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BIOLOGICAL ROLE OF HEAVY WATER IN LIVING ORGANISMS

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Researching of water and its relation with its biological structure reveals an exciting and promising area of studies that may lead us far in understanding of normal and pathological processes of life. Indeed, the structure of water proposes a new perspective on life itself. Until now water was regarded as a more or less neutral medium filling the space between the structural elements of the cell. «Water is an integral part of the living organism, and not just its environment». In such a way the role of water is characterized by well-known biophysicist Albert Szent-Györgyi in his book «Bioenergetics».

We will focus on a property of water, which has not yet been attached special importance. It will be a question of biological role of that small amount of heavy water which is contained in ordinary water and, therefore, in organisms. The vast majority of ordinary water molecules is H_2O with impurity of HDO molecules. Genuine heavy water has molecules of a type of D_2O (the difference in the oxygen isotopes is neglected in all cases).

When heavy water was discovered (1932), the study of its effect on biological objects when used in high concentrations began. Concentrated heavy water, in which all hydrogen atoms are replaced by deuterium, differs from ordinary water in physical properties. In ordinary water the presence of a small amount (0,0149%) of heavy water practically does not affect its physical properties. Compounds with normal and heavy hydrogen also differ from each other in chemical properties. Replacing of usual hydrogen by heavy one primarily leads to reducing of the rate of chemical reactions (by 3-10 times). Naturally, the replacement of a significant part of the hydrogen atoms with deuterium will lead to a slowdown in many chemical reactions that determine complex biological processes in cells, tissues and body as a whole.

Alotofliteratureisdevotedtosuchresearches.

However, it is already possible to make a conclusion that the most common physical and biological considerations indicate that a small change in concentration of heavy water in organism (even close to its “normal” value) can give a great biological effect. Indeed, if we look at a formula of any organic molecule, the eye catches the abundance of hydrogen atoms. Hydrogen atom as the lightest, the most agile and easily converted into H⁺ ion or forming an OH⁻ ion together with an oxygen atom plays a very special role in all biochemical processes in living organisms. The course of biological processes is inevitably linked with such physical phenomena as diffusion through porous membranes, isotope exchange reactions etc., which cause a separating isotope effect, and due to it the ratio between the number of H and D atoms in different parts of the body will be different from the average for the body. It is known that as time goes by heavy water accumulates in the body. This accumulation in specific organs may also go faster than the average for the body due to the above mentioned physical causes. And quite special effects can cause a substitution of deuterium atoms to hydrogen atoms in the molecules of DNA. Substitution in the DNA, for example, 10-25% of deuterium atoms by hydrogen atoms will not cause a catastrophe, of course (only about 0.015% of the hydrogen atoms are deuterium), since it will not change the general plan of development of the organism encoded in DNA (heavy hydrogen is still hydrogen), but this change could affect the pace of construction of new DNA molecules and thus the whole life of an organism. These assumptions about the possible mechanism of action of small changes in the concentration of heavy water opens the field for research in the broadest sense, beginning with general biological questions and ending with issues of gerontology and geriatrics. Experiences with low deuterium water are very accessible. Nature gives us such water in great amounts – this is first of all the snow water. Snow water (prepared with certain precautions) compared with usual river water or rain water contains not 0.0149% of heavy water, but approximately 1/4 less (i.e. about 0,012%). Water obtained from frost is similar in composition to the snow water. When ice melts, the first batch of melt water is deuterium-depleted, but later portions will approach to usual river water in the content of heavy water. For sustainable results it is possible to recommend only snow water.

Initial point in our researches was the evident feature in the distribution of the number of long-lived people (over 100 years) in regions of the USSR. If we count that the number of long-lived people per million inhabitants, in the Chechen-Ingush there will be 353 of them, in Yakutia - 324 people. On average there are only 81 of them in the RSFSR. These facts can be explained, in particular, by the fact that the Caucasian mountaineers drink water from the melted alpine glacier snow; inhabitants of Yakutia should use snow water because of rivers freezing during the long winter. In both cases, drinking water turns out to be deuterium depleted.

To test whether these effects are connected with concentration of heavy water, we organized a series of experiments. In experiments with tissue cultures (L. Korochkin (1962) Central research laboratory of Tomsk Medical Institute) was studied the effect of light and heavy water on cell cultures (Hel, Hel-2 cells, deuterium-6, 580), liver and fibroblasts of chick embryos. In this case, it was found that **culturing cells on heavy water dramatically accelerates the aging process** and leads to the death of culture on 3–4 day. The opposite effect was obtained when culturing these cells **on light (snow) water. The aging process was delayed**, even for 7-8 days did not come their degradation, but the cells showed signs of growth.

Serious experiments were carried out on mice (1959 – 1960). In the first series of experiments were used 150 adult (6 to 10 weeks) mice of clear line 0.57. Animals received **weighted water** as drinking water (3% of heavy water) instead of usual one and the usual food ration. Mice treated with usual served as control water. For several months the test animals maintained their weight, **remained active**, coat was brilliant and clean. Pregnancy and childbirth with normal amounts of mice - 5 – 6 – came in usual time (number of fruits within the specified limits has been calculated for 5,000 mice of previous generations which consumed regular water). Process of lactation did not change. **However, the weight of newborn mice was on average 20% less** than weight of control ones. At the same time markedly predominant male sex (about 75%). **Development** of the first [born] generation was **significantly inhibited**, despite good general condition and sufficient activity **very slow growth** was observed (at the age of 2 months the weight was 5.95 g when the weight of control ones - 15 - 17 g). The color of coat slightly changed - instead of shiny and black one as the parents has, the offspring had **hazy or brownish and dull coat**. Obviously puberty was late. Pregnancy occurred in only 8-week-old females, and accordingly, the childbirth at 11-week-old. **Males were not sexually active**, though they were held together with females.

The females of the second generation, despite the pregnancy, **were incapable of lactation**, and **newborns**, born in the number of 3-4, **died**. The second pregnancy ended in the same result, so the **third generation could not be obtained**.

Quite different results were obtained during the experiments carried out on the same line of mice and at the same feeding regime. 84 mice were taken in experiment. **Maintenance on snow water caused an increase in sexual activity in males**, and it maintained throughout the period of observation. The females obviously expressed multifetation (8 – 10 little mice with a high weight of offspring: 5.8 g in the experiment, 5.53 g in control and greater weight gain, the experimental animals - by 190% in 10 days, the control - 137%, within twenty days - 393% in the experimental, by 339% in the control group, and in the future, this growth rate was maintained). These observations were the basis for setting up the experiments on some animals with economic value (1960–1963). Results of these experiments were published in periodical press since 1961

(for example, the magazine «Agriculture of Siberia», № 7, 1961). These experiments were repeated with positive results in many agricultural institutions, state and collective farms on different animals and plants (i.e. snow water led to a significant increase in weight gain, egg production, fertility and resistance to diseases), if the following recommendations for the preparation of snow water were implemented: take the snow was not eroded, melt it in a sealed container to avoid evaporation loss, which leads to changes in the isotopic composition of water. It is necessary to mention one experiment with thoroughbred laying hens, giving up to 260 eggs per year. In this experiment the snow water didn't demonstrate a stimulating effect (carried out in the Scientific Research Institute of Poultry in Zagorsk). This is obviously due to the fact that egg-laying capacity of chickens is close to a biological limit.

These were the results of the first, intelligence cycle of experiments with snow water. Recent experiments were aimed at clarifying the mechanism of snow water action. There is also another point of view on the effect of snow water, well-established even before the discovery of the existence of heavy water in ordinary water. According to this point of view, the effect of snow water is connected with the fact that after the snow (or ice) is melted there still continue to exist some kind of "shatters" of ice crystals and their amount increases as the temperature of water reaches 0. And these fragments of the ice structure «ease» the biochemical reactions in organisms that consume snow (or ice) water, - like artificially prepared beeswax helps bees to quicker build honeycom.

Methodical requirements for experiments from both points of view are completely opposite. While from our point of view for some experiments we can bring snow water to a boil (or even distillate) and then cool it to room temperature, without changing its properties (for this purpose it is only necessary to minimize the loss of water through evaporation); from the point of view of structural theory boiling should completely eliminate the difference between snow and plain water.

There were also points of view that snow water may contain microelements from the atmosphere, nitrogen may be captured, and these impurities may be responsible for all effects of snow water.

Experiments with seeds, soaked in snow water were very revealing (1964–1965). If we split portion of the snow water into two parts, and add to one of them concentrated heavy water in such an amount (0.003%) to adjust its concentration to the normal concentration of it in the usual water, neither ice structure of snow water nor the chemical composition will change. We soaked barley seeds before sowing in there types of water: 1st variant – snow water without addition of heavy water; 2nd variant – snow water with addition of heavy water in the proportion stated

above, 3rd variant (control) – usual tap water. Soaking lasted about 8 hours. After the soaking the seeds were dried and sown in plots. In variant with snow water weight per plant exceeds the weight of control ones in the flowering stage by 13% in the phase of maturity - 25%, tillering has not changed. The variant with seeds soaked in snow water had advantages in characteristics of the head and grain. The variant with snow water exceeds the variant with tap water: in the length of the head – by 13%, in the number of spikelets per head – by 13%, in the absolute weight of grain – by 18%, in biological productivity – by 25%. The variant with snow water with the addition of heavy water and the variant with tap water gave similar results; in terms of spike and grain the variant with the addition of heavy water even lags behind a little.

Experiment with wheat was carried out in the field. In the fields of Tomsk regional experiment station two similar sections of one-quarter hectare have been allocated for wheat. On one dry seeds were sown, on other – seeds, presoaked in snow water. Soaking was performed on the day of seeding during one and a half hours. Seeds were dried to a state of flowability and sown by a seed drill. During the growing season the difference was imperceptible to the eye. In both variants a phase development advanced at the same time. And only before harvesting the area where the seeds were soaked in snow water the wheat looked visibly different. Plants significantly exceeded the controls in height, stalk's diameter and head's size. The length of the head on snow water increased by 43%, number of spikelets per head - by 28%, the absolute weight of seeds - 22%, biological yield - 56%.

The total yield from the “snow” area per hectare was 18,3 quintals, the control – 11 quintals. Experience with wheat was repeated in 1965 in the state farm “Tomsky” on a larger scale. The total area of experience was 82 hectares; wheat seeds soaked in snow water were sown on 10 hectares. The crop from the soaked seeds – 14 tons per hectare, from the dry seeds – 11,3 quintals (yield increase of 22%).

How can we explain the results of these experiments?

The fact that only short-term soaking of seeds in snow water causes changes in all subsequent development of the plants cannot be explained from the point of view of the structure of snow water. In the experiment with barley ice structure in snow water and in snow water with addition of heavy water was obviously identical, but the results were quite different.

From our point of view such explanation can be offered. During steeping snow water seeps into the seed and penetrates almost every molecule of grain, replacing the “old” hydrogen atoms with the “new” ones. As we have already said, the replacement in hydrogen bonds and spirals of DNA molecules or nucleotides themselves leads to speedy construction of new DNA molecules.

It is necessary in this regard to note that replacement of ordinary hydrogen by deuterium is not equal to reverse replacement, so far as it concerns structures under construction (both living and non-living). For example, the snow itself is depleted in deuterium because during the formation of snow from water vapor into the snow crystals water molecules not containing deuterium align firstly, as this gives the most "pure" structure with a minimum of impurities, which in this case are deuterium. This phenomenon is called "melting zone" and is used for the purification of germanium and other materials. In cruder form, this effect appears in the freezing salt water when the ice is bland, so also in the construction of the structures of living cells in the maximum "screening" of impurities that may violate the "purity" of the structure. As in our case such impurity is deuterium, depleting of DNA molecules of the grain in deuterium by soaking them in snow water, we'll create a system, which will be stable to the penetration of deuterium, - and this ensures a lasting effect of snow water on the body.

Microscopic alga *Chlorella* turned out to be a very convenient object for experiments with snow water, firstly, because experiments with it can be performed in the most "pure" conditions.

Experience (1965) was put in two versions: one - nutrient mixture was prepared on the pre-distilled snow water, in the second - a solution also was prepared on distilled snow water, but with the addition of 0.03 milliliters of concentrated heavy water for each liter of solution.

Medium prepared on plain distilled water served as the control. Thus, the second variant of water approached the concentration of deuterium in ordinary water. Since the first and second solutions are made of the same portion of snow water, the difference between them is only in the concentration of heavy water, there won't be any difference in chemical composition (if it is possible to speak about impurities in snow distilled water). The results of the last two experiments are shown in Table 1.

Table 1

	Weight of chlorella mass per unit volume in g.		
	Control	Pure snow water	Snow water with the addition of heavy water
First test	0,03141	0,006185	0,002300
	100%	196,6%	73,2%
Second test	0,00338	0,006083	0,002483
	100%	179,9%	73,4%

Thus, the addition of heavy water entirely eliminates the stimulating effect of pure snow water.

This experience very clearly emphasizes the role of the isotopic composition of water. Influence of heavy water concentration on sex of offspring was discovered in experiments with mice. To clarify this effect, experiments on fruit flies were conducted (1962–1965). Fruit flies were chosen because of the convenience of experimenting with them. Culture media, on which flies were grown, had different concentrations of deuterium. In particular, experiments were conducted with snow water, with simulated snow water (in DDW with 0 ppm concentrated heavy water was added in an appropriate amount); in addition the effect of more considerable concentration of heavy water was examined (2% and 3%). Control flies were grown in medium prepared with tap water. Total number of flies in each series reached 1500. The material was processed statistically. Table 2 shows the results of one experiment.

Table 2

	Tap water	Snow water	Artificial snow	Weighted water	
				2%	3%
Males, %	49,6	41,9	46,6	55,9	53,3
Females, %	50,5	53,1	53,4	44,1	46,7

Thus, the lower content of heavy water leads to a predominance of women in offspring.

Observations were made for the sex ratio in several successive generations of flies, grown on the snow water. Results obtained for 32000 mice are shown in table 3.

Table 3

	1 st generation	2 nd generation	3 rd generation	Control
Males	41,4	40,7	39,6	48,0
Females	58,6	59,3	60,4	52,0

However, if the flies, reared on the snow water for several generations, would be transferred on the tap water back, the next generations would give usual ratio of male and female mice. Thus, the experiments have shown quite clearly the biological effect almost entirely (if not entirely) dependent on the concentration of deuterium and water. This effect of snow water can be opposed to universal dampening effect of heavy water in large concentrations. Apparently, the concentration of deuterium in the snow water is biologically optimal (rather than one that normal

water has). 3rd cycle of work will be devoted to the elucidation of the finer details of heavy water action on the organism. One of the works of this cycle is finished. It has shown that heavy water accumulates in the ovaries of female white mice, and this accumulation of heavy water induces the change of morphology and function of the ovaries, which is expressed in death of oviducts, and this process extends not only on mature oviducts, but also on oviducts in the early phases of their development. The opposite pattern is observed under the influence of snow water. Were obtained statistically reliable figures showing the stimulation of egg cells, and this is due to an increase in fertility when consuming snow water. Necrobiotic processes in eggs has never been found. Thus, one of the ways of impact of snow water on organisms is revealed. This way might have been expected on the basis of considerations set out earlier in this article.

Translated by Deeva Maria