Form of Aromatics in Thermally Induced Reactions of Chemically Bonded RP-C18 Stationary Phase

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In continuation of the research on the thermally induced chemical transformation of the silica-based chemically bonded stationary phases (C18), the oxidative cleavage of the silicon–carbon bonds with hydrogen peroxide and potassium fluoride was utilized, followed by the gas chromatography coupled with mass spectrometry (GC–MS) study of the resulting products. These investigations allowed determination of the probable structures of certain thermal modification products as the various different alkyl derivatives of the phenylsilane ligands. Apart from aromatic compounds, the products with unsaturated bonds and carbonyl functionalities were found in the analyzed extracts. The analysis of the GC–MS chromatograms reveals that under the applied working conditions, the investigated process runs with relatively low yields.

Introduction

The starting point for this research was the results of applying the high-energy laser Raman spectrometry to the study of the aliphatic chemically bonded silica-based stationary phases reported in (1, 2). The authors found out that some stationary phases present an atypical band for aliphatic moieties in the Raman spectra at the frequency of 2285–1130 cm\(^{-1}\). Only chemically bonded stationary phases with the ligands composed of at least five carbon atoms (or a combination of carbon atoms and heteroatoms making a total number of five) were able to produce this surprising effect. Because the observed Raman band is typical of aromatic functionality, it was assumed that the observed Raman scattering originates from the products of the temperature-induced and silica-catalyzed structural transformation of the chemically bonded aliphatic ligands. These facts were reported as a novel approach to the estimation of the density of coverage of the silica gel matrix with aliphatic ligands, based on the predominant Raman band of an originally obscure origin in the wavenumber range from 2285 to 1130 cm\(^{-1}\) (2–5).

Further study has furnished additional experimental evidence that the light emitted from a high-power neodymium laser resulted in a substantial elevation of the samples’ temperature, which certainly affected the aliphatic chemically bonded stationary phases. Moreover, employing the absorption spectrometry in the UV and IR range, and also the differential scanning calorimetry, determination of the minimal temperature level that enables the aromatization process was accomplished (6). This study provided a preliminary spectroscopic characterization of the products formed at high temperature as well. Quantitative relationships between the intensity of Raman scattering, the UV absorption changes (as a result of thermal modification) and the content of alkyl groups were presented in (7, 8).

The first attempt to identify the products that might appear in the course of thermal modification of the chemically bonded stationary phases was the study with use of the high-performance liquid chromatography with the diode-array detection of the selected aliphatic chemically bonded stationary phases for thin-layer chromatography (TLC). This experiment only revealed the presence of the thermal modification products, which are cleaved from the silica matrix (and hence, can be further extracted) (9).

In the subsequent study, gas chromatography coupled with mass spectrometry (GC–MS) was employed to investigate the extracts from the different thermally processed TLC-type RP-C18 stationary phases, using accelerated solvent extraction (10). Different quantities of individual thermal modification products cleaved from the silica matrix (1-octadecene, hexadecanal, octadecanal and methyl n-hexadecyl ketone) were identified in the obtained extracts. The identified non-aromatic products of thermal modification evidently are the byproducts of the main process.

In continuation of the research on the thermally induced chemical transformation of the silica-based chemically bonded stationary phases (C18), the oxidative cleavage of the silicon–carbon (Si–C) bonds with hydrogen peroxide and potassium fluoride was utilized, followed by the GC–MS study of the resulting products. These investigations allowed determination of the probable structures of certain thermal modification products as the various different alkyl derivatives of the phenylsilane ligands.

The aim of this study was to investigate the products responsible for the growth of the signal in the UV, IR and the high-energy laser Raman spectroscopy, resulting from thermal modification of the chemically bonded stationary phases. The main purpose of the chromatographic studies was to determine structures of these thermal modification products which do not undergo spontaneous cleavage from the silica matrix in the course of heating the stationary phase but thanks to the oxidative cleavage of the Si–C bond they form respective unbound hydroxy-derivatives and become extractable with solvent. Structures of these derivatives can be estimated upon their respective mass spectra and the match with the mass spectral library data. Moreover, we intended to get quantitative insights into the yields of these components in the extracts, based on the measurements of the chromatographic bands areas.

Experimental

Reagents, materials and standards

A description of reagents and standards used in this study is given in Table I.

1-Octadecanol and 2,5-bis(1,1-dimethylethyl)-4-methylphenol (BHT) were diluted in dichloromethane at concentrations of 90 and 100 mg/L, respectively.
**Thermal treatment**

Sample 1 of the RP-C18 TLC stationary phase was placed in the sealed glass ampoule and heated at 170°C for 2 h, while Sample 2 was left unheated as a reference. The thermal treatment experiment was repeated three times. A comparison of thermograms (DSC) acquired under the nitrogen and air indicates that due to the presence of air, oxidation takes place only above 200°C (6). Due to that, the temperature used in our experiment in the presence of air should not affect the process.

**Oxidative cleavage of Si–C bond**

Oxidative cleavage of the C–Si bond and isolation of the cleaved products were done according to the modified procedure presented in (11). To the sample of the stationary phase (100 mg) in THF/methanol (1:1, 8 mL), potassium bicarbonate (35 mg), potassium fluoride (20 mg) and 30% aqueous hydrogen peroxide (0.05 mL) were added. The resulting mixture was stirred at 65°C for 12 h. Then the solvents were removed under the vacuum and the cleavage products were isolated by transferring the residue to a glass column filled with the flash grade silica gel (ca. 6 g), using methanol/dichloromethane, 1:9 (v/v) as mobile phase (the total volume of the mobile phase used was ca. 60 mL). The mobile phase was removed to dryness under the vacuum, the cleavage products were dissolved in 5 mL of dichloromethane and the obtained solutions were placed in the two 2.0-mL vials. These solutions were further analyzed by means of GC–MS.

**GC–MS analysis**

GC–MS detection was performed with an Agilent (Hewlett-Packard) gas chromatograph under the following working conditions:

- **Column**: a 30 m × 0.25 mm i.d. capillary column coated with a 0.25-μm film of HP-5MS. Carrier gas, helium. The oven temperature was maintained at 80°C for 1 min after injection, then programmed at 25°C min⁻¹ for 3.2 min and at 3°C min⁻¹ for 47.67 min. The final temperature, 325°C, was maintained for 5 min. The full-scan mass spectrometry was performed in the electron-impact ionization mode, electron energy 70 eV, with scanning from 50 to 300 m/z.
- Data were acquired and processed by means of the Hewlett-Packard Chemstation software and the Wiley–NBS Registry of Mass Spectral Data NIST02.L.

**Results**

The resulting extracts obtained from the samples of the RP-C18 stationary phase (unheated and heated at 170°C for 2 h), and subsequently subjected to the oxidative cleavage of the Si–C bond, were analyzed by GC–MS. The respective chromatograms are shown in Figure 1.

The chromatograms in all the examined cases repeatedly show the predominant band of 1-octadecanol. The retention time determined for the 1-octadecanol standard (90 mg/L in dichloromethane) equals 16.9 min and it matches that of the predominant peak in the investigated samples.

One of the reaction products, which is present in the extract obtained from the non-heated sample, is BHT. Identification of this reaction product was made based on the mass spectrum and the match with the data from the mass spectral library, and also upon its retention time compared with that of the standard (4R = 6.5 min).

The selected aromatic species showing the match rate not <90% are presented in Table II.

**Discussion**

In the search for a suitable method enabling structural characteristics of the products of the thermally induced processes occurring in stationary phases, a method based on certain chemical reaction was taken into the account. Namely, the method to cleave alkyl ligands from the silica gel surface as stable and easily characterizeable compounds under mild working conditions is given in the literature (11). This method is based on oxidative cleavage of the Si–C bond with hydrogen peroxide and potassium fluoride. Schematic representation of structural transformations that can occur in the course of the oxidative cleavage of the Si–C bond with the heated samples of the aliphatic chemically bonded stationary phase is given in Figure 2, where in the simplest case, –R can be 1-octadecyl group, but it can also be a product of more complex structural changes.

From the GC–MS study, we derived information on the probable structures and the relative contents of substances found in the investigated extracts. With the samples obtained from the different types of the RP-C18 stationary phase, the area of the 1-octadecanol band characterizes with a variation of ±12%, probably resulting from a non-quantitative nature of the process of the oxidative cleavage of the Si–C bond. Thus, 1-octadecanol can be regarded as an internal standard for semi-quantitative estimation of the other analytes present in the investigated samples. The concentration of 1-octadecanol derived from the parameters of the test chromatogram allows estimation of its quantity in the stationary phase samples as equal to ca. 300 μmol/g sample.

Presumably, the main thermally induced reaction product, which is BHT, is formed already at a temperature of 65°C applied in the oxidative cleavage of the Si–C bond. As a result of heating the RP-C18 stationary phase samples (at 170°C), followed by the cleavage of the Si–C bonds, the contents of BHT in the obtained extracts has more than doubled. Assuming linear characteristics of the MS detector over the whole investigated range of concentrations, the estimated BHT amounts derived from the stationary phase samples are ca. 50 μmol/g sample.

Structures tentatively attributed to the other chromatographic bands were determined exclusively on the basis of their mass spectra and the match with the mass spectral library data.

**Table I**

<table>
<thead>
<tr>
<th>Description</th>
<th>Purity</th>
<th>Company</th>
<th>Cat. number</th>
</tr>
</thead>
<tbody>
<tr>
<td>RP-18 F544A TLC stationary phase</td>
<td>–</td>
<td>Merck</td>
<td>115389</td>
</tr>
<tr>
<td>Potassium bicarbonate</td>
<td>Analytical grade</td>
<td>POCh</td>
<td>746640421</td>
</tr>
<tr>
<td>Potassium fluoride anhydrous</td>
<td>Analytical grade</td>
<td>Chempur</td>
<td>117414208</td>
</tr>
<tr>
<td>30% aqueous hydrogen peroxide</td>
<td>Analytical grade</td>
<td>POCh</td>
<td>885193111</td>
</tr>
<tr>
<td>Tetrahydrofuran</td>
<td>Analytical grade</td>
<td>POCh</td>
<td>278001118</td>
</tr>
<tr>
<td>Methanol</td>
<td>HPLC grade</td>
<td>Aldrich</td>
<td>34980</td>
</tr>
<tr>
<td>Dichloromethane</td>
<td>Analytical grade</td>
<td>Chempur</td>
<td>116284101</td>
</tr>
<tr>
<td>Silica gel 60</td>
<td>Flash grade</td>
<td>Merck</td>
<td>1.07734.1000</td>
</tr>
<tr>
<td>1-Octadecanol</td>
<td>99%</td>
<td>Aldrich</td>
<td>258768</td>
</tr>
<tr>
<td>BHT</td>
<td>Grade Ph Eur, JPE, NF, E 321</td>
<td>Merck</td>
<td>817074</td>
</tr>
</tbody>
</table>
Hence, possible discrepancies between the structures obtained from the match and the real structures of the respective analytes have to be anticipated. Thus, only the set of the selected aromatic species whose presence is essential to explain spectroscopic effects described in the literature (1–8) is presented in Table II. The entire chromatograms contain bands which can be attributed to the other aromatic substances, compounds containing unsaturated bonds or carbonyl groups and aliphatic alcohols.

Analyzing the entire chromatograms, we can notice certain regularities. For the selected functionalities (e.g., for aromatic compounds, those containing unsaturated bonds or carbonyl groups in their structure), concentrations are usually much higher in the extracts resulting from the cleavage of the Si–C bond from the heated stationary phase samples than from the unheated ones. This observation can be considered a support of the hypothesis that the aforementioned hydroxy-derivatives originate from the ligands formed from the chemically bonded octadecylsilane groups in the course of heating the stationary phase at 170°C. In most cases, respective signals in the UV, IR and Raman spectra of the above organic compound classes are consistent with the experimental characteristics of the stationary phases presented in (6–8). In the case of aliphatic alcohols, however, we do not observe any significant difference between their concentrations in the extracts resulting from the Si–C bond cleavage in the heated and unheated stationary phase samples. It can be an indication that the presence of aliphatic alcohols in the extracts is not strongly associated with the main thermally induced process.

The results of the GC–MS study suggest that the catalytic action of the silica surface allows a number of processes at a relatively low temperature already. Probably, the first stage is dehydrogenation of the chemically bonded alkyl groups, followed by a double bond oxidation, dehydration of vic-diols, rearrangements and cyclization.

The above characteristics of the chemical reactions, their products and substrates and the conditions that enable their progress indicate certain similarity to the geochemical and natural processes described in (12–16). These geochemical and natural processes generate alkyl aromatics with the long side chains, produced by the catalytic thermal reactions occurring at relatively low temperatures. Such compounds have been found in

Figure 1. Chromatograms of the dichloromethane extracts obtained in the course of the oxidative cleavage of the Si–C bond for the (a) unheated and (b) heated at 170°C for 2 h sample of the RP-18 chemically bonded stationary phase.

(a) Abundance

(b) Abundance

Hence, possible discrepancies between the structures obtained from the match and the real structures of the respective analytes have to be anticipated. Thus, only the set of the selected aromatic species whose presence is essential to explain spectroscopic effects described in the literature (1–8) is presented in Table II. The entire chromatograms contain bands which can be attributed to the other aromatic substances, compounds containing unsaturated bonds or carbonyl groups and aliphatic alcohols.

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the deposits of crude oil and coal in various different regions of the globe, and their concentrations are frequently reported as anomalous. The starting substrates for the geochemical production of the components of crude oil are the straight chain fatty acids. \( n \)-Alkanes could have been formed by decarboxylation of the straight chain fatty acids and their relatively high abundance in crude oil is typical. Alkylcyclohexanes and alkylbenzenes could have been formed by direct cyclization and aromatization of the straight chain fatty acids, yet this process requires much higher temperatures than decarboxylation. Thus, the presence of aromatic compounds in the deposits of crude oil is not as common as that of \( n \)-alkanes.

Conclusions

Heating of the RP-C18 TLC-type chemically bonded stationary phase at 170 °C causes conversion of a relatively small number of octadecylsilane groups, which leads to formation of unsaturated, carbonyl and aromatic products. Presence of these relatively small amounts of aromatic products is reflected in spectroscopic properties of the thermally processed stationary phases. These changes basically are the growth of the signal in the UV absorption and Raman spectrometry and also changes in the intensity of the IR absorption bands attributed to structural fragments which are substrates and products of this reaction. As these aromatic products remain uncleaved from the silica gel matrix, their GC–MS study is possible only after the oxidative cleavage of the Si–C bond.

References

3. Kowalik, G., Klama, B., Okopska, R., Kowalska, T.; Study of the solvolytic cleavage of octadecyl ligands from the surface of RP-18-type stationary phases in TLC. Part 3. TLC and Raman spectroscopic quantification of the extent of cleavage for buffer pH from 1 to 10;

![Table II](https://academic.oup.com/chromsci/article-abstract/52/9/1005/273616/1008)

<table>
<thead>
<tr>
<th>( t_R ) (min)</th>
<th>Structure matching the mass spectral library</th>
<th>Compound</th>
<th>% Match rate</th>
<th>Peak area vs. 1-octadecanol peak area</th>
</tr>
</thead>
<tbody>
<tr>
<td>5.4</td>
<td>6-tert-Butyl-2,4-dimethylphenol</td>
<td>92</td>
<td>0.00000</td>
<td>0.00141</td>
</tr>
<tr>
<td>6.5</td>
<td>Phenol, 2,4-bis(1,1-dimethylethyl)-</td>
<td>97</td>
<td>0.000237</td>
<td>0.00269</td>
</tr>
<tr>
<td>6.6</td>
<td>2,6-Bis(1,1-dimethylethyl)-4-methylphenol (BHT)</td>
<td>98</td>
<td>0.0313</td>
<td>0.0650</td>
</tr>
<tr>
<td>17.8</td>
<td>( \text{H}_2\text{C}-(\text{CH}<em>2)</em>{16}-\text{OH} )</td>
<td>1-Octadecanol</td>
<td>94</td>
<td>1</td>
</tr>
</tbody>
</table>

Figure 2. Schematic representation of chemical transformations occurring in the course of the oxidative cleavage of the Si–C bond for heated samples of the chemically bonded stationary phase.


7. Prus, W.; Changes in the UV absorption of silica containing aliphatic moieties after thermal modification, and the quantitative relationship between the changes and the organic group content; *Acta Chromatographica*, (2005); 15: 150–161.

8. Prus, W.; Relationship between Raman scattering intensity in the aromatic region and the thermally induced increase in the UV absorption of chemically bonded stationary phases; *Acta Chromatographica*, (2006); 16: 204–215.


