MOLECULAR MECHANISMS OF WATER DIFFUSION IN COLLAGEN-LIKE STRUCTURES

L.A. BULAVIN, O.YU. AKTAN

Taras Shevchenko Kyiv National University
(2, Academician Glushkov Prosp., Kyiv 03680; e-mail: aktanl@univ.kiev.ua)

We experimentally investigate the process of swelling of collagen-like structures in water at various temperatures by the example of gelatin. The diffusion coefficients for water in gelatin are obtained. The existence of the characteristic temperature $T_0 = 291$ K separating the temperature intervals with different diffusion patterns is established. At $T < T_0$, water molecules move in disordered regions of collagen-like structures; at $T > T_0$, they penetrate into ordered regions, which is accompanied by the break of transverse bonds between peptide chains.

1. Introduction

Mechanisms of water diffusion in low-molecular structures were investigated in a number of works (see, e.g., [1, 2]). How will water diffuse in high-molecular systems where water molecules are surrounded by chain molecules? As far as the authors know, the mechanisms of such a motion have not been till now discussed in the literature. The given work is devoted to the search of the answers to this question.

2. Choice of the Investigation Object

In this work, the system under investigation is presented by gelatin as the example of a collagen-like structure.

As is known, gelatin is the product of thermal denaturation of collagen protein [3]. This process represents physical-chemical transformations of collagen in the case of its interaction with water, whose temperature is equal to the boiling one. In the course of denaturation, peptide chains separate from collagen triple spirals and their length decreases. Thermal denaturation results in the formation of a two-component system consisting of water and separate peptide chains in the form of statistical coils. The cooling of the water solution of gelatin leads to its renaturation, i.e. separate peptide chains start to create spiral collagen-like structures. The latter form a polymer gel with the help of transverse bonds [4]. A structure similar to that of gelatin gel can be obtained due to the swelling of solid gelatin.

We chose gelatin as the investigation object due to the following two reasons.

The first one consists in the fact that collagen structures represent a constituent of living organisms. That is why the results of the given investigation can be useful from the viewpoint of the problem of the interaction of medical products with biological tissue.

The second reason lies in the practical interest in the question concerning the influence of water on the physical-chemical properties of gelatin. This is related to the fact that the process of interaction of low-molecular solutions and the mentioned biopolymer is widely used in pharmaceutical, food, and other branches of industry [5–8].

The use of gelatin is caused by its ability to form gel under certain conditions [9]. Gelatin gels are widely used in food and medicopharmaceutical industries [10, 11]. They also serve as protecting coatings in the case of the capsulation of small doses of various substances [12]. For technical purposes, gelatin is most often used as a binding material for shaping, as well as a component of adhesive mixtures. It is also applied in metallurgy, printing and textile industries, and in the production of plaster figures. In addition, gelatin is used when fabricating money and securities and as a constituent of paints in the case of the publishing of decorated books, magazines, or reproductions and in the priming of canvases for painting [13].

3. Experimental Results

We investigated the process of swelling of gelatin in water at various temperatures. The swelling coefficient was determined experimentally using the weighting method [14]. The experimental dependences of the swelling coefficient N on time t are presented in Fig. 1.

As one can see from the figure, the temporal dependences of the swelling coefficient at large t tend to some equilibrium (asymptotic) values. Figure 2 shows the temperature dependences of the equilibrium values of the swelling coefficients.
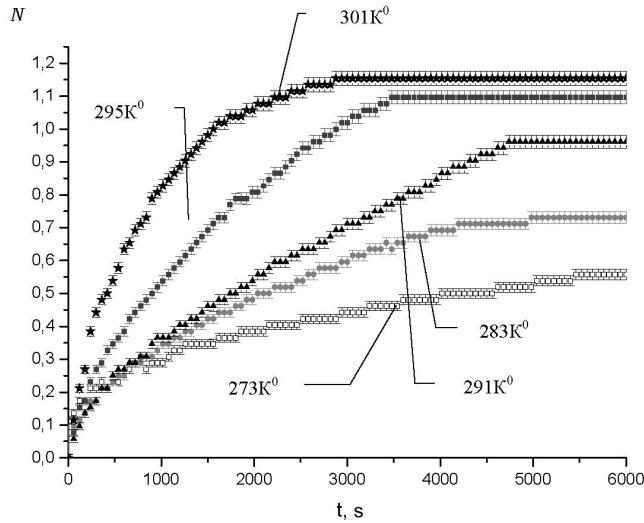
Fig. 1. Experimental temporal dependences of the swelling coefficient N

Fig. 2. Temperature dependences of the equilibrium swelling coefficients

The temporal dependences of the swelling coefficient were used for the determination of the diffusion coefficient of water in gelatin. In order to calculate the diffusion coefficient, we used the solution of the one-dimensional diffusion equation. When solving this equation, the water concentration at the boundaries of the sample was considered constant and equal to that corresponding to the equilibrium value of the swelling coefficient. The integration of this solution gave the total amount of water that diffused to the sample from outside. This integration results in the formula

$$N(t) = N_\infty \left(1 - \exp\left(-\frac{\pi^2 D}{\ell^2} t\right)\right), \tag{1}$$

where N_∞ denotes the asymptotic value of the swelling coefficient, D is the diffusion coefficient, and ℓ is the thickness of the sample.

This formula represents the theoretical temporal dependence of the swelling coefficient. Using it for processing the experimental data, we obtained the diffusion coefficients of water in gelatin at various temperatures. These values are presented in Table 1.

4. Discussion of the Experiment

As is known, the diffusion motion of particles represents a random walk. Such a motion consists in a sequence of elementary displacements of particles or a particle group. In the course of each of these elementary displacements, the system passes through the energy
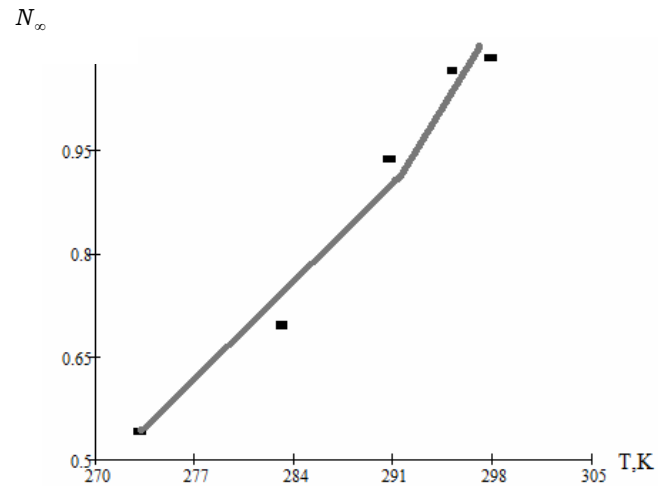
barrier

$$\Delta F = \Delta U - T\Delta S, \tag{2}$$

where ΔF, ΔU, and ΔS denote the free energy, activation energy, and entropy of activation, correspondingly, and T is the temperature.

The diffusion coefficient is determined by the formula

$$D = D_0 e^{\frac{\Delta S}{k}} e^{-\frac{\Delta U}{kT}}, \tag{3}$$

where k is the Boltzmann constant.

The coefficient D_0 is determined as

$$D_0 = \frac{\delta^2}{2\tau_0}, \tag{4}$$

where δ is the elementary displacement, and τ_0 is the time of the same order of magnitude as the period of thermal oscillations of a particle. Figure 3 shows the dependence $\ln D\left(\frac{1}{T}\right)$.

As one can see from the figure, this dependence represents a broken line consisting of two segments. The break of the line is observed at a temperature of 291 K. Each of these segments was used for the calculation of the activation energy and entropy presented in Table 2.

T a b l e 1. Diffusion coefficients of water in gelatin at various temperatures

Temperature T, K	Diffusion coefficient D, cm^2/s$\times10^{-6}$
274	0.86
283	1.31
291	1.31
295	1.61
301	2.44

Fig. 3. Dependence $\ln D \left(\frac{1}{T}\right)$

Fig. 4. Formation of supervacancies in collagen-like structures
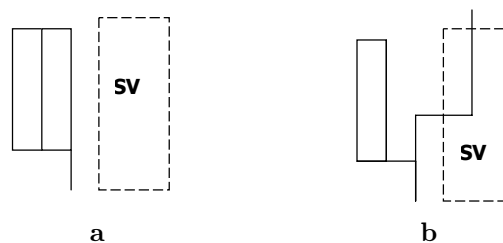
Fig. 5. Displacement of neighboring chains in the neighborhood of a supervacancy
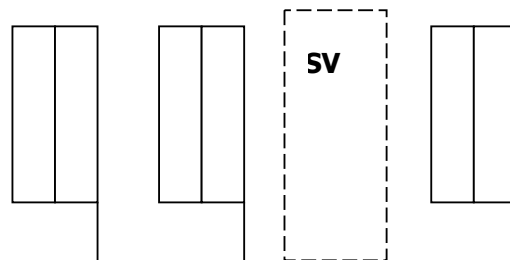
The quantity D_0 was calculated with the following parameters:

$$\delta = 10^{-8} \text{ cm}, \quad \tau_0 = 10^{-12} \text{ s.} \tag{5}$$

According to Fig. 3 and Table 2, the diffusion of water molecules in gelatin in the intervals below 291 K and above 291 K is of different nature. Establishing the diffusion mechanism in the temperature interval below 291 K, we based upon the fact that the diffusion coefficient for water in gelatin at 293 K practically coincides with the tabular value of the diffusion coefficient of water 10^{-5}cm^2/s [15]. This allows one to consider that this temperature interval is related to the water diffusion in disordered regions that are filled, first of all, with water in the course of swelling. Such an assumption is also confirmed by the value of the activation entropy which is of the order of k. As is known, such a value corresponds to the single diffusion, where an elementary displacement practically involves one particle.

As one can see from the table, the activation energy in the temperature interval above 291 K considerably increases and (what is more important) the activation entropy also rises by an order of magnitude. The latter point together with the increase of the activation energy testify to the fact that an elementary displacement

T a b l e 2. Diffusion parameters of water in the case of swelling of gelatin

Temperature, K	Above 291 K	Below 291 K
Activation energy, J	7.6×10^{-20}	1.2×10^{-20}
Activation entropy, J/K	22.8×10^{-23}	2.04×10^{-23}

involves a lot of particles – the diffusion becomes collective.

It is possible that, in this temperature interval, polymer chains start to participate in the diffusion, so that an elementary displacement involves a water molecule together with its polymer surrounding. Such a situation can arise in ordered regions of gelatin, where chains must "part," by letting a water molecule to pass. In this case, transverse bonds between the chains break. Thus, the temperature interval above 291 K corresponds to the motion of water molecules in ordered regions, which results in their disordering and the formation of a polymer gel.

To our mind, the specific mechanism of such an elementary displacement can be as follows. In [16–18], the authors introduced a concept of "supervacancy," being a structural defect. The specificity of supervacancies in collagen-like structures was considered in [19]. The appearance of supervacancies (SV) in such structures is depicted in Fig. 4, where the lines show peptide chains.

The origination of supervacancies allows the neighboring chains to move. In this case, an elementary displacement can be of the following character: a chain passes from position 1 (Fig. 5,*a*) to position 2 (Fig. 5,*b*) partially occupying the neighboring supervacancy.

Let a water molecule get to a region vacated by a chain, as it is shown in Fig. 6,*a*. In this case, in order
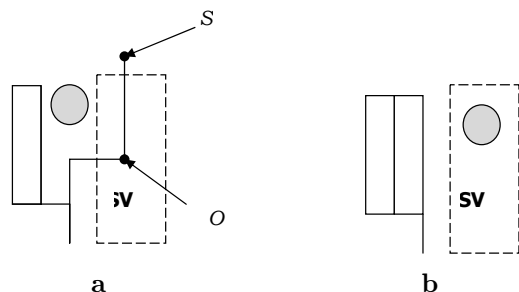
Fig. 6. Displacement of a water molecule and a polymer segment

that the molecule could pass to a supervacancy, the chain must get back from the position shown in Fig.6,*a* to its previous location simultaneously with the transition of water to the supervacancy (Fig. 6,*b*). Thus, the *OS* segment and the water molecule must simultaneously participate in the elementary displacement. Such a synchronous motion of the water molecule and the polymer segment with respect to each other substantially deforms the ordered region, which results in the increase of the activation entropy and the energy.

5. Conclusions

For collagen-like structures, there exists the characteristic temperature $T_0=291$ K that separates the temperature intervals with different diffusion patterns. At $T < T_0$, water molecules move in disordered regions of collagen-like structures; at $T > T_0$, they penetrate into ordered regions, which is accompanied by the break of transverse bonds between peptide chains.

1. L.A. Bulavin, T.V. Lokotosh, N.P. Malomuzh, and K.S. Shakun, Ukr. J. Phys. **49**, 556 (2004).
2. L.A. Bulavin, N.P. Malomuzh, and K.N. Pankratov, Zh. Struct. Khim. **47**, 54 (2006).
3. J. Koolman and K.H. Röhm, *Color Atlas of Biochemistry* (Thieme, New York, 2005).
4. M. Djabourov, J. Leblond, and P. Papon, J. de Phys. **49**, 333 (1988).
5. T.A. Dyakina, S.R. Derkach, and S.M. Levachev, Vestn. Mosk. Univ., Ser. 2, **45**, 58 (2004).
6. S. Iwamoto, M. Nakajma *et al.*, Coll. Surf. B **38**, 15 (2004).
7. K.A. Vaynberg, N.J. Wagner *et al.*, J. Coll. Interf. Sci. **205**, 131 (1998).
8. A. Seeboth, R.Wüstneck *et al.*, Coll. Polym. Sci. **268**, 286 (1990).
9. S.M. Pankov, *Gelatinous State of Polymers* (Khimiya, Moscow, 1974) (in Russian).
10. L.V. Safonova, Pishch. Tekhnol. **1**, 48 (1982).
11. E. Esposito, R. Cortesi *et al.*, Biomater. **20**, 9 (1995).
12. G. Digenis, T. Gold, and V. Shah, J. Pharm Sci. **83**, 15 (1994).
13. P.V. Kozlov and G.I. Burdygina, Polymer **24**, 651 (1983).
14. A. Bigi, S. Panzavolta, and K. Rubini, Biomater. **25**, 5675 (2004).
15. I.S. Grigor'ev and E.Z. Melikhov, *Physical Quantities* (Energoatomizdat, Moscow, 1991) (in Russian).
16. L.A. Bulavin, O.Yu. Aktan, and Yu.F. Zabashta, Vysokomol. Soed. A **44**, 1536 (2002).
17. L.A. Bulavin, O.Yu. Aktan, and Yu.F. Zabashta, Vysokomol. Soed. A **45**, 1684 (2003).
18. L.A. Bulavin, O.Yu. Aktan, and Yu.F. Zabashta, Vysokomol. Soed. B **47**, 690 (2005).
19. L.A. Bulavin and O. Yu. Aktan, Adv. Mol. Med. **1**, 103 (2005).

МОЛЕКУЛЯРНІ МЕХАНІЗМИ ДИФУЗІЇ ВОДИ В КОЛАГЕНОПОДІБНИХ СТРУКТУРАХ

Л.А. Булавін, О.Ю. Актан

Р е з ю м е

На прикладі желатини експериментально досліджено процес набрякання колагеноподібних структур у воді при різних температурах. Отримано значення коефіцієнтів дифузії води в желатині. Встановлено існування характерної температури $T_0 = 291$ K, яка розділяє температурні інтервали з різним характером дифузії. При $T < T_0$ молекули води рухаються в невпорядкованих областях колагеноподібних структур, при $T > T_0$ молекули води проникають у впорядковані області, що супроводжується розривом поперечних в'язей між пептидними ланцюгами.