ISOLATION AND STRUCTURAL CHARACTERIZATION OF THE ACETYLATED HETEROXYLAN FROM SISAL (*Agave sisalana*) AND ITS FATE DURING ALKALINE PULPING AND TCF/ECF BLEACHING

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ABSTRACT

The heteroxylan from sisal, an *O*-acetyl-(4-*O*-methylglucurono)xylan with a molecular weight (M_w) of 18 kDa, was isolated by extraction of peracetic holocellulose with Me_2SO and thoroughly characterized by wet chemistry, and NMR spectroscopy. The heteroxylan backbone is composed of (1→4)-linked β-D-xylopyranosyl units (Xyl*p*) partially ramified with terminal (1→2)-linked 4-*O*-methyl-α-D-glucuronosyl (MeGlc*p*A) and a small proportion of α-D-glucuronosyl (Glc*p*A) residues. Roughly 61mol% of Xyl*p* residues are acetylated (DS =0.70). During soda/AQ pulping, Glc*p*A and MeGlcA are partially removed or converted to 4-deoxy-β-L-*threo*-hex-4-enopyranosyluronic acid (HexA), though notable proportion of Glc*p*A maintained intact. The major part of acetyl groups was hydrolyzed during pulping. It was proposed that the predominantly low molecular weight xylan fraction was removed from pulp during bleaching. The profiles of uronosyl residues in xylans from TCF and ECF bleached sisal pulps were rather different.

I. INTRODUCTION

Sisal fibre is a hard fibrous material isolated from the leaves of sisal (*Agave sisalana*), a monocotyledonous plant from the *Agavaceae* family [1, 2]. Originally from Central America and Mexico, the major sisal growing countries are Brazil, Tanzania, and Kenya [2, 3]. The main applications of sisal fibres are the manufacture of ropes for boats, for agriculture needs, and for reinforcement of polymeric matrices [1-4]. Indeed, sisal fibres have certain characteristics that offer new opportunities for the papermaking due to their high tear resistance and folding endurance, high porosity and bulk, high absorbency and alpha cellulose content, making them an excellent raw material for special papers [2-6]. Sisal pulp is industrially produced employing soda AQ pulping.

The study on the hemicelluloses behavior during the pulping from sisal is important, because the partial degradation and dissolution of hemicelluloses is responsible for a significant consumption of pulping chemicals and a decrease of the pulp yield and papermaking properties of pulps [7-9]. Xylans are the major hemicelluloses in most of annual plants including those used for the papermaking. Unlike to xylans from other annual plants (kenaf, flax and jute), which structural features have been determined [10-14], the xylan from sisal never was structurally characterised. This information is important from both fundamental and practical point of view.

In this work, a heteroxylan isolated from sisal fibres was thoroughly characterized by wet chemistry and NMR spectroscopy techniques. Simultaneously, the fate of this heteroxylan during soda/AQ pulping and TCF and ECF bleaching has been studied.

II. EXPERIMENTAL

Sisal (*Agave sisalana*) leaf fibers and corresponding soda/AQ pulps (unbleached and ECF or TCF bleached pulps) were supplied by CELESA pulp mill (Tortosa, Spain). The bleaching sequences applied were D-EP and Q-P for ECF and TCF pulps, respectively. The samples were air-dried, milled using a knife mill and extracted with acetone in a Soxhlet apparatus for 8 h. Klason lignin was estimated according to Tappi standards [15]. Ash content was determined after calcinations 6 h at 575 ºC.The holocelluloses from sisal fibres and of unbleached pulp were obtained from extractives-free sawdust (5.0 g) by a previously published procedure [7]. The holocellulose yields were 69.7% and 94.7% for the sisal fibres and unbleached pulp, respectively. The isolation of acidic heteroxylans was carried out by two consecutive extractions with Me_2SO (1.5 g of holocelluloses) as previously described [7]. For the ECF and TCF bleached pulps the xylans were extracted directly from the pulp without previous preparation of holocelluloses.

The heteroxylan was subjected to Seaman hydrolysis and analysed by GC as alditol acetates [7, 16]. About 4.5 mg of a freeze-dried sisal fibers and pulps (unbleached, ECF and TCF bleached) were subjected to an acid methanolysis according to previously described procedure [17]. GC-MS analysis were performed on a Hewlett-Packard Gas Chromatograph 5890 equipped with a mass selective detector MSD series II, using Helium as carrier gas (35 cm/s), equipped with a DB-1 J&W capillary column (30 m×0.32 mm i.d.×0.25 µm film thickness). The column temperature program was from 175 ºC to at 4 ºC/min and then to 290 ºC at 12 ºC/min.

The detector (FID) temperature was 290 ºC. The SEC analysis for xylan samples has been carried out as described previously [7]. The injected volume was 100 μL.
One-dimensional ^{1}H NMR spectra and two-dimensional ^{1}H-^{1}H TOCSY spectra and ^{1}H-^{13}C HSQC for the xylan samples were acquired as described previously [7, 18]. The hexenuronic acid content was determined in sisal unbleached and TCF pulps by acid hydrolysis according to published procedure [19].

III. RESULTS AND DISCUSSION

The chemical compositions of sisal fibres, the unbleached soda/AQ pulp and TCF and ECF bleached pulps, are presented in Table 1. The sugar analyses of sisal fibres clearly showed that xylan is the principal hemicellulose and the second most abundant polysaccharide after cellulose. The structure of sisal xylan was elucidated based on sugar's analysis by methanolysis (Table 2), 1D and 2D NMR techniques (Figs.1 and 2). It was suggested that the backbone of sisal heteroxylan is composed of (1→4)-linked β-D-xylopyranosyl units (Xyl*p*) partially ramified with terminal (1→2)-linked 4-*O*-methyl-α-D-glucuronosyl (MeGlc*p*A) and a small proportion of α-D-glucuronosyl (Glc*p*A) residues. According to assignments of ^{1}H and ^{13}C signals in ^{1}H-^{1}H TOCSY and ^{1}H-^{13}C HSQC spectra (Fig. 1) the specific regions of partially acetylated β-D-xylopyranosyl units were defined in the ^{1}H NMR spectra (Fig. 2). This allowed the integration of protons from particular structural fragments and their quantification. Roughly 61mol% of Xyl*p* residues are acetylated: 3-*O*-acetylated (39 mol%), 2-*O*-acetylated (13mol%) or 2,3-di-*O*-acetylated (9 mol%) and possesses the substitution degree of 0.70. Almost all Xyl*p* linked at *O*-2 with MeGlc*p*A were 3-*O*-acetylated in (9 mol%). The molecular weight (M_W) of sisal xylan was about 18 kDa, as revealed by SEC analysis.
During the soda/AQ pulping roughly half of xylan was dissolved in liquor. The molecular weight in the remained xylan of pulp decreased to 10 kDa reflecting the alkali-induced degradation. The xylan suffered significant deacetylation (about 95%) and the major part of uronic moieties (about 75%) were converted to 4-deoxy-β-L-*threo*-hex-4-enopyranosyluronic acid (hexenuronic acid or HexA) (Fig. 2). The HexA content was 60.6 mmol/kg of pulp. The balance of uronic moieties in initial xylan and in xylan remained in pulp showed its removal of about 30% during the pulping.
During TCF and ECF bleaching, the xylan of sisal pulp was almost completely deacetylated and possessed slightly higher molecular weight (12 kDa in TCF pulp and 14 kDa in ECF pulp), when compared to this in unbleached pulp. The last fact was explained by predominant elimination of low molecular weight xylan fractions during bleaching procedures. Notable proportion of MeGlc*p*A (about one third) in unbleached pulp was converted to HexA residues during the TCF bleaching. The degradation of HexA with chlorine dioxide in ECF bleaching led to the drastic diminishing of uronic moieties in xylan, whereas the major part of HexA in TCF bleached pulp maintained intact.

Table 1. Chemical composition of sisal fibers and corresponding unbleached and ECF /TCF bleached pulps (%).

Component	Sisal fibers	Unbleached pulp	TCF pulp	ECF pulp
Ash	1.0	1.0	0.4	0.4
Extractives	7.0	0.3	0.1	0.1
Water-soluble	2.3	0.7	0.6	0.4
Klason lignin	5.9	0.7	-	-
Holocellulose	69.7	94.7	-	-
Sugars				
Rha	0.7	0.7	tr	tr
Ara	1.3	tr	tr	tr
Xyl	20.0	19.0	19.4	20.6
Man	0.8	-	-	-
Gal	1.0	tr	tr	tr
Fuc	0.5	-	-	-
Glc	75.7	80.4	80.6	79.4

[tr] traces.

Table 2. Sugar's composition (%) of xylans isolated from sisal fibers and pulps determined by acid methanolysis.

	Rha	Ara	Xyl	Man	Gal	Glc	GalA	GlcA	4-*O*-MeGlcA
Sisal fibers	0.8	0.6	81.6	tr	0.9	1.1	3.5	0.5	11.0
Unbleached pulp	tr	-	99.3	-	-	0.2	-	-	0.5
TCF pulp	tr	-	98.9	-	-	0.9	-	-	0.2
ECF pulp	tr	-	98.4	-	-	1.3	-	-	0.3

[tr] traces

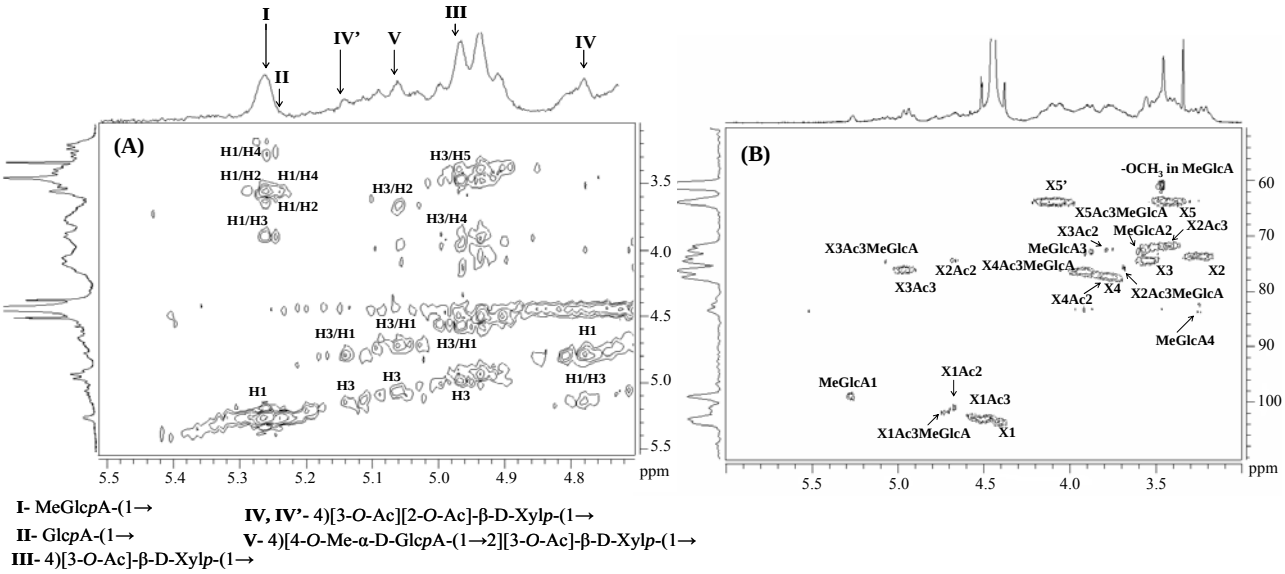

Figure 1. Anomeric region of TOCSY (A) and HSQC (B) spectra (D_2O, 25 ºC) of heteroxylan from sisal.

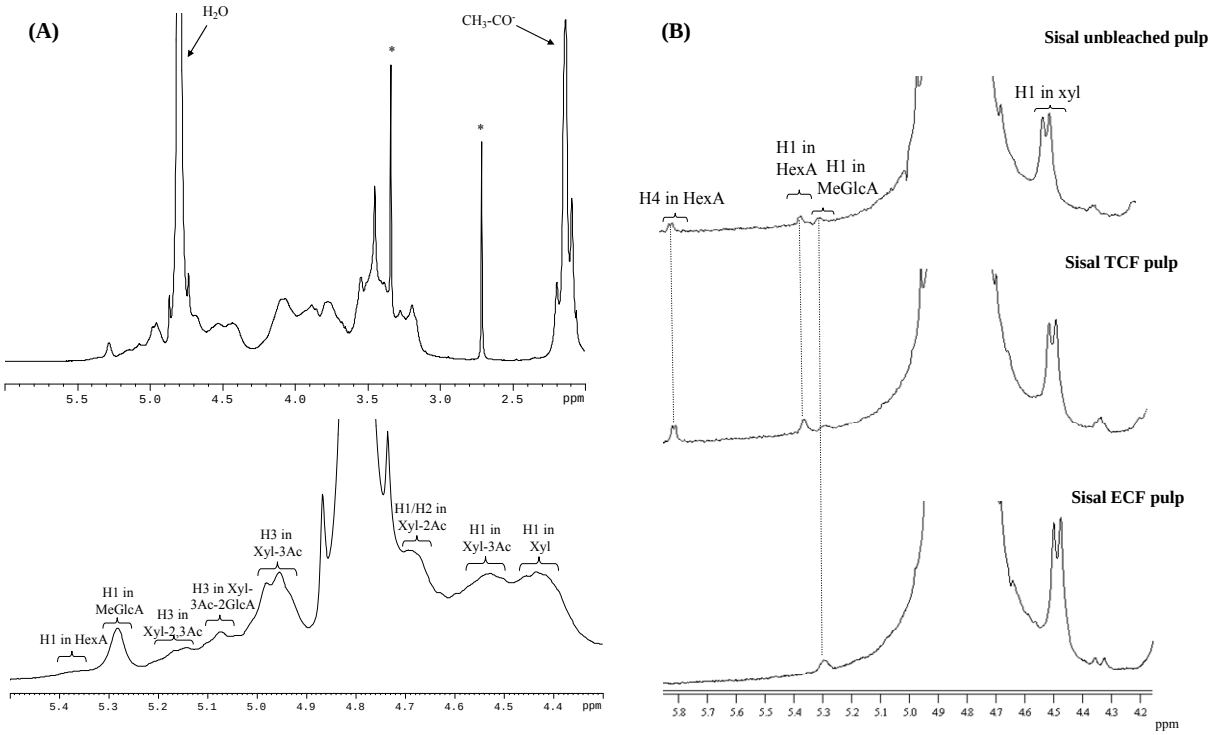

Figure 2. ^1H NMR spectrum (D_2O, 30 ºC) of heteroxylan from sisal fibres (A, top image) and the expanded region of anomeric protons (A, bottom image). ^1H NMR spectra of heteroxylans isolated from sisal unbleached pulp and from TCF and ECF bleached pulps (B). Solvent impurities are designated by asterisk.

IV. CONCLUSIONS

The heteroxylan isolated from sisal fibers has been characterized and its behaviour during soda/AQ pulping and TCF/ECF bleaching has been studied. The heteroxylan backbone is composed by (1→4)-linked β-D-xylopyranosyl units (Xyl*p*) partially ramified with terminal (1→2)-linked 4-*O*-methyl-α-D-glucuronosyl (MeGlc*p*A) and a small proportion of α-D-glucuronosyl (Glc*p*A) residues. Roughly 61mol% of Xyl*p* residues in sisal xylan are acetylated. The major proportion of acetyl groups were attached at *O*-3 of Xylp residues (39

mol%) followed by those attached at *O*-2 (13 mol%) and both at *O*-2 and at *O*-3 (9mol%). The average acetylation degree (DS) of heteroxylan was 0.70 and the molecular weight (M_w) of 18 kDa. After the pulping and bleaching the acetyl groups were removed almost completely. Uronic acids were also partially removed during the pulping or converted to HexA in a large extent. HexA revealed to be relatively stable during TCF bleaching and were predominant among uronic moieties of xylan. Since all HexA were degraded during ECF bleaching, the final pulp contained a xylan with rather small amount of uronosyls (MeGlc*p*A residues).

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