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Journal of Chromatography A, 807 (1998) 241–251

JOURNAL OF CHROMATOGRAPHY A

Analysis and characterization of naphthenic acids by gas chromatography–electron impact mass spectrometry of *tert.*-butyldimethylsilyl derivatives

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Received 21 April 1997; received in revised form 27 January 1998; accepted 28 January 1998

Abstract

The renewed interest in naphthenic acid (NA) as a wood preservative has driven the need for analytical techniques to characterize commercial supplies of NA. The compositional heterogeneity of NA makes analytical characterization extremely difficult. Fluoride ion chemical ionization mass spectrometry (FI-MS) has proven to be an effective technique in NA characterization. However, FI-MS is very complicated and expensive to perform. In this paper, an alternative to the FI-MS technique is presented which offers similar results with a more widely available bench-top electron impact (EI) mass spectrometer. By derivatization of NA components to their *tert.*-butyldimethylsilyl analogs, the extent of molecular fragmentation is greatly decreased and strong base peaks representing the unfragmented NA constituents are obtained in the EI spectra. Molecular mass and tentative structures can be deduced based on sites of unsaturation.

Keywords: Fluoride ion chemical ionization mass spectrometry; Mass spectrometry; Detection, GC; Naphthenic acids; Carboxylic acids

1. Introduction

Naphthenic acid (NA), as commercially derived from crude oil, is a complex mixture of monocarboxylic acids containing one or more alkyl-substituted alicyclic rings (naphthenes), with lesser amounts of aliphatic carboxylic acids [1]. Little, or no, aromatic compounds are present in pure supplies. Naphthenic acids have long been important as biomarkers and geochemical indicators for petroleum operations [2]. The ability of NA to complex copper, forming copper naphthenate (CNA), has led to increased interest in this product as an effective and environmentally safe wood preservative [3]. Recently, however, the performance of CNA as a wood decay fungicide has been called into question due to the compositional variations among commercial supplies of NA [4]. Therefore, it is critical to develop methods to evaluate batches of NA from different sources.

The compositional heterogeneity of NA makes it difficult to characterize by commonly used chromatographic or spectroscopic techniques. Small amounts of thousands of types of alicyclic acids exist in any given NA supply. Further complicating the problem is the lack of consistency among synthetic

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0021-9673/98/$19.00
PII: S0021-9673(98)00085-5

naphthenic acid supplies. Synthetic products may contain substantial quantities of mineral and crude oils and fatty acids. Even supplies from a single manufacturer or feed stock are often significantly different in composition [4]. Even users of NA traditionally have relied on the amount of titratable acids, or acid number, in the supply as the sole indicator of product quality. As a result, some supplies, while having a correct acid number, contain only a small NA component.

Attempts to characterize NA by gas chromatography (GC) and GC–mass spectrometry (GC–MS) have been reported [4–6]. These GC techniques have proven useful for identifying the presence of some synthetic additives. Electron impact mass spectrometry (EI-MS) and chemical ionization mass spectrometry (CI-MS) provide some information about NA impurities, such as polycyclic aromatic hydrocarbons and phenolic compounds. However, due to the extensive fragmentation of aliphatic molecules in the mass spectrometer, limited molecular mass and structural information can be obtained about NA components. Nuclear magnetic resonance data has been used to determine CH_2/CH_3 ratios and the aromatic content of NA samples, but the technique cannot provide structural information on complex mixtures without preliminary sample fractionation [7].

Dzidic et al. [8] have developed a technique which utilizes negative chemical ionization with a nitrogen trifluoride reagent gas and mass spectrometric detection (FI-MS) to characterize NA. In FI-MS, a fluoride anion is generated from the NF_3 reagent gas which abstracts the acidic proton from the carboxylic acid to form hydrogen fluoride and a singly charged carboxylate ion ($RCOO^-$). This technique significantly reduces the complexity of mass spectrum because only a single ion is generated for each compound present. This ion represents the molecular mass of the acid, less one mass unit. Isomeric compounds, however, cannot be individually determined. Since negative chemical ionization techniques cannot be performed on many of the benchtop MS instruments commonly found in analytical laboratories, FI-MS has been used only in specialized laboratories.

This paper describes a simple alternative to FI-MS for the analysis of NA mixtures which can be performed on any bench-top GC–EI-MS system, precluding the need for special reagent gases or instrument hardware. The technique is based on the analysis of *tert.*-butyldimethylsilyl (t-BDMS) derivatives of NA. The t-BDMS derivative is obtained by reaction of *N*-methyl-*N*-(*tert.*-butyldimethylsilyl) trifluoroacetamide with the acidic proton on alcohols or carboxylic acids. For each proton exchanged, the t-BDMS group adds 114 to the molecular mass of the molecule. The derivative is very stable and has been used to enhance resolution in some difficult gas chromatographic analyses [9,10].

The t-BDMS derivative also affords a sensitivity enhancement in applications using EI-MS due to the prominence of the $[M-C_4H_9]^+$ base peaks [11,12]. Although molecular ions and secondary fragmentation ions are present, fragmentation is predominantly directed toward the $[M-C_4H_9]^+$ ion, corresponding to the $Si(CH_3)_2$ adduct of the parent compound. By derivatization of NA to its t-BDMS analog, it was possible to obtain molecular mass and structural information for NA components from the prominent $[M-C_4H_9]^+$ base peaks. Relative percentage composition of individual components was also determined for several NA supplies.

2. Experimental

2.1. Chemicals

Seven NA samples were obtained from several domestic and international suppliers (sources cannot be identified due to confidentiality requirements). *tert.*-Butyldimethylsilyl derivatizations were prepared with *N*-methyl-*N*-(*tert.*-butyldimethylsilyl) trifluoroacetamide which contained 1% t-BDMS-chloride (MTBSTFA, Regis, Morton Grove, IL, USA). Other chemicals were obtained from Aldrich (Milwaukee, WI, USA). Standard solutions of NA and model compounds were prepared in dichloromethane. No fractionation or cleanup procedures were performed on samples prior to analysis.

2.2. Derivatizations

Derivatizations were performed by adding 100 μl of the MTBSTFA reagent to 100 μl of a NA

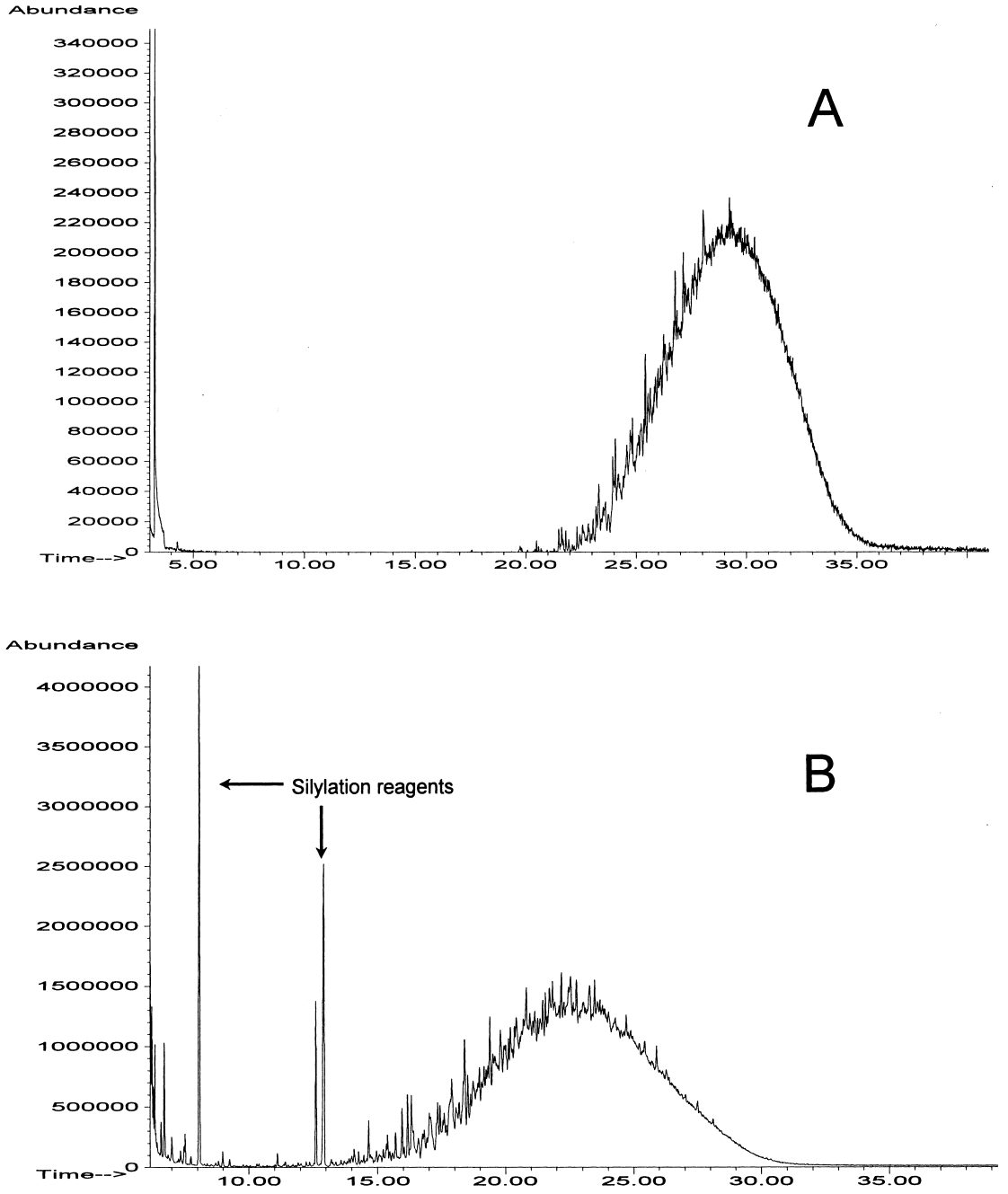

Fig. 1. Total ion chromatograms of a NA supply in the acid (A) and t-BDMS derivatized (B) form. GC temperature program modified from A to shorten analysis time. Time in min.

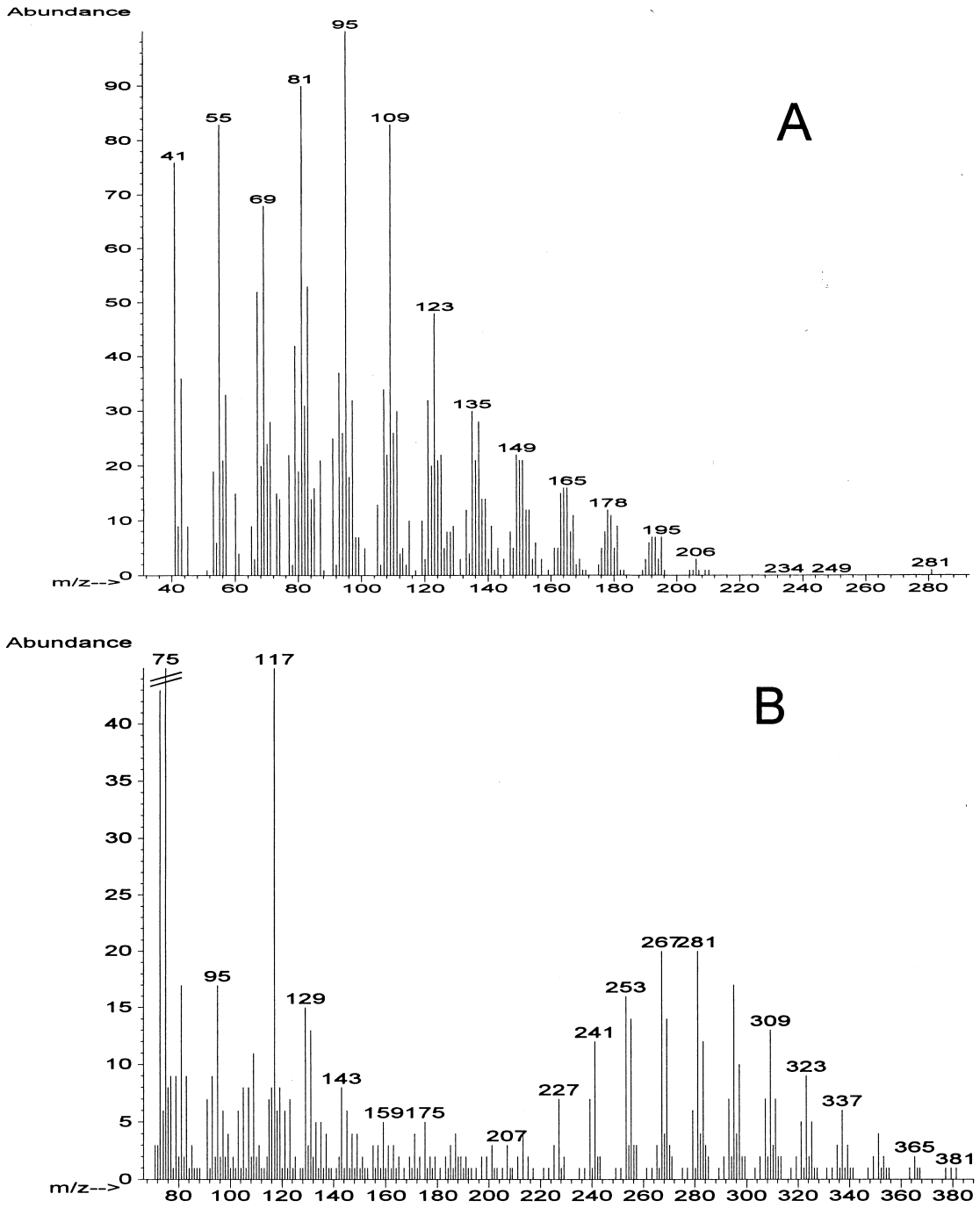

Fig. 2. Averaged spectra of the NA region of Fig. 1A,B. Ions of $m/z>200$ are scarce in underivatized NA (A) but are very abundant in t-BDMS derivatized NA (B).

standard (5 mg/ml) in methylene chloride. Samples were then heated to 60°C for 20 min to generate the t-BDMS derivative.

2.3. Instrumentation

Derivatized samples were analyzed by GC–EI-MS. A Hewlett-Packard 5890 Series II gas chromatograph equipped with a 30 M DB-5MS capillary column (J&W Scientific, Folsom, CA, USA) was used for sample introduction into the mass spectrometer. The GC run program began at an initial temperature of 100°C (60°C for underivatized samples) held for 3 min, ramped to a final temperature of 300°C at 8°C/min and held for 10 min. The GC–MS transfer line was held at 310°C. A Hewlett-Packard Model 5971 mass-selective detector (MSD) operated in the EI ionization mode (70 eV) was used to obtain all mass spectra. The instrument was autotuned using perfluorotributylamine and operated at 1.2 scans/s from m/z of 70–550. The MS remained off for the initial 6 min (3 min for underivatized samples) of the run to allow solvent and derivatization reagent to pass through the system.

3. Results and discussion

3.1. Comparison of free acids and t-BDMS derivatives

A chromatogram of an underivatized NA is provided in Fig. 1A. Commercial products consist of thousands of naphthenic acid congeners and fatty acids which all elute together, resulting in a large ''hump'' with very few resolvable peaks. Fig. 1B contains a sample chromatogram for a t-BDMS derivative of NA. The MTBSTFA reagent introduces additional peaks into each chromatogram which elute well before the derivatized NA components.

Fig. 2A,B show the averaged mass spectral data for the acid and t-BDMS forms of NA, respectively. Data was taken and averaged between ~1 min before and after the NA ''hump'' in the chromatogram. As can be seen, there is a significant increase in the number and intensity of mass fragments greater than ~200 m/z for the t-BDMS derivatives. Large m/z ions and molecular ions are very scarce in the EI spectrum of the underivatized NA.

Analysis of underivatized NA by GC–MS showed that phenolic compounds were present in some of the NA supplies obtained. Analysis of t-BDMS deriva-

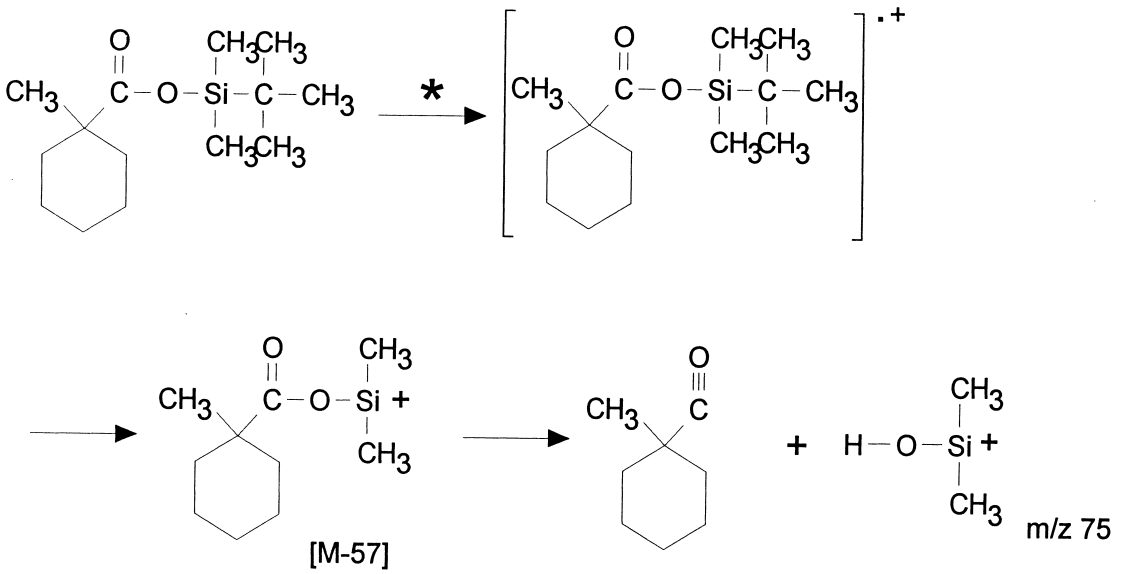

Fig. 3. Fragmentation pathways leading to the predominant ions in t-BDMS derivatives of naphthenic acids.

tives indicated that not all substituted phenols could be derivatized under the conditions described above. Some slightly hindered hydroxy functions have been reported to resist derivatization even after several hours [13]. However, phenolic compounds can easily be identified and quantified under normal EI-MS conditions without derivatization.

3.2. Spectral interpretation

Fig. 3 describes the fragmentation pathway of the t-BDMS derivatives which is consistent with that described elsewhere [14]. Several model compounds have been chosen to demonstrate spectral interpretation. Camphoric acid, 1-methyl-1-cyclohexanecarboxylic acid, and decanoic acid were derivatized to their t-BDMS analogs and analyzed. Fig. 4A–C show the predominance of the $[M-C_4H_9]^+$ base-peak ion for each compound, corresponding to the $[naphthenate+dimethylsilyl]^+$ ion. For all model compounds, this ion results from preferential cleavage of the *tert.*-butyl group. By subtraction of 57 (C_2H_5Si) from the $[M-C_4H_9]^+$ base peak, the exact molecular mass of each compound can be obtained.

Although dicarboxylic acids are not known to be present in NA to any great extent, camphoric acid was used to further demonstrate the stability of the $[M-C_4H_9]^+$ ion. This dicarboxylic acid undergoes derivatization at both carboxyl groups for a total molecular mass of 428. However, Fig. 4C shows that the most abundant fragment ion is the $[M-C_4H_9]^+$ molecule at 371 m/z. This is similar to the FI-MS technique in which only the one proton is abstracted from the dicarboxylic acid.

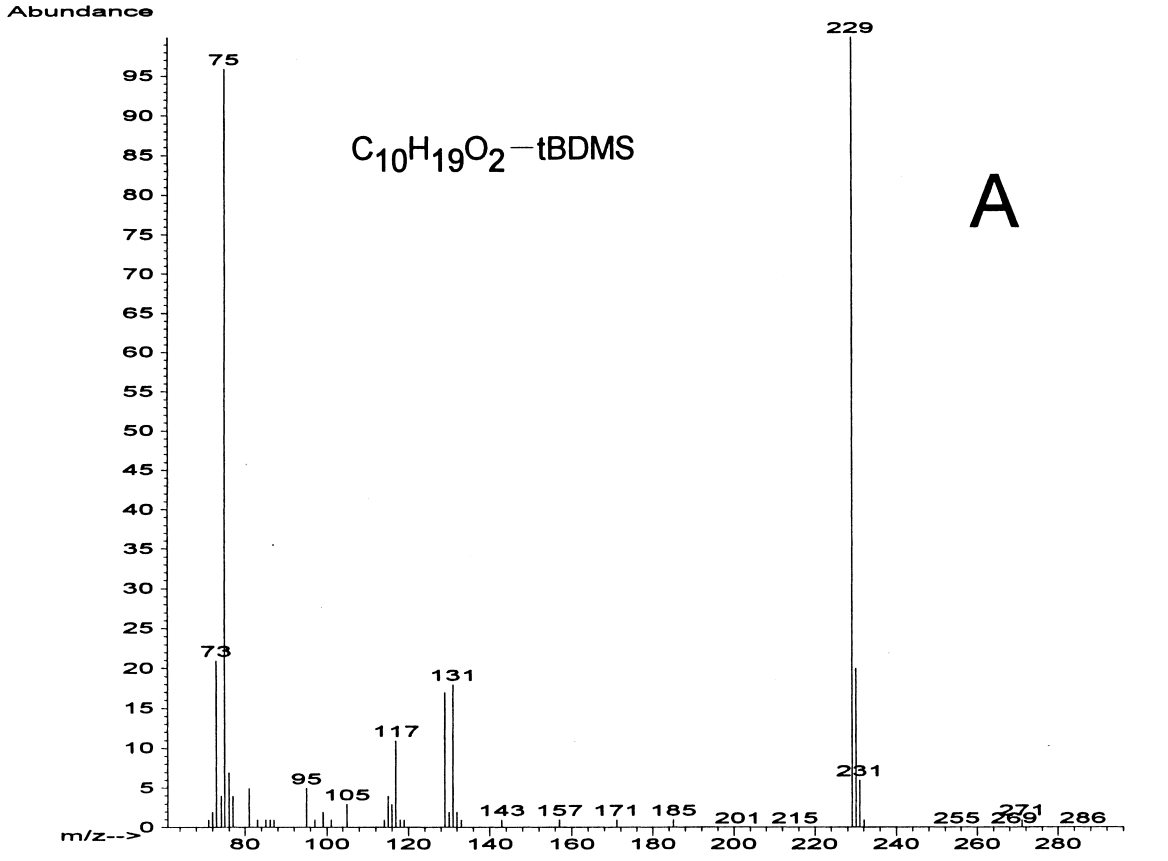

Fig. 4. Scans for t-BDMS derivatives of decanoic acid (M_r 172, A), 1-methyl-1-cyclohexanecarboxylic acid (M_r 142, B) and camphoric acid (M_r 200, C).

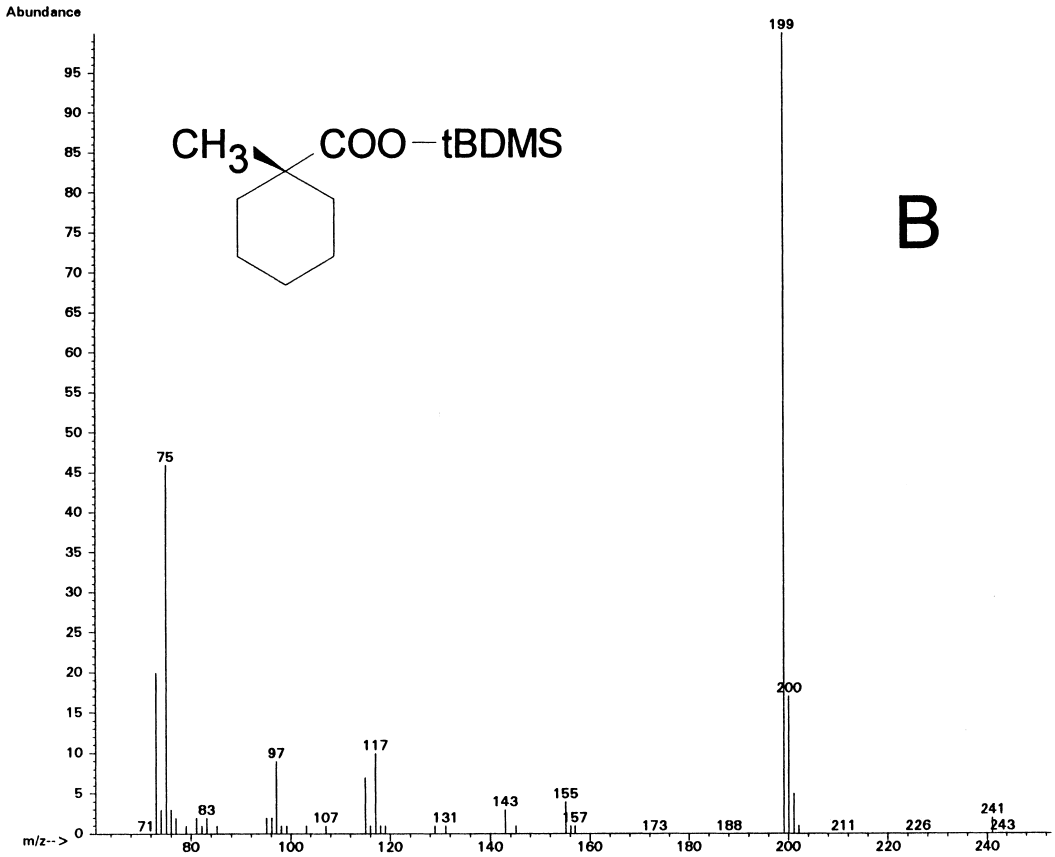

Fig. 4. (*continued*)

3.3. Structural information

The t-BDMS derivatives are very useful in determining structures of compounds eluting within the large, unresolved NA peak. For NA derivatives, it is assumed that the large ions (above m/z~200) represent the base peak ions for each individual compound. These ions, which represent the ion $[M-C_4H_9]^+$, can be used to calculate the molecular mass for each acid as previously described. From the molecular mass, a reasonable formula can be given based on the general form $C_nH_{2n-z}O_2$ for alicyclic monocarboxylic acids. In this formula, z represents two times the number of rings in the molecule. Therefore, structures with the correct molecular mass and number of rings can be posed for each $[M-C_4H_9]^+$ ion in the mass fragmentogram. Indeed, it is possible to catalog the base peaks expected for any compound fitting the definition of NA; this list is shown in Table 1. Note that $z=0$ corresponds to acyclic species and thus is not a NA. Fig. 5 shows structures posed for a single scan from the NA region in Fig. 1B.

Tables 2 and 3 indicate percentage composition of each compound found in two supplies of naphthenic acid determined by the relative abundance of each NA ion. Only the peaks at the characteristic NA retention time were integrated. It was assumed that all z homologs have equal ionization sensitivities. Unlike the NA product listed in Table 2, the product listed in Table 3 apparently contains little or no NA. Its bulk composition is straight chain fatty acid with $z=0$. The amount of titratable acids in each supply were similar, however. If this acid number was used

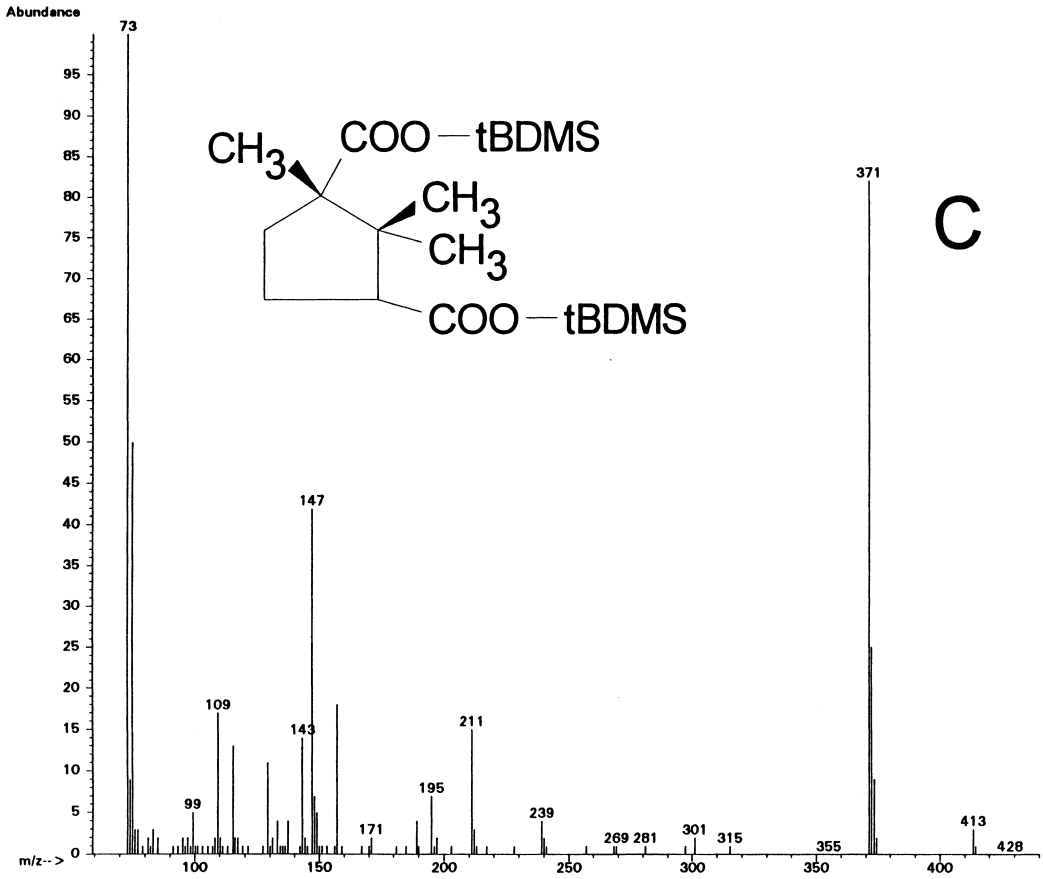

Fig. 4. (*continued*)

as the sole quality indicator, as is common in the forest products industry, the difference in NA composition would not be known.

The data and structural information available is very similar to that obtained by Dzidic et al. using the fluoride ionization technique. Both the FI-MS and EI-MS techniques described have similar limitations regarding isomeric compound identification and spectral complication due to the presence of hydroxy, dicarboxylic and olefinic acids. The main difference in data interpretation between the two techniques is that FI-MS generates only the $[M-1]^-$ ion and the EI-MS generates a predominant $[M-C_4H_9]^+$ ion, with a few less-abundant ions.

With the EI-MS technique, it is difficult to identify the $[M-C_4H_9]^+$ ions of $m/z<200$ due to the presence of other fragment ions. These would include all molecules with a carbon number of 8 or less. This is of minor consequence, since >97% of the acids in NA are of carbon number 9 or greater. It is also not possible to identify individual isomers with the same molecular formula. Interference from C_nH_{2n+2} saturated hydrocarbons at $m/z>200$ is unlikely due to the extensive molecular fragmentation of these molecules that occurs under EI conditions.

For detailed work with the EI-MS method, it is important to be aware of the 5% natural abundance of ^{30}Si. For refined NA, this would most influence the mole percent determinations of *z*-series molecules which follow the most abundant series. For the NA in Table 2 these would be *z* numbers 4 and 6.

Table 1
Expected [M−57] base peak ions for a homologous series of t-BDMS derivatized naphthenic acids

Carbon No.	z Number						
	0	2	4	6	8	10	12
6	173	171	169	167	165	163	161
7	187	185	183	181	179	177	175
8	201	199	197	195	193	191	189
9	215	213	211	209	207	205	203
10	229	227	225	223	221	219	217
11	243	241	239	237	235	233	231
12	257	255	253	251	249	247	245
13	271	269	267	265	263	261	259
14	285	283	281	279	277	275	273
15	299	297	295	293	291	289	287
16	313	311	309	307	305	303	301
17	327	325	323	321	319	317	315
18	341	339	337	335	333	331	329
19	355	353	351	349	347	345	343
20	369	367	365	363	361	359	357
21	383	381	379	377	375	373	371
22	397	395	393	391	389	387	385
23	411	409	407	405	403	401	399
24	425	423	421	419	417	415	413
25	439	437	435	433	431	429	427
26	453	451	449	447	445	443	441
27	467	465	463	461	459	457	455
28	481	479	477	475	473	471	469
29	495	493	491	489	487	485	483
30	509	507	505	503	501	499	497
31	523	521	519	517	515	513	511
32	537	535	533	531	529	527	525
33	551[a]	549	547	545	543	541	539

[a] NA – The quadrapole mass-selective detector has an upper mass range limit of m/z 550. This ion will not be detected.

However, for applications where analyses are being conducted solely for quality control purposes, isotope correction is not a large concern.

4. Conclusion

The extensive molecular fragmentation of alicyclic NA compounds in electron impact mass spectrometers makes it difficult to obtain molecular mass and structural information from the complex spectra by standard methods. FI-MS has greatly aided NA characterization, but the technique itself is very complicated and expensive to perform. In this report, an EI-MS technique has been presented which generates data similar to FI-MS and is easily performed on any bench-top GC–MS system under normal operating conditions. The purity of NA supplies can be easily assessed and the percentage composition of specific NA components determined. The technique should prove most useful for maintaining a quality control history of commercial supplies of NA, such as in the wood preservation industry.

Acknowledgements

The research presented here was funded by the Federal Highway Administration and the United

Table 2
Percent determination of NA components by z and carbon numbers (pure supply)

Carbon No.	z Number							Percent by carbon number
	0	2	4	6	8	10	12	
10	0.85	2.5	0.92	0.18	0.20	0.16	0.22	5.05
11	0.88	4.2	2.6	0.23	0.18	0.11	0.12	8.37
12	0.90	4.9	5.5	0.46	0.25	0.14	0.05	12.25
13	0.86	4.9	7.0	1.2	0.35	0.22	0.04	14.49
14	0.82	4.2	7.0	2.0	0.42	0.29	0.05	14.73
15	0.72	3.4	5.9	2.5	0.55	0.32	0.11	13.52
16	0.56	2.5	4.6	2.4	0.62	0.31	0.15	11.13
17	0.39	1.7	3.0	1.7	0.56	0.28	0.17	7.91
18	0.28	1.2	2.0	1.1	0.42	0.23	0.15	5.40
19	0.23	0.80	1.3	0.68	0.27	0.17	0.11	3.53
20	0.15	0.51	0.74	0.38	0.16	0.11	0.07	2.11
21	0.05	0.29	0.39	0.18	0.07	0.04	0.01	1.03
22	0.01	0.14	0.17	0.06	0.01	0.00	0.00	0.38
23	0.00	0.05	0.05	0.00	–	0.00	–	0.11
Percent by z number	6.71	31.33	41.18	13.05	4.07	2.40	1.25	100.00

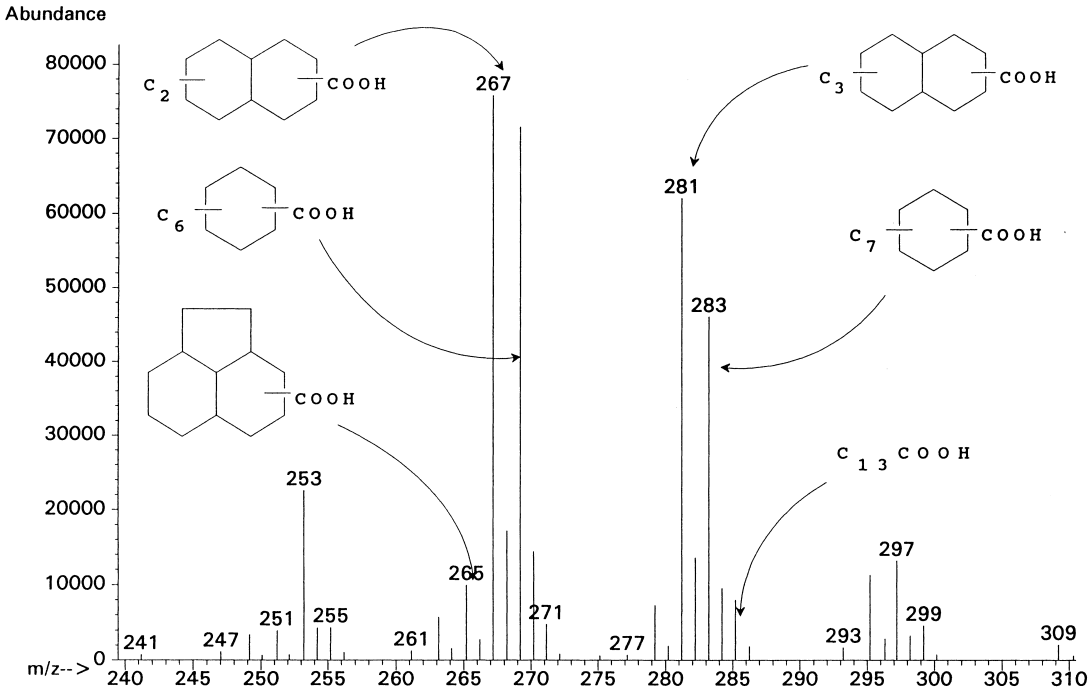

Fig. 5. Single GC–MS scan at 21.5 min from the derivatized NA in Fig. 1B.

Table 3
Percent determination of NA components by *z* and carbon numbers (synthetic supply)

Carbon No.	*z* Number							Percent by carbon number
	0	2	4	6	8	10	12	
10	15	1.3	0.22	0.02	0.04	0.03	0.37	17.00
11	4.4	1.4	0.59	0.02	0.03	0.01	0.89	7.38
12	1.1	1.4	1.2	0.09	0.05	0.02	0.29	4.16
13	2.2	1.6	1.7	0.26	0.09	0.02	0.11	5.97
14	3.2	1.5	2.0	0.43	0.11	0.04	0.17	7.39
15	4.1	1.2	1.3	0.52	0.14	0.07	0.25	7.61
16	1.3	0.87	1.1	0.53	0.17	0.09	0.31	4.33
17	1.1	0.73	0.87	0.48	0.20	0.11	0.19	3.66
18	1.5	0.77	0.75	0.41	0.20	0.11	0.17	3.87
19	19	1.8	0.67	0.34	0.17	0.13	0.16	21.78
20	3.3	0.64	0.53	0.27	0.13	0.14	1.4	6.37
21	0.68	0.41	0.37	0.19	0.09	0.12	0.26	2.12
22	1.1	0.29	0.24	0.11	0.51	4.1	0.06	6.36
23	0.35	0.20	0.15	0.04	0.02	0.22	0.06	1.04
24	0.19	0.10	0.08	0.00	0.00	0.01	0.00	0.38
25	–	0.02	0.01	–	0.01	0.11	–	0.16
26	0.00	–	–	–	–	0.01	–	0.01
27	0.00	–	–	–	–	0.01	–	0.01
28	0.36	0.00	–	–	–	–	–	0.36
29	0.06	–	–	–	–	–	–	0.06
Percent by *z* number	58	14	12	3.7	2.0	5.3	4.7	100.00

States Department of Agriculture Forest Products Laboratory, under a research joint venture agreement, 95-RJVA-2622. The authors would like to thank Dr. Roger Pettersen of the Forest Products Laboratory, Madison, WI, USA, for his review of this work.

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