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Gas chromatography–mass spectrometry method for the simultaneous determination of wood extractive compounds in quaking aspen

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Abstract

We have developed a rapid gas chromatography–mass spectrometry (GC–MS) method for the detailed compositional analysis of 70 underivatized wood extractive components present in quaking aspen (*Populus tremuloides* Michx.). Forty-four compounds were unequivocally identified by retention time and mass spectral comparison with standards. An additional 26 chromatographic peaks were assigned to broad chemical classes using retention time and mass spectra features. The results were compared to the respective *tert*.-butyldimethylsilyl derivatized wood extractives profile, and it was determined that derivatization was unnecessary for the GC–MS analysis of the target compounds.

Keywords: *Populus tremuloides*; Wood; Derivatization, GC; Steryl esters; Sterols; Phenolic compounds; Fatty acids; Flavonoids; Triterpenes; Lipids; Terpenes

1. Introduction

Wood extractives are defined as compounds that can be extracted from wood by means of both polar and non-polar solvents [1]. They include a large variety of compounds grouped into general classes including terpenoids, fats and waxes and their components, and phenolics, as well as other minor components not formally classified into these three groups, such as carbohydrates, peptides and inorganic compounds. In the processing of aspen (*Populus tremuloides* Michx.) for pulp and paper, resinous extractives, including fatty acids, waxes, fatty alcohols, triterpenes/sterols, steryl esters and glycerides [2], tend to form pitch deposits on equipment surfaces in mills, or become imbedded in the final product along with accumulated dirt particles [3,4]. Furthermore, extractive compounds such as certain phenolics, sterols and fatty acids may be responsible for acute and chronic toxicities observed in aspen woodpile leachates and pulp/paper mill effluents [5,6]. Thus, rapid procedures for the identification and quantification of problematic wood extractive components in commercially valuable

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pulpwood species such as aspen are desirable. Such procedures could be used to (a) track pitch-generating extractives in chip seasoning and in pulp processing, (b) monitor toxic or bioactive compounds in process effluents and the environment, and (c) select from standing trees superior clones, families or hybrids with appropriate extractives contents.

Gas chromatography (GC) has been the traditional technique for the analysis and isolation of the more volatile terpenoids [7]. Improved technology in GC fused-silica capillary columns has enabled the use of much shorter (<10 m) columns allowing for the analysis of higher-molecular-mass and less volatile components such as triglycerides and steryl esters [2]. Several studies on the characterization of wood extractives show that capillary GC is by far the most convenient and comprehensive technique available for separating out the individual components in wood extractives for subsequent identification [2,4,8–10].

Derivatization of wood extractives for GC analysis is usually carried out to improve compound resolution, and can be achieved by methylation, acetylation or silylation [11]. However, derivatization procedures increase sample preparation time, reagent costs, and introduce a degree of error into any quantitative analysis. Gas chromatographic analysis of lipophilic wood extractives such as steryl esters, triglycerides and waxes has been performed without any prior derivatization due to the lack of free derivatization sites on these lipids [12]. To our knowledge, no published methods exist which allow convenient simultaneous analysis of several classes of compounds implicated in both pitch and toxicity issues for commercially important hardwoods such as aspen. In this work, we present a gas chromatography–mass spectrometry (GC–MS) method for the detailed analysis of important wood extractives in aspen that requires no prior derivatization or fractionation.

2. Experimental

2.1. Sample preparation

Mature aspen trees located in northeastern British Columbia, Canada were non-destructively sampled by the removal of a 10 mm increment core at breast height (~1.3 m from base) across the entire diameter of each stem. Each sampled core was freeze–dried for 24 h, ground into wood meal through a 2 mm screen, and 5 g sub-samples were weighed into Soxhlet thimbles and Soxhlet extracted for exactly 8 h at a rate of 4–5 cycles/h using 200 ml of certified ACS-grade acetone. The acetone extracts (≈200 ml) were then concentrated to ≈10 ml on a rotary evaporator (<30°C water bath), passed through pasteur pipettes packed with glass wool and transferred to pre-tared 16-ml screw cap vials. Each extract residue (≈10 ml) was then further concentrated to <1 ml by passing a stream of nitrogen into the vial while heating (<40°C), freeze–dried for exactly 24 h (~$2\cdot10^{-1}$ mbar, −45°C) and weighed (±0.01 mg). Sub-samples of the freeze–dried (fd) extract material were then suspended at 5.00 mg/ml in an internal standard solution of 0.250 mg/ml heptadecanoic acid (99%; Sigma, St. Louis, MO, USA) in certified ACS-grade acetone. Finally, these solutions were sonicated for 60 min.

2.2. Preparation of tert.-butyldimethylsilyl (TBDMS) derivatives

A 200-μl volume of stock sample solution (containing 1 mg of extractives) was added to a 2.0-ml amber auto-sampler vial, and the solution was dried with a stream of nitrogen while heating (<40°C). The residue was then resuspended in 100 μl of HPLC-grade acetonitrile and 100 μl of *N*-methyl-*N*-(*tert*.-butyldimethylsilyl)trifluoroacetamide (silylation reagent; Sigma) was added. The reaction mixture was capped for 20 min at room temperature, then diluted with 1 ml of HPLC/GC–MS-grade dichloromethane.

2.3. Gas chromatography–mass spectrometry

A Varian 3800 gas chromatograph coupled to a Varian Saturn 2000 ion trap mass spectrometer (Walnut Creek, CA, USA) was used for the analysis. GC was carried out on a DB-XLBitd capillary column (10 m×0.25 mm, 0.25 μm film; J&W Scientific, Folsom, CA, USA) with a deactivated 1078 fritted splitter inlet sleeve (Restek, Brockville, Canada) and a Varian 1079 injector. The MS electron multiplier voltage was set at 2150 V and an ioniza-

tion time of 25 000 μs was used, running in the electron impact (EI) mode, with transfer line, ion trap and manifold temperatures of 350°C, 250°C and 50°C, respectively. The data acquisition sampling frequency was 60 Hz for a scan range of 50 to 650 m/z.

Samples were injected via a Varian 8200 autosampler fitted with a 10-μl syringe. For the underivatized samples, 1 μl was injected with a split of 1:60 and an injector temperature of 320°C. A continuous flow-rate of 1.6 ml/min of chromatographic-grade helium was used. The temperature profile started at 50°C for 3 min, followed by a 10°C/min ramp to 340°C, which was held for 36 min, then finally a 10°C/min ramp to 360°C, the thermal maximum of the column. For the TBDMS derivatives, 2 μl was injected with a split of 1:20 and an injector temperature of 300°C. A continuous flow-rate of 1.6 ml/min was used with a temperature profile starting at 50°C for 3 min followed by a 6°C/min ramp to 340°C, which was held for 20 min, then finally a 10°C/min ramp to 360°C.

2.4. Identification and quantification of compounds

Eluted compounds were identified by unequivocal matches with mass spectra of the 1998 National Institute of Standards and Technology (NIST '98) mass spectral library. Some cases were confirmed with standards (Sigma). For some chromatographic peaks, complete identification was not possible from the mass spectra obtained in the total extractives chromatogram. Such peaks were classified into the chemical classes, fatty acids, flavonoids, sterols/triterpenes, diglycerides, waxes, steryl/triterpene esters and triglycerides using broad mass spectral features along with retention time similarities within each class. Finally, the steryl/triterpene esters that eluted in the total extractives GC–MS profile were initially identified using additional chromatographic techniques prior to GC–MS analysis, as outlined in Serreqi et al. [13].

Once identified or classified, each compound was then quantified using a quantitation ion based peak area [$A_{\mathrm{QI}(X)}$] that was directly proportional to the concentration (C_X) for that compound in the sample, as seen in Eq. (1):

$$C_X = \left[A_{\mathrm{QI}(X)}/A_{\mathrm{QI(I.S.)}}\right] \cdot \left[\mathrm{QIR}_{(X)} \cdot C_{\mathrm{I.S.}}\right] / \left[F_X \cdot \mathrm{QIR}_{(\mathrm{I.S.})}\right] \quad (1)$$

where F_X = the response factor (RF) for a particular analyte (X) measured on a particular detector relative to the internal standard (I.S.); QIR (quantitation ion ratio) = the total ion intensity of the 50–650 m/z mass spectrum divided by the sum of intensities of the quantitation ion(s).

The QIR of a particular compound was calculated from the mass spectrum for that compound in the NIST library or from the mass spectrum of the standard compound. However, if the compound was not successfully identified by mass spectrometry, but only classified, an estimate of the QIR was obtained directly from the sample mass spectrum. In every case, A_{QI} needed to be converted to A_{RIC} since the reconstructed ion chromatogram (RIC) area response factors may be very similar for chemically similar groups. Chemical class specific RIC response factors were calculated relative to heptadecanoic acid using representative standard compounds (99%, Sigma, except stigmsta-5,22-dien-3β-ol which was 95%; Aldrich, Milwaukee, WI, USA) for each broad chemical class as illustrated in Table 1.

3. Results and discussion

3.1. GC–MS analysis of underivatized extractives

The underivatized acetone extractives of aspen were analyzed by GC–MS using a DB-XLB low polarity column. This column features excellent inertness for active compounds and a high temperature limit of 360°C making it an ideal column for the analysis of underivatized semi-volatile wood extractives. Furthermore, the separation of target compounds in aspen extractives was maximized by optimization of temperature programs and carrier gas flow conditions. The optimized GC–MS trace is presented in Fig. 1 with the major peaks and compound classes highlighted. A more complete list of identified compounds is given in Table 2, with the structural formulas for trivial compounds presented in Fig. 2. The separation of the high-boiling-point lipophilic constituents such as steryl/triterpene esters

Table 1
Chemical class-specific response factors used for quantitation of components in aspen extractives

Chemical class	Compound used for RF determination	RF
Monoaryl phenolics	Benzoic acid	2.3±0.1
Fatty acids/alcohols	Heptadecanoic acid	1[a]
Flavonoids	2-Phenyl-4H-1-benzopyran-4-one	1.65±0.04
Diglycerides	Glycerol dioctadecanoate	0.6±0.1
Sterols/triterpenes	Stigmsta-5,22-dien-3β-ol	0.98±0.09
Waxes	Octadecyl octadecanoate	3.6±0.2
Steryl/triterpene esters	Cholest-5-en-3β-yl octadecanoate	4.0±0.3
Triglycerides	Glycerol trioctadecanoate	4.2±0.6

[a] Internal standard set at 1; Note: error presented with average RF represents the 95% confidence interval for $n=7$.

and triglycerides shown in Fig. 1 was far superior to that seen in several studies utilizing GC for similar compounds [8,9,14].

Forty-four compounds have been identified and an additional 26 chromatographic peaks were classified in the single chromatogram. These latter components were classified into fatty acid, flavonoid, sterol/triterpene, diglyceride, wax, steryl/triterpene ester and triglyceride classes based on general mass spectral features characteristic of each particular chemical class. The broad mass spectral patterns, were complementary to retention time grouping, which if used alone, would have undoubtedly lead to false classifications due to the overlapping of retention zones

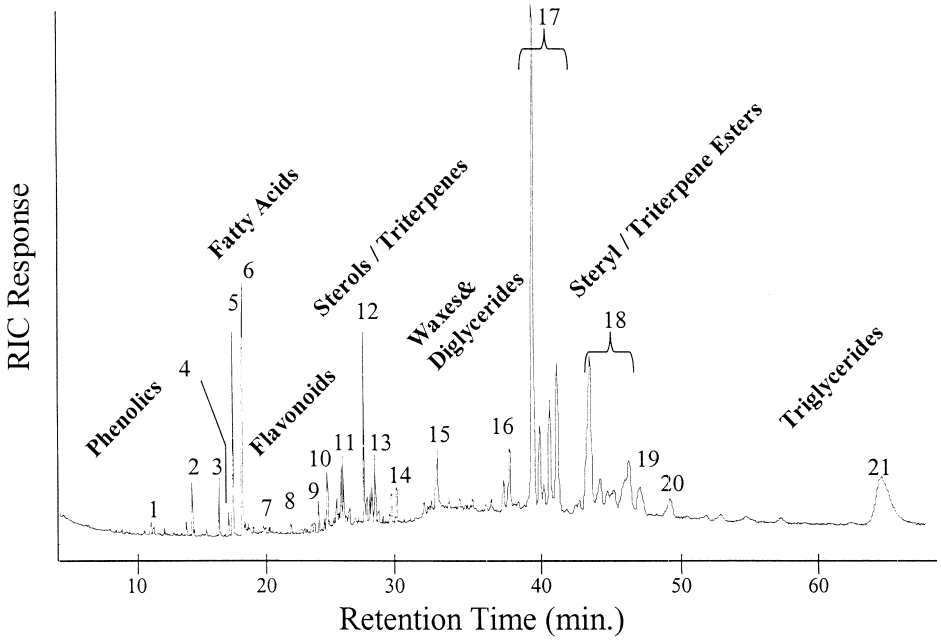

Fig. 1. Typical profile for underivatized aspen wood acetone extractives by GC–MS (1 μl at 5.00 mg/ml). 1=4-Hydroxybenzoic acid; 2=3-(4-hydroxy-3-methoxyphenyl)-2-propen-1-ol; 3=hexadecanoic acid; 4=4-(3-hydroxy-1-propenyl)-2,6-dimethoxyphenol; 5=internal standard (heptadecanoic acid); 6=(*Z*,*Z*)-9,12-octadecadienoic acid; 7=eicosanoic acid; 8=docosanoic acid; 9=4′,5-dihydroxy-7-methoxyflavanone; 10=4′,5,7-trihydroxyflavanone; 11=stigmast-5-en-3β-yl acetate; 12=stigmast-5-en-3β-ol; 13=24-methylcycloart-24,(24[1])-en-3β-ol; 14=unidentified sterols/triterpenes; 15=various waxes; 16=unidentified steryl/triterpene esters; 17=hexadecanoate (C16:0) steryl/triterpene esters; 18=octadecanoate (C18:0) steryl/triterpene esters; 19=unidentified triglyceride; 20=tirucalla-7,24-diene-3β-yl eicosanoate; 21=(9*Z*,12*Z*)-glycerol-tri-9,12-octadecadienoate.

Table 2
List of all compounds identified and quantified in a single GC–MS analysis for aspen 10 mm increment core extractives

Compound	Retention time (min)	Identification method/MS fragments with relative intensities	RF	Amount (mg/kg fd wood)[a]
Monoaryl phenolics				
Benzoic acid	6.87	AS/105(100), 122(79)	2.3	25
1-Ethyl-4-hydroxybenzene	6.92	NIST/107(100), 122(59)	2.3	7.3
3-(2-Hydroxyphenyl)-2-propenoic acid	7.82	NIST/91(100), 120(89)	2.3	7.6
2-Hydroxybenzyl alcohol	8.53	NIST/78(100), 106(64)	2.3	6.2
4-Hydroxy-2-methylacetophenone	9.27	NIST/150(93), 135(100), 107(73)	2.3	3.6
4-Hydroxybenzoic acid	11.88	NIST/121(100), 138(73)	2.3	44
3-(4-Hydroxyphenyl) propanoic acid	13.33	NIST/107(100), 166(40)	2.3	9.4
3-(4-Hydroxy-3-methoxyphenyl)-2-propen-1-ol	14.62	NIST/180(78), 137(100)	2.3	$1.1 \cdot 10^2$
3-(4-Hydroxy-3-methoxyphenyl)-2-propenal	14.75	MSF/178(100), 161(39), 147(53), 135(67), 107(62), 77(61)	2.3	14
3-(4-Hydroxyphenyl)-2-propenoic acid	15.08	NIST/164(100), 147(61)	2.3	27
4-(4-Hydroxy-3-methoxyphenyl)-2-propenoic acid	15.95	NIST/194(100), 133(38)	2.3	3.8
4-(3-Hydroxy-1-propenyl)-2,6-dimethoxyphenol	17.25	MSF/210(100), 193(18), 182(41), 167(84), 154(27), 77(23)	2.3	68
3-(4-Hydroxy-3,5-dimethoxyphenyl)-2-propen-1-al	17.32	NIST/208(100), 165(67)	2.3	15
Fatty acids/alcohols				
9-Oxononanoic acid	11.62	NIST/155(8), 144(6), 83(100)	1	$1.9 \cdot 10^2$
Hexadecanoic acid	16.60	AS/256(17), 129(89), 73(100)	1	$4.2 \cdot 10^2$
(Z,Z)-9,12-Octadecadienoic acid	18.25	AS/280(3), 109(25), 67(100)	1	$1.5 \cdot 10^3$
Octadecanoic acid	18.47	AS/284(14), 129(77), 55(100)	1	70
Eicosanoic acid	20.20	AS/312(28), 269(34), 129(81), 57(100)	1	52
Docosanoic acid	21.82	AS/340(35), 297(35), 87(100)	1	71
Tetracosanoic acid	23.35	MSF/368(33), 325(22), 269(25), 185(35), 129(71), 55(100)	1	84
1-Docosanol	24.23	NIST/111(53), 97(100), 83(79)	1	70
Flavonoids				
4′,5-Dihydroxy-7-methoxyflavanone	23.83	NIST/285(100), 167(79)	1.65	$2.6 \cdot 10^2$
4′,5,7-Trihydroxyflavanone	24.48	AS/271(100), 153(75)	1.65	$3.7 \cdot 10^2$
3,5,7-Trihydroxy-4′-methoxyflavone	25.69	NIST/300(100), 257(30)	1.65	61
3,4′,5,7-Tetrahydroxyflavone	26.42	NIST/286(100), 229(14)	1.65	60
Sterols/triterpene alcohols				
Stigmast-5-en-3β-ol	27.10	AS/397(76), 213(100)	0.98	$1.4 \cdot 10^3$
Oleanen-3β-ol	27.32	NIST/218(33), 203(100)	0.98	88
Tirucalla-7,24-diene-3β-ol	27.35	Other[b]/412(24), 394(100), 241(38)	0.98	$1.2 \cdot 10^2$
Cycloart-24-en-3β-ol	27.58	NIST/426(17), 408(26), 394(100)	0.98	$1.4 \cdot 10^2$
12-Ursen-3β-ol	27.65	AS/426(8), 218(100), 203(80)	0.98	$1.7 \cdot 10^2$
24-Methylcycloart-24,(24^1)-en-3β-ol	27.90	NIST/422(82), 408(89), 407(100)	0.98	$1.9 \cdot 10^2$
Citrostadienol	27.97	NIST/328(13), 285(100)	0.98	68
Steryl/triterpene esters				
Stigmast-5-en-3β-yl acetate	25.60	NIST/396(100), 382(36)	4.0	$2.7 \cdot 10^2$
Tirucalla-7,24-diene-3β-yl hexadecanoate	39.55	Ref. [13]/394(100), 241(22)	4.0	$1.7 \cdot 10^3$
12-Oleanen-3β-yl hexadecanoate	40.08	Ref. [13]/218(40), 203(100)	4.0	$3.4 \cdot 10^2$
20(29)-Lupen-3β-yl hexadecanoate	40.82	Ref. [13]/410(26), 203(100)	4.0	$4.4 \cdot 10^2$
12-Ursen-3β-yl hexadecanoate	41.40	Ref. [13]/218(100), 203(62)	4.0	$7.4 \cdot 10^2$
Tirucalla-7,24-diene-3β-yl octadecanoate	43.70	Ref. [13]/394(100), 241(18), 109(25)	4.0	$7.5 \cdot 10^2$
12-Oleanen-3β-yl octadecanoate	44.45	Ref. [13]/218(38), 203(100)	4.0	88
20(29)-Lupen-3β-yl octadecanoate	45.62	Ref. [13]/410(20), 392(19), 203(100)	4.0	34
12-Ursen-3β-yl octadecanoate	46.20	Ref. [13]/218(100), 203(82)	4.0	$1.6 \cdot 10^2$
Tirucalla-7,24-diene-3β-yl eicosanoate	49.50	Ref. [13]/218(100), 203(82)	4.0	$1.4 \cdot 10^2$
Lipids				
α-Tocopherol	25.82	NIST/431(100), 165(77)	0.98	37
(9Z,12Z)-Glycerol tri-9,12-octadecadienoate	65.07	AS/600(100), 340(32)	4.2	$7.7 \cdot 10^2$

[a] Average of 50 sampled 10 mm increment cores.

[b] Mass spectral match with sterol derived from the base hydrolysate of corresponding steryl ester identified in Serreqi et al. [13]; AS = authentic standard; MSF = mass spectral fragmentography; NIST = National Institute of Standards and Technology (mass spectral library).

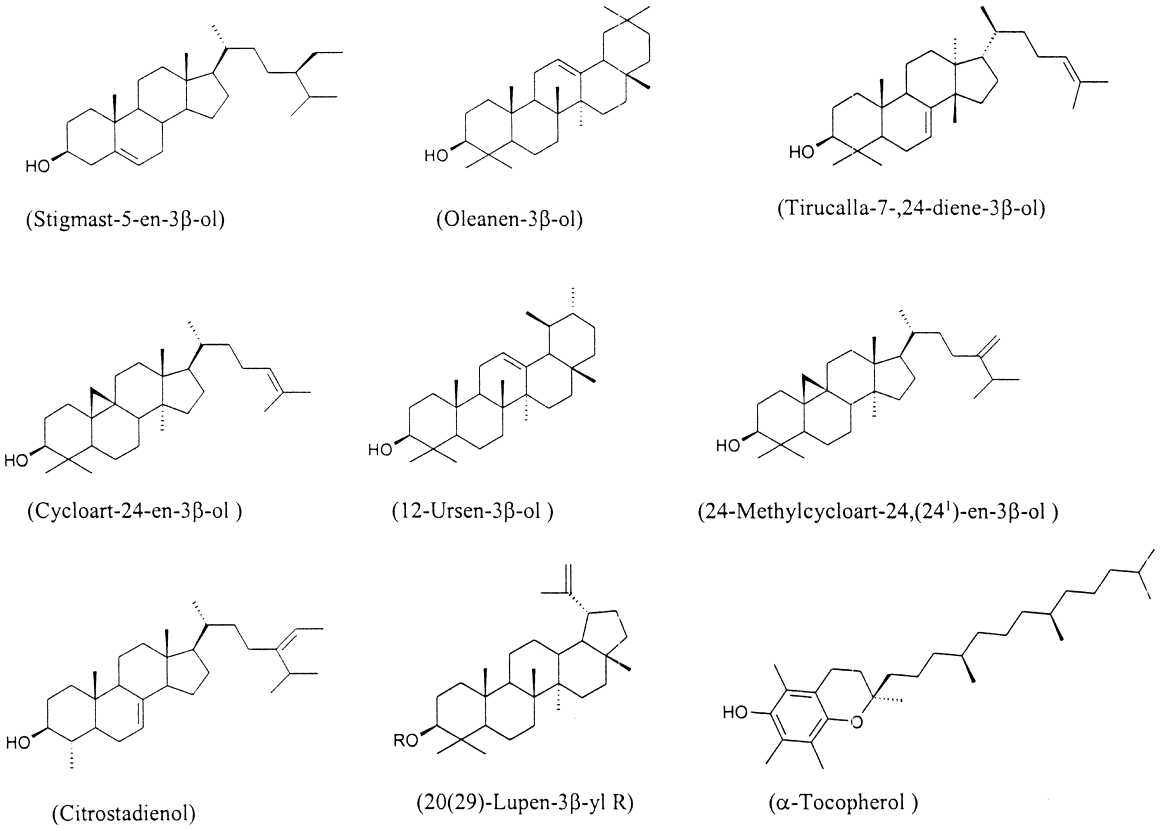

Fig. 2. Structural formulas for compounds with trivial names listed in Table 1.

shown for several adjacent chemical classes in Table 2 (see retention times). Ultimately, those compounds implicated in pitch formation during the pulping of aspen (i.e., fatty acids, sterols/triterpenes, steryl esters, waxes and triglycerides) are particularly well resolved and represented in quantifiable amounts.

Gutiérrez et al. [12] determined the effect of column length on the resolution and detection of eucalyptus extractives by GC. They concluded that one must seek an optimum of maximized resolution (brought about by an increase in column length) versus minimized exposure of the analyte to high temperatures (brought about by a decrease in column length). The GC method developed in this study utilizes a moderate temperature ramp of 10°C/min with a relatively short column (10 m), together with a long temperature hold at 340°C to allow for the elution of high-molecular-mass and boiling point compounds such as steryl/triterpene esters and triglycerides. This method was sufficient to provide high resolution without severe decomposition of thermally labile components near the end of the run. Also, to ensure that no material eluted after 70 min, an analysis using a longer final temperature hold at 340°C was performed on a few samples. Additionally, to assess the reproducibility of analysis by the GC–MS method, a standard solution containing 0.25 mg/ml of heptadecanoic acid and 0.25 mg/ml cholest-5-en-3β-yl hexadecanoate in acetone was injected several times prior to sample analysis. The results of nine consecutive trials indicated that the instrumentation error was within 10%.

As a faster (<70 min) GC method may be desirable for rapid and routine monitoring of various deresination treatments on aspen wood chips or logs, including biological treatments (i.e., Cartapip 97 developed by Blanchette et al. [15]) and seasoning, GC–MS runs of underivatized extractives were

performed using double the original temperature ramp rates. Although the resolution of low-molecular-mass components (i.e., phenolics, fatty acids and sterols/triterpenes) was reduced, the main pitch-forming compounds in aspen, the steryl esters and waxes, were still well resolved by this fast method in under 30 min. However, the triglyceride, (9*Z*,12*Z*)-glycerol tri-9,12-octadecadienoate, was undetectable using this fast method.

3.2. Composition of aspen extractives

The most abundant material extracted from the cores was the steryl/triterpene esters and waxes at an average of over 5 g per kg of freeze–dried core material (Fig. 3). Also high in abundance were the sterols/triterpenes, glycerides, and fatty acids, occurring at an average of over 2 g/kg in aspen cores. Some unique compounds, not classified into any of the major compound classes, were found in this work. 9-Oxononanoic acid, not previously reported in the literature to occur in aspen, was found in significant amounts (189 mg/kg fd wood). Nugent et al. [16] reported that oxidized fatty acid material may be present in an unknown fraction (~0.2% of dry wood) of aspen wood acetone extractives. Thus, 9-oxononanoic acid may be the oxidized derivative of (*Z*,*Z*)-9,12-octadecadienoic acid, which was also found in large amounts in this study and has a reactive double bond on the 9-carbon. In addition, α-tocopherol (vitamin E), previously reported to occur in aspen bark [17], was found in relatively minor amounts (37 mg/kg) in every sample. α-Tocopherol is thought to prevent the autooxidation of unsaturated lipids in plants [18].

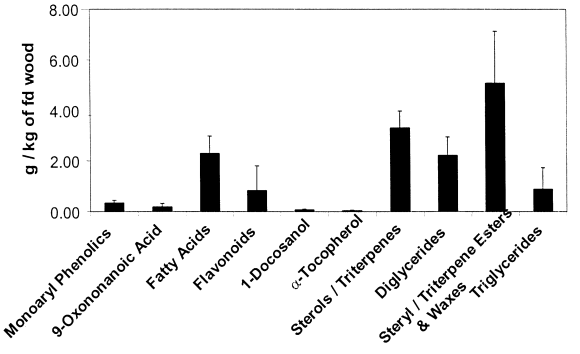

Fig. 3. The average amounts of several chemical classes found in the acetone extracts of aspen wood core (10 mm) samples based on GC–MS quantitation. (Note: error bars represent standard deviation).

3.3. Comparison to TBDMS-derivatized extractives

Components that may not be detected via the underivatized extractives method include compounds with a normal boiling point of greater than 320°C, the isothermal temperature of the gas chromatograph injector. Compounds such as sterols, triterpenes and some phenolics, which are of interest in this work, may be bound up as high-molecular-mass glycosides in aspen and other hardwoods [19], and hence may not be seen in the underivatized chromatogram. Fig. 4 illustrates the *tert*.-butyldimethylsilyl derivatized extractives chromatogram. TBDM-silylation was used since the resultant TBDMS derivatives retained most of the desirable features of trimethylsilyl ethers, yet they were much more stable under hydrolytic conditions [20]. This is important, considering that wood extractives are generally hydrophobic, and trace amounts of water are inevitably present during the silylation.

A comprehensive evaluation of all the detected components in the derivatized chromatogram revealed that, with the exception of the monosaccharides (peaks 2 and 5 in Fig. 4) there were no newly identified components as a result of the silylation, which are of relevance to most pitch and toxicity studies. Thus, we concluded that derivatization is an unnecessary extra step in the preparation of samples for the analysis of extractives in aspen.

4. Conclusions

Aspen wood extractives contain a wide range of compound classes which does not necessarily lend itself well to single analysis. However, the GC–MS method developed allowed for the accurate quantification and characterization of up to 70 components in over eight chemical classes in aspen wood acetone extracts, with minimal sample preparation. Forty-four compounds were identified, and 26 chromatographic peaks were classified (e.g., fatty acids, flavonoids, sterols/triterpenes, diglycerides). The

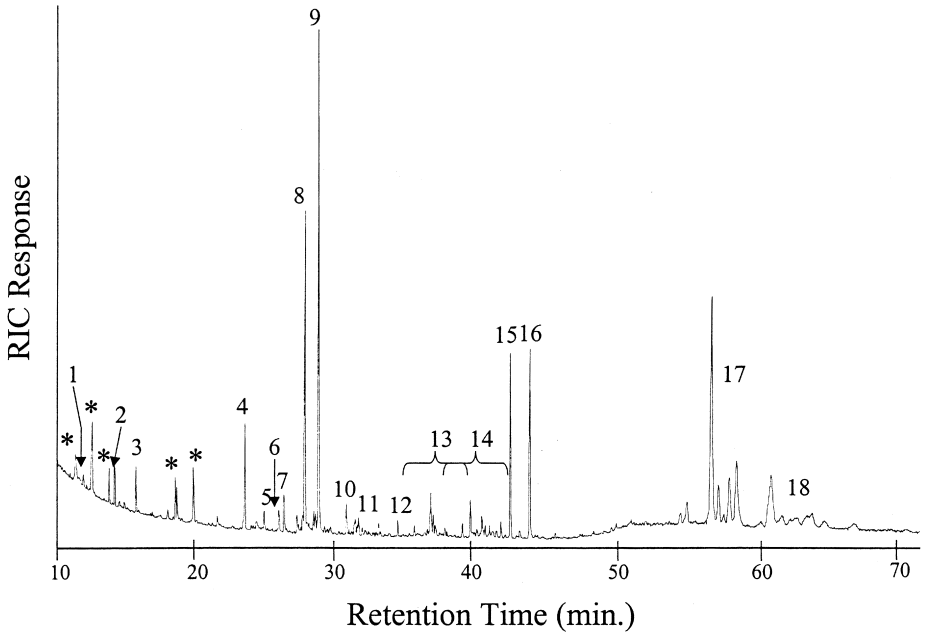

Fig. 4. Typical profile for TBDMS-derivatized aspen wood acetone extractives by GC–MS [2 μl at approx. 0.83 mg/ml (depends on derivatization yield)]. *Silylation contaminants (seen in blank); 1=TBDMS–benzoic acid; 2=monosaccharide; 3=unknown; 4=TBDMS–4-hydroxybenzoic acid; 5=monosaccharide; 6=TBDMS–1,9-nonanedionic acid; 7=TBDMS–hexadecanoic acid; 8=internal standard; 9=TBDMS–(*Z*,*Z*)-9,12-octadecadienoic acid; 10=unknown; 11=TBDMS–eicosanoic acid; 12=TBDMS–docosanoic acid; 13=TBDMS–C23:0–C26:0 fatty acids; 14=various underivatized and some TBDMS-derivatized sterols/triterpenes; 15=TBDMS–4′,5,7-trihydroxyflavanone; 16=TBDMS–4′,5-dihydroxy-7-methoxyflavanone; 17=underivatized hexadecanoate steryl/triterpene esters; 18=underivatized octadecanoate steryl/triterpene esters.

total extractives profile by GC–MS accounted for an average of 56% of the mass of total acetone extractives in aspen, and, most importantly, all the components that were targeted in this study were quantifiable in a single chromatogram. The method would be useful for monitoring both pitch-causing compounds and environmentally problematic components to improve process efficiency, product quality and environmental monitoring and protection in pulp mills.

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References

[1] D. Fengel, G. Wegener, in: Wood Chemistry, Ultrastructure, Reactions, Walter de Gruyter, Berlin, 1983, p. 132.
[2] B.B. Sitholé, J.L. Sullivan, L.H. Allen, Holzforschung 46 (1992) 409.
[3] L.H. Allen, Pulp Paper Can. 89 (1988) 87.
[4] B.B. Sitholé, L.H. Allen, in: E.L. Back, L.H. Allen (Eds.), Pitch Control, Wood Resin and Deresination, Tappi Press, Atlanta, GA, 2000, p. 289, Chapters 12–13.
[5] B.R. Taylor, J.S. Goudey, N.B. Carmichael, Environ. Toxicol. Chem. 15 (1996) 150.
[6] T.R. Zacharewski, K. Berhane, B.E. Gillesby, B.K. Burnison, Environ. Sci. Technol. 29 (1995) 2140.
[7] P.M. Dey, J.B. Harborne (Series Eds.), Methods in Plant Biochemistry, Vol. 7, Academic Press, San Diego, CA, 1991, pp. 1, 331, 369.
[8] J. Del Río, A. Gutiérrez, F.J. González-Vila, F. Martín, J. Romero, J. Chromatogr. A 823 (1998) 457.
[9] F. Orså, B. Holmbom, J. Pulp Paper Sci. 20 (1994) 361.
[10] P. Charlet, G. Lenone, B. Joseleau, P. Chareyre, in: Proceedings of the 9th International Symposium on Wood and Pulp Chemistry, Montreal, June 1997, p. 15-1.

[11] J. Peng, R. Leone, A.N. Serreqi, C. Breuil, Tappi J. 82 (1999) 204.
[12] A. Gutiérrez, J.C. del Río, F.J. González-Vila, F. Martín, J. Chromatogr. A 823 (1998) 449.
[13] A.N. Serreqi, R. Leone, L.F. Del Rio, S. Mei, M. Fernandez, C. Breuil, J. Am. Oil Chem. Soc. 77 (2000) 413.
[14] A.F.A. Wallis, R.H. Wearne, Appita J. 52 (1999) 295.
[15] R.A. Blanchette, R.L. Farrell, Y. Hader, J.E. Merritt, R.A. Snyder, P.A. Wendler, Eur. Pat. 0387, 187, AZ (1991).
[16] H.M. Nugent, L.H. Allen, H.I. Bolker, Trans. Tech. Sect., CPPA 3 (1977) 103.
[17] V.B. Nekrasova, Lesokhim. Podsochka 6 (1982), abstract only – translated into English.
[18] L.P. Miller (Ed.), Phytochemistry, Inorganic Elements and Special Groups of Chemicals, Vol. III, Van Nostrand Reinhold, New York, 1973, p. 195;
L.P. Miller (Ed.), Phytochemistry, Inorganic Elements and Special Groups of Chemicals, Vol. III, Van Nostrand Reinhold, New York, 1973, p. 327.
[19] R.G. Jonasson, B. Fuhr, B.B. Sitholé, L.H. Allen, in: Proceedings of the 9th International Symposium on Wood and Pulp Chemistry, Montreal, June 1997, p. 43-1.
[20] M.A. Quilliam, K.K. Ogilvie, K.L. Sadana, J.B. Westmore, J. Chromatogr. A 194 (1980) 379.