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Spin-dependent absorption of water molecules

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Abstract

The effects of spin state of water molecules on its absorption on lyophilized DNA, lysozyme and some inorganic sorbents were studied. It was shown that the absorption rates of ortho and para water from vapor differ noticeably. The para isomer binding with preparations is distinctly faster than that of the ortho isomer in all cases. Clear-cut distinction in the sorption kinetics is determined by the difference in quantum statistics for spin isomers, which in its turn can give rise to remarkable differences in physico-chemical properties of ortho and para water. This finding opens a wide field of activity in studying fundamental and applied problems relating to the role of the spin state of water molecules in physics, chemistry, biology and medicine.

Keywords: Water; Spin isomer; Ortho; Para; Sorption; Properties

1. Introduction

All processes in cells occur in an aqueous medium. Water molecules are either directly (chemical reactions with involvement of water, water as ligands etc.) or indirectly (hydrophobic interactions [1]) involved in many biochemical processes. However, up to the present, the role of the spin in the formation of physical and chemical properties of water has been underestimated yet.

Analogous to hydrogen molecules, water molecules take on two different spin states—ortho (parallel nuclear spins of hydrogen atoms) and para (opposite spins). In water at the temperature above 50 K, these spin isomers are presented in a ratio of 3/1. It is notable that quantum mechanics laws prevent spin isomers from interconversion upon collision of molecules and due to radiative transition [2]. Spin isomers of water molecules remain unaltered in a gas phase for a long time. Thus, vapor can be regarded as a mixture of two different gases—ortho water and para water. Moreover, spin conversions virtually do not take place in a solid phase [3]. The ortho/para (OP) ratio is maintained at least for several months in ice. Spin isomers can be long-lived even in a liquid phase at rather low temperatures or if exchange effects are restricted.

To assess the energy of interaction of water molecules with different spins between each other or with other molecules is a quite difficult task. Nevertheless, it might seem that the highest possible (hypothetical upper limit) contribution of a spin to intermolecular interaction cannot exceed the values of energies of spin–spin and spin–orbit interactions in the same molecules. As a rule, this value does not exceed 10^{-24} erg [4], which is many orders lower than the energy of thermal motion kT ($\sim 4 \times 10^{-14}$ erg). Therefore, a spin-only interaction cannot essentially affect the intermolecular interactions. However, though it being paradoxical, the difference in the binding of spin isomers can be significant if, for no other reason, ortho and para water could be separated by gas chromatography at room temperature [3].

In order to estimate the real difference in their intermolecular interaction, spin isomers absorption on the preparation of interest is best suited. Until recently, studies of this kind have been restrained due to difficulties in

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controlling the spin state of water. The problem has been solved with the design of a unique spectrometer, which can operate over frequency ranges 6 to 40 cm^{-1} where rotational lines of water molecules are located. Taking into account that radiation transitions are allowed only between levels of the same spin-modification, different lines in the spectrum correspond to ortho and para isomers. The frequencies of such transitions are known [5]. The density of spin isomers in the cell can be determined by measuring radiation absorptions at corresponding frequencies. Thus, at present, monitoring the quality of water in ortho and para states is not very complicated at least in a gaseous phase [3].

This study is devoted to the investigation of the influence of the spin state of water on the rate of its absorption on lyophilized preparations of DNA, lysozyme and some inorganic sorbents. The aim of the study is to show the existence of selective binding of spin isomers of water by biological macromolecules and to assess how common is such selectivity relative to the sorbents used.

2. Experimental procedures

Egg white lysozyme and salmon DNA were used in experiments. Concentrations of DNA and lysozyme in water solutions were determined spectrophotometrically using extinction coefficients $E^{1\%}_{260\ \mathrm{nm}}=200$ [9] and $E^{1\%}_{278\ \mathrm{nm}}=26.7$ [10], respectively. Preparations were lyophilized for no less than 24 h. Bulbs with the lyophilized preparations were stored sealed and prior to an experiment were connected to a measuring cell. After that the bulbs was vacuumized and stored for no less than 2 h. The operating pressure in the system was ~0.5 Torr.

Quantity of other sorbents in the cell was determined by weight. Preliminarily all inorganic substances were ignited at 120 K under vacuum during 2 h.

The concentration of ortho and para water in the measuring cell was determined on the rotational absorption lines 0_{00}–1_{11}–37.1 cm^{-1} (para water) and 3_{03}–3_{12}–36.5 cm^{-1} (ortho water) [5]. The accuracy of measuring the absorption was 3 to 4%.

3. Results

The most direct and conceptually simple way to measure the binding rate constants of spin isomers with sorbent is a dynamic mode. If water vapor is supplied at a constant rate to a preliminarily pumped-out cell containing a sorbent, the density of every spin isomer will increase asymptotically reaching a value at which the rate of supply of this spin isomer will be equal to the rate of its binding to the sorbent. Due to a linear dependence of the rate on the density of the corresponding component (first-order reaction), the approach to the steady state behavior will be exponential. The argument of exponent is equal to the product of the rate constant and time; the reciprocal of the constant rate is characteristic time of absorption (see "Supplementary information"). Thus, the characteristic time for every isomer can be estimated from the curves of dependency of its density in the cell on time using the best-fit procedure. Water vapor can be supplied to the cell at a constant rate via a tiny gauging inlet connecting the measuring cell to the bulb with water. Any microscopic leakages in the vacuum cell (if exist) can change the rate of water vapor supply, but cannot influence the measured characteristic time.

It should be noted that the sensitivity of the spectrometer used is quite high. The density of ortho and para water in a dry glass cell with the total pressure 0.5 Torr is sufficient for a stable recording of spectra. Fig. 1B shows the dependence of density of two isomers in vapor versus time in the presence of 100 mg DNA. Water vapor was supplied at a constant rate via a calibrated inlet (Fig. 1A). Characteristic absorption times for ortho and para water, as well as their ratio, are also shown. As seen from the figure, the binding of the para water molecules with the DNA preparation proceeds much faster than that of ortho water. Similar experiments were performed in the presence of a different amount of DNA (Table 1). As should be expected, characteristic absorption time depends greatly on the sorbent amount used due to a change of the absorbing surface. However, the ratio of absorption times (the binding

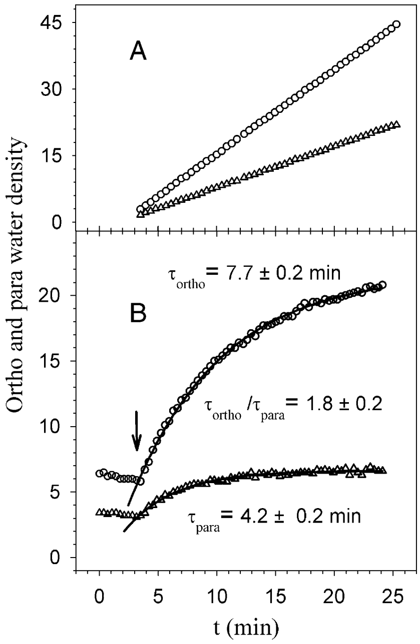

Fig. 1. Dependence of density of ortho (circle) and para (triangle) water in a gas phase on time (B) at a continuous supply of water vapor to a cell containing 100 mg of lyophilized DNA. The density is shown in conventional units. Water vapor was supplied at a constant rate. The top figure (A) shows the change of densities of ortho and para water in a gaseous phase in the cell without sorbent. Arrows indicate the beginning of water vapor supply to the cell. τ_{ortho} and τ_{para} are characteristic times ($\tau=1/k$, where k is the constant of absorption rate) of ortho and para isoforms absorption, respectively. Solid lines are the best fits of experimental curves.

Table 1
Results of experiments on binding spin isoforms of water molecules by different preparations

	Amount and conditions	τ_{ortho}, min	τ_{para}, min	τ_{ortho}/τ_{para}
DNA	14 mg	16.8 ± 0.8	9.2 ± 0.8	1.8 ± 0.27
	254 mg	4.0 ± 0.16	2.2 ± 0.2	1.8 ± 0.25
	100 mg	7.7 ± 0.2	4.2 ± 0.14	1.83 ± 0.1
Lysozyme	202 mg	4.8 ± 0.13	2.1 ± 0.13	2.3 ± 0.2
	100 mg	15.5 ± 0.5	8.0 ± 0.3	1.95 ± 0.14
Collagen	22 mg	26.9 ± 1.2	17.8 ± 2.3	1.5 ± 0.2
$MgSO_4$	3.1 g	8.3 ± 0.4	3.9 ± 0.3	2.1 ± 0.3
CaO	3.0 g	11.6 ± 0.7	6.3 ± 0.6	1.8 ± 0.2
Zeolite	Burnt, 270 °C	4.1 ± 0.2	2.2 ± 1.4	1.9 ± 0.25
	Damp	11.7 ± 0.7	12.3 ± 1.4	0.95 ± 0.16

τ_{ortho} and τ_{para} are characteristic times of absorption of ortho and para isoforms.

selectivity) remained almost constant through all experiments and was 1.8 ± 0.3 (Table 1). Thus, DNA shows preference for para water as judged by the absorption rate. A greater preference for para water was shown by lyophilized protein lysozyme as well (Table 1). In this case, the binding selectivity was 2.3 ± 0.2.

To clarify whether the absorption selectivity is a specific property of biological molecules or it is inherent to inorganic sorbents as well, similar experiments were also performed with properly dried zeolite, $MgSO_4$ and CaO. In all cases, selectivity varies from 1.5 to 2.3 (Table 1). Thus, the selectivity of binding ortho and para water molecules is not a specific property of DNA and protein and might be quite common. The binding of the para component by all sorbents studied occurs much faster than that of the ortho component.

It should be noted that the selectivity of binding the ortho and para components in all the preparations studied decreases with the growth of their humidity. For example, there is almost no selectivity in zeolite saturated with water vapor (0.95 ± 0.15). Similar results are observed for lyophilized DNA and protein preparations.

4. Discussion

The main result of our studies is a clear-cut distinction of the binding rate of water spin isomers. On the assumption that an energetic barrier determines the rates of absorption, the difference in the magnitude of such barriers for ortho and para water can be estimated. Since subject to the sorbent used the selectivity of lyophilized preparations varies from 1.5 to 2.3, the difference in the free energy of these barriers estimated as $kT\ln(\tau_{ortho}/\tau_{para})$ makes up from 0.4 to 0.8 kT (0.2–0.5 kcal/mol). This value exceeds the energy of spin–spin and spin–orbit interactions of many orders [4]. Obviously, such a great activation energy difference cannot be explained by spin-only interactions of water molecules with the environment. Therefore, there should be an indirect mechanism to provide this phenomenon.

One of the results of spin conversion of water molecules is a changed set of allowed rotational energy levels [6]. This is a consequence of the requirement of antisymmetry for the complete wave function of water molecule [7]. In particular, only a para water can reach the ground rotational level with a zero-point rotational energy (the molecule is not rotating). Since the field close to the sorbent surface is strongly anisotropic, the molecule in the ground state takes on certain advantage for absorption. The non-rotating para-molecule is oriented due to this field so that its energy of interaction with the surface would be maximal. As a result, such molecules will be most likely absorbed by the surface. All ortho-molecules have non-zero rotational energies. For them, the energy of interaction with the surface is averaged due to rotation and the attraction may be much lower [6]. This hypothesis proposed earlier by Tikhonov and Volkov [3] is in good agreement with the fact that selectivity of absorption of ortho and para water depends only a little on the nature of the sorbent used (see the Table). Indeed, we used sorbents with various chemical compositions and different mechanisms of binding water molecules. For example, $MgSO_4$ can form a crystalline hydrate whereas CaO can be involved in the reaction with the formation of $Ca(OH)_2$. Nevertheless, for all the preparations used, the differences in selectivity are quite insignificant (from 1.8 to 2.3). This suggests that the differences in the rate of binding ortho and para water depend first of all on the properties of the molecules per se, rather than on the properties of the sorbent used.

The hypothesis is quite suitable from the point of view of observed effects, but no doubt it should be verified experimentally. There is another hypothesis that tries to explain spin-dependent absorption of water molecules using the binary rotation theory [8]. In any case, remarkable difference in the binding rates of ortho and para water makes us to be prudent in estimating a possible difference in their equilibrium binding constants. It is not excluded that the differences in the binding energies can also reach a value of the order of kT. This may require revision of some physical and chemical phenomena from the point of view of actual two-component water. However, this guess-work should be treated with great prudence, because our data show that the selectivity of the binding of ortho and para water decreases with an increase in the sorbent humidity. It is probable that in real cellular environment, the effect of the binding selectivity might be also negligible.

5. Conclusions

Inasmuch as the interaction of the molecule spin with the medium is negligible, prominent differences in the sorption kinetics for water spin isomers may be determined by the difference in their quantum statistics. As it was mentioned above, allowed rotational state for spin isomers is different. It seems probable that differences in the statistics can also lead to appreciable differences in equilibrium thermodynamic parameters. Since interconversion between spin isomers is a quite slow process even in a liquid phase, water can be regarded as a mixture of two liquids with close but not the same properties. Both the physico-chemical properties of these substances as well as the kinetics and thermodynamics of chemical reactions in which water is involved can be different. Unfortunately, most physico-chemical and biophysical studies do not take this into account. For example, strictly speaking, all the data on NMR studies of water relate only to ortho water in a mixture of spin isomers. Para water has no intrinsic magnetic moment and is “silent” in NMR experiments. In studying the thermodynamics and kinetics of chemical reactions, the two-component composition of water is also neglected. However, the data obtained do suggest remarkable differences in physico-chemical properties of ortho and para water. This opens a wide field of activity in studying fundamental and applied problems relating to the role of the spin state of water molecules in physics, chemistry, biology and medicine.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bpc.2005.07.003.

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