A New Approach to the Analysis of Phthalate Esters by GC/MS

Application

Gas Chromatography/Mass Spectrometry

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Authors

Cameron George
Agilent Technologies, Inc.
91 Blue Ravine Road
Folsom, California 95630-4714
USA

Harry Prest
Agilent Technologies, Inc.
1601 California Avenue
Palo Alto, California 94304-1111
USA

Abstract

A new instrumental method for the determination of 29 phthalate esters, including six recently banned from baby toys by the European Union, using positive chemical ionization and retention-time locking is described. Positive chemical ionization provides a high degree of selective ionization for the phthalates, primarily producing spectra in which the protonated molecule (M+1) is the base peak. This provides easy discrimination among the phthalates on the basis of their molecular weight, while retention-time locking increases confidence in the identification of the various isomers.

In this approach, both pure compounds and technical mixtures are considered. Although this work focuses on the more commonly used 1,2-substituted esters, the 1,3-isomers and 1,4-isomers are also characterized.

The combination of positive chemical ionization and retention-time locking makes the method rugged, durable and applicable to a wide variety of matrices.

Introduction

The widespread use and manufacture of plastics have made the phthalate esters one of the most ubiquitous classes of compounds in our everyday environment. These "plasticizers" increase polymer flexibility due to their function as intermolecular "lubricants". Because they are additives and not reagents, they are not chemically bound in the polymer and are available to leach from the matrix. Phthalates are also components of cosmetics, detergents, building products (flooring, sheeting, films), lubricating oils, PCB substitutes, carriers in pesticide formulations and solvents. Consequently, the potential for human exposure is very high. Toxicological studies have linked some of these compounds to liver and kidney damage, and to possible testicular or reproductive-tract birth defect problems, characterizing them as endocrine disruptors. Scientists at the U.S. Centers for Disease Control have, for the first time, documented human exposure to phthalates by determinations of the monoester metabolites in human urine [1]. Their work leads to the conclusion that "phthalate exposure is both higher and more common than previously suspected."

Of particular concern were the significantly higher concentrations of the dibutyl phthalate metabolite in urine of women of childbearing age (20-40 years) than in other portions of the population.

The presence of phthalate esters in polyvinyl chloride (PVC) toys has generated the most controversy. While regulators in Greece have completely banned soft PVC toys, Austria, Denmark, Finland, France, Germany, Norway and Sweden have unilaterally banned phthalates in PVC toys for children under three years old. In December of 1999, the European Union (EU), concerned with a “serious and immediate risk” to children, placed an emergency ban on six of the phthalate esters in soft PVC toys and childcare products meant to be placed in the mouths of children under the age of three [2]. None of the six banned phthalates may exceed 0.1% by weight.

These heightened concerns suggest the need for an improved method of detecting and characterizing phthalate esters which is applicable to a wide variety of matrices. This application note describes such an analytical method.

Phthalate Structure and Mass Spectra

The three primary structures of phthalates are shown in Figure 1. Although there are three possible positions for the ester linkages, the most commonly used phthalates are based on the 1,2-benzenedicarboxylic acid structure (top). There are an infinite number of possible alkyl side chains, (R) and an infinite number of combinations of the side groups (R and R'). For example, the diisononyl phthalate consists of an array of compounds due to the isomeric branched-chain alkyl groups on both side chains.

For phthalate esters with saturated alkyl side chains (without oxygen), the most intense peak in the electron impact (EI) ionization mass spectrum at 70 eV is always at *m/z* 149 due to the rapid formation and stability of the ion shown in Figure 2. (The only exception is $R=R'=CH_3$ where the base peak is at *m/z* 163).

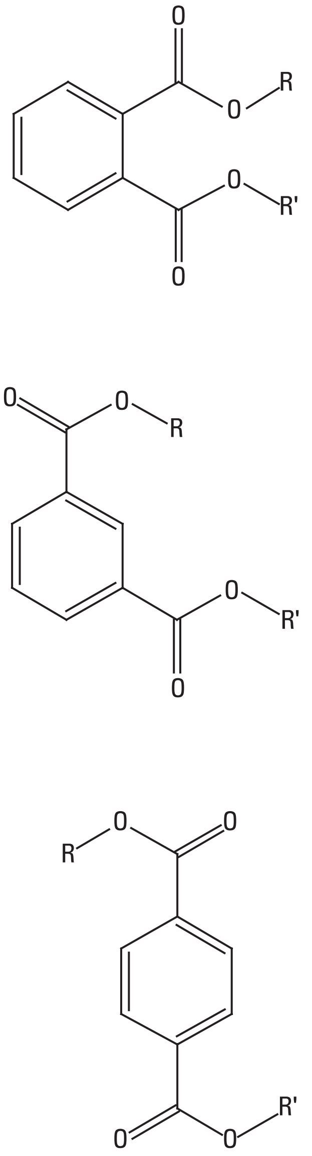

Figure 1. Phthalic ester (top) or the 1,2-benzenedicarboxylic acid ester, isophthalic ester (middle) or the 1,3-benzenedicarboxylic acid ester, and terephthalic ester (bottom) or the 1,4-benzenedicarboxylic acid ester. R and R' represent alkyl side chains which may be branched and contain oxygen.

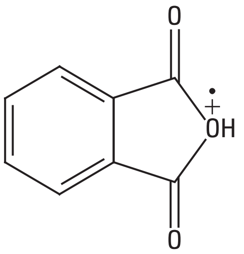

Figure 2. The most abundant ion in the mass spectra of the phthalate esters with saturated alkyl side chains; *m/z* 149. The exception is for dimethyl phthalate where both R and R' are CH_3 and so the H on the oxygen is replaced by CH_3 and consequently *m/z* 163 becomes the base peak.

Invariably, the molecular ion is very weak or altogether absent; other fragments that provide information on the phthalate identity are also of very low abundance. As an example, consider the EI mass spectrum of dibutyl phthalate, one of the six banned by the EU, and bis(4-methyl-2-pentyl) phthalate in Figure 3. Identifying fragments have relative intensities of less than 10%. Gas chromatography provides some separation of the phthalates, but with the array of possible isomers and essentially a single identifying ion (i.e., *m/z* 149), distinguishing the individual phthalates of concern is difficult. More confident identification of the phthalates is possible using chemical ionization mass spectrometry in conjunction with retention-time locking (RTL).

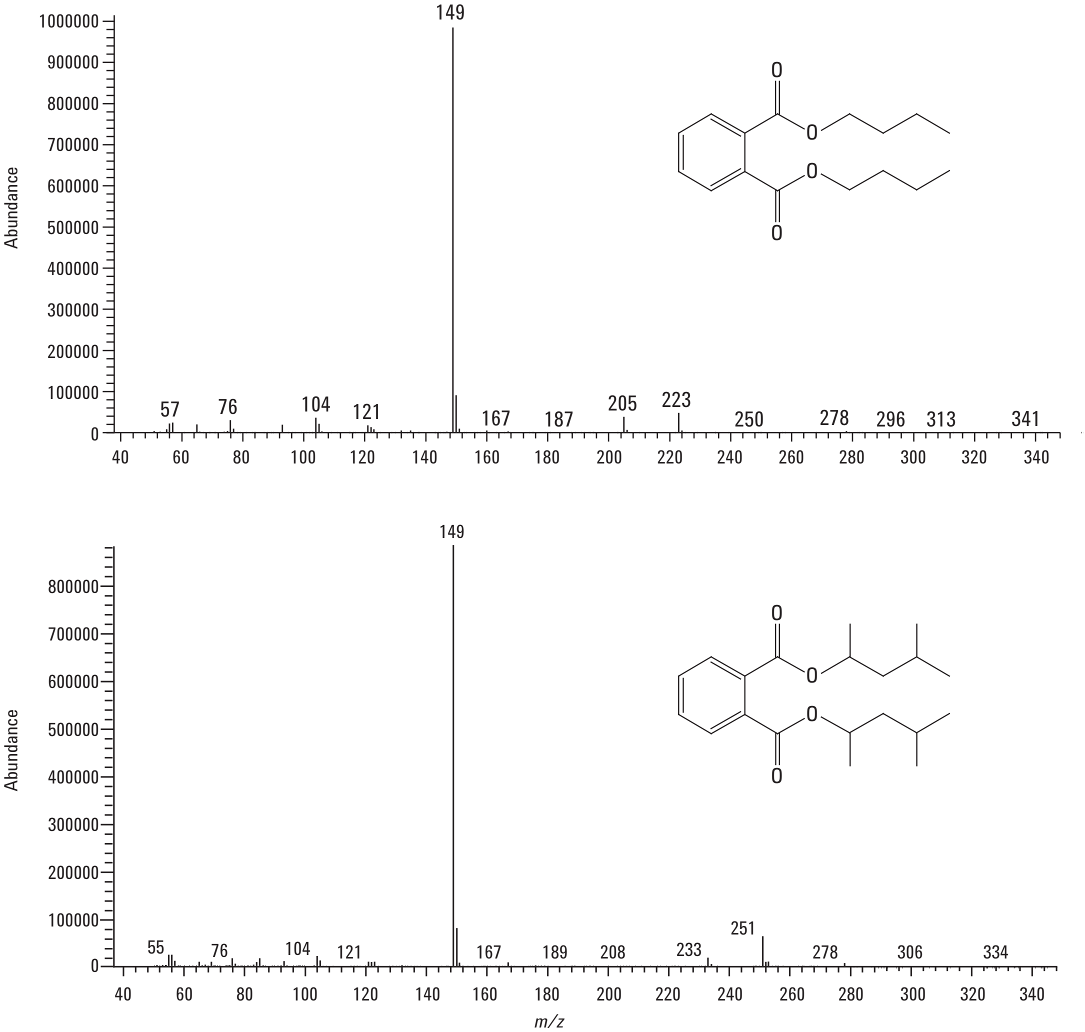

Figure 3. Electron impact ionization mass spectra of di-n-butyl phthalate (upper panel) and bis(4-methyl-2-pentyl) phthalate (lower panel) from *m/z* 50 to 350 at 70 eV. Notice the lack of intense fragments and molecular ions. The molecular weights are 278 and 334 g/mole, respectively.

Retention-time locking allows compound retention times achieved on any one Agilent 6890 gas chromatograph (GC) to be replicated to within a few seconds on any other Agilent 6890 gas chromatograph (GC) applying the same GC method [3-5]. RTL is a powerful approach to compound identification. RTL allows the creation of compound acquisition methods and quantitation databases that can be reproduced in any laboratory, anywhere, because a compound can have a universally fixed and reproducible retention time. It is important that RTL be applied in conjunction with the appropriate detection scheme and sample reparation methods.

Chemical ionization provides a more selective form of ionization than electron impact [6]. By judicious choice of the reagent gases, the degree of compound fragmentation can be controlled to a certain extent. In positive chemical ionization, methane reagent gas usually provides more fragmentation than gases of higher proton affinity such as ammonia. Less fragmentation would be helpful in identifying the phthalates. Instead of all phthalates generating a single, similar ion, positive ionization can provide phthalate ester molecular weights.

Experimental

Phthalate esters were obtained from Ultra Scientific (North Kingstown, RI), AccuStandard (New Haven, CT), and ChemServices (West Chester, PA) as neat compounds and mixtures. Dilutions were made in isooctane (Burdick and Jackson Grade, VWR Scientific).

The configuration and operating parameters of the Agilent 6890Plus GC (standard 120V or "faster ramping" 220V), 7683 Automatic Liquid Sampler and 5973N MSD with CI option used for acquiring the data are given in the following tables. PCI reagent gas purities were 99.99% or higher.

Injection Parameters		
Injection Mode	Pulsed Splitless	
Injection Port Temperature	300°C	
Pulse Pressure & Time	25.0 psi	1.00 min
Purge Flow & Time	20.0 mL/min	3.00 min
Gas Saver Flow & Time	20.0 mL/min	3.00 min

Oven Parameters		
Temperature Program	80°C	1.00 min
50.00°C/min	200°C	0.00 min
15.00°C/min	350°C	2.00 min
Oven Equilibrium Time	0.25 min	
MSD Transfer Line Temp	325°C	

Column Parameters		
GC column (122-5532)	DB-5MS 30 m; 0.25 mm i.d.; 0.25 µm film	
Initial Flow & Mode	1.2 mL/min	Constant Flow
Detector & Outlet Pressure	MSD	Vacuum

Mass Spectrometer Parameters	
Tune Parameters	PCI Autotune (NH3)
Electron Multiplier Voltage	Autotune + 400V
Solvent Delay	4.00 min
Scan Parameters	194 - 550 *m/z*
Quadrupole Temperature	150°C
Source Temperature	250°C
Ammonia Gas Flow (MFC setting)	0.5 mL/min (10%)

Miscellaneous Parts		
Septa	5182-0739	BTO septa (400°C)
Liner	5062-3587	Deactivated 4 mm i.d. single taper
GC column ferrule	5181-3323	250 µm Vespel
MSD interface ferrule	5082-3508	0.4 mm i.d. graphitized Vespel

Results and Discussion

As expected using methane as the reagent gas, the PCI mass spectra of the phthalates show ions corresponding to the protonated molecule $[M+H]^+$ and adducts $[M+C_2H_5]^+$ and $[M+C_3H_5]^+$. Because of the relatively vigorous fragmentation produced by methane, the spectra of the dialkyl phthalate esters still resemble that produced in EI. In most cases, the fragment at *m/z* 149 is the base peak, however ions at *m/z* M+1, M+29 and M+41 are relatively intense with $[M+H]^+$ from 10% to 30% (Figure 5). The dialkyl phthalate spectra also show a fragment corresponding to loss of one of the alkyloxy side chains to produce an ion shown in Figure 4. This is the most intense fragment for the dimethyl and diethyl phthalates and for the dibutyl and dipentyl (diamyl) phthalates, about 75% of the 149 base peak. As the length of ester alkyl chain increases, the intensity of this fragment decreases. (Apparently, in the dialkyl isophthalates, loss of the alkyl side chain not accompanied by the oxygen may be a preferred route.)

Although positive chemical ionization with methane provides more information than EI on phthalate identity, the methane reagent is still rather unselective in ionization and will produce more chemical noise in the background, complicating identification in complex matrices.

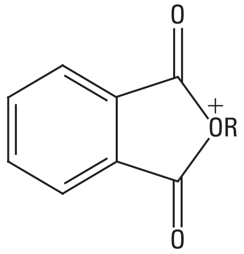

Figure 4. One of the most intense fragments in the methane PCI spectra of the phthalate esters is formed by loss of one of the alkyloxy side groups.

Applying ammonia as the reagent gas in PCI to reduce chemical noise and enhance identification of the phthalates is a more useful approach. The relatively gentle ionization produces protonation of the dialkyl phthalates, with *m/z* M+1 the base peak in their spectra. When combined with retention-time locking, identification of phthalates becomes further simplified. Compare the spectra of the di-n-butyl phthalate acquired using methane versus ammonia as the reagent gas (Figure 5). The protonated molecule is the single dominant peak in the ammonia PCI mass spectrum of the di-n-butyl phthalate, and the adduct at *m/z* 296 ($[M+NH_4]^+$) is relatively small.

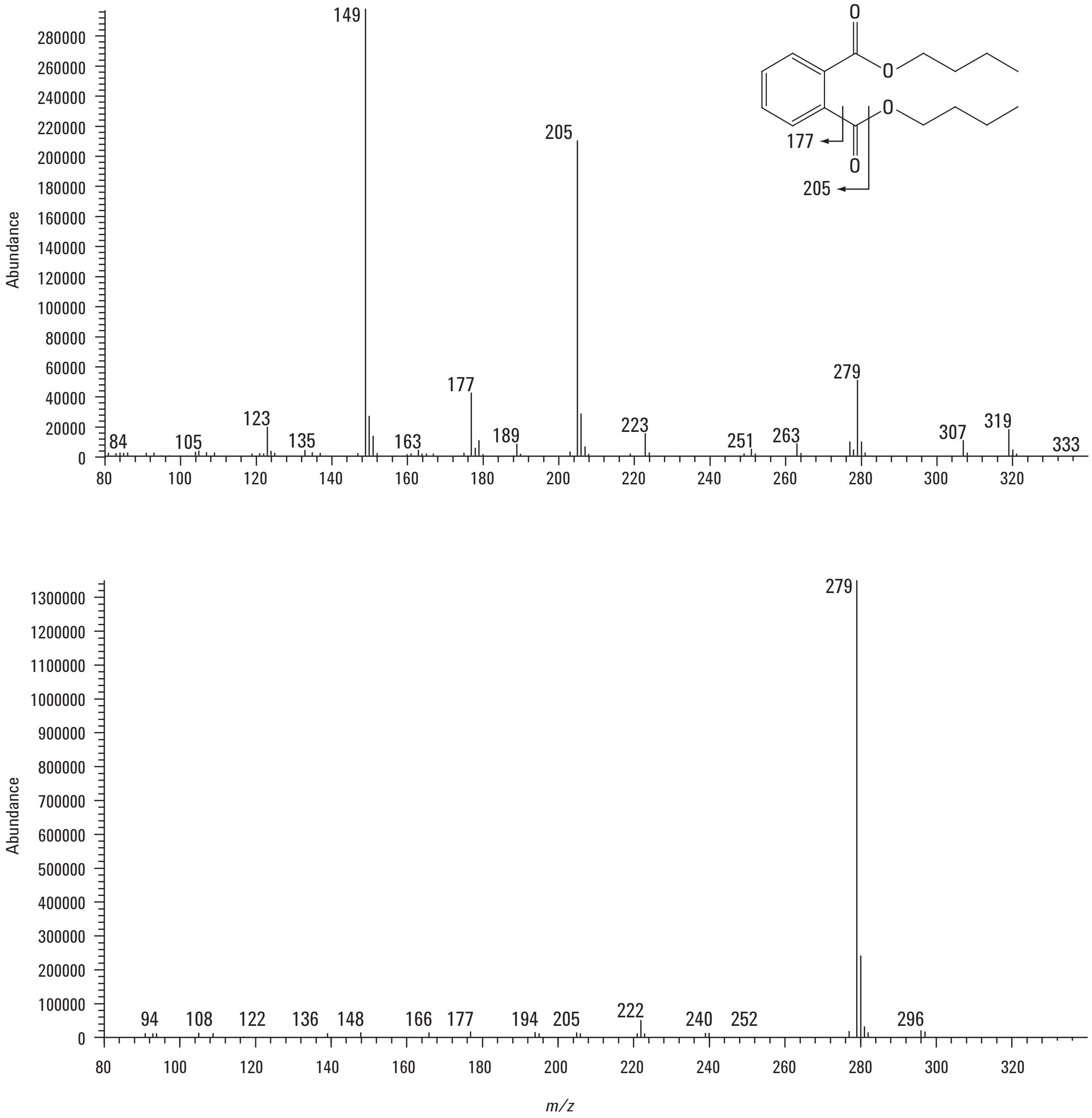

Figure 5. PCI methane (upper panel) and ammonia (lower panel) mass spectra of di-n-butyl phthalate. The PCI methane mass spectrum shows substantial fragmentation but relative to the EI spectrum in Figure 3, high abundance for the higher *m/z* ions such as the protonated molecule at *m/z* 279. The ion at *m/z* 205 is generated by loss of an oxybutyl fragment; a process described in Figure 4. The PCI-ammonia mass spectrum consists almost completely of the protonated molecule.

This implies an easy method for identification. Whereas the EI spectra of the phthalates most frequently result in a base peak at *m/z* 149, the dialkyl phthalate PCI-ammonia spectra have base peaks at *m/z* = M+1. All dialkyl phthalates molecular formulas can be expressed as

$$C_8H_6O_4(CH_2)_y\ (CH_2)_x.$$

These phthalates have (nominal) molecular masses given by

$$M = 166 + (x + y)\cdot 14, \text{ or}$$

$$M = 166 + (w)\cdot 28,$$

where x and y are the side chain lengths, and the second formula applies to symmetrical side chains (i.e., x = y = w). For example, di-n-butyl phthalate has x = y = 4, and therefore a (nominal) molecular mass of 278 which produces *m/z* 279 as the base peak. Interestingly, the PCI-ammonia spectra of the dialkyl isophthalates and terephthalates appear to have base peaks at *m/z* M+18 due to $[M{+}NH_4]^+$. Because of the greater steric access to the ester linkages, adduct formation may be preferred.

Table 1 gives the phthalate names, CAS numbers, molecular formula, nominal molecular mass, base peak in the PCI-ammonia spectrum and the RTL elution times. These retention times are "locked" relative to diphenyl phthalate, which has been chosen as the RTL locking compound and locked to elute at 9.450 min. Notice that the branched chain isomers elute prior to their straight chain forms on this column phase.

Table 1. Phthalate compound names, Chemical Abstracts Services numbers (CAS), molecular weights (M. Wt.), molecular formulas, nominal base peak in the PCI-ammonia spectrum and retention time (RT) in minutes. Retention times are locked relative to diphenyl phthalate (9.450 min). Retention time ranges are given for the isoalkyl phthalate technical mixtures. Phthalates banned by the EU are indicated by an asterix*. Benzyl benzoate is included since it is used as a surrogate in U.S. Environmental Protection Agency Method 8061.

Name	CAS	M. Wt.	Molecular Formula	Base Peak	RT (min)
dimethyl phthalate	131-11-3	194	$C_8H_4O_4(CH_3)_2$	195	4.32
dimethyl isophthalate	1459-93-4	194	$C_8H_4O_4(CH_3)_2$	212	4.54
diethyl phthalate	84-66-2	222	$C_8H_4O_4(C_2H_5)_2$	223	4.81
diethyl terephthalate	636-09-9	222	$C_8H_4O_4(C_2H_5)_2$	240	5.06
benzyl benzoate	120-51-4	212	$C_{14}H_{12}O_2$	230	5.62
diisobutyl phthalate	84-69-5	278	$C_8H_4O_4(C_4H_9)_2$	279	5.95
di-n-butyl phthalate*	84-74-2	278	$C_8H_4O_4(C_4H_9)_2$	279	6.40
bis(2-methoxyethyl) phthalate	117-82-8	282	$C_8H_4O_4(C_2H_4OCH_3)_2$	283	6.57
diamyl phthalate	131-18-0	306	$C_8H_4O_4(C_5H_{11})_2$	307	6.94
bis(2-ethoxyethyl) phthalate	605-54-9	310	$C_8H_4O_4(C_2H_4OC_2H_5)_2$	311	7.13
butyl benzyl phthalate*	85-68-7	312	$C_8H_4O_4(C_4H_9)(CH_2C_6H_5)$	313	8.42
diphenyl phthalate	84-62-8	318	$C_8H_4O_4(C_6H_5)_2$	319	9.45
diphenyl isophthalate	744-45-6	318	$C_8H_4O_4(C_6H_5)_2$	319	10.30
dicyclohexyl phthalate	84-61-7	330	$C_8H_4O_4(C_6H_{11})_2$	331	9.32
bis(4-methyl-2-pentyl) phthalate	146-50-9	334	$C_8H_4O_4(CH_3C_5H_{10})_2$	335	6.93
diisohexyl phthalates	146-50-9	334	$C_8H_4O_4(C_6H_{13})_2$	335	7.55 - 8.28
dihexyl phthalate	84-75-3	334	$C_8H_4O_4(C_6H_{13})_2$	335	8.34
dibenzyl phthalate	523-31-9	346	$C_8H_4O_4(CH_2C_6H_5)_2$	347	10.51
hexyl-2-ethylhexyl phthalate	75673-16-4	362	$C_8H_4O_4(C_2H_5C_6H_{12})(C_6H_{13})$	363	8.84
bis(2-n-butoxyethyl) phthalate	117-83-9	366	$C_8H_4O_4(C_2H_4OC_4H_9)_2$	367	8.98
bis(2-ethylhexyl) phthalate*	117-81-7	390	$C_8H_4O_4(C_2H_5C_6H_{12})_2$	391	9.32
di-n-octyl phthalate*	117-84-0	390	$C_8H_4O_4(C_8H_{17})_2$	391	10.28
dioctyl isophthalate	137-89-3	390	$C_8H_4O_4(C_8H_{17})_2$	408	10.84
diisononyl phthalates*	28553-12-0	418	$C_8H_4O_4(CH_3C_8H_{17})_2$	419	9.40 - 11.10
dinonyl phthalate	84-76-4	418	$C_8H_4O_4(C_9H_{19})_2$	419	11.19
diisodecyl phthalates*	26761-40-0	446	$C_8H_4O_4(CH_3C_9H_{18})_2$	447	10.16 - 11.86
didecyl phthalate	84-77-5	446	$C_8H_4O_4(C_{10}H_{21})_2$	447	12.05
diundecyl phthalate	3648-20-2	474	$C_8H_4O_4(C_{11}H_{23})_2$	475	12.87
didodecyl phthalate	2432-90-8	502	$C_8H_4O_4(C_{12}H_{25})_2$	503	13.65
ditridecyl phthalate	119-06-2	530	$C_8H_4O_4(C_{13}H_{27})_2$	531	12.01 - 13.81

Technical formulations of the isoalkyl phthalates tended to contain substantial amounts of the straight chain isomer, which may convolute quantitation as well as peaks that may be construed as originating from nonequivalent side chains i.e., x ≠ y in equation 1). These impurities can be detected as M±14 around the mass of the nominal isomer. For example, technical grade diisononyl phthalate contains compounds that generate ions at *m/z* 391 (minor), 405, 433, and 447 in addition to the nominal diisononyl phthalate compound at *m/z* 419. The "gentle" ionization of ammonia reagent gas, the elution times and the study of the spectra of other pure isomers, such as the dinonyl phthalate, suggest that these fragments are not formed by the PCI process but are due to these different alkyl side chain impurities (Figure 6). To demonstrate the utility of the PCI-ammonia compared to conventional EI analysis, consider the chromatograms presented in Figure 7. The EI spectra of the phthalates produce *m/z* 149 as the base peak for all the phthalates present; distinguishing ions are minor constituents (<10% relative intensity), making identification complicated. However, by examining the appropriate PCI-ammonia ions, the various phthalates are easily distinguished.

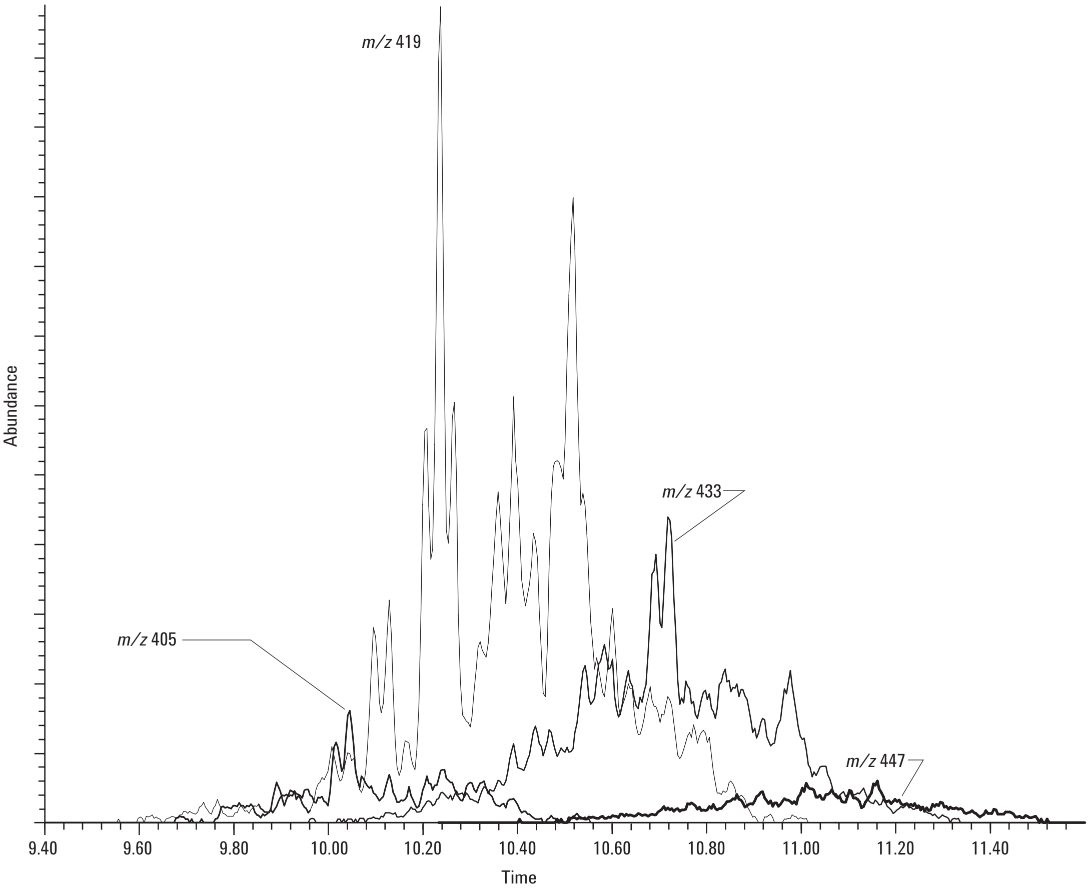

Figure 6. PCI-Ammonia extracted ion chromatogram of technical diisononyl phthalate. The diisononyl appears as the major component at *m/z* 419 while ions at *m/z* 405, 433, and 447 indicate alkyl chains shorter by one CH_2 unit and longer by one and two CH_2 units respectively.

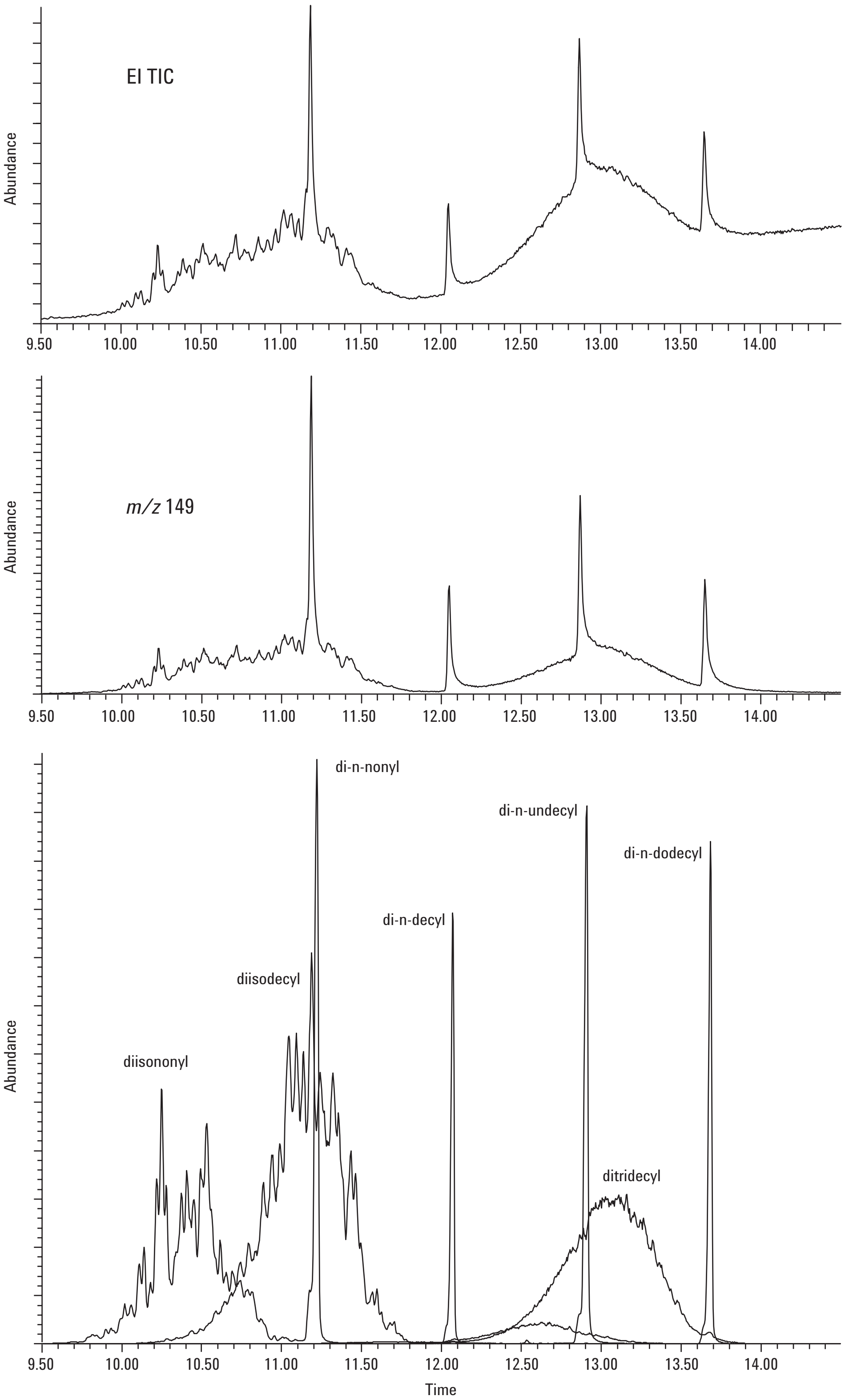

Figure 7. Chromatograms of dinonyl, diisononyl, didecyl, diisodecyl, diundecyl, didodecyl, ditridecyl phthalate esters in EI (upper panel), EI as an extracted ion chromatogram at *m/z* 149 (middle panel), and PCI-extracted ion chromatogram with ions selected for the individual phthalate classes as given in Table 1. The EI information is insufficient to identify coeluting phthalates. For example, the dinonyl and diundecyl phthalates are "buried under" the signals from the isodecyl and ditridecyl phthalates.

Conclusions

Applying GC - electron impact (EI) mass spectrometry to the determination of phthalates requires full chromatographic separation. The EI spectra of the phthalates are distinguished only by ions of very low intensity. In EI, the phthalates produce a single common ion (*m/z* 149) as the most intense spectral peak, regardless of the alkyl side chain substitution. Applying tandem mass spectrometry (i.e., EI/MS/MS) gains nothing, because there is a common parent ion, and therefore any daughter ions would also be non-unique. However, the combination of positive chemical ionization with retention-time locking allows even complex mixtures of phthalates to be characterized. Ammonia reagent gas produces the protonated molecule as the base peak, which immediately allows the phthalates to be distinguished on the basis of their substitution. PCI is also an advantage in complex matrices, where the non selective ionization of EI produces a high chemical background. This method should therefore be suitable for use in phthalate determinations in environmental media, plastics, cosmetics and many other matrices.

"Locking" the retention time enhances confidence in the characterization of the various phthalate isomers on the basis of their definitive retention time. This is especially helpful for determinations using selected ion monitoring (SIM), since SIM groups need not be edited after column maintenance [4]. The data in Table 1 facilitate the development of a SIM method. The extension of the method to phthalates which elute at higher temperatures (>350°C) is also easily accomplished.

References

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