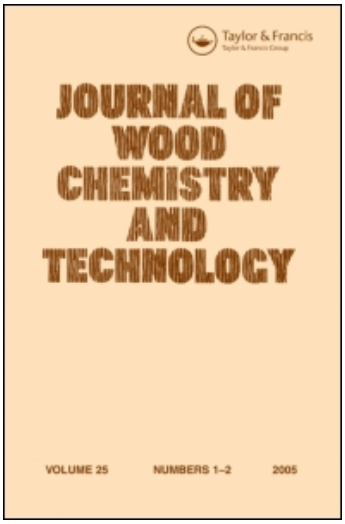

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COMBINED HPLC ANALYSIS OF ORGANIC ACIDS AND FURANS FORMED DURING ORGANOSOLV PULPING OF FIBER HEMP

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ABSTRACT

During organosolv pulping of fiber hemp (*Cannabis sativa L.*) with a mixture of ethanol/water, delignification is catalyzed by released acetic acid and formic acid in the effluent.
The major sources of acetic acid are the acetyl groups, as determined by means of the acetyl balance, whereas formic acid is mainly formed as degradation product of polysaccharides. Trace amounts of other short chain organic acids and furans, formed from cellulose and hemicellulose, are also present in the effluent.
A relatively simple and reliable HPLC method is described to analyze simultaneously the carboxylic acids and furans quantitatively. Acetyl and formyl contents of hemp core (4.3% and 0.2% respectively) and hemp bast (1.3% and 0.2%) are also analyzed with HPLC after an alkaline saponification.

INTRODUCTION

Autocatalyzed Organosolv Pulping of Fiber Hemp

Organosolv pulping of lignocellulosic materials is a promising pulping process. Especially, autocatalyzed organosolv pulping fulfills conditions of

stricter environmental regulations, because of relative small scale pulping, easy recovery of chemicals and byproducts. Delignification of woods and annual plants is actually catalyzed by organic acids liberated during autocatalyzed organosolv pulping.

In this paper the release of acids and furans in the effluent during autocatalyzed organosolv pulping of fiber hemp (*Cannabis sativa L.*) with ethanol/water (60 : 40 v/v) in batch reactors, was studied by means of a combined HPLC method to gain a better understanding of this process. Organosolv and saponification experiments have been performed with hemp, hardwoods, xylan, xylose, cellulose and lignin for identification of the main sources of the components released in the effluent.

Degradation Products Formed during Delignification of Lignocellulosic Material

Short chain organic acids, like acetic acid, formic acid, glyoxylic acid and hydroxy-acids such as lactic acid and glycolic acid are formed during delignification of lignocellulosic material. Acetic acid and formic acid are the main volatile acids released during hydrothermal and organosolv degradation of wheat straw and wood[1,2,3].

Acetyl esters bound to noncellulosic polysaccharides are the major sources of acetic acid. The amount of acetyl groups in hardwoods is about 3 - 5% and 1 - 2% in softwoods[4,5]. In hardwoods these groups occur on 70 - 80% of xylosyl residues, whereas in softwoods they are both attached to mannose and glucose units of glucomannan[6].

The formyl content does not exceed 0.04% for most untreated woods[4]. Higher quantities of formic acid found in the effluent of treated lignocellulosic material probably result from degradation of polysaccharides. Acetyl and formyl groups are more easily released in alkaline media than in acidic solvents[2,4,7].

During organosolv delignification of woods and annual plants removal and degradation of hemicellulose also takes place. Hemp core hemicellulose, mostly consisting of xylan (TABLE 1), is hydrolyzed to pentoses (xylose) and may be converted to furfural, aldehydes, ketones or organic acids[7]. In turn furfural will be degraded by acids to produce formic acid and a polymeric black resin after condensation[8].

The absolute amount of cellulose in hemp core decreases only slightly during organosolv delignification in both flow-through or batch reactors, whereas about 50% of the hemicellulose is removed (pulping time: 2.5 hours, temperature: 195 °C, ethanol/water: 60 : 40 v/v)[9]. Cellulose and hemicellulose may be depolymerized to oligomers and finally monosaccharides. Glucose and other hexoses are convertible to 5-hydroxy-methylfurfural (HMF) in an acidic medium. These reactions are thermally activated. HMF can be degraded to levulinic acid and formic acid[8,10,11].

Breakdown of lignin may also result in the release of short chain organic acids[2].

Analytical Methods for Determination of Organic Acids and Furans

Many methods have been developed for the analysis of short chain organic acids in diverse samples. These methods include volumetric[4], isotachophoretic[6], spectrophotometric[12] and enzymatic determinations, as well as capillary electrophoresis[13], gas liquid chromatography (GLC)[14,15] and high performance liquid chromatography (HPLC)[15-19].

Volumetric and spectrophotometric methods are non specific and rather time-consuming. Enzymatic analysis is also slow compared to chromatographic methods.

GLC is commonly used for the analysis of short chain carboxylic acids. A major problem is that a flame ionization detector cannot be used, because this detector has a very low response for formic acid, unless this acid is first

TABLE 1

Chemical Composition of Fiber Hemp, Variety Compolti Sargaszaru (% w/w)[30]

Component	Bast fiber	Core fiber
Hemicellulose#		
Rhamnose	0.4	0.5
Arabinose	0.7	0.3
Xylose	1.5	16.4
Mannose	2.3	1.2
Galactose	1.7	0.6
Glucose	5.5	3.1
Cellulose	61.2	34.1
Lignin	3.0	21.8
Uronic acids	1.8	3.3
Proteins	2.9	3.3
Ash	5.9	4.5
Extractives##	11.9	8.9

= Calculated as polysaccharides.
= After successive soxhlet extraction with ethanol/benzene, ethanol and hot water.

derivatized. Furthermore, some of the organic acids analyzed in this study are not volatile enough, so they must also be derivatized. Therefore a complex sample pretreatment must be used.

HPLC has overcome the disadvantages of the previous described methods. Several ion-exchange, ion-exclusion, ion-pair and reverse-phase chromatographic determinations have been applied for the quantitative analysis of organic acids[15-19]. Detection is performed with refractive index (RI), conductivity or ultraviolet (UV) detectors at 206 - 220 nm. When using a RI detector only an isocratic elution gives a stable background signal.

The acetyl and formyl content of lignocellulosic material can be determined by GLC or HPLC after saponification[20] or by distillation and titration[3]. Månsson et al[21,22] have determined the amount of O-acetyl and other O-acyl groups in cellulosic material and wood after aminolysis with pyrrolidine.

The formed acylamides are analyzed by GLC. Bethge and Lindström[23] found that acetyl groups in wood can be split off with sodium ethoxide and analyzed by GLC after conversion to their benzyl esters. The GLC methods are rather complicated, time-consuming and often hazardous chemicals are used.

Furfural and HMF can be determined by spectrophotometric and chromatographic[24] methods. The former determinations do not differentiate between HMF and furfural. To overcome this problem Tu et al[25] have developed a method for the simultaneous analysis of both furans by derivative spectrophotometry using the reaction with 2-thiobarbituric acid. Scott[26] has used chloride to increase the differences between the spectra of these furans.

Furans can be rapidly analyzed with HPLC without derivatization on reversed-phase based columns and UV detection at 280 - 285 nm. HMF and furfural will also be retained on an ion-exchange stationary phase[7,27,28], so that the analysis of short chain organic acids and furans can simply be combined.

HPLC Methods Used for the Analysis of Organic Acids and Furans

First, this paper describes a method for the determination of the acetyl and formyl content of lignocellulosic materials by means of HPLC after an alkaline saponification. Release of acyl groups was performed in 0.6 M sodium hydroxide in 2-propanol/water to prevent depolymerization of hemicellulose and cellulose[20]. Therefore all polysaccharides will be precipitated and no interference in the HPLC analysis occurs.

Secondly, organic acids and furans in the rather complex organosolv effluents are analyzed on a strong cation-exchange HPLC column. This column is packed with sulfonated styrene divinylbenzene gels and can be used for the separation and determination of organic acids, alcohols, aldehydes and nitriles[29]. Besides short chain organic acids and furans also lignin, phenolic acids, proteins and other extractives may be present in the effluent. To protect the column for fouling, components like lignin, proteins and several extractives are removed from the effluent by acidic precipitation.

EXPERIMENTAL

Chemicals

Milli-Q water was made by means of a Reagent Grade Water Purification System (Millipore, Milford, MA, U.S.A.). Ethanol, acetic acid, formic acid, lactic acid, sulphuric acid, furfural, 2-propanol (IPA), 5-hydroxymethyl-furfural (HMF), phenoxyacetic acid, glycolic acid, sodium hydroxide, cellulose native, D(+)-xylose, ethyl acetate and fumaric acid were all purchased from Merck (Darmstadt, F.R. Germany). Levulinic acid was obtained from Janssen Chimica (Geel, Belgium). Xylan (from oat spelts), cellulose acetate and D-glucuronic acid were obtained from Sigma (St. Louis, MO, U.S.A.). Helium was purchased from Van Veenendaal (Amerongen, The Netherlands). Lignin used was Alcell™ lignin from mixed hardwoods (Repap Technologies, Valley Forge, PA, U.S.A.).

Raw Materials

Air dried fiber hemp stems (variety Compolti sargaszaru; TABLE 1) consisting of about 35% bast fibers and 65% core fibers, were cracked in a wheel stem brake machine and separated into core and bast fractions. Hemp core chips were sieved at sizes between 0.42 and 10 mm, whereas bast fibers and whole stems were cut to 4.1 mm length.

Impregnation of the biomass was carried out with ethanol/water (60 : 40 v/v) by boiling under reflux during 15 minutes.

For determination of the acetyl and formyl content, air dried hemp core, hemp bast, hemp whole stem, hemp organosolv pulps, yellow poplar (*Populus robusta*) and aspen (*Populus tremuloides*) were milled to pass a 0.42 mm sieve.

Organosolv Pulping of Hemp

Autocatalyzed organosolv pulping of hemp core (HC) chips, hemp bast (HB) and hemp whole stems (HS) with ethanol/water (60 : 40 v/v) was

performed in a 4 litre rocking batch reactor. The experiments were carried out at 195 °C. The reactor was heated with silicone oil and de-aerated at 75 °C. The average heating up and cooling down time was 60 minutes. During the process, samples of 10 ml were taken, by which the release of acids and furans could be monitored at any time in the pulping process. A liquor to hemp raw material (l/r in l/kg) ratio for HC, HS and HB of 10, 9 and 7 was used respectively[9].

Organosolv Model Compound Degradation

Batch reactors of 100 ml were used for organosolv degradation of model compounds during 180 minutes at 195 °C. The reactors were heated and shaken in an oil bath. At about 75 °C the reactors were de-aerated and the average heating up and cooling down time was 15 minutes.

To get more information about the possible sources of the organic acids and furans, mono- and polysaccharides and lignin were used in the same amounts as occurring in hemp raw materials (TABLE 1). The effluent pH after autocatalyzed organosolv pulping of hemp bast and core with ethanol/water (60 : 40 v/v) was found[9] to be 5.2 and 4.8 respectively. In model compound degradation experiments the pH was lowered before pulping by addition of acetic acid.

HPLC Analysis of the Acetyl and Formyl Content of Raw Materials and Pulps

The validity of the method used was checked by recovery measurements with cellulose acetate and ethyl acetate of known acetyl content. Saponification conditions were varied as described below.

Samples of 60 mg from raw materials and pulps were suspended in 2 ml solvent consisting of 0.6 (0.4) M sodium hydroxide in 2-propanol (IPA)/water (1 : 1 v/v). During 2 hours the suspension was shaken (300 r.p.m.) at ambient temperature. The mixture was then centrifuged 10 minutes at 4200 r.p.m.. The

clear supernatant was diluted 1 : 1 (v/v) with approximately 3.5 g/l phenoxyacetic acid (Internal Standard = IS) dissolved in 1.0 M sulphuric acid. After mixing, the solution was centrifuged and the clear supernatant was filtered through a 0.2 μm PTFE-filter (Altech, Deerfield, IL, U.S.A.).

Samples of 250 mg from hemp bast and hemp core fibers were saponified in 5 ml 2.0 M sodium hydroxide in water during 24 hours at ambient temperature. The same experiments were carried out in 15 ml batch reactors during 2 hours at 170 °C. To the 2.0 M sodium hydroxide extracts 2.0 M sulphuric acid with IS was added and the previous described sample pretreatment was used. The standard mixtures of acids were all made in the same extraction solvent as used.

A HPLC system (Pharmacia LKB, Uppsala, Sweden) consisting of one HPLC pump 2248, column oven 2155, autosampler 2157 and a variable wavelength monitor 2141 was used combined with a differential refractometer 410 (Waters, Milford, MA, U.S.A.). Sample injection of 10 μl was performed using a 100 μl sample loop. The column used was a Shodex Ionpak KC-811 (300 x 8 mm I.D.; Shodex, Tokyo, Japan) packed with strong cation-exchange resin gels fitted with a Fast Fruit Juice Guard-PAK™ Precolumn (Waters, Milford, MA, U.S.A.). The eluent was 0.1% (v/v) phosphoric acid, helium degassed and an isocratic elution was performed with a flow of 1 ml/min during 30 minutes. The column temperature was held at 60 °C and detection was carried out with UV 210 nm and RI.

Effluent Analysis of Short Chain Organic Acids and Furans with HPLC

The organosolv effluents from pulping of fiber hemp and model compound degradation experiments were collected and each effluent was centrifuged at 4200 r.p.m. during 10 minutes. The supernatant was diluted 1:1 (v/v) with an IS-solution of circa 3.5 g/l phenoxyacetic acid in 1.0 M sulphuric acid. After mixing, this solution was centrifuged 10 minutes at 4200 r.p.m.. The clear

supernatant was filtered through a 0.2 µm PTFE-filter. The standard mixtures of acids and furans were also prepared in ethanol/water (60 : 40 v/v) and the same sample pretreatment was used. A sample volume of 10 µl was injected on the same column used for the acetyl and formyl content determination.

The eluent was 0.025% (v/v) phosphoric acid, helium degassed and used at a flow of 1 ml/min. The column temperature was controlled at 40 °C. Duration of a single run was 40 minutes and detection was performed with UV and RI. After 16 minutes, UV detection was switched from UV 210 nm (acids analysis) to UV 280 nm (furans determination). The refractive index detector was thermostated at 35 °C.

Quantities of organic acids and furans were calculated with a computer integrator according to the internal standard method. For the measurement of the biomass content, from which the components were probably formed, stoichiometric factors were used as shown in TABLE 2.

RESULTS AND DISCUSSION

The main released compounds besides lignin and hemicellulose fragments in the effluent from organosolv pulping of hemp have been determined to elucidate the catalytic character of this process. The acyl contents of hemp raw materials were analyzed by means of HPLC as well as short chain carboxylic acids in the effluent. Since polysaccharide degradation is also catalyzed by acids, the release of furfural and HMF was monitored as well.
Furthermore, the amounts of acetic acid (acetyl) found in raw material, effluent and pulp give an acetyl balance. Together with model compound degradation experiments, formation of acidic products and furans were studied.

Acetyl and Formyl Content Analysis of Raw Materials and Pulps with HPLC

In the concentration range of 0.01 g/l - 3 g/l (UV 210 nm detection) and 0.03 - 3 g/l (RI detection) all acids analyzed show an excellent linearity with

TABLE 2

Conversion Factors

Compound	Calculated as	Factor
Formic acid	Formyl	29/46
Acetic acid	Acetyl	43/60

correlation coefficients of 0.99. Phenoxyacetic acid gives a similar linearity for concentrations ranging from 0.5 - 4 g/l. The response factors calculated with regard to phenoxyacetic acid are constant (rel. std. dev. < 5%).

HPLC results for the saponification of hemp core and bast with 0.6 M sodium hydroxide in IPA/H_2O are illustrated in FIGURE 1 and FIGURE 2.

The recoveries of acetyl groups released from cellulose acetate and ethyl acetate are good (TABLE 3). Detection with RI and UV 210 nm gives similar results. As saponification with 0.6 M NaOH gives a slightly higher recovery of acetyl groups, than extraction with 0.4 M NaOH, the former was used as the saponification solvent.

Saponification of hemp with different sodium hydroxide concentrations at ambient temperature gives no significant difference in the acetyl or formyl content (TABLE 4). Thus the release of these acyl groups is probably complete after extraction during 2 hours. Also lactic acid in quantities of 0.1 - 0.2% (w/w) was found.

Saponification at 170 °C strongly increases the amounts of released acetic acid, formic acid (TABLE 4) and lactic acid (4 - 5% w/w) in comparison with the amounts released at ambient temperature. This must be ascribed to sugar degradation, which is in good agreement with the findings of Oefner et al[7]. They found a 10 - 23% (w/w) yield of acids (formic, acetic, lactic and glycolic acid) based on xylose breakdown in 0.1 M NaOH at 180 - 220 °C.

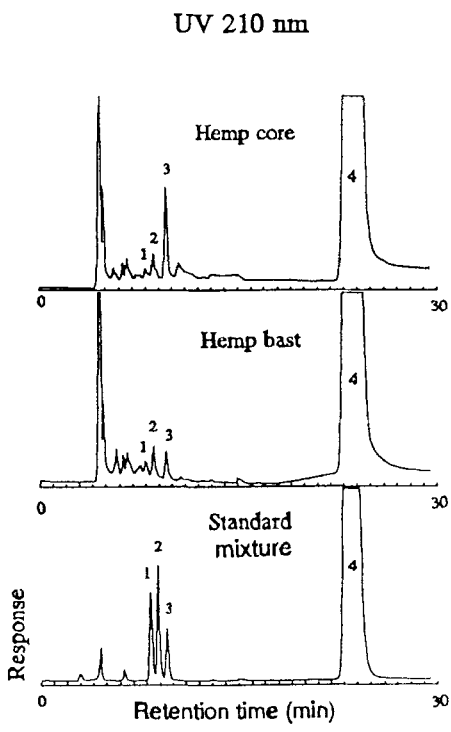

FIGURE 1:
Chromatogram of acids released by saponification of hemp in IPA/H_2O (UV 210 nm)
Peak number: 1= Lactic acid; 2= Formic acid; 3= Acetic acid; 4= Phenoxyacetic acid (IS).

TABLE 5 shows the acetyl and formyl content of several raw materials investigated. Analysis was performed by saponification with 0.6 M sodium hydroxide in IPA/H_2O (1:1 v/v). Similar results are obtained with RI and UV 210 nm detection.

The acetyl content of hemp core is within the range of 3 - 5% found for hardwoods[4,5] (TABLE 5), whereas the acetyl content of hemp bast is

RI

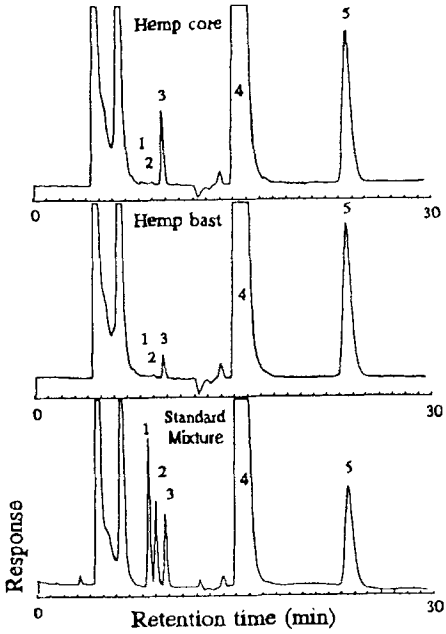

FIGURE 2:
Chromatogram of acids released by saponification of hemp in IPA/H_2O (RI)
Peak number: 1= Lactic acid; 2= Formic acid; 3= Acetic acid; 4= IPA;
5= Phenoxyacetic acid (IS). The first two peaks are from sulphuric acid.

TABLE 3

Acetyl Determination of Samples with Known Acetyl Contents

Sample	NaOH (M) in IPA/H_2O	n	Acetyl mean (% w/w)			
			Known/ Calculated	Found	Std.dev.	Recovery
UV210 detection:						
Cellulose acetate	0.4	2	38.3	35.2	1.0	91.8
Cellulose acetate	0.6	3	38.3	35.8	0.8	93.5
Ethyl acetate	0.6	3	48.9	47.0	3.1	96.3
RI detection:						
Cellulose acetate	0.4	2	38.3	36.3	1.0	94.8
Cellulose acetate	0.6	3	38.3	36.6	0.4	95.5
Ethyl acetate	0.6	3	48.9	48.4	2.7	99.0

TABLE 4

Saponification Experiments of Hemp Core and Hemp Bast (RI detection)

Sample	NaOH (M)	n	Solvent	Temp. (°C)	Time (hour)	Acetyl content mean (% w/w)	Acetyl content std. dev.	Formyl content mean (% w/w)	Formyl content std. dev.
Hemp bast	0.6	6	IPA/H_2O	Ambient	2	1.27	0.04	0.18	0.02
	2.0	2	H_2O	Ambient	24	1.29	0.00	0.15	0.00
	2.0	2	H_2O	170	2	2.09	0.08	1.29	0.02
Hemp core	0.4	2	IPA/H_2O	Ambient	2	4.16	0.11	0.23	0.01
	0.6	6	IPA/H_2O	Ambient	2	4.31	0.17	0.25	0.04
	2.0	2	H_2O	Ambient	24	4.08	0.09	0.20	0.00
	2.0	2	H_2O	170	2	6.42	0.19	1.38	0.16

TABLE 5

Acetyl and Formyl Content of Raw Materials (% w/w; n=3)

Sample	Acetyl Mean (%) UV210	Std. dev.	Acetyl Mean (%) RI	Std. dev.	Formyl Mean (%) UV210	Std. dev.	Formyl Mean (%) RI	Std. dev.
Hemp bast	1.24	0.02	1.27	0.04	0.20	0.02	0.18	0.02
Hemp core	4.34	0.19	4.31	0.17	0.27	0.09	0.25	0.04
Aspen	2.98	0.11	3.12	0.10	0.01	0.00	0.11	0.00
Yellow poplar	2.73	0.04	2.85	0.05	0.03	0.00	0.07	0.00
Xylose	0.00	0.00	0.07	0.01	2.09	0.03	2.42	0.16
Xylan	0.05	0.00	0.00	0.00	0.01	0.00	0.00	0.00
Glucose	0.00	0.00	0.00	0.00	1.08	0.02	1.18	0.06
Glucuronic acid	0.15	0.00	0.00	0.00	1.35	0.03	1.44	0.01

comparable with the 1 - 2% determined for softwoods[4,5]. The amounts of formyl groups in hemp are higher than in hard- and softwoods. In all lignocellulosic materials analyzed acetyl groups are the main acyl groups. The alkaline degradation of polysaccharides to acetic and formic acid under these analytical conditions can be neglected (TABLE 5). The acetyl balance for autocatalyzed organosolv pulping of hemp will be discussed later in this paper.

Analysis of Organic Acids and Furans in Organosolv Effluents with HPLC

The acids analyzed show good linearity for concentrations ranging from 0.01 - 6 g/l (UV 210 nm detection) and 0.03 - 6 g/l (RI detection). HMF and furfural are determined in the concentration range of 0.1 - 400 mg/l (UV 280 nm detection), which give similar linearity. Chromatograms of representative effluent fractions taken during autocatalyzed organosolv pulping of hemp core and bast are shown in FIGURE 3 and 4.

The amounts of acids (RI) and furans (UV 280 nm) formed during autocatalyzed organosolv pulping of hemp are shown in FIGURES 5 - 7. In these figures the first 60 minutes represent the heating up time, whereas during the last 60 minutes the reactor was cooled to ambient temperature.

Detection of formic acid at UV 210 nm consequently gives higher values than calculation with RI data. This is due to co-elution of fumaric acid (t_r= 8.8 min.; $\varepsilon_{UV\ 210}$= 126) with formic acid (t_r= 9.0 min.; $\varepsilon_{UV\ 210}$= 1). Even trace amounts of fumaric acid will give a much higher UV 210 nm response caused by the relatively large extinction coefficient, while contribution to the RI signal is negligible. This conclusion is confirmed by a HPLC analysis of the effluent with two serial connected columns (Shodex Ionpak KC-811; eluent: 0.025% (v/v) phosphoric acid; flow: 0.8 ml/min; temperature: 65 °C), which give a baseline separation of these acids. The concentration of fumaric acid decreases with increasing pulping time and at the end of the organosolv experiments performed the amount released can be neglected.

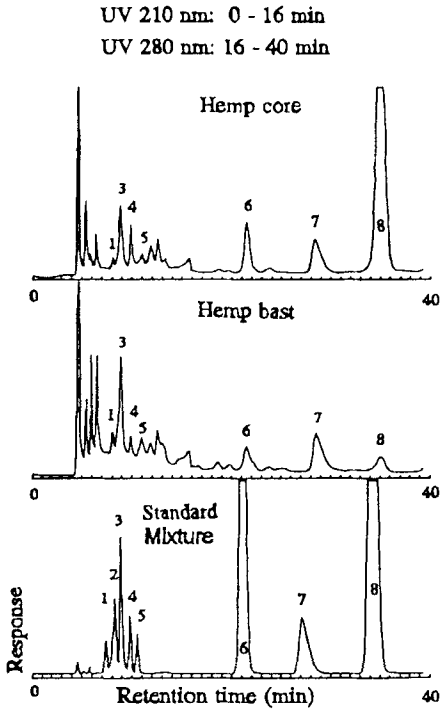

FIGURE 3:
Chromatogram (UV) of compounds formed during autocatalyzed OS pulping of hemp
Peak number: 1= Glycolic acid; 2= Lactic acid 3= Formic acid; 4= Acetic acid; 5= Levulinic acid; 6= HMF; 7= Phenoxyacetic acid (IS); 8= Furfural
Wavelength changing at 16 minutes is preceded by an autozero of the UV signal.

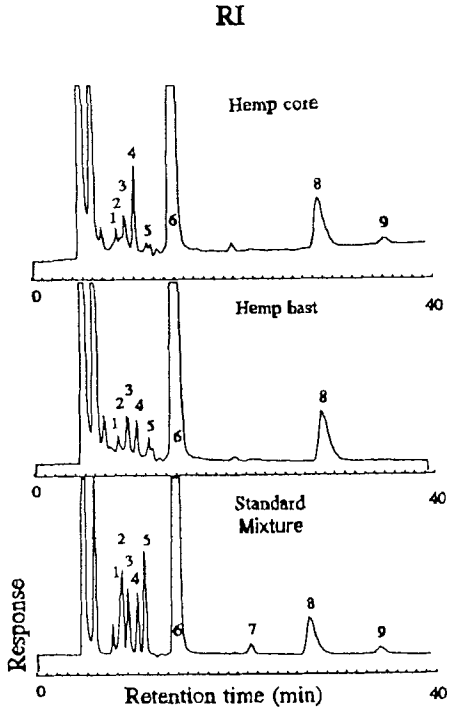

FIGURE 4:
Chromatogram (RI) of compounds formed during autocatalyzed OS pulping of hemp Peak number: 1= Glycolic acid; 2= Lactic acid; 3= Formic acid; 4= Acetic acid; 5= Levulinic acid; 6= Ethanol; 7= HMF; 8= Phenoxyacetic acid (IS); 9= Furfural The first two peaks are from sulphuric acid.

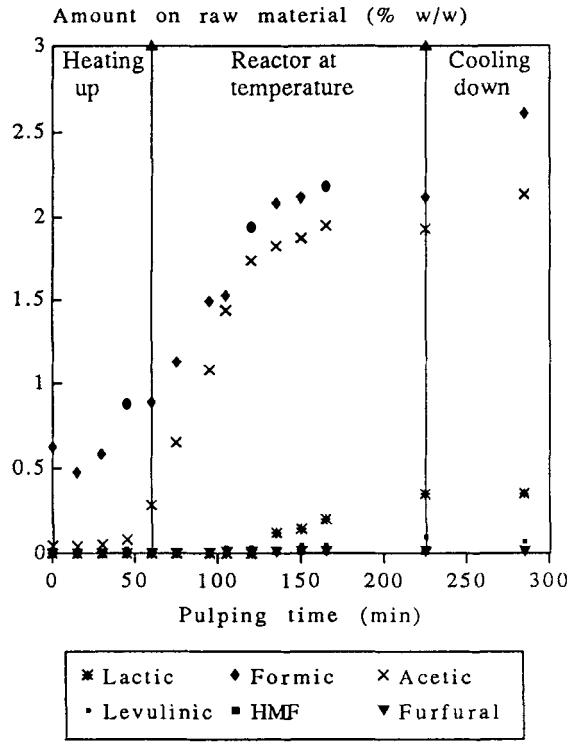

FIGURE 5:
Compounds released during autocatalyzed organosolv pulping of hemp bast (4 l.; 60% ethanol; 195 °C; l/r = 7).

Acetic acid and formic acid are the main volatile acids formed (FIGURES 5-7). The results are comparable with those found for five hardwoods[2]. Acetic acid (pK_a= 4.77) and formic acid (pK_a= 3.80) mainly account for lowering of the effluent pH. Equal concentrations of formic acid are found in the effluents of HB, HC and HS (FIGURES 5-7). The amounts of glycolic, levulinic and lactic acid remain relatively small. Minor degradation of hemicellulose and cellulose to furfural and HMF occurs under the conditions studied (FIGURES 5-7). However, these furans may polymerize or can be further degraded to short

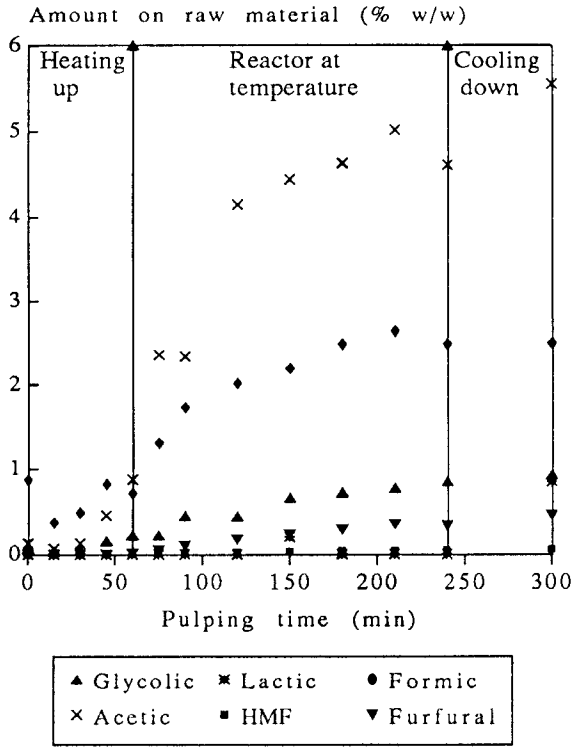

FIGURE 6:
Compounds formed during autocatalyzed organosolv pulping of hemp core (4 l.; 60% ethanol; 195 °C; l/r = 10).

chain acids. The amounts of most compounds analyzed in the effluents increase with increasing pulping time.

From about 150 °C the amount of acetic acid strongly increases, indicating thermally activated hydrolysis of acetyl groups (FIGURE 8). Formic acid is very quickly released during impregnation to amounts (0.6% w/w) higher than the formyl groups (0.2% w/w) present in hemp bast (FIGURE 8). Transfer of the impregnated biomass to the reactor can lead to evaporation of the volatile formic acid, causing dips in FIGURE 5 and 6. During autocatalyzed

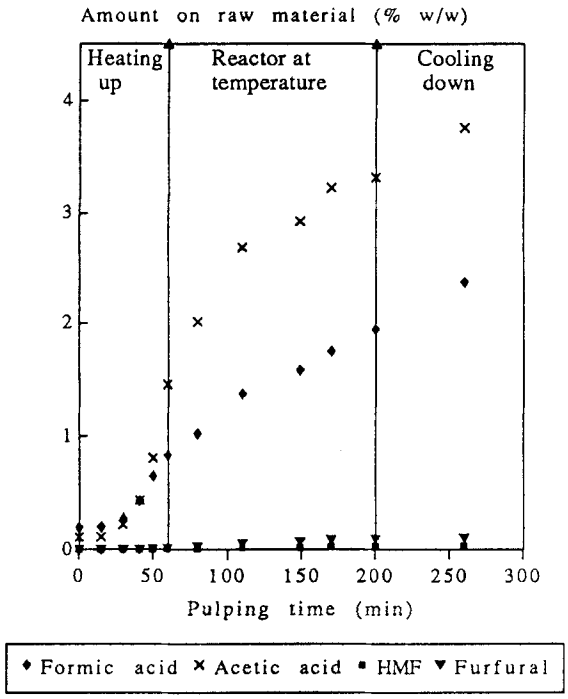

FIGURE 7:
Compounds formed during autocatalyzed organosolv pulping of hemp whole stem (4 l.; 60% ethanol; 195 °C; l/r = 9).

organosolv pulping of hemp, formyl groups are almost completely removed to amounts lower than 0.02% (w/w). Mainly other sources than ester groups are responsible for the release of formic acid.

The results shown in TABLE 6 indicate that the amounts of acetic acid (calculated as acetyl) in the effluents mainly result from the release of acetyl groups present in hemp raw material. However, during extended pulping of HB (see TABLE 6; HB: 285 min) acetic acid is not only formed from released acetyl groups, but also to a lesser degree from degradation reactions as studied in the next section.

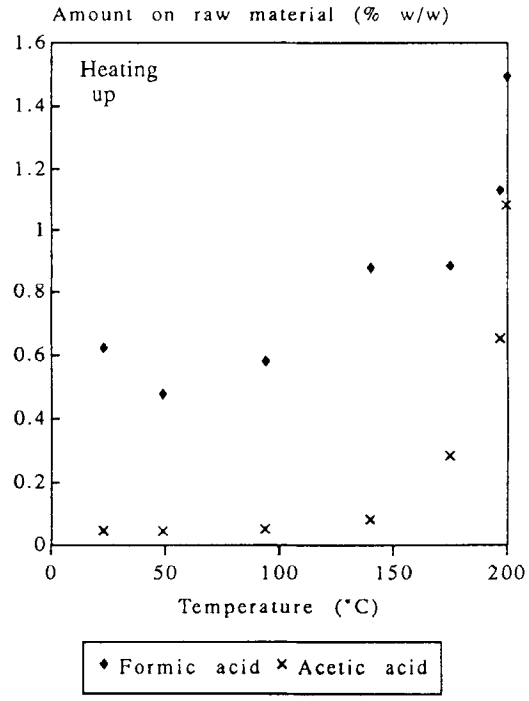

FIGURE 8:
Influence of temperature on the release of acids during the heating up phase of organosolv pulping of hemp bast (4 l.; 60% ethanol; l/r = 7).

TABLE 6

The Acetyl Balance of Autocatalyzed Organosolv Pulping of Hemp

Sample	Total pulping time (min)	Acetyl (% w/w on raw material)		
		Raw material	Effluent	Pulp
HB	285	1.27	1.53	0.06
HB	180	1.27	1.07	0.12
HC	300	4.31	3.98	0.41
HC	270	4.31	n.d.	0.47
HS	260	3.15	2.70	0.20

n.d. = not determined.

Organosolv Model Compound Degradation

In TABLE 7 the results of the model experiments are shown. Acetic acid concentrations of respectively 0.041 g/l and 0.013 g/l were used to lower the solvent pH to 4.8 (HC) and 5.2 (HB). Even without addition of acetic acid, xylose is degraded to relative high quantities of furfural (TABLE 7). The presence of acids will only increase the furfural formation from xylose[7]. In this model study, the accessibility of xylose is optimal and no prior degradation of xylan to the monosaccharide is necessary. Even so, amounts of furfural found in the effluent during pulping of hemp can be explained by the degradation of xylan.

Formic acid and all other acids analyzed, except levulinic acid (TABLE 7), can also be formed from xylan (xylose) during organosolv pulping of hemp. In spite of the different xylan content of hemp raw materials (TABLE 1) equal quantities of formic acid are found in their effluents. Thus the concentration of xylan in the raw material is not the determining factor for the production of this acid. The xylan used in this study contains approximately 15% glucose and 10% arabinose residues. The formed acids (TABLE 7) cannot be ascribed only to degradation products of xylan but also the other sugar residues will contribute.

Glucose is one of the sources for formation of HMF (TABLE 7). As native cellulose is contaminated with other sugar residues, like xylose (4% w/w)[30] and mannose (1% w/w)[30], the released acids and furans in these experiments may partly be formed from these residues. Under the organosolv conditions used for pulping of hemp, HMF may be formed from cellulose and hemicellulose after depolymerization to glucose. This furan is rather stable under these conditions, because formation of levulinic acid in the effluents hardly occurs.

The organic acids and furans formed during model pulping of Alcell lignin (TABLE 7) may be released from formyl groups, acetyl groups, xylan and glucan (0.14%, 0.16%, 0.20%[30] and 0.10%[30] w/w respectively). This lignin from

TABLE 7

Amounts (% w/w; n=2) of Acids and Furans Formed during Organosolv Model Pulping in 100 ml Batchreactors (195 °C; 60% Ethanol; 3 Hours)

Sample[#]	pH initial	Glycolic UV210	Lactic UV210	Formic RI	Acetic RI[###]	Levulinic RI	HMF UV280	Furfural UV280
Xylose	-[##]	1.71	4.51	0.91	0.69	0.00	0.02	15.87
Xylan	4.8	1.31	1.04	1.26	0.54	0.00	0.18	1.95
Xylan	5.2	0.82	0.97	0.79	0.00	0.00	0.11	1.41
Cellulose	4.8	0.18	0.41	0.12	0.07	0.01	0.75	0.31
Cellulose	5.2	0.17	0.53	0.28	0.04	0.00	0.78	0.32
Lignin	-[##]	0.00	0.00	0.28	0.00	0.00	0.02	0.08

[#] = Concentrations used as occurring in hemp raw materials (TABLE 1); pH = 4.8 for model experiment of hemp core and pH = 5.2 for hemp bast.

[##] = No addition of acetic acid. Amounts used as present in hemp core.

[###]= Released amounts corrected for acetic acid addition.

mixed hardwoods is not representative of the native lignin occurring in hemp fibers, and Alcell lignin is a product of a pulping process. Even so, high amounts of acids and furans formed from hemp lignin are not expected, but this has not been tested with the model compound experiments performed.

Model experiments with formic acid and acetic acid in ethanol/water show only a very slight decrease of the concentration of acetic acid, probably due to ester formation. Both HMF and furfural are degraded under these conditions, without addition of acids, to formic and acetic acid (1% w/w based on both furans). The presence of acids may increase the breakdown of these furans.

CONCLUSIONS

Development of this HPLC analysis enables the simultaneous determination of short chain organic acids and furans in organosolv effluents with one relatively simple and reliable method.

Hemp core and hemp bast fibers contain respectively 4.3% and 1.3% (w/w) acetyl groups and the formyl content in these raw materials is approximately 0.2% (w/w).

Both acetic acid and formic acid are the main acids responsible for lowering the effluent pH and accomplishing autocatalyzed organosolv pulping of hemp. The thermally released acetyl groups in hemp are the most important source of acetic acid, whereas formic acid is mainly formed by degradation of polysaccharides and in much lesser degree from formyl groups. During autocatalyzed organosolv pulping of hemp core, bast and whole stem, equal amounts of formic acid (about 2.5% w/w at the end of the pulping processes) are released in the effluent. In contrast, the concentrations of acetic acid strongly differ according to the acetyl content of the raw materials.

Also relatively small amounts of glycolic acid, lactic acid, fumaric acid, levulinic acid, HMF and furfural are found in the effluents. Polysaccharides are probably the main sources of these compounds. The furans formed indicate degradation of hemicellulose and cellulose. Breakdown of lignin to short chain organic acids and furans has not been elucidated.

The acids and furans analyzed are quite stable in a mixture of ethanol/water (60 : 40 v/v) under the pulping conditions used.

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