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Solution properties of water-insoluble rye-bran arabinoxylan

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An arabinoxylan, extracted from rye bran, was measured by viscosity and light-scattering techniques in different solvents as a function of time over a period of more than three years The freshly isolated polysaccharide is soluble in DMSO as a double stranded complex. In the course of time, clusters of about seven strands are formed as an intermediate structure which further aggregates unspecifically to very large clusters, which after about three years comprise more than 800 chains These clusters can be partly broken up with the well-known complexing solvents for cellulose: cuoxam, cadoxen and the iron tartrate alkali complex FeTNa. A substructure of clusters with seven aggregated strands is obtained. DMSO is not capable of disrupting the large clusters and it is a poorer solvent than DMSO containing 10% water. Although used to extract the xylan, 1 N NaOH solution is not a good solvent and clusters of about 30 chains were found. The molecular weight of the individual single-stranded xylan was found from the carbanilate derivative which gave a DP_W of 423.

INTRODUCTION

In recent years heteroxylans from various cereals have aroused keen interest because of their importance as dietary fibre components (van Soest & McQueen, 1973) as well as the undeniable role they play in dough development and their beneficial effects on loaf properties (McCleary, 1986; Meuser & Suckow, 1986). While most of the studies have focused on the primary structure, there have been a few attempts to establish relationships between molecular structure and the physico-chemical properties of these polysaccharides. Systematic studies in this direction have been started on hemicelluloses from wheat and rye flours (D'Appolonia & MacArthur, 1975; Lineback *et al.*, 1977; Clacco & D'Apolonia, 1982; Izydorczyk *et al.*, 1991, 1992; Theander & Aman, 1979), but none of those from bran fractions.

The preparation of true solutions of heteroxylans is often a difficult task because of the poor solubility of most heteroxylans in aqueous and aprotic solvent systems, even when originally isolated by water extraction. This makes molecular weight determination and characterization of solution properties difficult. Earlier works on birchwood glucuronoxylan solutions in DMSO (Timell, 1964) revealed marked discrepancies in light-scattering (LS) data, and extremely high molar masses in comparison to data obtained by sedimentation methods. No further attempts have been made since then to improve the LS measurements using modern techniques.

In previous papers on rye-bran hemicellulose, the isolation (Hromadkova & Ebringerova, 1987), general

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chemical structure (Hrodmakova *et al.*, 1987; Ebringerova *et al.*, 1989) and some rheological properties (Hrodmakova & Ebringerova, 1992) of the water-insoluble heteroxylan have been reported. In this paper the solution properties of this polysaccharide in aqueous and aprotic media were measured by static and dynamic light scattering and viscometry and are discussed at some length.

EXPERIMENTAL

Materials

The arabinoxylan (AX-I) was isolated by extraction of the chlorite holocellulose that was prepared for delipidated, de-starched and de-pectinated rye bran with 4–5% NaOH and precipitated from the alkaline extract by acidification to pH 5 (Hromadkova & Ebringerova, 1987). The polysaccharide was reprecipitated from alkaline solution by acidification, followed by dialysis and lyophilization. The product was insoluble in cold and hot water. Some general characteristics are given in Table 1.

In spite of the low glucuronic acid content, the sample was transformed into the free acid form by treating the material with 0·1 M HCl in 90% ethanol (Kohn *et al.*, 1986).

Light scattering

The samples were dissolved in the various solvent systems as described in the Section Solvents at concentrations of at most 5 mg/ml. A dilution series of four lower concentrations was made, and the solutions were then filled through millipore filters of 0·45 μm pore size directly into the cylindrical light-scattering cells. This technique was applicable to aqueous media and DMSO solutions but not to the complexing solvents cuoxam, cadoxen and FeTNa. In these cases filtration was not possible, and the solutions of various concentrations were optically clarified by ultracentrifugation for half an hour at 12 000 rpm. For this purpose the scattering cells containing the solutions were suspended in an aqueous CsCl solution and centrifuged (floating technique (Dandliker & Kraut, 1956)).

Table 1. General characteristics of AX-I

Sugar compositions (mol. %)	
Xylose	85·6
Arabinose	12·0
Glucose	1·7
Ara:Xyl, molar ratio	1:7·14
Uronic acid (%)	0·7
$[\alpha]_D$, in 2% NaOH	−107°
$[\eta]$ in DMSO (dl/g)	2·65
DP_η (from $[\eta]$ in DMSO; LeBel *et al.*, 1963*a*)	633
M_η	93 700
$[\eta]$ in cadoxen (dl/g)	2·37
DP_η (from $[\eta]$ in cadoxen; Wikström, 1968)	741
M_η	106 700
M_n (osm. in DMSO)	26 650

Static light-scattering measurements were performed with SOFICA photogoniometers which were equipped with a HeNe (5 mW) laser or an Ar ion laser (10 mW). The measurements performed in Teltow were made manually, and the results had to be evaluated separately by special software. Later both laboratories, in Teltow and in Freiburg, used computer-driven SOFICA instruments which had thoroughly redesigned electronics (Baur, 1990), and to which an on-line evaluation software was attached for the measured data, applying either the Zimm-plot, modified Guinier-plot or Berry-plot versions.

Measurements were made at 20°C in the angular region between 25° and 150° and in steps of 5° (in some cases at steps of 2·5° up to 45°). The cuoxam solutions developed a deep blue colour, and hence an absorption correction had to be applied. This was achieved by dividing the scattering intensities by the corresponding absorptions. The cadoxen solutions are colourless and offered no problems; the lightly green-coloured FeTNa solutions afforded correction only when the blue Ar line was used.

In some cases dynamic light-scattering measurements were also carried out. Here the ALV photogoniometer (ALV-Langen, Hessen, Germany) was used, equipped with an ALV 3000 autocorrelator/structurator. Only the last measurements have been made with the new ALV 5000 correlator, which allowed far more accurate measurements than possible with the ALV 3000 correlator. Measurements were made partly with an instrument that was equipped with a Kr ion laser of the 2000 series supplied by Spectra Physics and partly with one that had an Ar ion laser as the light source, and with which several lines could be selected. These two ALV goniometers were designed for simultaneous static and dynamic LS and allowed a cross-check of the separately performed static LS measurements. Because of the time needed for the recording of one correlogramme, the dynamic LS measurements were made only at steps of 10° covering the range from 30 to 150°. Details of the instrumental set-up are given in the paper by Bantle *et al.* (1982).

Refractive index increments

The refractive index increments were taken from the literature (Gralén, 1944; Valtasaari, 1957; Henley, 1969; Huglin, 1972), where we assumed the same values as were reported for cellulose. The data are collected in Table 2.

Solvents

The preparation of the complexing solvents is crucial and is described in detail as follows.

Table 2. Refractive indices n_0 and refractive index increment dn/dc of xylans in various solvents. The required values of (dn/dc) at the HeNe line of 632·8 nm were not measured directly but taken as those at 546 nm

Solvent	n_0	[dn/dc]		Reference
		$\lambda_0 = 546$ nm	$\lambda_0 = 436$ nm	
DMSO	1·479	0·066	0·067	Huglin (1972)
DMSO/H_2O (90/10)	1·482	0·074		Huglin (1972)
1M NaOH	1·340	0·142	0·142	Huglin (1972)
Cuoxam	1·347		0·233	Gralén (1944)
Cadoxen/NaOH	1·361		0·184[a]	Henley (1969)
FeTNa	1·384		0·244	Valtasaari (1957

[a]Interpolated from measurements at 546 mm and 436 mm for $\lambda_0 = 457·9$ nm (Kr-ion laser).

Cadoxen (according to Henley, 1969)
155·7 ml ethylenediamine, p.a.; 360 ml H_2O; bidestille; 50 g CdO, p.a.: From freshly distilled ethylenediamine (en) and water at 28%, ethylenediamine solution was prepared under cooling in an ice-water/NaCl mixture ($T < 0$ C). Finely ground CdO was slowly added within 24 h under stirring at that temperature. The slightly turbid solution was then centrifuged for half an hour at 20 000 rpm in a Beckman ultracentrifuge to remove $Cd(OH)_2$ traces that were formed as colloidal particles. The Cd content was determined complexometrically (Jander, 1986) to amount to 7·8% Cd. By adding NaOH-en-H_2O (28% en content) the solution was brought to a composition of 5% (w) Cd, 28% (w) ethylenediamine and 0·35 M NaOH. After dissolution of the xylan the solvent was diluted with water 1:1.

A slight variation was used by the Teltow group who used a 24% (w) ethylenediamine composition.

Cuoxam
5 g $Cu(OH)_2$, tech. grade (Aldrich), 500 ml NH_3 solution, 25%, p.a.: The solution was prepared at 5 C in deeply brown-dyed vessels, which were flushed with argon before preparation. The solution was kept at 5 C until use. Because of the high oxygen and light sensitivity it is essential to work under an argon atmosphere and to exclude light.

FeTNa (according to Valtasaari, 1957)
121·206 g $Fe(NO_3)_3 \cdot 9H_2O$; 207·000 g $Na_2(C_4H_4O_6) \cdot 2H_2O$; 96·0 g NaOH: In a 1·5 litre beaker the tartrate is first dissolved by heating in 400 ml of water. The dissolution of the nitrate in water is preferably done just before mixing to avoid hydrolytic decomposition. The mixing of the solutions is carried out under vigorous stirring with light excluded. The stirring is continued until all the tartrato-ferric acid possibly precipitated has been re-dissolved. The beaker is then placed in an ice-water bath, and when the solution has attained a temperature of 10–15 C the addition of alkali is begun. The sodium hydroxide is dissolved in water to 200 ml. During the exothermic complex-forming reaction the addition must be slow enough to avoid raising the temperature by more than 15–20 C. About 75 ml of the alkali solution is consumed in this reaction and its completion is indicated by a shift of the reddish-brown colour of the solution to yellowish green; the rest of the alkali is added rapidly, disregarding any possible slight rise in temperature. The solution is finally transferred into a one-litre flask (containing 5 g of sodium tartrate prescribed by Jayme & Bergmann (1956) for stabilization) and adjusted with water to the correct volume. The addition of water causes hydrolytic decomposition, which is why the volume adjustment should be done as quickly as possible. The solution is now allowed to stand for at least 24 h, after which it is filtered through a No. 4 glass filter. The solution can be stored without any precautions against daylight. Sometimes during long storage a small precipitate of ferric hydroxide forms and should be removed by filtration. In every case, this filtered solution was again centrifuged at 12 000 rpm for half an hour.

RESULTS

The results of the light-scattering measurements are collected in Table 3, together with 'particle' weight, obtained from the intrinsic viscosity ([η]) using the relationships of LeBel *et al.* (1963*a*) and Wikström *et al.* (1968) in DMSO and cadoxen, respectively. Also included are results reported for the carbanilate derivative (Mertz, 1991) measured in dioxane by light scattering. There is a wide spread in data for the particle weight that results partly from the solvent and the pretreatment of the solution but to a large extent also depends upon the time that has elapsed from the date of extraction.

The lowest particle weights of comparable size in both solvents, DMSO and cadoxen, were obtained from the intrinsic viscosity measured shortly after the material was extracted, purified and precipitated.

Table 3. Particle weight M_w, aggregation number x, radius of gyration R_g, hydrodynamic radius R_h, second virial coefficient A_2 and the structure-sensitive parameter $\rho = R_g/R_h$ for AX-I in various solvents

Solvent	$M_w \times 10^{-3}$	Aggregation number x*	R_g (nm)	$A_2 \times 10^5$ (Mol cm^3/g^2)	R_h (nm)	$\rho = R_g/R_h$
DMSO ($M\eta$)[a]	94	1·5				
DMSO[b]	643	10·3	164	14·1		
DMSO[c]	1290	20·6	237	30·4		
DMSO 48 h[c]	199	3·2	194	34·2		
DMSO[d]	481	7·7	70·1	−4·9	42·1	1·67
DMSO (heated)[d]	343	5·5	49·7	16·5	31·8	1·56
DMSO/H_2O (90/10)[d]	162	2·6	40·9	75·4		
DMSO/H_2O (90/10)[e]	52 200	834	268	0·05		
1M NaOH[d]	1850	29·6	173	13·4	122	1·41
Cadoxen (M_η)[a]	110	1·8				
Cadoxen[e]	17 200	275·0	209	−0·36		
Cadoxen/NaOH (1:2)[e]	13 000	208·0	307	0·88		
Cadoxen ½h[e]	530	8·5	154	−15·2		
Cadoxen 2h[e]	363	5·8	143	15·8		
FeTNa[e]	482	7·7	185	−81·0		
Cuoxam[e]	485	7·7	120	0·68		
Carbanilate[f]	62·6					

[a] 1989 [η] measured in Bratislava; M_η calculated according to LeBel (1963*a*,*b*).
[b] 1989 LS measured in Teltow.
[c] 1990 LS measured in Teltow.
[d] 1991 LS measured in Freiburg.
[e] 1993 LS measured in Freiburg.
[f] 1991 LS from carbanilate derivative in dioxan. M_w given in this table refers to the degree of polymerization × 148 (= M_0, the mass of the repeating unit).
* $x = M_w/M_{w\,carb}$, where $M_{w\,carb} = 62{\cdot}6 \times 10^3$ is the molar mass of AX-I obtained from the carabanilate derivative.
The notation added to the name of solvent is explained as follows:
(90/10): DMSO/H_2O (v/v).
48 h: time of dissolution under rigorous shaking.
heated: the solution was heated up to 80°C and kept there for 1 h. After cooling to 20°C a series of four more dilute solutions were prepared and filtered.
½h or 2 h: solutions were not filtered but centrifuged in a Beckman Ultracentrifuge for half an hour or two hours at 15 000 rpm, respectively.

DMSO was most efficient as a solvent if 10% water was added, and the solution was heated at 80°C for one hour prior to measurement. In this case, almost the same particle weight was obtained as deduced from viscosity measurements. (The difference may arise from the different averages, since $M_\eta < M_W$ for broad molecular weight distributions.) DMSO containing 10% water, was not an adequate solvent for samples aged for 2 years (see Table 3). Similar observations were made by LeBel *et al.* (1963*b*) several years ago.

Most interesting is the solution behaviour of this arabinoxylan in complexing solvents which have been long recognized as solvents for cellulose. Cuoxam was shown (Seger, 1993; Seger & Burchard, 1994) to appear as a derivatizing solvent for cellulose and to dissolve this $\beta(1-4)$ glucan completely down to a molecular level. Cadoxen and FeTNa seem to have a similar solvation power but do not appear to derivatize the OH-groups on C_2 and C_3. Cadoxen was used previously in SEC experiments with xylans (Eriksson *et al.*, 1968; Bobleter & Schwald, 1985), and the Mark–Houwink relationship was established for birchwood glucuronoxylans (Wikström, 1968) as $[\eta] = 9{\cdot}2{\cdot}10^{-3}DP_w^{0.84}$ (cm^3/g). SEC on 'Separon Hema S' columns (Eremeeva & Ebringerova, 1991) of AX-I solutions in cadoxen/water = 1/1 yielded an apparently very narrow distribution curve. Such apparent narrow distributions may be the result of aggregation where more dense structures are obtained with a small range of hydrodynamic volumes, which, however, would correspond to a wide molar mass distribution. The present LS-measurements, in fact, indicate aggregation, and the time correlation function of the dynamic LS revealed a rather broad size distribution (see Fig. 4). Typical LS measurements are presented in Figs 1 to 3.

When applied to the AX-I xylan all these complexing solvents cause similar behaviour which also agrees with the LS-measurement in DMSO made half a year after isolation of the sample (bold solvents in Table 3). It is instructive to compare also the mean radii of the particles and the ratios of the radius of gyration to the hydrodynamic radius. Figure 5 shows a plot of R_g as a

function of M_w on a double logarithmic scale. One notices a far more expanded structure in the complexing solvents than for the materials of low molar particle weight in DMSO.

The time effect and influence of the pretreatment of solutions become particularly noticeable with the DMSO solutions. Between the measurements of (a) to (b) and (b) to (c) (see Table 3) there were time shifts of about 3 months. One notices that in the same laboratory (Teltow) the particle mass increased by a factor of 2 during that time. A similar tendency was observed in the Freiburg laboratory, with a DMSO/water ratio of 90/10, where between the two measurements a time of $2\frac{1}{2}$ years had elapsed.

The high M_w values clearly indicate strong aggregation, which could be only partly broken up by either a longer exposure of the solution to shear forces (shaking) or by heating the solution to 80°C. (A higher temperature could not be applied because of solvent decomposition that starts at about 90°C.) The effect of heating cannot directly be compared with that from the exerted shear force because of the time shift of about half a year. The extremely large particle weight as found in the Teltow laboratory with cadoxen is probably caused by a slightly lower ethylenediamine content (24%) than was used in the Freiburg laboratory (28%).

As may be noticed from the Tables, the measurements of the xylans, particularly in the metal complexing solvents, involved comparatively high experimental errors. No systematic error determinations were made in this study, but the accuracy can be estimated in these cases to amount to the following: $M_w \pm 15$–20%; $R_g \pm 10$–12%; $R_h \pm 5$–8% and $A_2 \pm 20$–30%. The cadoxen, cuoxam and FeTNa solutions had to be centrifuged, mainly because traces of colloidal metalhydroxides had to be removed. When centrifuging the solutions in beakers no detectable amount was seen at the bottom. Thus, when the solutions were centrifuged in the scattering cells applying the floating technique (Dandliker & Kraut, 1956) the maximum amount removed is estimated to be no more than 3%.

DISCUSSION

The large number of measurements with the same material and the large variations in the values for M_w and R_g raises the question as to what would be the molar mass of an isolated, non-aggregated structure. Therefore, the sample was transformed in to the carbanilate derivative and measured in dioxane (Mertz, 1991). From this, a degree of polymerization was derived which corresponded to a molar mass of the non-substituted chain of $M_w = 62{\cdot}6{\cdot}10^3$. This allows an estimation of the number x of aggregated strands which are given in column 3 of Table 3. Most frequent (6 from a total of 16) is a complex of about 7 strands followed by a dimerization of chains (4 from 16). Evidently these are the most stable structures. The 7-chain complex is obtained in particular with the complexing solvents. Dimerization was found only with freshly isolated xylans.

Figure 5 exhibits rather strange and unexpected

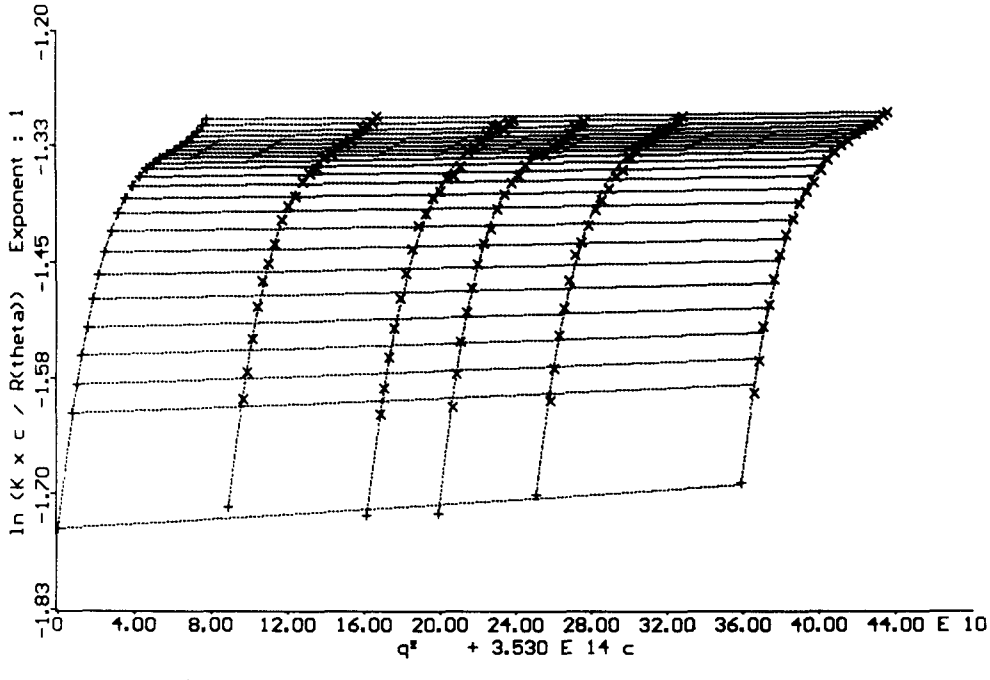

Fig. 1. Plot of $\ln(Kc/R_0)$ against $q^2 + kc$ (Guinier-plot) for AX-I in DMSO/water (9/1). Measurements were made three years after isolation: $q = (4\pi n_0/\lambda_0)\sin(\theta/2)$ with n_0 the solvent refractive index, λ_0 the wavelength of the laser beam and θ the scattering angle.

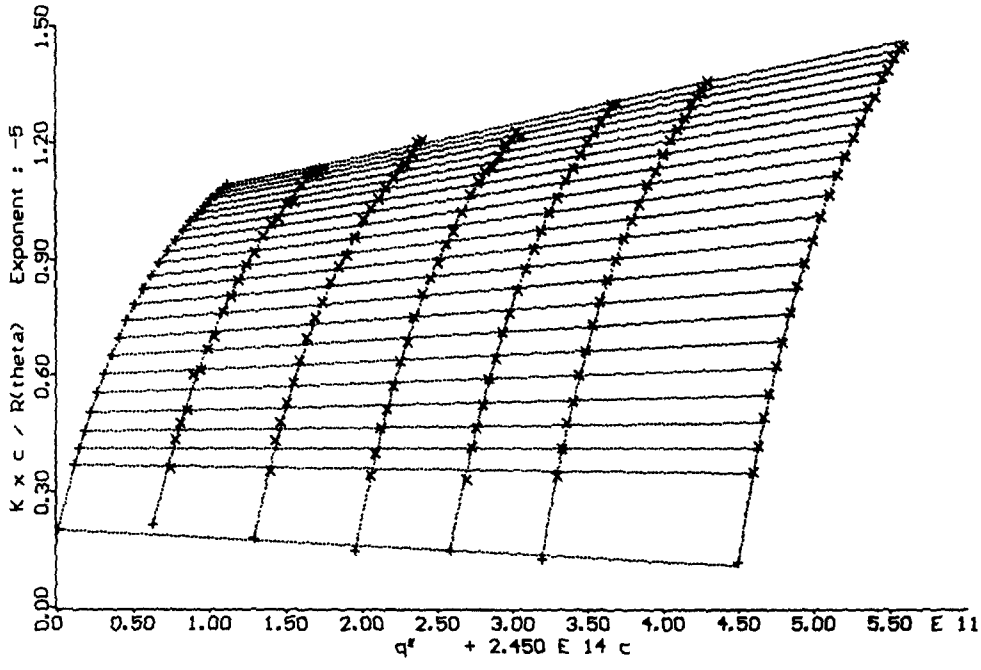

Fig. 2. Plot of KC/R_0 against $q^2 + kc$ (Zimm-plot) for AX-I in cadoxen. Measurements were made three years after isolation.

behaviour. The effects of ageing and of the different solvents could have been expected to have different influences on the structure of the aggregates. In contrast, all the points in Fig. 5 appear to form one common curve if three points (in brackets) are disregarded. One of these three points corresponds to a cadoxen solution that was centrifuged for 2 h. Here a certain amount may have been removed by sedimentation such that the actual concentration in the cell was reduced by about 10%. Evidently, this sedimentation did not cause a marked fractionation since the radius of gyration decreased only slightly. A second off-line point corresponded to a high molar particle weight found in cadoxen with 24% ethylene diamine content, probably as a result of the poorer solvation power compared with 28% ethylene diamine content. A much stronger aggregation combined with a denser packing of the segments is the result. Addition of NaOH causes the structure to swell and partly to disintegrate. 1 M NaOH solution is apparently not a good solvent, and strong aggregation combined with a densely packed structure is obtained. This may be the reason for the third off-line point and

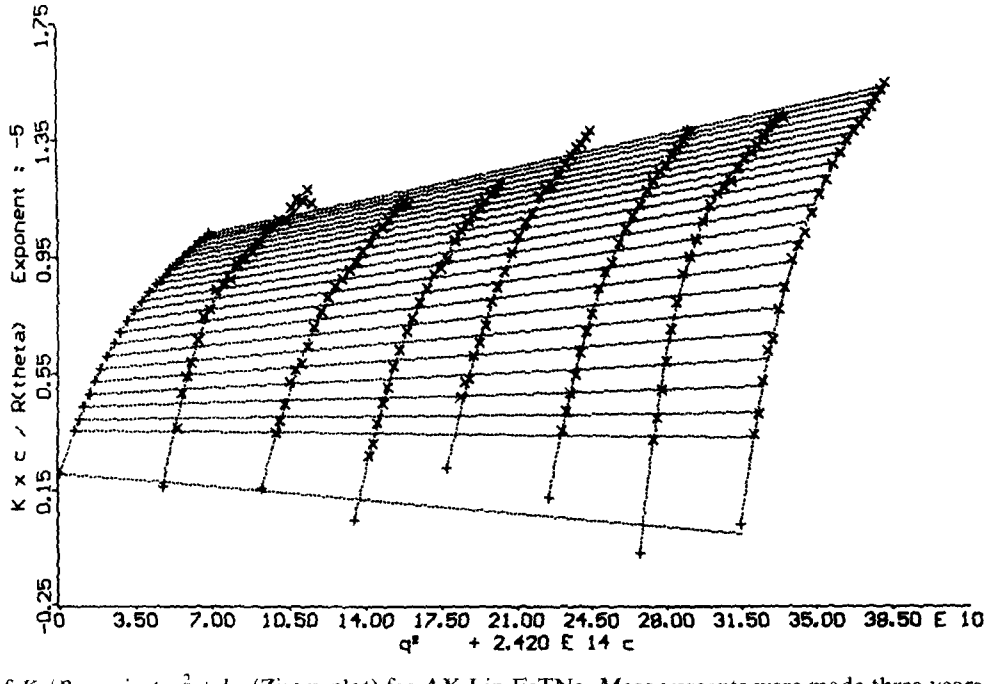

Fig. 3. Plot of Kc/R_0 against $q^2 + kc$ (Zimm-plot) for AX-I in FeTNa. Measurements were made three years after isolation.

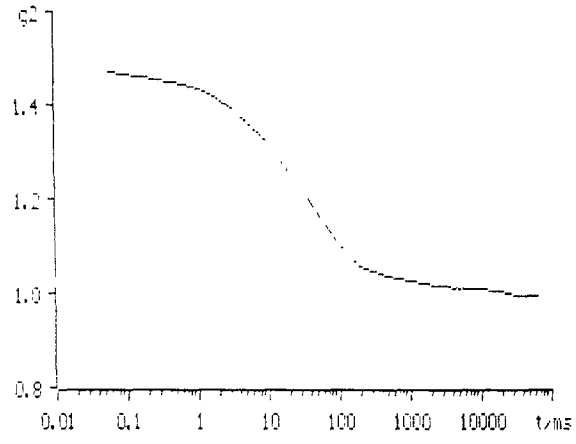

Fig. 4. Time correlation function $g_2(t) = <i(0)\,i(t)> / <i(0)>^2$ for AX-I in cadoxen at a scattering angle of 30°. The relaxation distribution is very broad and spans over two decades in time.

this observation may give a hint as to the origin of the strong tendency to aggregate (see below).

In a few cases the hydrodynamic radius R_h was measured by dynamic light scattering, using the Stokes–Einstein relationship $D_z = kT/(6\pi\eta_0 R_h)$, where D_z is the translational diffusion coefficient (z-average). The ratio $\rho = R_g/R_h$ of the radius of the gyration to the hydrodynamic radius is a sensitive measure of the structure (Burchard *et al.*, 1980). A decrease is predicted from values of $\rho = 1{\cdot}8$–$2{\cdot}0$ for polydisperse ($M_w/M_n = 2$) flexible linear chains when branched or densely packed clusters are present. On the other hand, an increase in ρ is predicted and found with linear chains of increasing chain stiffness. Table 3 shows in fact lower values for the aggregate comprising about six chains, but this value increased for a structure with more than six aggregated chains. This observation suggests that a

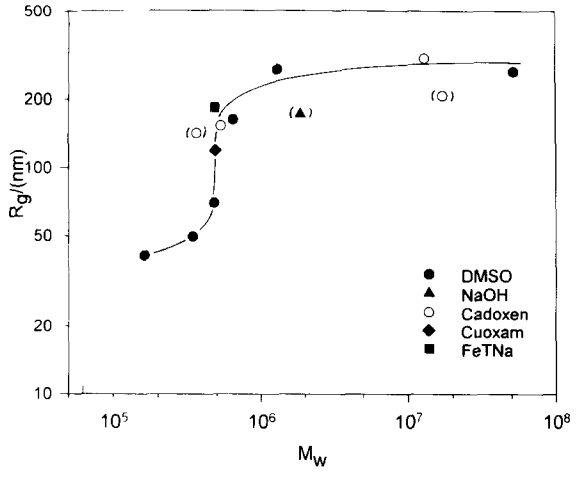

Fig. 5. Radius of gyration as a function of particle weight for AX-I in several solvents. The single, non-aggregated chain has $M_w = 62{\cdot}6 \times 10^3$ (derived from the percarbanilated sample measured in dioxane (Mertz, 1991); its position is marked by a vertical line on the abscissa.

stiffening of originally fairly flexible xylan chains occurred, probably caused by a lateral (side-by-side) aggregation.

This conjecture is supported by the unusual molar mass dependence of the particles radii of gyration. Figure 5 reveals a strong increase in the particle dimensions at low aggregation numbers until fairly stable complexes containing approximately 7–8 chains is reached. On further aggregation only a weak additional increase is observed. The first section of the curve (at low x) is indicative of the already-mentioned stiffening on aggregation; the second section of the curve is easily explained by lateral aggregation of fairly inflexible cylinders, which causes almost no change in the radius of gyration so long as the cross-section thickness of the resulting cylinders remains smaller than the length.

Taking all these observations together we can now draw a picture of the structure in the native tissue and what is happening when the xylan is extracted from the tissue. Evidently, individual chains or at most dimerized chains are extracted from the cell walls of rye bran, which suggests that these xylans are not strongly aggregated there, but probably stabilized due to the presence of water and other components in the cell wall. Here we may recall that 1 M NaOH induced a dense aggregate structure, which indicates that this solvent causes a strong dehydration. The extracted chains are not in thermodynamic equilibrium, and a more stable structure is attained when about seven strands come together to form what may be called a fringed micelle. This process is accompanied by a stretching or stiffening of the chains.

Further support for such a structure was drawn from the γ-radiolytic fragmentation of the dry AX-I sample in the solid state (Ebringerova *et al.*, 1989). Similar results to that expected for semicrystalline polymers (Sohma, 1988) were found. Cleaving of the chains occurred preferentially in the amorphous regions. The water-insoluble fractions after irradiation have a lower content of arabinosyl side chains compared to that of the original polysaccharide and a higher degree of order, as indicated by IR spectroscopy. In contrast, the water-soluble fractions carried more arabinosyl substituents and may originate from regions with a low degree of order. The insolubility of the low-branched arabinoxylan in water and the formation of fringed micelles in the complexing solvents are due to the tendency to form interchain aggregations through strong hydrogen bonds in the low-substituted and thus ordered regions (Blake & Richards, 1971).

The initial step in aggregation seems to be a dimerization which could be understood as the result of uronic acid dimerization that will occur for weak electrolytes already in weak acidic media (pH 5). Such dimerization will not significantly change the internal mobility of the chains, since the unimer carries on average only three uronic acid groups.

The expanded 7-chain cluster is still not in the thermodynamic equilibrium and proceeds without further stiffening of the aggregates under the formation of H-bonds. The stabilizing bonds are evidently very strong such that the clusters cannot be broken up once the H-bonds have been formed on storing the dry material.

The incomplete solubilization of the heteroxylans in DMSO is disadvantageous in the homogeneous reaction carried out in this medium. Methylation of the low-branched xylan AX-I in the system DMSO–solid NaOH–CH_3I (Table 4) with various amounts of added water had shown a maximum in yield as well as in methoxyl content at about 10% water. Also the yield and degree of methylation of pectins (Odonmazhig *et al.*, 1992), glucuronoxylan (Odonmazhig *et al.*, 1990) and other low-branched heteroxylans were enhanced under these methylation conditions.

CONCLUSION

The xylan AX-I, and probably other low-branched xylans, are extracted from the cell walls either as single strands or at most dimerized strands. These structures have a high tendency to aggregate further even in the dry state, i.e. after the material was precipitated. Initially, metastable clusters consist of about seven strands with a variation of ±1, two strands on average. This stage of aggregation is accompanied by substantial stiffening of the chains. This intermediate state of aggregation is mainly achieved with the complexing solvents cuoxam, cadoxen and FeTNa. The mixture of DMSO with water in the ratio 90/10 is evidently a better solvent than pure DMSO. Over a period of years, the intermediate clusters slowly aggregate further and unspecifically without any limitation, and eventually the whole material may become insoluble. The radius of gyration of these clusters increase only weakly with the particle weight. From this finding a fringed micelle model is suggested for the clusters where several chains aggregate side by side. It is worth emphasizing that once a large aggregate was formed, the favourable DMSO/H_2O = 90/10 mixture loses its strength and cannot break up the aggregate. The complexing solvents, however, still keep their power, but nevertheless only dissolve the xylans down to a 6–7 stranded bundle. This observation gives further evidence for these bundles staying in a rather stable state, which might be caused by a highly ordered arrangement of the chains.

Table 4. Methylation of arabinoxylan AX-I (Ebringerova, 1993)

DMSO:H_2O (v/v)	Yield[a] (mg/100 mg)	OCH_3 (%)
100:0	70	33·1
95:5	94	34·4
90:10	118	38·4
85:15	81	31·2

[a]The methylated product was obtained by lyophilization of the non-dialysible portion of the reaction mixture.

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REFERENCES

Bantle, S., Schmidt, M. & Burchard, W. (1982). *Macromolecules*, **15**, 1604–9.
Baur, G. (1990). Special development.
Blake, J.D. & Richards, G.N. (1971). *Carbohydr. Res.*, **18**, 11–21.
Bobleter, O. & Schwald, W. (1985). *Das Papier*, **39**, 437–41.
Burchard, W., Schmidt, M. & Stockmayer, W.H. (1980). *Macromolecules*, **13**.
Clacco, C.F. & D'Appolonia, B.L. (1982). *Cereal Chem.*, **59**, 96–100.
Dandliker, W.B. & Kraut, J. (1956). *J. Am. Chem. Soc.*, **78**, 2380–4.
D'Appolonia, B.L. & MacArthur, L.A. (1975). *Cereal Chem.*, **52**, 230–9.
Ebringerova, A. (1993). Unpublished data.
Ebringerova, A., Kacurakova, M., Hromadkova, Z. & Pruzinec, J. (1989). *J. Radioanl. Nucl.*, **136**, 695–8.
Ebringerova, A., Kacurakova, M., Hromadkova, Z. & Pruzinec, J. (1989). *J. Radioanl. Nucl. Chem. Lett.*, **136**, 73–9.
Eremeeva, T. & Ebringerova, A. (1991). *Application Note F321.* Tessek Ltd, Praha, CSFR.
Eriksson, K.E., Peterssen, B.A. & Steenberg, B. (1968). *Svensk Papperstidn.*, **71**, 695–8
Gralén, N. (1944). PhD Thesis. Uppsala University, Sweden.
Henley, D. (1969). *Arkiv Kemi*, **18**, 327–92.
Kromadkova, Z. & Ebringerova, A. (1987). *Nahrung*, **31**, 149–56.
Hromadkova, Z. & Ebringerova, A. (1992). *Food Hydrocoll.*, **6**, 437–42.
Hromadkova, Z., Ebringerova, A., Petrakova, E. & Schraml, J. (1987). *Carbohydr. Res.*, **163**, 73–9.
Huglin, B.H. (1972). In *Light Scattering from Polymer Solutions*. Academic Press, London, UK, Ch. 16.
Izydorczyk, M.S. & Billarderis, C.G. (1992). *Carbohydr. Polym.*, **17**, 237–47.
Izydorczyk, M.S., Billarderis, C.G. & Bushuk, W. (1991). *Cereal Chem.*, **68**, 139–44.
Jander, G. (1986). *Massanalyse*, 14th edn. De Gruyter, Berlin, Germany.
Jayme, G. & Bergmann, W. (1956). *Das Papier*, **10**, 307–12.
Kohn, R., Hromadkova, Z. & Ebringerova, A. (1986). *Coll. Chem. Czech. Commun.*, **51**, 2250–8.
LeBel, R.G., Goring, D.A.I. & Timell, T.E. (1963a). *J. Polym. Sci.*, **C2**, 9–28.
LeBel, R.G., Goring, D.A.I. & Timell, T.E. (1963b). *J. Polym. Sci.*, **C2**, 29–48.
Lineback, D.R., Kakuda, N.S. & Tsen, C.C. (1977). *J. Food Sci.*, **42**, 461–7.

McCleary, B.V. (1986). *Int. J. Biol. Macromol.*, **8**, 349–54.
Mertz, P. (1991). Diploma Thesis. Freiburg University, Germany.
Meuser, F. & Suckow, P. (1986). In *Chemistry and Physics of Baking*, eds. J.M.V. Blanchard, P.J. Frazier & T. Gailard. The Royal Society of Chemistry, Burlington House, London, UK, pp. 42–61.
Odonmazhig, P., Badgaa, D., Ebringerova, A., Mihalov, V & Alföldi, J. (1990). *Carbohydr Res.*, **198**, 163–7.
Odonmazhig, P., Badgaa, D., Ebringerova, A. & Alföldi, J. (1992) *Carbohydr. Res.*, **226**, 353–8.
Seger, B. (1993). Diploma Thesis. Freiburg, Germany.
Seger, B. & Burchard, W. (1994). *Makromol. Chem.; Macromol. Sympos.*, **83**, 291–310.
van Soest, J.P. & McQueen, R.W. (1973) *Proc. Nutr. Soc.*, **32**, 123–30.
Sohma, J. (1988). *Pure Appl. Chem.*, **55**, 1595–601.
Theander, O. & Aman, P. (1979). *Swed. J. Agric. Res.*, **9**, 97–106.
Timell, T.E. (1964). *Adv. Carbohydr. Chem.*, **19**, 247-99.
Valtasaari, L (1957) *Paperi ja Puu*, **39**, 243–52.
Wikström, R (1968) *Svensk Papperstidn.*, **71**, 399–404.