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Abstract

Full Text

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ON TWO MODIFICATIONS OF GELATIN

(Presented by Academician P. A. Rehbinder, 10 IX 1963)

As early as 1915, Smith ([1]) proposed considering gelatin as a protein having two modifications: α and β.

It was thereby assumed that α-gelatin (otherwise the sol form) is stable at high temperature, whereas β-gelatin (or the gel form) is stable at low temperature. The transition α β-gelatin is fully reversible and occurs when the temperature changes between 35 and 15°.

The existence of two modifications of gelatin is indicated by the fact that gelatin at temperatures above 35° has a specific optical rotation approximately 2-2.5 times lower than at temperatures below 15°.

Further confirmations were subsequently obtained for the existence of two modifications of gelatin with characteristic properties and structure. Thus, for example, it was established that the α β transition can be carried out at constant temperature by changing the pH of the medium. The accompanying changes in a number of physical and colloidal properties of gelatin (optical rotation, swelling, viscosity, isoelectric point) were explained by keto-enol rearrangement ([2],[3]). It was further shown that the α β transition is accompanied by a characteristic change in the X-ray patterns of gelatin; the α form gives an amorphous picture, whereas the β form gives the X-ray pattern of ordinary gelatin with crystalline interferences ([4]). The difference in the structure of α- and β-gelatin is also indicated by a study of their infrared spectra: in the α form the absorption band of NH vibrations corresponds to 3310 cm^{-1}, and in the case of the β form to 3330 cm^{-1} ([5]).

Fig. 1. Kinetics of swelling of gelatin films: α-gelatin (1), β-gelatin (2)

It is extremely important, as was noted already in early works, that gelatin in the α form (i.e., at $t > 35^{\circ}$) reacts quite differently to changes in pH and to the presence of electrolytes than gelatin in the β form (i.e., at $t < 15^{\circ}$). Suffice it to say, for example, that α-gelatin exhibits a minimum of specific optical rotation at the isoelectric point, which is weakly expressed, whereas β-gelatin at the isoelectric point shows a sharp maximum ([6]).

The experimental data presented for the most part leave no doubt; as for their unambiguous interpretation, until recently it was in fact impossible. This was explained by the lack of the necessary information on the molecular structure of gelatin. At present, represen–

it becomes possible to bring substantial clarity to this question thanks to major modern achievements in the study of the structure of the protein molecule and its fragments–polypeptide chains. An attempt at such a generalization was made by us ([7]) and led to the conclusion that the molecule of α-gelatin has the conformation of a randomly coiled coil (Random Coil), while the molecule of β-gelatin is a rod-like helix (Helix) of the proline type. New proof of the existence of these two conformations was provided by experiments on fixing gelatin with chromium salts in the coil conformation and separately in the helix conformation ([8]).

Table 1

Object	Film area, cm^2	Film area, cm^2
	α-gelatin	β-gelatin
Initial gelatin	2.0	2.0
After swelling	7.6	2.5
After drying	6.1	2.0

In this work new facts are presented that characterize the structure and properties of gelatin in the α- and β-conformations. To obtain α- and β-gelatin, a solution of standard photographic gelatin, heated to 40–45°, was poured onto two organic-glass plates in a definite amount per unit area of the plate. After this, one of the plates was placed in a thermostat at 50°, and the other in a desiccator with calcium chloride, kept in a refrigerator at +5°. To accelerate drying at 5°, an air stirrer driven from outside was placed in the desiccator. At the end of drying, films of gelatin were obtained in the α- and β-conformations, respectively; these then served as the object of further investigation. First, the kinetic curves of swelling of the films were recorded; swelling was determined by the gravimetric method at room temperature. The results are given in Fig. 1 and clearly show the difference in the behavior of α- and β-gelatin. The helical conformation (β-gelatin) of the polypeptide chains forms a more compact structure, which slows the swelling process; however, this process does not stop and reaches limiting values only after 20 h. Gelatin in the coil conformation (α-gelatin) has a looser structure, which is quite understandable, and shows rapid absorption of water at the beginning and attainment of the limit already after 80 min, with approximately twofold lower water uptake than in β-gelatin. Measurements of the density of α- and β-gelatin are in full agreement with these data. It was found that α-gelatin has a density $d_{20^\circ} = 1.340$, while β-gelatin has $d_{20^\circ} = 1.394$. The following observation was made during the above-described experiments on measuring the swelling kinetics. It turned out that α- and β-gelatin have completely different swelling "geometry."

Observing films of α- and β-gelatin, we found that the film dried at 5° increases almost not at all in area but becomes thicker, whereas the film obtained at 45° visibly increases in length and width and at the same time becomes thinner. After 12 h of swelling, the films were dried at room temperature, and it turned out that the α-gelatin film had changed its area relatively little after drying, whereas the β-gelatin film regained its former dimensions. This is illustrated by Table 1.

It was also noted, so far by qualitative signs, that films of α- and β-gelatin have different mechanical properties. Thus, the α-gelatin film had noticeably greater brittleness. X-ray photographs of α- and β-gelatin were also taken; they proved to be different, as had already been known earlier ([4]).

The facts presented here once again confirm the existence of two modifications of gelatin and the sharp difference in their physicochemical properties. Taking into account previous works ([7], [8]), we can represent with sufficient reliability the molecular structure of α- and β-gelatin and control it, using optical rotation, density measurement, and X-ray diffraction as methods for monitoring the conformation of the molecules.

Of the numerous consequences arising from all that has been said, only one practical conclusion will be given. It consists in the fact that in all work involving gelatin it is necessary to take into account that its solutions and gels at room temperature (18–20°) are a mixture of the α- and β-modifications. It is therefore clear that the study of any property of gelatin will be indeterminate and difficult to explain. Everything is greatly simplified if one works either at 5–10° or at 40–45°.

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