking down of hydrogen bonds. H₂O molecules being out of equilibrium positions begin to “fall down” into the adjacent structural voids and for a time held up there, as cavities correspond to the relative minimum of potential energy. This leads to an increase in the coordination number, and the formation of lattice defects. The coordination number (the number of nearest neighbors) during the transition from ice to melt water varies from 4,4 at $+1,5^{\circ}\text{C}$ to 4,9 at $+80^{\circ}\text{C}$.

At present time 14 crystalline modifications of ice are known, each of them has its own structure and a character of disposition of hydrogen atoms (Table 8). In the crystalline structure of natural ice I_h hydrogen bonds are oriented towards the tetrahedron apexes at strictly defined angles equal to $109^{\circ}5'$ (in liquid water this angle is $104^{\circ}5'$) (Mosin & Ignatov, 2013). In ice structures I_c, VII and VIII this tetrahedron is nearly the same as a regular 4 triangular tetrahedron. In ice structures II, III, V and VI the tetrahedrons are noticeably distorted. In ice structures VI, VII and VIII two intercrossing systems of hydrogen bonds are distinguished. In the centre of the tetrahedron is located O-atom, at each of the two vertexes – H-atom, which electron takes part in formation of a covalent bond with an electron pair of O-atom. The rest two vertexes of the tetrahedron are occupied by two pairs of non-shared electrons of O-atom not participating in formation of covalent bonds, but participating in formation of two intermolecular hydrogen bonds with two neighbor H₂O molecules.

Table 8: Ice crystal modifications and their physical characteristics

Modification	Crystal structure	Hydrogen bond lengths, Å	Angles H–O–H in tetragonals, °
I _h	Hexagonal	2.76	109.5
I _c	Cubic	2.76	109.5
II	Trigonal	2.75–2.84	80–128
III	Tetragonal	2.76–2.8	87–141
IV	Rhombic	2.78–2.88	70.1–109
V	Monoclinic	2.76–2,87	84–135
VI	Tetragonal	2.79–2.82	76–128
VII	Cubic	2.86	109.5
VIII	Cubic	2.86	109.5
IX	Tetragonal	2.76–2.8	87–141
X	Cubic	2.78	109.5
XI	Hexagonal	4.50	90.0
XII	Tetragonal	4.01	90.0
XIII	Monoclinic	7.47	90–109.7
XIV	Rhombic	4.08	90.0

Notes:

I_h – natural hexagonal ice; I_c – cubic ice.

The carcasses of hydrogen bonds allocate H₂O molecules in form of a spatial hexagon network with internal hollow hexagonal channels. In the nodes of this network O-atoms are orderly organized (crystalline state), forming regular hexagons, while H-atoms have various positions along the bonds (amorphous state). When ice melts, its network structure is destroyed: H₂O molecules begin to “fall down” into the network hollows, resulting in a denser structure of the liquid – this explains why water is heavier than ice. The hydrogen bonding explains other anomalies of water (anomalies of temperature, pressure, density, viscosity, fluidity etc.). According to theoretical calculations, at the melting of the ice it breaks about 15% of all hydrogen bonds (Mosin & Ignatov, 2011); by further heating to $t = +40$ °C breaks down about half of hydrogen bonds in water associates. In the water vapor heated at $t \geq +100$ °C the hydrogen bonds are absent.

The reports about the evidence on the existence of analogs of crystal structures in water have become more frequent in scientific literature nowadays (Michaelides & Morgenstern, 2007). As computer modeling showed, H₂O molecules after being placed in carbon nanotubes under high pressure and low temperatures formed the crystalline nanostructures like DNA double helix (Bai *et al.*, 2006). In modeling experiments water got frozen inside the carbon nanotubes with diameter 1.35–1.90 nm, with pressure 10–40000 atm and temperature -23 °C. The hydrogen bonds in an ice crystal got distorted leading to the formation of a double-walled helix. The inner wall of this structure represents a four-fold twisted helix, while the outer consists of four double helices, resembling a helix of DNA molecule (Figure 6). While being frozen at 17 K on hydrophobic surfaces of Cu, Ag and their salts, water became crystallized into two-dimensional ice hexamer-nanocrystal, consisting of six hexagonally attached H₂O molecules (Figure 7).

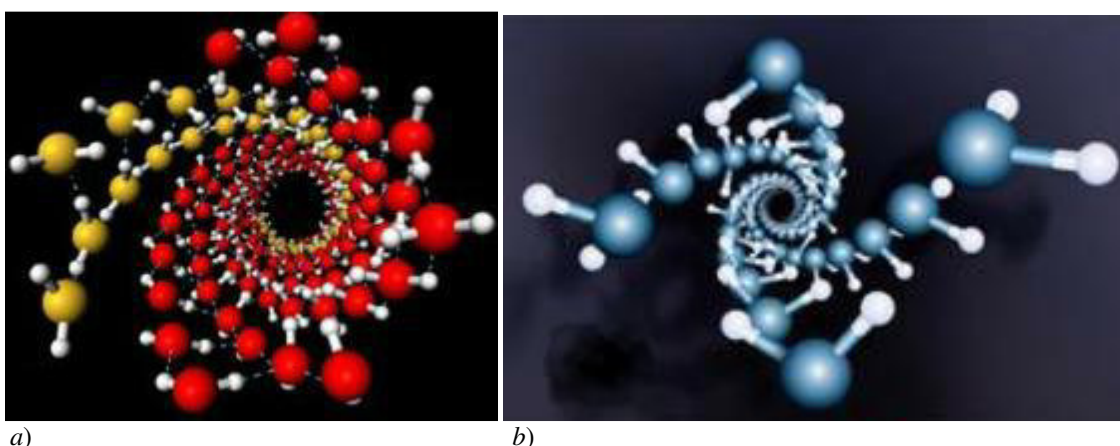


Figure 6: Spiral structures of water nanocrystals in carbon nanotubes according to computer simulations (Bai *et al.*, 2006) (diameter of nanotubes 1.35–1.90 nm; pressure 10–40000 atm; temperature -23 °C): a) – a general view of the crystal structure of water in nanotubes, b) – the inner wall of the structure of water.

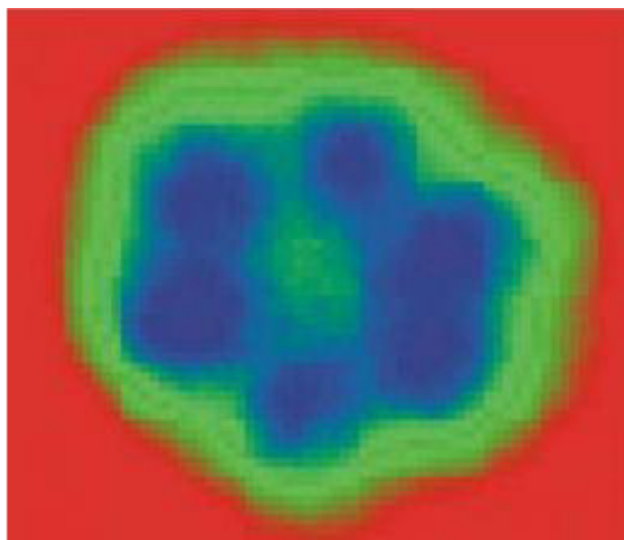


Figure 7: Image of hexamer nanocrystal of ice water (average size ~1 nm) consisting of six hexagonally attached H₂O molecules (blue colour) obtained by a scanning tunneling microscope after freezing water to 17 K on the hydrophobic metallic plates of Cu and Ag (Michaelides & Morgenstern, 2007).

3.7. Deuterium content in mountain and melt water

The preliminary analyses of water from various water sources, carried out by us, show that the mountain and melt water as the result of natural isotope purification contains less amount of deuterium than the tap water. This type of water also contains ions of Ca²⁺, Mg²⁺, Na⁺, HCO₃⁻ and SO₄²⁻. The content of K⁺ and N⁺ cations in the melt water are <30 mg/l, Mg²⁺ – <10 mg/l, Ca²⁺ – <40 mg/l, the content of SO₄²⁻ – <100 g/l, HCO₃⁻ <90 mg/l, Cl – less than 70 mg/l, total rigidity ≤ 4 mEq/l, the total mineralization ≤ 0.2 g/l, pH – 6.5–7.0 at 25 °C. The degree of natural purification of melt water from impurities makes up ~50–60%. The concentration of salts of rigidity – Ca²⁺, Mg²⁺, Fe²⁺, Fe³⁺, heavy metals and organochlorine compounds, as well as heavy isotopes, including deuterium in melt water is less that of ordinary portable water. This fact is important because some authors consider the hardness of the water to be among the main factors in cardiovascular diseases (Pocock *et al.*, 1981; Rubenowitz *et al.*, 1999; Marque *et al.*, 2003). However, mild correlation was further proven that water hardness could not be a decisive factor for human longevity. Analysis of water from various sources of Russia and Bulgaria show that the mountain water contains on average ~4–5% less deuterium in form of HDO, than the river water and sea water. In natural waters, the deuterium content is distributed irregularly: from 0.02–0.03 mol.% for river and sea water, to 0.015 mol.% for water of Antarctic ice – the most purified from deuterium natural water containing deuterium in 1.5 times less than that of seawater. According to the international SMOW standard isotopic shifts for D and ¹⁸O in sea water: D/H = (155.76±0.05)·10⁻⁶ (155.76 ppm) and ¹⁸O/¹⁶O = (2005.20±0.45)·10⁻⁶ (2005 ppm) (Lis *et al.*, 2008). For SLAP standard isotopic shifts for D and ¹⁸O in seawater: D/H = 89·10⁻⁶ (89 ppm) and for a pair of ¹⁸O/¹⁶O = 1894·10⁻⁶ (1894 ppm). In surface waters, the ratio D/H = ~(1.32–1.51)·10⁻⁴, while in the coastal seawater – ~(1.55–1.56)·10⁻⁴. Waters of other underground and surface water sources contain varied amounts of deuterium (isotopic shifts) – from δ = +5,0 D,%, SMOW (Mediterranean Sea) to δ = -105 D,%, SMOW (Volga River). The natural waters of CIS countries are characterized by negative deviations from SMOW standard to (1.0–1.5)·10⁻⁵, in some places up to (6.0–6.7)·10⁻⁵, but there are observed positive deviations at 2.0·10⁻⁵. The content of the lightest isotopomer – H₂¹⁶O in water corresponding to SMOW standard is 997.0325 g/kg (99.73 mol.%) and for SLAP standard – 997.3179 g/kg (99.76 mol.%).

The thawed snow and glacial water in the mountains and some other regions of the Earth also contain less deuterium than ordinary drinking water. On average, 1 ton of river water contains 150–200 g deuterium. The average ratio of H/D in nature makes up approximately 1:5700. According to the calculations, the human body throughout life receives about 80 tons of water containing in its composition 10–12 kg of deuterium and associated amount of heavy isotope ¹⁸O. That is why it is so important to purify drinking water from heavy isotopes of D and ¹⁸O.

We have obtained results on growth and adaptation to D₂O of various cells of prokaryotic and eukaryotic organisms (Mosin & Ignatov, 2012; Mosin & Ignatov, 2013d). Our studies have shown that animal cells are able to withstand up to 25–30% D₂O, plants – up to 50–60% D₂O, and protozoa cells are able to live on ~90% D₂O. Further increase in the concentration of D₂O for these groups of organisms leads to cellular death. On the contrary, deuterium depleted water with the decreased deuterium content (60–100 ppm) has beneficial

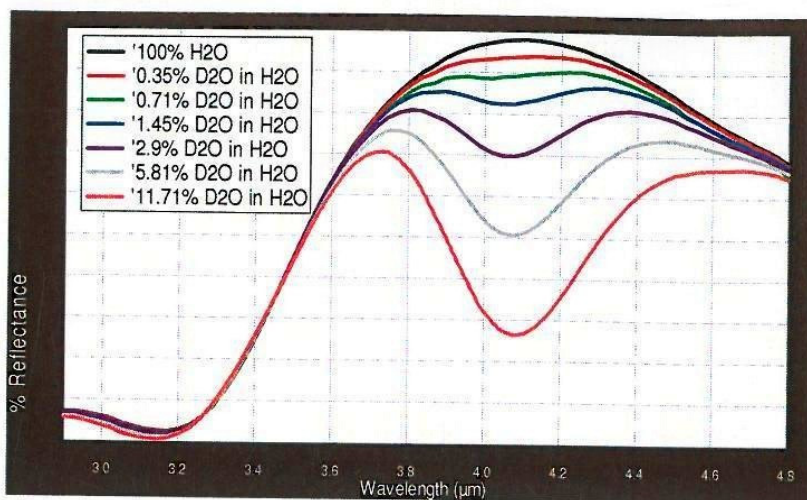
effects on metabolism.

As demonstrated in IR-spectra of water of different content of deuterium measured on a device Infra Spec VFA-IR, that at $\lambda = 4.1 \mu\text{m}$; even at low concentrations of deuterium of 0.35 and 0.71%, there is observed a decline in the local maximums relative to the local extremum of 100% pure water (the local maximums in IR-spectra reflect vibrational-rotational transitions in the ground electronic state because at changing the atomic mass of H-atoms and D-atoms in H_2O molecule their interaction will also change, although the electronic structure of the molecule and its ability to form the H-bonds, however, remains the same; with the substitution with deuterium the vibrational-rotational transitions are changed, that is why it appears other local maximums in IR-spectra. The result is reliable regarding the content of deuterium in natural waters from 0.015–0.03%.

The local maximums in IR-spectra reflect vibrational-rotational transitions in the ground electronic state; the substitution with deuterium changes the vibrational-rotational transitions in H_2O molecule that is why it appears other local maximums in IR-spectra. In the water vapor state, the vibrations involve combinations of symmetric stretch (ν_1), asymmetric stretch (ν_3) and bending (ν_2) of the covalent bonds with absorption intensity (H_2O) $\nu_1; \nu_2; \nu_3 = 2671; 1178.4; 2787.7 \text{ cm}^{-1}$. For liquid water absorption bands are observed in other regions of the IR-spectrum, the most intense of which are located at $2100, \text{ cm}^{-1}$ and $710\text{--}645 \text{ cm}^{-1}$. For the D_2O molecule these ratio compiles $2723.7, 1403.5$ and 3707.5 cm^{-1} , while for HDO molecule – $2671.6, 1178.4$ and 2787.7 cm^{-1} . HDO (50 mole% H_2O + 50 mole% $^2\text{H}_2\text{O}$; ~50 % HDO, ~25 % H_2O , ~25 % D_2O) has local maxima in IR-spectra at $3415 \text{ cm}^{-1}, 2495 \text{ cm}^{-1}, 1850 \text{ cm}^{-1}$ and 1450 cm^{-1} assigned to OH^- -stretch, OD^- -stretch, as well as combination of bending and libration and HDO bending respectively.

In the IR-spectrum of liquid water absorbance band considerably broadened and shifted relative to the corresponding bands in the IR-spectrum of water vapor. Their position depends on the temperature (Ignatov & Mosin, 2013b). The temperature dependence of individual spectral bands of liquid water is very complex (Zelmann, 1995). Furthermore, the complexity of the IR-spectrum in the area of OH^- stretching vibration can be explained by the existence of different types of H_2O associations, manifestation of overtones and composite frequencies of OH^- groups in the hydrogen bonds, and the tunneling effect of the proton (for relay mechanism) (Yukhnevitch, 1973). Such complexity makes it difficult to interpret the spectrum and partly explains the discrepancy in the literature available on this subject.

ANALYSIS



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Figure 9: The typical IR-spectra of water with varying content of deuterium

In liquid water and ice the IR-spectra are far more complex than those ones of the vapor due to vibrational overtones and combinations with librations (restricted rotations, e.g. rocking motions). These librations are due to the restrictions imposed by hydrogen bonding (minor L_1 band at 395.5 cm^{-1} ; major L_2 band at 686.3 cm^{-1} ; for liquid water at 0°C , the absorbance of L_1 increasing with increasing temperature, while L_2 absorbance decreases but broadens with reduced wave number with increasing temperature (Brubach et al., 2005). The IR spectra of liquid water usually contain three absorbance bands, which can be identified on absorption band of the stretching vibration of OH^- group; absorption band of the first overtone of the bending vibration of the molecule HDO and absorption band of stretching vibration of OD^- group (Max & Chapados, 2009). Hydroxyl group OH^- is able to absorb much infrared radiation in the infrared region of the IR-spectrum. Because of its polarity, these groups typically react with each other or with other polar groups to form intra- and intermolecular hydrogen bonds. The hydroxyl groups, which are not involved in formation of hydrogen bonds,

usually produce the narrow bands in IR spectrum, while the associated groups – broad intense absorbance bands at lower frequencies. The magnitude of the frequency shift is determined by the strength of the hydrogen bond. Complication of the IR spectrum in the area of OH stretching vibrations can be explained by the existence of different types of associations of H₂O molecules, a manifestation of overtones and combination frequencies of OH groups in hydrogen bonding, as well as the proton tunneling effect (on the relay mechanism).

Assignment of main absorption bands in the IR-spectrum of liquid water is given in Table 8. The IR spectrum of H₂O molecule was examined in detail from the microwave till the middle (4–17500 cm⁻¹) visible region and the ultraviolet region – from 200 nm⁻¹ to ionization limit at 98 nm⁻¹ (Walrafen, 1972). In the middle visible region at 4–7500 cm⁻¹ are located rotational spectrum and the bands corresponding to the vibrational-rotational transitions in the ground electronic state. In the ultraviolet region (200 to 98 nm⁻¹) are located bands corresponding to transitions from the excited electronic states close to the ionization limit in the electronic ground state. The intermediate region of the IR-spectrum – from $\lambda = 570$ nm to $\lambda = 200$ nm corresponds to transitions to higher vibrational levels of the ground electronic state.

Results of IR-spectroscopy with device Infra Spec VFA-IR show that at 4.1 μm , even at low concentrations of deuterium of 0.35 and 0.71%, there is observed a decline in the local maximums relative to the local maximum of 100% pure water (the local maximums in IR-spectra reflect vibrational-rotational transitions in the ground electronic state because at changing the atomic mass of hydrogen and deuterium atoms in the water molecule their interaction will also change, although the electronic structure of the molecule and its ability to form H-bonds, however, remains the same; with the substitution with deuterium the vibrational-rotational transitions are changed, that is why it appears other local maximums in IR-spectra. These results are shown in Figure 9. The result is reliable regarding the content of deuterium in natural waters from 0.015–0.03%.

Table 8: The assignment of main frequencies in IR-spectra of H₂O and D₂O

Main vibrations of liquid H ₂ O and ² H ₂ O				
Vibration(s)	H ₂ O (t = +25 °C)		D ₂ O (t = +25 °C)	
	ν , cm ⁻¹	E_0 , M ⁻¹ cm ⁻¹	ν , cm ⁻¹	E_0 , M ⁻¹ cm ⁻¹
Spinning ν_1 + deformation ν_2	780–1645	21.65	1210	17.10
Composite $\nu_1 + \nu_2$	2150	3.46	1555	1.88
Valence symmetrical ν_1 , valence asymmetrical ν_3 , and overtone $2\nu_2$	3290–3450	100.65	2510	69.70

At further transition from H₂O monomers to H₄O₂ dimer and H₆O₃ trimer absorption maximum of valent stretching vibrations of the O–H bond is shifted toward lower frequencies ($\nu_3 = 3490$ cm⁻¹ and $\nu_1 = 3280$ cm⁻¹) (Eisenberg & Kauzmann, 1969) and the bending frequency increased ($\nu_2 = 1644$ cm⁻¹) because of hydrogen bonding. The increased strength of hydrogen bonding typically shifts the stretch vibration to lower frequencies (red-shift) with greatly increased intensity in the infrared due to the increased dipoles. In contrast, for the deformation vibrations of the H–O–H, it is observed a shift towards higher frequencies. Absorption bands at 3546 and 3691 cm⁻¹ were attributed to the stretching modes of the dimer [(H₂O)₂]. These frequencies are significantly lower than the valence modes of ν_1 and ν_3 vibrations of isolated H₂O molecules at 3657 and 3756 cm⁻¹ respectively). The absorption band at 3250 cm⁻¹ represents overtones of deformation vibrations. Among frequencies between 3250 and 3420 cm⁻¹ is possible Fermi resonance (this resonance is a single substitution of intensity of one fluctuation by another fluctuation when they accidentally overlap each other). The absorption band at 1620 cm⁻¹ is attributed to the deformation mode of the dimer. This frequency is slightly higher than the deformation mode of the isolated H₂O molecule (1596 cm⁻¹). A shift of the band of deformation vibration of water in the direction of high frequencies at the transition from a liquid to a solid state is attributed by the appearance of additional force, preventing O–H bond bending. Deformation absorption band in IR-spectrum of water has a frequency at 1645 cm⁻¹ and very weak temperature dependence. It changes little in the transition to the individual H₂O molecule at a frequency of 1595 cm⁻¹. This frequency is found to be sufficiently stable, while all other frequencies are greatly affected by temperature changes, the dissolution of the salts and phase transitions. It is believed that the persistence of deformation oscillations is stipulated by processes of intermolecular interactions, e.g. by the change in bond angle as a result of interaction of H₂O molecules with each other, as well as with cations and anions.

Thus the study of the characteristics of the IR spectrum of water allows to answer the question not only on the physical parameters of the molecule and the covalent bonds at isotopic substitution with deuterium, but also to make a certain conclusion on associative environment in water. The latter fact is important in the study of structural and functional properties of water associates and its isotopomers at the isotopic substitution with deuterium. The substitution of H with D affects the stability and geometry of hydrogen bonds in an apparently rather complex way and may, through the changes in the hydrogen bond zero-point vibration energies, alter the conformational dynamics of hydrogen (deuterium)-bonded structures of DNA and proteins in D₂O (Cleland, 1976). It may cause disturbances in the DNA-synthesis, leading to permanent changes on DNA structure and consequently on cell genotype (Thomson, 1960; Katz, 1960; Den'ko, 1970; Török *et al.*, 2000).

4. Conclusion

The composition of various samples of water from Bulgarian water springs: the melt water from Glacier Rosenloui, Swiss Alps, Vortex Power Spring water as well as human blood serum of people with excellent health and cancer patients was studied by IR, NES and DNES-methods. In frames of the research 415 people living in the municipalities of Teteven, Yablanitza, Ugarchin, Lukovit, Lovech district; Dolni Dabnik, Pleven district, Kuklen, Pleven district (Bulgaria), where is lived the largest number of long lived people and their siblings, were also investigated regarding the water consumption. The research conducted by us shows that the direct relationship of man and nature – clean air, fresh mountain water, natural food from eco-farms and physical activity explains the difference between the larger number of long lived people and centenarians who live in the mountain regions of Bulgaria and Russia and their high average number. The natural mountain water and melt water with unique chemical composition, the pH value and less amount of deuterium seems to be one of the most important factors for human health. In Bulgaria, most long lived people and centenarians live in the Rhodope Mountains, while in Russia – in Dagestan and Yakutia. It worth to note that IR-spectrum of mountain water is most similar to the IR-spectrum of blood serum of healthy group of people with a local maximum at $\lambda = 8,95 \mu\text{m}$. The similar spectral characteristics possess mountain water from Teteven and other Bulgarian sources. 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 results of chemical composition of water from Glacier Rosenloui, Swiss Alps, as well as the results on influence of Vortex Power Spring apparatus on tap water with changes in the DNES-spectrum and permanent oxidation levels.

Acknowledgements

The authors wish to thank to the sponsors of the project “Nature, Ecology, Longevity” Aquachim (Ass. Prof. Borislav Velikov), eco hotel “Zdravetz” (Todor Burdzhiev), Bulgarian Association of Activated Water (Dipl. Eng. Atanas Atanasov), Lieselotte Eder, Marita Schirra-Saar, Roland Saar, Dr. Pascal Boesinger, Dipl. Eng. Enrico Bauer, Zohra and Manfred Dome, Paul Kleindienst. The authors wish thank for the collaboration the personal of Vortex Power Company (Matthias Mend, Alexander Class, Peter Zuecker) for the research of the melt mountain water from Glacier Rosenloui. The authors also thanks Alexander Ignatov – assistant of the research.

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