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GEL FORMATION OF CURDLAN-TYPE POLYSACCHARIDE IN DMSO-H_2O MIXED SOLVENTS

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Curdlan-type polysaccharide was found to form a thermoreversible gel in DMSO with the addition of water. The sol-gel transition was also reversible with respect to mole fraction of DMSO. Drastic changes in light scattering intensity, viscosity and optical rotation of the polysaccharide in DMSO with the addition of water were observed around the sol-gel transition point. The gelation mechanism of the polysaccharide in DMSO-H_2O mixed solvents is discussed.

Some mutants of Alcaligenes faecalis var. myxogenes 10C3K were found to produce a gel-forming β-1,3-glucan which is called "curdlan-type polysaccharide".[1,2) This gel-forming polysaccharide,which is soluble in an alkali solution but not in a neutral or acid solution, is swollen and forms an elastic gel from a heterogeneous state when its aqueous suspension (at $pH<12$) is heated above approximately 55°C. The gelation mechanism has not been clarified yet, because of difficulty in conformational studies by spectroscopic methods.

The authors tried to make a gel of curdlan-type polysaccharide from a homogeneous solution, and found that the polysaccharide could form a fairly transparent and elastic gel from a homogeneous solution in a certain dimethyl sulfoxide (DMSO) content of DMSO-H_2O mixed solvent. The gelation in the mixed solvent is convenient for surveying the mechanism by the physicochemical methods. The properties of the gel with DMSO-H_2O mixed solvents and some physicochemical changes accompanied with the gelation are described in this paper.

Curdlan-type polysaccharide used in this investigation is polysaccharide 13140 which is produced by a strain (NTK-U, IFO 13140) of Alcaligenes faecalis var. myxogenes 10C3K. Polysaccharide 13140 was supplied from Takeda Chem. Ind.,Ltd. The degree of polymerization of the polysaccharide is 467.

Shimazu Model PG-21 Light Scattering Photometer, JASCO Model J-5 Spectropolarimeter and Carl Zeiss Model A-1 Polarimeter were used at 25°C for determining the light scattering intensity, ORD and specific rotation at 578 nm respectively. The intrinsic viscosity was measured with an Ostwald viscometer at 25°C. The gel strength was determined with an Okada-type Gel Strength Tester, and was indicated in g/cm^2 unit.

Polysaccharide 13140 dissolves in DMSO without accompanying any gelation.

On addition of water, the solubility of this polysaccharide decreases with an increase in the water content, and becomes insoluble in below approximately 0.6 mole fraction of DMSO. In such a DMSO-H_2O mixed solvent, the polysaccharide was found to show a gelation from a homogeneous solution.

In the DMSO mole fraction range from 0.06 to 0.63, the polysaccharide suspended in the mixed solvent was found to be solubilized by heating and to form a gel when the homogenized solution was cooled. The sol-gel transition temperature is shown in Fig.1. The sol-gel transition is thermo-reversible in this DMSO concentration range, while the gelation of aqueous suspension (at $pH<12$) of the polysaccharide is thermo-irreversible. In the mixed solvent containing 0.62 mole fraction of DMSO, for instance, the polysaccharide-DMSO-H_2O system is in a sol state above 55°C and in a gel state below 55°C as is shown in Fig.1. The gel strength was varied markedly with the DMSO concentration. The effect of DMSO on the gel strength is presented in Fig.2. The polysaccharide concentration is fixed at 13.5 mg/ml. The gel strength of the gel which is formed in the DMSO mole fraction range from 0.06 to 0.63 increases with an increase in the DMSO concentration, reaching the maximum at approximately 0.35 DMSO mole fraction and decreasing up to 0.63.

On the other hand, in below 0.06 DMSO mole fraction the polysaccharide suspended in the DMSO-H_2O mixed solvent was swollen but not solubilized by heating, and formed a gel during heating. This gelation is quite agreed with that of the polysaccharide suspended in water (at $pH<12$). The gelation is thermo-irreversible. Marked decrease in gel strength was observed with an increase in the DMSO concentration as was shown in Fig.2.

According to these properties, the polysaccharide 13140 - DMSO - H_2O system can be classified into the following three groups:

(1) Thermo-irreversible gel being formed in the range from 0 to approximately 0.06 mole fraction of DMSO,

(2) Thermo-reversible gel in the range from approximately 0.06 to 0.63 mole fraction of DMSO, and

(3) Homogeneous solution in the range from approximately 0.63 to 1.0 mole fraction of DMSO.

The polysaccharide-DMSO-H_2O system changes its state from a homogeneous solution (sol) to a thermo-reversible gel with the addition of water. This sol-gel transition was reversible with respect to the DMSO concentration.

As DMSO is a good solvent for the polysaccharide 13140, DMSO may be tightly solvated to the polysaccharide, which prevents the intermolecular interaction of the polysaccharide chains. With the addition of water, however, the polysaccharide-DMSO interaction should be weakened in such a way that the polysaccharide chains may readily interact to form the three-dimensional network which is required in gelation.

The ORD curve of the polysaccharide in DMSO was negatively normal, whereas in the presence of approximately 0.4 mole fraction of water, rotation angle in the curve became positive. The specific optical rotation ($[\alpha]^{25}_{578}$) with mole fraction of DMSO is shown in Fig.3. Drastic change in $[\alpha]^{25}_{578}$ was observed around 0.63 mole fraction of DMSO, i.e. the gelation point. These results indicate that a conformational change was accompanied with the gelation. As was pointed by Ogawa and his co-worker,$^{3)}$

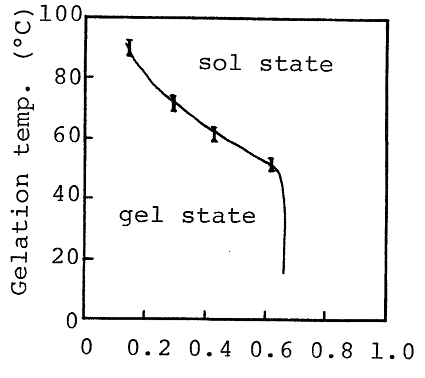

Fig.1 Gelation temperature of the polysaccharide-DMSO-H_2O system. The polysaccharide content is fixed at 5 mg/ml.

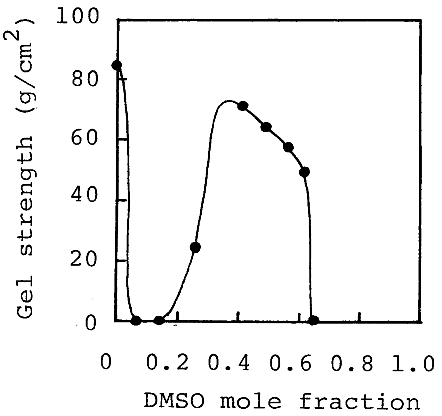

Fig.2 Gel strength of the polysaccharide-DMSO-H_2O system. Gel strength was determined with 13.5 mg/ml polysaccharide gels at 25°C.

the polysaccharide 13140 takes a disordered form in DMSO. On the other hand, model building in the computer showed that the β-1,3-glucan chain could form triple-helices.4) Therefore, with the addition of water,the polysaccharide may change its conformation from a disordered form to an ordered one (partly helical form) in DMSO-H_2O mixed solvents.

DMSO concentration dependence of reduced light scattering intensity at 45° (R_{45}) and intrinsic viscosity ([η]) are shown in Figs.4 and 5 respectively. These physicochemical properties changed sharply around 0.63 mole fraction of DMSO. Ogawa and his co-workers surveyed the NaOH dependence of the viscosity, optical rotation, and birefringence of the polysaccharide, and the changes in these physicochemical properties were observed around 0.24N NaOH. These changes were attributed to a conformational transition. The anomalous changes in R_{45} and [η] in the polysaccharide 13140-DMSO-H_2O system, which are accompanied with the gelation, resemble the changes reported by Ogawa.5) The marked increase in R_{45} with addition of water may be caused by the aggregation of the polymer at the gelation. Gradual increase in [η] with decreasing DMSO up to approximately 0.6 DMSO mole fraction also indicates that the aggregation of the polymer should occur. The abrupt decrease in [η], however, at the gelation point cannot be explained by the aggregation of the polymer. An aggregation which is accompanied with a conformational change may occur at the gelation point.

Such drastic changes in the physicochemical properties of polymers have not been reported yet for the gelation of polysaccharides. The present results (Figs.3-5) clearly showed that the aggregation of the polysaccharide occured in the gelation process. For the gelation, this aggregation should allow the polysaccharide chains to be cross-linked in a three-dimensional network. The detail discussion of the gelation mechanism is difficult because of insufficient data in spectroscopic results. The following description, however, may be approved.

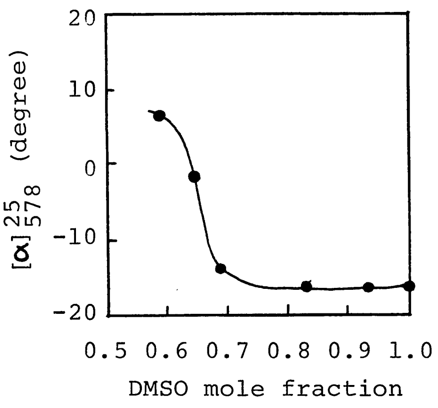

Fig.3 The DMSO concentration dependence of the specific rotation angle at 25°C ($[\alpha]^{25}_{578}$).

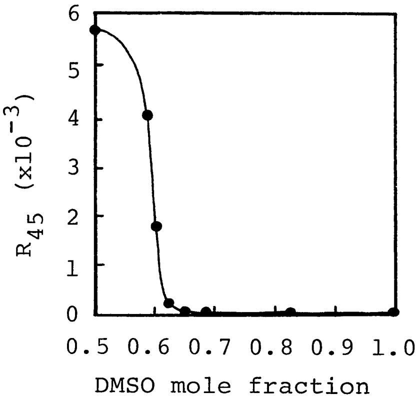

Fig.4 The DMSO concentration dependence of light scattering intensity (R_{45}) at 25°C.

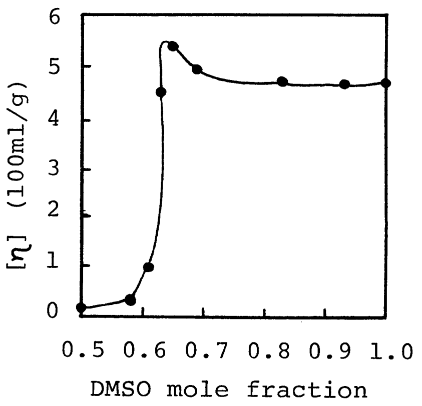

Fig.5 The DMSO concentration dependence of the intrinsic viscosity ($[\eta]$) at 25°C.

As was suggested by the results in ORD and specific optical rotation at 578 nm, a helical part of the polysaccharide may be formed in a gel state. These helical parts of the polysaccharide chain may be cross-linked intermolecularly for the network formation and may be responsible for the gel formation.

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