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Note

Observations on the crystallization and melting of maltopentaose hydrate

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Abstract

The crystallization of maltopentaose from concentrated aqueous mixtures was studied by differential scanning calorimetry, X-ray diffraction and polarised light microscopy. Under the conditions of study it was observed that maltopentaose crystallized as a hydrate, with single crystals assembling into spherulitic structures. The crystalline hydrate melted at 78 °C with an enthalpy of fusion of 53 Jg^{-1}. © 1997 Elsevier Science Ltd. All rights reserved.

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The crystallization behaviour of starch polysaccharides, and their oligosaccharides, is important to industrial usage and relevant to the biological assembly of the starch granule. Crystalline polymorphism of the starch polysaccharides has been examined by X-ray fibre diffraction studies [1] and more recently by electron diffraction on single crystals obtained from amylose and linear dextrins [2,3]. On crystallization from water, the amylose chain can adopt left-handed parallel stranded double helical conformations, which pack into monoclinic and hexagonal arrays for the A and B forms, respectively [3,4]. Single helical conformations can also be obtained on crystallization from aqueous solutions containing iodine or small hydrophobic solutes such as short chain fatty acids or alcohols. The minimum chain length required for the formation of the A and B crystalline forms is greater than 9 units [5,6]. Although the different polymorphic forms of D-glucose and maltose [7,8] are well known [9], there is less information on the crystallization behaviour of maltooligomers ranging in degree of polymerization from 3–9 units. These oligomers readily form stable glasses on drying concentrated aqueous solutions at room temperature [9,10]. Methyl α-maltotrioside crystallizes as a tetrahydrate in which the chain conformation adopted is similar to that found in the A and B forms of amylose [11]. Similarly *p*-nitrophenyl α-maltohexaoside was crystallized as a hydrated molecular complex with $Ba(I_3)_2$, with the carbohydrate chain forming a double helical arrangement [12]. Recently, in a study on the water sorption behaviour of amorphous maltooligomers, ranging in DP from 3 to 7, we observed that maltopentaose, as a result of its crystallization, displayed unusual behaviour. This note is a report of that study.

The water sorption behaviour of maltooligomers can give information on the strength of the interaction between the carbohydrate and water [13]. The

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Fig. 1. Light micrograph of maltopentaose hydrate spherulites (~ 10–50 μm in size) when viewed between crossed polars.

generally observed behaviour for amorphous carbohydrates is that the amount of water sorbed increases sharply as the relative humidity, RH, approaches 100%. A preparation of maltopentaose behaved differently with a plateau in the amount of water sorbed (~ 12% (w/w) at an RH > 70%). This indicated either a crystallization during the sorption process with the formation of a crystalline hydrate, or the sorption of water into a crystalline lattice — disrupted as a result of drying — to reform the crystalline hydrate. The latter interpretation was favoured as another preparation of maltopentaose behaved in a comparable way to other amorphous maltooligomers.

In an attempt to crystallize maltopentaose from aqueous solution, conditions of crystallization were chosen similar to those used for the preparation of α-D-glucose [14]. Crystallization from the 67% (w/w) solution took several days at 2 °C. Initial observations showed the presence of needle-like crystals which then aggregated into spherulites (Fig. 1). It is interesting to note that linear fragments of starch can form spherulitic structures which in some respects are reminiscent of the structure of the native granule.

Examination of the melting of the crystalline material showed a limited premelting starting at ~ 40 °C which was followed by a sharp endothermic transition with a peak maximum, T_f, of 77–79 °C. This compares with a T_f of 158 °C for D-glucose and 123 °C for maltose monohydrate (polymorph not specified) [15]. The enthalpy of fusion, ΔH_f, for the transition was 53 Jg^{-1} compared with a reported 179 Jg^{-1} for D-glucose, 126 Jg^{-1} for maltose monohydrate [15], and 42 Jg^{-1} for a highly crystalline spherulitic form of amylose of the B polymorph of starch [16]. It might be expected that the T_f of crystalline carbohydrates would generally increase with increasing molecular mass. However, the tendency of higher oligomers to form hydrates, and to form intramolecular hydrogen bonds can modify this expectation [9]. After melting, the maltopentaose/water mixture was quenched to −20 °C and on rescanning in the calorimeter a sharp increase in heat capacity, indicative of a glass transition, with a midpoint of 21 °C was observed. As the glass transition temperature, T_g, of dry maltopentaose is 168 °C, the observed depression in T_g of 147 °C indicates a water content of the crystalline hydrate of 14% (w/w) [10]. For comparison, the calculated water contents of hepta- and octa-hydrates would be 13.2 and 14.8%, respectively. A characteristic of oligosaccharide hydrates [9] is that the water forms a network structure with the sites of occupancy having different affinities for water molecules, with the extent of occupancy depending on ambient conditions of water vapour pressure and temperature.

The wide angle X-ray powder diffraction pattern of the crystalline hydrate of maltopentaose is shown in Fig. 2 (background subtracted) where X-ray inten-

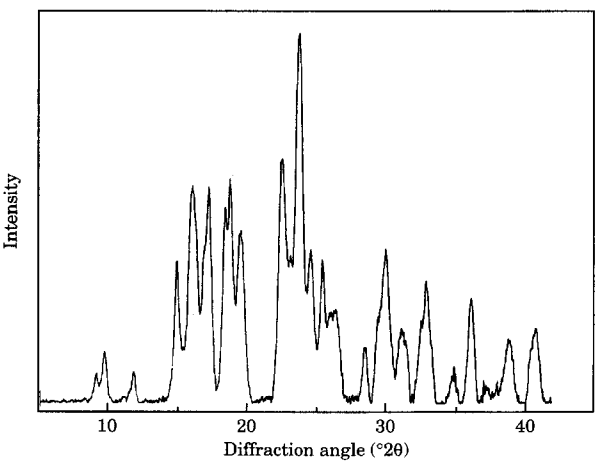

Fig. 2. X-ray diffractogram of crystalline maltopentaose hydrate.

Table 1
Crystallographic parameters from X-ray powder diffraction of maltopentaose hydrate

Angle (° 2θ)	d-spacing (nm)	Intensity [a]
9.13	0.968	w⁻
9.78	0.903	w
11.86	0.746	w
14.89	0.595	m
15.87	0.558	s⁻
17.19	0.516	s⁻
18.31	0.484	s⁻
18.73	0.473	s⁻
19.60	0.453	m
22.45	0.396	s⁻
23.71	0.375	s
24.58	0.362	m
25.39	0.350	m
26.39	0.337	m⁻
28.53	0.312	w⁺
29.37	0.304	w⁺
29.99	0.298	m
31.06	0.288	w
31.57	0.283	w⁻
32.88	0.272	m
34.84	0.257	w
36.12	0.248	m⁻
37.15	0.242	w⁻
38.91	0.231	w⁺
40.38	0.233	w⁻
40.77	0.221	w⁺

[a] s = strong; m = medium; w = weak.

sity is plotted as a function of diffraction angle 2θ. Although some of the reflections match those of the A and B polymorphic forms of starch, overall the pattern is quite different. The diffraction pattern is sharp, well-resolved and extends to wide angle. This indicates that the material obtained is highly crystalline. The crystallographic parameters obtained from analysis of the X-ray pattern are given in Table 1. In conclusion, analysis of the crystalline structure of disaccharides has often served as a model for the interpretation of the fibre diffraction data of related polysaccharides. In particular, information is gained on the pattern of hydrogen bonding which is not available from the fibre diffraction studies [9]. In this note we have demonstrated that maltopentaose forms a crystalline hydrate thus extending the limited range of oligosaccharides which are known to crystallize.

1. Experimental

Maltopentaose was obtained from Sigma. On storage in a moist atmosphere (70% RH) at 25 °C for 10 days the dried product had sorbed 12 ± 1.0% (w/w) water to form a crystalline solid as assessed by X-ray diffraction. Maltopentaose could also be crystallized from a concentrated aqueous solution, 67% (w/w), at 2 °C. Glacial acetic acid was layered on the surface of the undercooled liquid with crystallization being initiated at this interface and then propagating into the bulk. The purity of the crystalline products was checked by ion-exchange chromatography on a CarboPac PA-100 column equipped with a Dionex pulsed amperometric detector [13]. No evidence of maltopentaose hydrolysis under the crystallization conditions used was obtained.

Calorimetry.—Differential scanning calorimetry over the temperature range −20 to 120 °C was carried out using a Perkin–Elmer DSC 7 as described [13]. The instrument was calibrated from the melting of indium and the melting temperature, T_m, and the enthalpy of fusion, ΔH_f, obtained using the standard software.

X-ray diffraction.—X-ray diffraction measurements were carried out using CuKα_1 radiation of wavelength 0.154 nm. The diffractometer was a Philips Scientific PW 1820 vertical goniometer with an Anton Paar TTK camera. Data were collected and stored on a personal computer using Philips PC-APD (Version 3.6b) automated powder diffraction software. The sample was scanned over the range 5.0–42.0° 2θ, at a speed of 0.01° 2θ per second, with a step size of 0.02°. Identification of peak positions was accomplished using the Peak Search facility of PC-APD. This automatically locates peaks in a crystalline diffraction pattern by detecting minima in the second derivative of the diffractogram. The standard peak search settings were used, except that the minimum peak width was changed from 0.0 to 0.2° 2θ, causing a slight smoothing of data, but reducing the detection of 'phantom peaks' arising from instrumental noise.

Acknowledgements

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