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SHORT COMMUNICATION

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Studies of phase transformations in the gelatine-water system using near-IR spectroscopy

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Abstract Near-IR spectroscopy studies of gel formation mechanism are reported for gelatin-water systems. Relative absorbance in 1.4-μm and 1.9-μm bands was shown to be sensitive to the helix to coil phase transition in the pre-melting region. The formation mechanism of a metastable "coil" phase is in agreement with DSC and TGA data obtained in parallel experiments on the 10% gelatin gel.

Key words Gelatin gels · Phase transformations · Near-IR spectroscopy

Introduction

As gelatin gels are widely used for industrial and scientific purposes, considerable interest has been evoked in studies of structure and phase transitions in these systems. The process of gel formation is generally considered to be governed by a competition between intra- and intermolecular interactions [1, 2], resulting in the formation of polymer networks with inclusions of water [3, 4].

Studies of gel formation in water-gelatin systems have been reported in a number of papers, and the results obtained using various methods (e.g., X-ray diffraction [5], calorimetry [6–8], measurements of optical rotation [9], and viscosity [10, 11]), have shown that the gelatinization process proceeds in two steps with the coil to helix transition passing through the formation of an intermediate metastable phase.

It is widely accepted that the gelatin macromolecule in the unmelted state exists in the form of an expanded chain (helix); upon heating to pre-melting temperatures, it rolls itself into a "permeable", and then into an "impermeable" coil [12–14]. The process is reversible, and these stages are observed in the inverse order upon cooling [15].

Phase transitions in gelatin gels, as in other biocolloidal systems, are generally accompanied by the formation or splitting of hydrogen bonds and the resulting changes in the associated states of water [8]. It would be natural to expect these changes to be reflected in the IR bands related to vibrations of the hydroxyl groups.

However, there is practically no information on IR spectroscopy studies of such systems. One of possible reasons could be very high absorption intensity with samples of experimentally controllable thickness – in the conventional IR region (4000–400 cm^{-1}) the corresponding peaks are merged together, yielding just one broad band of low informativity.

The situation is quite different for bands in the near-IR region (0.7–2.5 μm) that correspond to combined frequencies of deformational and antisymmetric valence vibrations of water molecules. These bands have been repeatedly used for studies of systems and an organic substance can form a common network of hydrogen bonds [16–19]. In particular, clear changes were reported in [20] in the relative optical density of 1.4-μm and 1.9-μm bands due to phase transformations of hydrated phospholipids.

In the present work, we endeavored to use near-IR spectroscopy as a tool for investigation of the gel formation mechanism in water-gelatin mixtures.

Experimental

Spectra of 10% water-gelatin gels were recorded using spectrophotometers SPECORD–751-R (conventional IR region) and

HITACHI 330 (near-IR region). In the latter case, the measurements were carried out in a thermostabilized cell at 30–50 μm layer thickness. To interpret the temperature dependences of the spectra obtained, we used, as in [20], the values of relative absorbance $\Delta A = A_m - A_0$, where A_m is the measured absorbance (optical density) at the peak maximum, and A_0 is "background" absorbance in the region adjacent to the peak Absorbance $A = \lg (I_0/I)$, where I_0 and I are intensities of falling and transmitted light, accordingly. In parallel series of experiments, the same water-gelatin gel samples were studied by differential scanning calorimetry (DSC) and thermogravimetric analysis (TGA) using a METTLER TA 3000 thermoanalytical system.

Results and discussion

Figure 1 shows near-IR spectra of a freshly prepared 10% gelatin gel sample at different temperatures. The corresponding ΔA vs temperature plots are shown in Fig. 2. Clear changes in ΔA are observed within the temperature ranges 25–27 °C and 32–34 °C. In the second case, the sharp fall of ΔA is obviously due to the gel melting. As for the first range, the observed increase in ΔA can be attributed to the formation of a new metastable phase of low transparency. Measurements for a similar gelatin gel sample that had been kept for 5 days showed that it became less opaque (ΔA two times lower); the character of temperature-induced transitions has, however, remained unchanged.

Absorption intensity in the conventional IR region was, as expected, so large that no conclusions could be made concerning effects of temperature upon the spectrum. The sequence of transitions suggested by near-IR spectroscopy has been confirmed by thermal analysis (Fig. 3). TGA shows no sharp changes of mass at the transition points. This implies that possible chemical transformations yielding volatile products do not contribute significantly to the processes under study. As for DSC thermograms, two steps are clearly observed when the sample is heated in an unsealed crucible. The first step is exothermic (metastable state), and the second step is endothermic (gel melting). It should be noted that when heating is carried out in a hermetically sealed crucible, the metastable state is not observed, presum-

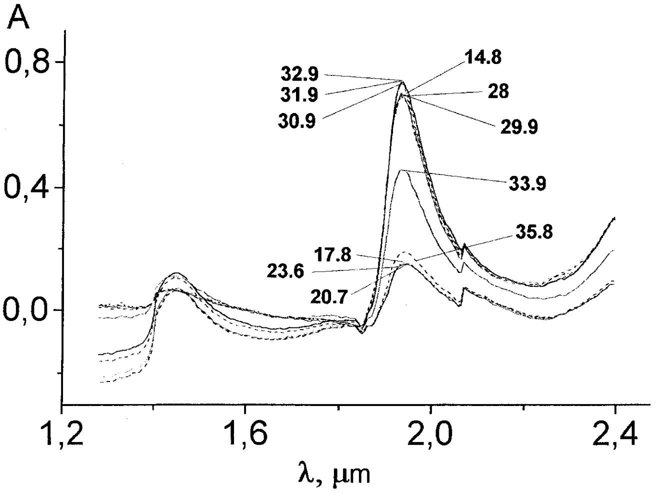

Fig. 1 Near-IR spectra of 10% gelatin gel (thickness of thermostabilized layer is 30 μm) at different temperatures in the range 15–36 °C

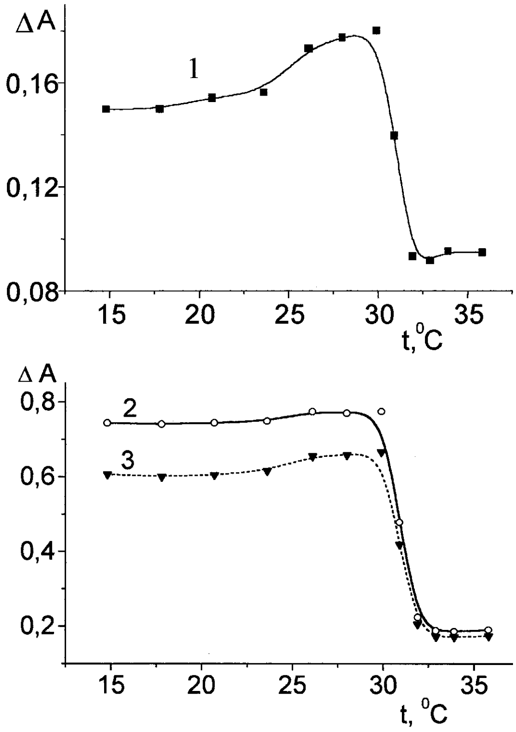

Fig. 2 Relative absorbance ΔA for the 10% gelatin gel of Fig. 1 as function of temperature Curve 1: $\Delta A = A_m(1.44\ \mu m) - A_0(1.80\ \mu m)$; Curve 2: $\Delta A = A_m(1.88\ \mu m) - A_0(1.80\ \mu m)$; Curve 3: $\Delta A = A_m(1.88\ m) - A_0(2.22\ \mu m)$

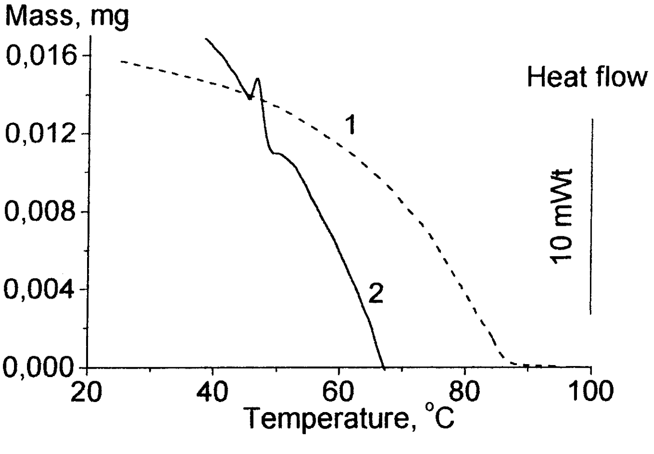

Fig. 3 TGA (1) and DSC (2) thermograms for 10% gelatin gel

ably due to a pressure increase in the closed system hindering such phase transition.

Obviously, transformations "impermeable coil – permeable coil – helix" are accompanied by restructuring of hydrogen bonds inside the macromolecules or between them. This restructuring is reflected by changes in the near-IR spectrum of the water-gelatin gel. In the temperature range where the metastable "coil" state is presumably formed, the relative absorbance is somewhat higher than in the unmelted state and significantly higher than after the melting has been completed. This fully agrees with a natural assumption that structural units of the macromolecule are only poorly ordered in the "coil" state.

It seemed interesting to compare our results with NMR data [15]. The estimated temperatures of transitions "impermeable coil – permeable coil" (~308 K) and "permeable coil – helix" (~301 K) are in good agreement with our results. However, an essential peculiar feature of our approach should be noted. The parameters measured in NMR experiments [15] included chemical shift, half-width, and intensity of the 0.93 m.p. line with respect to the reference tetramethylsilane. This line was assigned to overlapping signals of Val, Leu, Ile methyl groups. In our studies, the measured parameter was relative intensity of 1.4 m and 1.9 m bands related to vibrations of OH groups in different associated states of water. Good agreement of the two sets of data shows that spectrophotometry in the near-IR region, as an analytical criterion of the phase state, is no less adequate than the high resolution NMR spectra. In the latter case [15] the temperature range of the "intermediate" phase corresponding to the permeable coil is detected as the peak width on the differential curve (i.e., as the region of the steep rise of the 0.93 m.p. line intensity). In our experiments (relative intensity of the near-IR bands + DSC thermograms), we could clearly define both upper and lower limits of the "intermediate" phase – each of these limits was reflected both in the corresponding intensity jumps and calorimetric peaks. One can draw a certain analogy with mesomorphic phase transitions in thermotropic liquid crystalline mixtures [21], where DSC gives wide (smeared) transition peaks, while polarization microscopy shows clear temperature limits of the biphasic region.

Conclusions

The optical absorption spectra in the near IR-range, in parallel with DSC thermograms for the same systems, have been studied for water-gelatin mixtures. Comparison of our results with available high resolution NMR data shows that studies of temperature-dependent near IR-spectra can be considered as a sufficiently simple, convenient, and informative method for investigation of gel formation processes in water-gelatin mixtures.

References

1. Szucs M (1989) In: Photogr Gelatin, 5th IAG Conference, part 2, p 317
2. Hermel H, Wappler HJ, Wuestneck R, Wetzel R, Buder E, Huettner A, Herbrich H (1989) J Int Rec Mater17:207
3. Carvalho W, Djabourov M (1997) Rheol Acta 36:591
4. Veis A (1964) The macromolecular chemistry of gelatin. Academic Press, New York London
5. Hirojumi S, Nobuaki N (1989) Kyochu Daigaku Chuo Bunseki Senta Hokoku 7:29
6. Tseretely GI, Smyrnova OI (1991) Vysokomol Soedin Ser A 33: 2243; Chem Abs (1992) 113:236,197e
7. Bohidar Himadri B, Jena Sidartha SJ (1993) Chem Phys 98:8970
8. Kristiansen J, Hvidt A (1990) Acta Chem Scand 44:144
9. Djabourov M, Leblond J (1987). In: ACS Symp Paris, Ser 350, p 211
10. Levi SM (1960) Kolloidn Zh 22:599
11. Izmailova VN, Rebinder PA (1974) Structure formation in protein systems. Nauka, Moscow (in Russian); Chem Abs (1975) 82:B73,471g
12. Zimm B, Bragg J (1959) J Chem Phys 31:526
13. Tsvetkov VN, Eskin VE, Frenkel SY (1964) Structure of macromolecules in solutions. Nauka, Moscow (in Russian)
14. Doi M, Edwards SF (1986) Theory of polymer dynamics. Academic Press, New York
15. Krasovskii AN, Nikolaev BP, Shlyakov AM, Mnatsakanov SS (1995) Vysokomol Soedin 37: 273; Chem Abs (1995) 123:147,219v
16. Lindberg JJ, Haila T (1980) Acta Chem Scand B 34:757
17. Salder A, Horsch I, Lawson E (1984) Anal Biochem 138:44
18. Davies A, Miller C (1988) Appl Spectrosc 42:703
19. Hollenberg J, Iffr J (1982) J Phys Chem 86:1938
20. Panikarskaya VD, Lisetski LN (1998) Functional Mater 5:610
21. Van Hecke GR (1985) J Phys Chem 89:2058