

# Vortex Power Spring Water: Physical-Chemical Qualities of this Water compared to Mountain and Melt Water from Bulgaria, Russia and Glacier Rosenlaui 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 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 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<sub>2</sub>O 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  $\lambda=8.95$   $\mu m$ . 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<sub>2</sub>O 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<sub>2</sub>O molecules in the process of cluster formation was measured by the DNES method compiles  $-0.1067 \pm 0.0011$  eV. It was shown that mountain water is among the most important factors for human longevity and human health. The variety of ions (K<sup>+</sup>, Na<sup>+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>, Mn<sup>2+</sup>, Fe<sup>2+</sup>, Fe<sup>3+</sup>, Zn<sup>2+</sup>, SO<sub>4</sub><sup>2-</sup>, CI, HCO<sub>3</sub>, CO<sub>3</sub><sup>2</sup>), 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<sub>2</sub>/dm<sup>3</sup>) relative to the control (0.9±0.09 mg O<sub>2</sub>/dm<sup>3</sup>).

Keywords: deuterium, longevity, mountain water, IR, NES, DNES, Glacier Rosenlaui, 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<sub>2</sub>O 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<sub>2</sub>O molecules, binding them into water associates (clusters) with the general formula (H<sub>2</sub>O)<sub>n</sub> 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  $\rm H_2^{16}O$ , which molecules are formed by  $^{1}H$  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 –  $\rm HD^{16}O$ ,  $\rm HD^{17}O$ ,  $\rm HD^{18}O$ ,  $\rm D_2^{16}O$ ,  $\rm D_2^{17}O$ ,  $\rm D_2^{18}O$ , while 3 configuration are formed by isotopomers of oxygen –  $\rm H_2^{16}O$ ,  $\rm H_2^{17}O$ ,  $\rm H_2^{18}O$ .



The aim of this research was studying the various samples of water from Bulgarian water springs: the melt water from Glacier Rosenlaui, 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  $D_2O$  (99.9 atom%) received from the Russian Research Centre "Isotope" (St. Petersburg, Russian Federation). Inorganic salts were preliminary crystallized in  $D_2O$  and dried in vacuum before using.  $D_2O$  distilled over KMnO<sub>4</sub> with the subsequent control of deuterium content in water by <sup>1</sup>H-NMR-spectroscopy on Brucker WM-250 device ("Brucker", Germany) (working frequency – 70 MHz, internal standard –  $Me_4Si$ ) and on Brucker Vertex ("Brucker", Germany) IR spectrometer (a spectral range: average IR – 370–7800 cm<sup>-1</sup>; visible – 2500–8000 cm<sup>-1</sup>; the permission – 0,5 cm<sup>-1</sup>; accuracy of wave number – 0,1 cm<sup>-1</sup> on 2000 cm<sup>-1</sup>).

# 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  $^{0}$ 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±7 nm. The device measures the angle of evaporation of water drops from 72.3 $^{0}$  to 0 $^{0}$ . The DNES-spectrum was measured in the range of -0.08–0.1387 eV or  $\lambda$  = 8.9–13.8  $\mu$ m 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 H<sub>2</sub>O 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 O<sub>2</sub>/dm<sup>3</sup>).

# 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 Brucker Vertex ("Brucker", Germany) IR spectrometer (a spectral range: average IR – 370–7800 cm<sup>-1</sup>; visible – 2500–8000 cm<sup>-1</sup>; the permission – 0,5 cm<sup>-1</sup>; the accuracy of wave number – 0,1 cm<sup>-1</sup> on 2000 cm<sup>-1</sup>) 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).

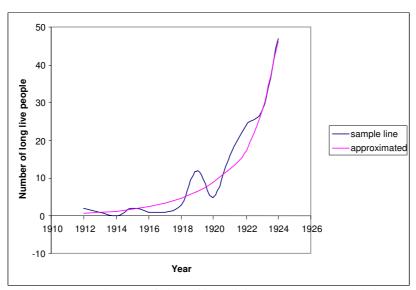


Figure 1: Interrelation between the year of birth of long living people (age) and their number in Teteven municipality, Bulgaria.

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.



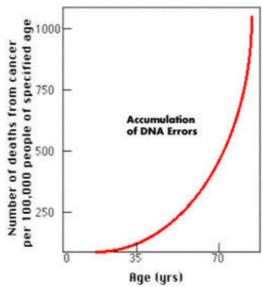


Figure 2: Interrelation between age and the number of cancer patients (Orgel, 1963).

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  $\theta$  decreases discreetly to 0, and the diameter of 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  $f(\theta)$ . 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  $H_2O$ -molecules (Antonov, 1995).

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

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

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

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

$$\theta = \arccos\left(-1 - 14.33E\right),\tag{2}$$

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  $\Delta f(E) = f$  (samples of water) – f (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  $H_2O$  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  $H_2O$  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<sub>2</sub>O 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:

$$\bar{E} = -0.1067 \pm 0.0011 \text{ eV}$$
 (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  $+36.6^{\circ}$ 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.

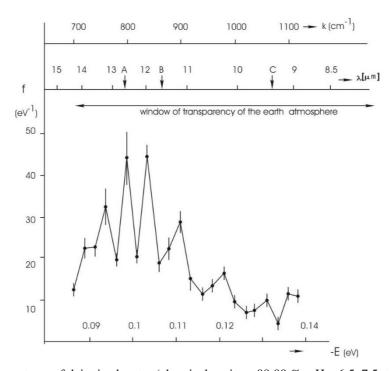


Figure 3: The NES-spectrum of deionized water (chemical purity -99.99%; pH -6.5-7.5; total mineralization -200 mg/l; electric conductivity -10  $\mu$ S/cm): the horizontal axis shows the energy of the H...O hydrogen bonds in the associates -E (eV); the vertical axis - the energy distribution function -f (eV<sup>-1</sup>); k - the vibration frequency of the H–O–H atoms (cm<sup>-1</sup>);  $\lambda$  - wavelength ( $\mu$ m)

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 blood serum and 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  $E = -9.1 \pm 1.1$  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  $\mu$ m. 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  $\mu$ m (Krasnov, Gordetsov, 2009). The resulting peak at  $\lambda = 8.95 \, \mu m$  in IR-spectrum (Ignatov, 2012) approaching the peak at  $\lambda = 8.85 \, \mu m$ monitored by Russian researchers. In the control group of healthy people the average value of the energy distribution function f(E) at  $\lambda = 8.95 \,\mu \text{m}$  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  $f(E) \sim 24.1 \text{ eV}^{-1}$ . 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 H<sub>2</sub>O 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 DNESspectrum of blood serum of healthy people at  $\lambda = 8.95 \mu m$ , 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<sup>2+</sup> 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<sup>2+</sup> 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 Rosenlaui, 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<sup>-1</sup> 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, Rosenlaui, 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  $H_2O$  molecules according to energy f(E). The function of distribution of  $H_2O$  molecules according to energy f(E) for tap water in Teteven is  $11.8\pm0.6~eV^{-1}$  (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

Bulgaria) and local extremums in NES-spectra of water					
Indicators	Results of the	Norm			
	research				
	(mg/l)				
Sodium (Na <sup>+</sup> )	0.96	< 200			
Calcium (Ca <sup>2+</sup> )	100.4	< 150			
Magnesium (Mg <sup>2+</sup> )	12.65	< 80			
Iron (Fe)	0.016	< 0.2			
Manganese (Mn <sup>2+</sup> )	0.0018	< 0.2			
Zinc (Zn <sup>2+</sup> )	0.18	<4.0			
Sulfates (SO <sub>4</sub> <sup>2</sup> -)	81.8	< 250			
Chlorides (Cl <sup>-</sup> )	3.96	< 250			
Carbonates (CO <sub>3</sub> <sup>2-</sup> )	< 2.0	-			
Hydrocarbonates (HCO <sub>3</sub> <sup>-</sup> )	184.0	-			
Other values	Results				
Active reaction (pH)	7.9	6.5-9.5			
	alkaline				
Electroconductivity	536.8 μS/cm	< 2000			
Hardness of water	16.5 dH	<33.7			
	hard				
Local	36.9	>24.1			
extremum*					
eV <sup>-1</sup>					
at (-0.13620.1387 eV)					

<sup>\*</sup>Function of distribution of H<sub>2</sub>O 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

and local extremums	-	
Indicators	Results of the	Norm
	research	
	(mg/l)	
Sodium (Na <sup>+</sup> )	4.5	< 200
Calcium (Ca <sup>2+</sup> )	55.5	< 150
Magnesium (Mg <sup>2+</sup> )	2.28	< 80
Iron (Fe)	0.0127	< 0.2
Manganese (Mn <sup>2+</sup> )	0.0014	< 0.2
Zinc (Zn <sup>2+</sup> )	0.006	<4.0
Sulfates (SO <sub>4</sub> <sup>2</sup> -)	16.9	< 250
Chlorides (Cl <sup>-</sup> )	3.4	< 250
Carbonates (CO <sub>3</sub> <sup>2</sup> -)	< 2.0	-
Hydrocarbonates (HCO <sub>3</sub> <sup>-</sup> )	118.0	-
Other values	Results	
Active reaction (pH)	7.4	6.5-9.5
	alkaline	
Electroconductivity	285.0 μS/cm	< 2000
Hardness of water	7.9 dH	<33.7
	slightly hard	
Local	40.1	>24.1
extremum*		
$eV^{-1}$		
at (-0.13620.1387 eV)		
CILLO	1 1 1'	

<sup>\*</sup>Function of distribution of H<sub>2</sub>O 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	Norm
	research	
	(mg/l)	
Sodium (Na <sup>+</sup> )	7.6	< 200
Calcium (Ca <sup>2+</sup> )	3.5	< 150
Magnesium (Mg <sup>2+</sup> )	0.63	< 80
Iron (Fe)	0.007	<0.2
Manganese (Mn <sup>2+</sup> )	0.002	<0.2
Zinc $(Zn^{2+})$	0.007	<4.0
Sulfates (SO <sub>4</sub> <sup>2-</sup> )	26.8	< 250
Chlorides (Cl <sup>-</sup> )	3.00	< 250
Carbonates (CO <sub>3</sub> <sup>2-</sup> )	< 2.0	=
Hydrocarbonates (HCO <sub>3</sub> <sup>-</sup> )	21.3	=
Other values	Results	
Active reaction (pH)	5.93	6.5-9.5
	normal	
Electroconductivity	536.8 μS/cm	< 2000
Hardness of water	1.4 dH	<33.7
	soft	
Local	59.3	>24.1
extremum*		
$eV^{-1}$		
at (-0.13620.1387 eV)	CIV.O. 1. 1	1 C(T)

<sup>\*</sup>Function of distribution of H<sub>2</sub>O molecules according to energy f(E).

Table 4: The composition of mountain melt spring Glacier Rosenlaui, Swiss Alps and local extremums in NESspectra of water

Indicators	Results of the	Norm
	research	
	(mg/l)	
Sodium (Na <sup>2+</sup> )	0.53	< 200
Calcium (Ca <sup>2+</sup> )	8.7	< 150
Magnesium (Mg <sup>2+</sup> )	0.6	< 80
Iron (Fe)	0.106	<0.2
Manganese (Mn <sup>2+</sup> )	0.0023	<0.2
Zinc (Zn <sup>2+</sup> )	0.009	<4.0
Sulfates (SO <sub>4</sub> <sup>2-</sup> )	8.4	< 250
Chlorides (Cl <sup>-</sup> )	< 1.0	< 250
Carbonates (CO <sub>3</sub> <sup>2</sup> -)	< 2.0	=
Hydrocarbonates (HCO <sub>3</sub> <sup>-</sup> )	36.0	=
Other values	Results	
Active reaction (pH)	7.3	6.5-9.5
	alkaline	
Electroconductivity	82.3 μS/cm	< 2000
Hardness of water	<1.4 dH	<33.7
	soft	
Local	70.1	>24.1
extremum*		
eV <sup>-1</sup>		
at (-0.13620.1387 eV)		

<sup>\*</sup>Function of distribution of H<sub>2</sub>O molecules according to energy f(E).

Table 5 shows the results on water composition in field area of Dolni Dabnik. The maximum peak in NESspectra of Dolni Dabnik water (eV<sup>-1</sup>) is detected at (-0.1362–-0.1387 eV), in water from Danubian Plain is detected at 23.2 eV<sup>-1</sup>, and in water from Thracian Valley – at -21.3 eV<sup>-1</sup>. In the water samples from Danubian Plain and Thracian Valley there are data for presence of nitrites (NO<sub>2</sub><sup>-</sup>), nitrates (NO<sub>3</sub><sup>-</sup>), ammonia (NH<sub>4</sub><sup>+</sup>),



phosphates (HPO<sub>4</sub><sup>2</sup>-) 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	Norm		
	research			
	(mg/l)			
Sodium (Na <sup>+</sup> )	14.2	< 200		
Calcium (Ca <sup>2+</sup> )	103.3	< 150		
Sulfates (SO <sub>4</sub> <sup>2-</sup> )	19.2	< 250		
Magnesium (Mg <sup>2+</sup> )	64.0	< 80		
Chlorides (Cl <sup>-</sup> )	9.2	< 250		
Carbonates (CO <sub>3</sub> <sup>2</sup> -)	< 2.0	-		
Hydrocarbonates (HCO <sub>3</sub> <sup>-</sup> )	184.4	-		
Other values	Results			
Active reaction (pH)	7.3	6.5-9.5		
	alkaline			
Hardness of water	29.1 dH	<33.7		
	Very hard			
Local	23.2	>24.1		
extremum*				
eV <sup>-1</sup>				
at (-0.13620.1387 eV)				

Table 6 shows the optimal chemical composition of water, hardness, the local extremum (eV<sup>-1</sup>) 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	Results of melt
	mountain water	water
	(Bulgaria)	(Russia)
	(mg/l)	(mg/l)
Sodium(Na <sup>+</sup> ) + Potassium	6.1	< 30
$(K^{+})$		
Calcium (Ca <sup>2+</sup> )	29.5	< 50
Magnesium (Mg <sup>2+</sup> )	1.5	< 10
Iron (Fe)	0.083	-
Manganese (Mn <sup>2+</sup> )	0.0017	_
Zinc (Zn <sup>2+</sup> )	0.007	_
Sulfates (SO <sub>4</sub> <sup>2-</sup> )	21.9	< 100
Chlorides (Cl <sup>-</sup> )	3.2	< 70
Carbonates (CO <sub>3</sub> <sup>2</sup> -)	< 2.0	ı
Hydrocarbonates (HCO <sub>3</sub> <sup>-</sup> )	69.7	< 100
Other values	Results	
Active reaction (pH)	6.7	6.5-7.0
	normal	
Electroconductivity	410.9 μS/cm	< 2000
Hardness of water	4.65 dH	<33.7
	Moderately soft	
Total mineralization (g/l)	0.132	< 0.3
Local	49.7	>24.1
extremum*		
$eV^{-1}$		
at (-0.13620.1387 eV)		



# 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<sup>-1</sup>) in spectra of water samples as a function of distribution of H<sub>2</sub>O molecules according to energy f(E) at  $\lambda = 8.95 \mu m$  shows the analogue peak at analogous values of f(E), E and  $\lambda$ , which was detected in water with Ca<sup>2+</sup> ions inhibiting the growth of cancer cells. Magnesium (Mg<sup>2+</sup>), zinc (Zn<sup>2+</sup>) and manganese (Mn<sup>2+</sup>) 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<sup>2+</sup> in plasma. An inverse correlation was also found between the content of Zn<sup>2+</sup> ions in plasma relative to age. The prevalence of Zn<sup>2+</sup> deficiency is increased with age; with normal Zn<sup>2+</sup> 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),  $\beta$ -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  $\mu$ g. For the  $Na^+$  content the norm according to the WHO is less than 20  $\mu$ g.

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 ( $HCO_3 \cdot MgCO_3 \cdot MgC$ 

# 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

Table 7. The local extremums in NES spectra of water samples of different origin						
Sources and	Local	Local	[%,(-	[%,(-	[%,(-	[%,(-
Types of	extremum*	extremum**	$E_{value}$ )/(- $E_{total}$	$E_{value}$ )/(- $E_{total}$	$E_{value}$ )/(- $E_{total}$	$E_{value}$ )/(- $E_{total}$
Waters	at (-	at (-	value)]	value)]	value)]	value)]
	0.1112	0.1362	-0.1112 eV	-0.1137 eV	-0.1162 eV	-0.1187 eV
	0.1137 eV)	0.1387 eV)				
	eV <sup>-1</sup>	eV <sup>-1</sup>				
	norm	norm				
	(>24.1)	(>24.1)				
1.Zlatishko-	46.1±2.3	36.9±1.9	4.1	7.0	4.0	7.4
Tetevenska						
Mountain						
(Bulgaria)						
2.Vasiliovska	12.0±0.6	44.9±2.2	1	2	5	6.2
Mountain						
(Bulgaria)						
3.Rhodops	53.3±1.6	59.3±3.0	11.0	3.6	5.5	13.0
Mountain (Eco						
hotel						
Zdravetz)						
(Bulgaria)						
4. Deionized	70.6±3.5	47.1±2.4	5.9	11.8	0	11.8
Water with						
Shungite						
(Bulgaria)						
5. Deionized	0	50.1±2.0	0	0	0	12.5
Water with						
Zeolite						
(Bulgaria)						
6. Danubian	25±1.25	23.2±1.7	0	6.3	0	3.1
Plain,						
Sadovetz						
(Bulgaria)						
7. Tap Water	16.0±0.8	16.0±0.8	4.0	0	0	4.0
Teteven						
(Bulgaria)						
8. Glacier	129.0±6.3	70.1±3.5	22.6	9.7	6.5	12.9
Rosenlaui						
(Switzerland)						
9. Vortex	88.8±4.4	74.1±3.4	7.4	14.8	7.4	11.1
Power Water						
(Switzerland)						
10. Deionized	38.7±1.9	32.3±1.6	3.2	6.4	4.8	3.2
Water	30.7=1.7		5.2			
		I	I		I	

Notes:

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  $74.1\pm3.7$  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<sub>3</sub>) 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  $88.8\pm4.4$  eV $^{-1}$ . The result of tap water of Teteven was observed within a statistical error or  $16.0\pm0.8$  eV $^{-1}$  for -0.11 eV and -0.1387 eV. The measurement of the DNES spectrum for Vortex Power Spring water was  $72.8\pm3.6$  eV $^{-1}$  in the range (-0.1112-0.1137 eV). The result in the range (-0.1362-0.1387 eV) of DNES

<sup>\*</sup>The values (E= -0.1112...-0.1137 eV) correspond to the wavelengths ( $\lambda$ =10.91–11.91 $\mu$ m).

<sup>\*\*</sup>The values (E= -0.1362...-0.1387 eV) correspond to the wavelengths ( $\lambda$ =8.95–9.10  $\mu$ m).



spectrum was 58.1±2.9 eV<sup>-1</sup>. 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  $\lambda = 8.95~\mu m$  was detected at an average value of 75.3±3.8 eV<sup>-1</sup>. In the group of people in critical condition this value was 24.1±1.2 eV<sup>-1</sup>. 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±3.7 eV<sup>-1</sup>. 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 [%, (- $E_{value}$ )/(- $E_{total\ value}$ ] 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...-0.1137 eV) and (-0.1362...-0.1387 eV). The average energy ( $E_{H...O}$ ) of hydrogen H...O-bonds among individual  $H_2O$  molecules in Vortex Power Spring water was measured at E=-0.1195 eV. The result for the control sample (deionized water) was E=-0.1102 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±0.0011 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 eV. Thus, the result for Vortex Power Water measured with the DNES method was  $\Delta E$ =-0.008±0.0011 eV.

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

Another important physical parameter was calculated with using the NES method – the average energy ( $E_{H...O}$ ) of H...O-bonds between  $H_2O$  compiled –  $0.1067\pm0.0011$  eV.

According to the analysis of various water samples by the NES and DNES methods can be drown 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  $(H_2O)_n$  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 Rosenlaui there was the increasing of surface tension regarding the control samples;
- The redistribution of H<sub>2</sub>O 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 1–3 kcal/mol or 0.043–0.13 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 eV is decreasing. In this group of tested people the result obtained by the DNES method was  $-\Delta E = -5.5 \pm 1.1$  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 Wartburg

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±0.0011 eV. The result for tap water as a control sample from the Danubian plain water with a local extremum at (-0.1362–0.1387 eV) was 12.5 eV<sup>-1</sup>. The result for tap water after the treatment of the water sample from Danubian plain by Vortex Power apparatus was 75 eV<sup>-1</sup> with a local extremum at (-0.1362–0.1387 eV).

The result before using of Vortex Power Spring apparatus on permanent oxidation of water was  $0.9\pm0.09$  mg  $O_2/dm^3$ . After the usage of Vortex Power Spring apparatus the result was  $1.26\pm0.13$  mg  $O_2/dm^3$ . Thus, there is statistically reliable result measured at  $0.36\pm0.04$  mg  $O_2/dm^3$  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 (mg  $O_2/dm^3$ ).

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--0.1387~eV) - 75  $eV^{-1}$  The result of NES – (-0.1246~eV)

The research of Tap Water from Danubian plain: Local extremum at (-0.1362--0.1387 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 (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 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<sup>\delta+</sup>-O<sup>\delta</sup>. 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 ↔ 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<sub>2</sub>O 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<sub>2</sub>O molecule to approach the hydrogen atom of another molecule at very close distance without experiencing any strong electrostatic repulsion. The H<sub>2</sub>O 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<sub>2</sub>O 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:

- a) 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.
- b) On the line of O-atom there can be disposed only one proton H<sup>+</sup>;
- c) 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.
- d) 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<sub>2</sub>O 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  $\pm$  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:  $(H_2O)_2 = H_2O^-HOH$ .

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 H<sub>2</sub>O 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 H<sub>2</sub>O 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 H<sub>2</sub>O molecules (Figure 4) (Saykally, 2005). It was also estimated the possible number of hydrogen bonds (100) depending on the number of H<sub>2</sub>O molecules (250) in clusters (Sykes, 2007). O. Loboda and O.V. Goncharuk provided data about the existence of icosahedral water clusters consisting of 280 H<sub>2</sub>O molecules with the average size up to 3 nm (Loboda & Goncharuk, 2010). The ordering of H<sub>2</sub>O 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  $\Delta H$  minus the change in entropy  $\Delta S$  (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 H<sub>2</sub>O 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 H<sub>2</sub>O 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  $H_2O$ molecules in the formation of cluster associates with the formula  $(H_2O)_n$  compile -0.1067  $\pm$  0.0011 eV. As the energy of hydrogen bonds between H<sub>2</sub>O molecules increases up to -0.14 eV, the cluster formation of water becomes "restructuring". In this case, the redistribution of energies among the individual  $H_2O$  molecules occurs.

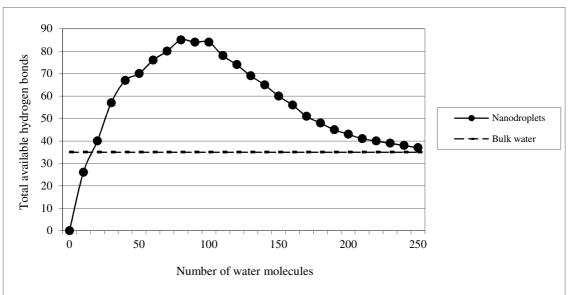


Figure 4: The total number of hydrogen bonds depending on the number of H<sub>2</sub>O molecules in clusters.

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  $H_2O$  molecules (Tokmachev *et al.*, 2010). Associates can be perceived as unstable groups (dimmers, trimmers, tetramers, pentamers, hexamers etc.) in which  $H_2O$  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  $H_2O$  molecules may vary from 2 to 6. In 2000 it was deciphered the structure of the



water trimmer, 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  $H_2O$  molecules (Figure 5a–c). Subsequently, it was shown that  $H_2O$  molecules capable of hydrogen bonding by forming the structures representing topological 1D rings and 2D chains composed from numerous  $H_2O$  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.

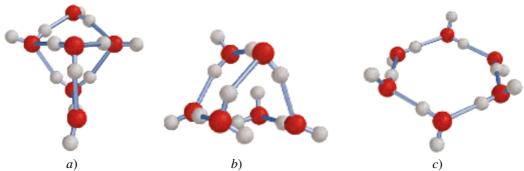


Figure 5: Computer calculations of small water cluster structures (a-c) with general formula  $(H_2O)_n$ , where n = 6 (Tsai & Jordan, 1993).

#### 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<sub>h</sub>, hexagonal lattice) is usually much cleaner than water, because the solubility of all substances (except NH<sub>4</sub>F) 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 "icelike" 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 H<sub>2</sub>O 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 H<sub>2</sub>O molecule with four neighboring H<sub>2</sub>O 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 °C leads to a loss of the biological activity. The storage of melted water at +22 0C 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. H<sub>2</sub>O 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  $^{\circ}$ C to 4,9 at +80  $^{\circ}$ 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  $104^{0}5$ ) (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 verticess – H-atom, which electron takes part in formation of a covalent bond with an electron pair of O-atom. The rest two vertices 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  $H_2O$  molecules.



Table 8: Ice crystal	modifications	and their phy	vsical ch	aracteristics

Modification	Crystal structure	Hydrogen bond lengths,	Angles H–O–H in
		Å	tetragonals, <sup>0</sup>
$I_h$	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  $H_2O$  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:  $H_2O$  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  $t = +40~^{\circ}C$  breaks down about half of hydrogen bonds in water associates. In the water vapor heated at  $t \geq +100~^{\circ}C$  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,  $H_2O$  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  $^{0}C$ . 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 helixes, 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  $H_2O$  molecules (Figure 7).

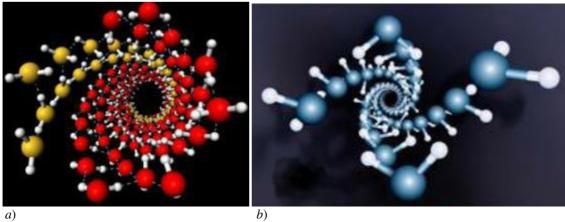


Figure 6: Spiral structures of water nanocrystals in carbon nanotubes according to computer simulations (Bai *et al.*, 2006) (diameter of nanotubes 1.35–1.90 nm; pressure 10–40000 atm; temperature -23  $^{0}$ C): *a*) – a general view of the crystal structure of water in nanotubes, *b*) – the inner wall of the structure of water.



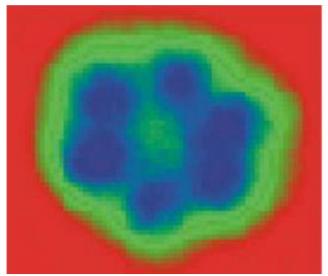


Figure 7: Image of hexamer nanocrystal of ice water (average size ~1 nm) consisting of six hexagonally attached H<sub>2</sub>O molecules (blue colour) obtained by a scanning tunneling microscope after freezing water to 17 K on the hydrophobic metallic plates of Cu and Ag (Michaelides & Morgenstern, 2007).

#### 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  $Ca^{2+}$ ,  $Mg^{2+}$ ,  $Na^+$ ,  $HCO_3^-$  and  $SO_4^{2+}$ . The content of  $K^+$  and  $N^+$  cations in the melt water are <30 mg/l,  $Mg^{2+}$  <10 mg/l,  $Ca^{2+}$  <40 mg/l, the content of  $SO_4^{2-}$  <100 g/l,  $HCO_3^-$  <90 mg/l,  $CI^-$  - less than 70 mg/l, total rigidity  $\leq 4$  mEq/l, the total mineralization  $\leq 0.2$  g/l, pH - 6.5 - 7.0 at  $25^{-0}C$ . The degree of natural purification of melt water from impurities makes up ~50–60%. The concentration of salts of rigidity – Ca<sup>2+</sup>, Mg<sup>2+</sup>, Fe<sup>2+</sup>, Fe<sup>3+</sup>, 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  $^{18}$ O in sea water: D/H =  $(155.76\pm0.05)\cdot10^{-6}$  (155.76 ppm) and  $^{18}$ O/ $^{16}$ O =  $(2005.20\pm0.45) \cdot 10^{-6} (2005 \text{ ppm})$  (Lis *et al.*, 2008). For SLAP standard isotopic shifts for D and <sup>18</sup>O in seawater: D/H =  $89 \cdot 10^{-6} (89 \text{ ppm})$  and for a pair of  $^{18}\text{O}/^{16}\text{O} = 1894 \cdot 10^{-6} (1894 \text{ ppm})$ . In surface waters, the ratio D/H =  $\sim$ (1.32–1.51) 10<sup>-4</sup>, while in the coastal seawater  $\sim$  (1.55–1.56) 10<sup>-4</sup>. Waters of other underground and surface water sources contain varied amounts of deuterium (isotopic shifts) – from  $\delta = +5,0$  D,%, SMOW (Mediterranean Sea) to  $\delta = -105$  D,%, SMOW (Volga River). The natural waters of CIS countries are characterized by negative deviations from SMOW standard to (1.0–1.5) 10<sup>-5</sup>, in some places up to (6.0–6.7) 10<sup>-5</sup>, but there are observed positive deviations at  $2.010^{-5}$ . The content of the lightest isotopomer –  $H_2^{16}O$  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 <sup>18</sup>O. That is why it is so important to purify drinking water from heavy isotopes of D and <sup>18</sup>O.

We have obtained results on growth and adaptation to  $D_2O$  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%  $D_2O$ , plants – up to 50–60%  $D_2O$ , and protozoa cells are able to live on ~90%  $D_2O$ . Further increase in the concentration of  $D_2O$  for these groups of organisms leads to cellular death. On the contrary, deuterium depleted water with the decreased deuterium content (60–100 ppm) has benefitial



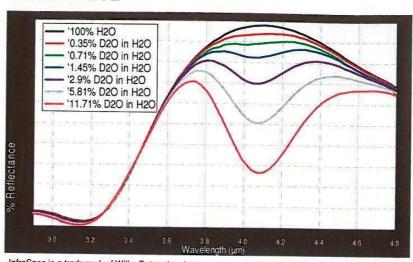
effects on metabolism.

As demonstrated in IR-spectra of water of different content of deuterium measured on a device Infra Spec VFA-IR, that at  $\lambda=4.1~\mu m$ ; 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<sub>2</sub>O 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  $(v_1)$ , asymmetric stretch  $(v_3)$  and bending  $(v_2)$  of the covalent bonds with absorption intensity  $(H_2O)$   $v_1;v_2;v_3=2671$ ; 1178.4; 2787.7 cm<sup>-1</sup>. For liquid water absorption bands are observed in other regions of the IR-spectrum, the most intense of which are located at 2100, cm<sup>-1</sup> and 710–645 cm<sup>-1</sup>. For the  $D_2O$  molecule these ratio compiles 2723.7, 1403.5 and 3707.5 cm<sup>-1</sup>, while for HDO molecule – 2671.6, 1178.4 and 2787.7 cm<sup>-1</sup>. HDO (50 mole%  $H_2O + 50$  mole%  $^2H_2O$ ; ~50 % HDO, ~25 %  $H_2O$ , ~25 %  $D_2O$ ) has local maxima in IR-spectra at 3415 cm<sup>-1</sup>, 2495 cm<sup>-1</sup> 1850 cm<sup>-1</sup> and 1450 cm<sup>-1</sup> 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 (Zelsmann, 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<sub>2</sub>O 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<sub>1</sub> band at 395.5 cm<sup>-1</sup>; major L<sub>2</sub> band at 686.3 cm<sup>-1</sup>; for liquid water at 0 °C, the absorbance of L<sub>1</sub> increasing with increasing temperature, while L<sub>2</sub> 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 H<sub>2</sub>O 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 absorbtion bands in the IR-spectrum of liquid water is given in Table 8. The IR spectrum of  $H_2O$  molecule was examined in detail from the microwave till the middle (4–17500 cm<sup>-1</sup>) visible region and the ultraviolet region – from 200 nm<sup>-1</sup> to ionization limit at 98 nm<sup>-1</sup> (Walrafen, 1972). In the middle visible region at 4–7500 cm<sup>-1</sup> 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<sup>-1</sup>) 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  $\mu$ 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 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 H<sub>2</sub>O and D<sub>2</sub>O

Main vibrations of liquid H <sub>2</sub> O and <sup>2</sup> H <sub>2</sub> O					
Vibration(s)	$H_2O (t = +25  {}^{0}C)$ $D_2O (t = +25  {}^{0}C)$		$t = +25  {}^{0}C$		
	v, cm <sup>-1</sup>	E <sub>0</sub> , M <sup>-1</sup> cm <sup>-1</sup>	v, cm <sup>-1</sup>	E <sub>0</sub> , M <sup>-1</sup> cm <sup>-1</sup>	
Spinning $v_1$ + deformation $v_2$	780–1645	21.65	1210	17.10	
Composite $v_1 + v_2$	2150	3.46	1555	1.88	
Valence symmetrical $v_1$ , valence asymmetrical $v_3$ , and overtone $2v_2$	3290-3450	100.65	2510	69.70	

At further transition from H<sub>2</sub>O monomers to H<sub>4</sub>O<sub>2</sub> dimmer and H<sub>6</sub>O<sub>3</sub> trimmer absorption maximum of valent stretching vibrations of the O-H bond is shifted toward lower frequencies ( $v_3 = 3490 \text{ cm}^{-1}$  and  $v_1 = 3280 \text{ cm}^{-1}$ cm<sup>-1</sup>) (Eisenberg & Kauzmann, 1969) and the bending frequency increased ( $v_2 = 1644 \text{ cm}^{-1}$ ) 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<sup>-1</sup> were attributed to the stretching modes of the dimmer [(H<sub>2</sub>O)<sub>2</sub>]. These frequencies are significantly lower than the valence modes of  $v_1$  and  $v_3$  vibrations of isolated  $H_2O$  molecules at 3657 and 3756 cm<sup>-1</sup> respectively). The absorption band at 3250 cm<sup>-1</sup> represents overtones of deformation vibrations. Among frequencies between 3250 and 3420 cm<sup>-1</sup> 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<sup>-1</sup> is attributed to the deformation mode of the dimmer. This frequency is slightly higher than the deformation mode of the isolated H<sub>2</sub>O molecule (1596 cm<sup>-1</sup>). 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<sup>-1</sup> and very weak temperature dependence. It changes little in the transition to the individual H<sub>2</sub>O molecule at a frequency of 1595 cm<sup>-1</sup>. 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 H<sub>2</sub>O 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  $D_2O$  (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 Rosenlaui, 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  $\lambda = 8,95$ μm. 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<sub>2</sub>O molecules and the distribution function of H<sub>2</sub>O 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 IRspectrum 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<sup>+</sup>, Na<sup>+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>, Mn<sup>2+</sup>, Fe<sup>2+</sup>, Fe<sup>3+</sup>, Zn<sup>2+</sup>, SO<sub>4</sub><sup>2-</sup>, Cl<sup>-</sup>, HCO<sub>3</sub><sup>-</sup>, CO<sub>3</sub><sup>2-</sup>), 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<sup>2+</sup>, Mg<sup>2+</sup>, Zn<sup>2+</sup> and Mn<sup>2+</sup> in composition of water on human organism and DNES-spectra of water. There are results of chemical composition of water from Glacier Rosenlaui, 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.

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