Quantitative analyses of fullerene and polycyclic aromatic hydrocarbon mixtures via solvent-free matrix-assisted laser desorption/ionization mass spectrometry

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We explore the feasibility of reliable quantitative matrix-assisted laser desorption/ionization mass spectrometry (MALDI-MS) analyses via solvent-free sample preparation, as this procedure provides the unique convenience of being applicable also to insoluble samples. As quantitative MALDI measurements are even more complicated for species ionized by cation attachment, we investigated model systems, such as polycyclic aromatic hydrocarbons (PAHs) and fullerenes, which undergo photoionization and do not require additional cationizing salts. Our quantitative approach rests upon applying the standard-addition method in MALDI for the quantitative characterization of binary mixtures. Two different systems are tested. Set 1 is composed of hexakis(dodecyl)hexabenzocoronene and hexakis(dodecyl)hexaphenylbenzene, which represent the product and precursor of a cyclodehydrogenation reaction, and Set 2 is a mixture of C60 and C70 fullerenes. In Set 1, severe anomalies could be detected due to a strong influence of the matrix/analyte ratio on the correlation between signal intensity and analyte amount. This can be related to the strong intermolecular interactions among the hexabenzocoronene (HBC) aromatic cores hampering the desorption step and to intermolecular charge transfers, which influence the ionization probability. Minor interferences to the quantitative MALDI characterization are encountered in the analysis of C60 and C70 fullerenes. The spherical shapes of C60 and C70 buckyballs prevent strong aggregation. Thus, no molecule-dependent anomalies in their desorption-photoionization behaviour are recognized. Copyright © 2008 John Wiley & Sons, Ltd.

The acquisition of reliable quantitative information via matrix-assisted laser desorption/ionization mass spectrometry (MALDI-MS) represents a big challenge. The qualitative characterization of polymers and macromolecules is straightforward, as the MALDI approach is sensitive, fast and flexible.1–3 Despite these potential benefits, quantitative determinations by MALDI are complicated by poor reproducibility of this technique due to the non-uniform nature of the matrix/analyte mixture, different procedures of sample preparation or instrumental parameters. Here we examine the benefits of an internal standard as a means of overcoming these difficulties. It has been reported that the use of standards labeled with stable isotopes, such as 2H, 13C, 15N, and 18O, delivers good results in terms of quantification accuracy.4,5 In addition, the use of structurally modified compounds as internal standards has been successfully employed for the relative quantification of low molecular weight compounds.6 To date, however, standard molecules with properties resembling those of the analyte have been used to quantify a single component in a mixture. The method proposed here also has the aim of allowing quantification of both components of a binary mixture by using one of them as an internal standard. This is of special value for a slightly soluble or insoluble component not obtainable as a pure sample.

Most quantitative MALDI-based investigations have used the traditional solvent-based technique for sample preparation. However, this approach often leads to the so-called hot-spot formation due to severe inhomogeneities caused by solvent evaporation, leading to a large variation in relative signal intensities. Recently, it has been shown that improved homogeneity can be achieved by employing an alternative preparation technique, which avoids the use of a solvent, the so-called solvent-free sample preparation.7,8 This technique not only overcomes the most encountered problems of solubility, miscibility, and segregation effects during crystallization resulting from unfavorable analyte and matrix polarities, but also allows the characterization of insoluble samples.9 The many benefits of this preparation technique
and the introduction of a standard molecule could offer the possibility of more reliable quantitative characterization by MALDI-time-of-flight (TOF)MS for either soluble or insoluble samples.

As quantitative MALDI characterization is also affected by interferences caused by the type and the concentration of additional cationizing salts for the ionization of the analytes, this work is centered on the MALDI-based quantitative analyses of polycyclic aromatic hydrocarbons (PAHs) and fullerenes, representative of samples which undergo photo-ionization during MALDI and desorb as radical cations (M⁺).

For this investigation we have chosen hexabenzocorone (HBC) among the PAH family, which is a graphite-like molecule characterized by the presence of an extended aromatic π system. The overlap of the π orbitals from neighboring molecules allows the formation of columnar aggregates, which have promising electronic properties for novel organic electronic devices.10–13 The most important issue for improved electronic properties is the absence of impurities and solvent-free MALDI-MS has already been used to detect traces of byproducts or starting materials, which arise from incomplete reactions during synthesis.6 Here, we explore the possibility of obtaining not only qualitative but also quantitative information for a model system composed of the HBC-C₁₂ molecule and its precursor (hexakis(dodecyl)hexaphenylbenzene HPB-C₁₂) in the cyclodehydrogenation reaction (HPB-C₁₂ → HBC-C₁₂). Our approach rests upon using HPB-C₁₂ (the starting material) as the internal standard while applying the standard-addition method15 to the MALDI analyses, in order to develop a general procedure for measuring the yield of reactions from binary mixtures when only one of the two components involved is available as a pure sample.

Furthermore, the feasibility of quantitative MALDI analyses by employing the traditional solvent-based sample preparation method for the MALDI analyses is tested, in order to compare results from the two different preparation techniques.

In addition, quantitative MALDI-based analyses of fullerene mixtures are performed, as they can be characterized by independent methods to evaluate the reliability of the MALDI-based approach.

**EXPERIMENTAL**

**Materials**

The syntheses of hexakis(dodecyl)hexabenzocorone (HBC-C₁₂) and its precursor hexakis(dodecyl)hexaphenylbenzene (Set 1) have been reported elsewhere.14 C₆₀ and C₇₀ fullerene (Set 2) were obtained from Sigma-Aldrich (Munich, Germany) and used without further purification. The molecular structures of the analytes are reported in Scheme 1.

![Scheme 1. Structures of the analytes.](attachment:scheme1.png)
7,7,8,8-Tetracyanoquinodimethane (TCNQ, 98%) was purchased from Acros Organics (Geel, Belgium) and used without further purification. Tetrahydrofuran (THF, for analysis, ACS) was obtained from Acros Organics. Fullerite, a commercially available fullerene mixture, was purchased from Sigma-Aldrich.

**MALDI instrument and calibration**

MALDI analyses were performed using a Bruker Reflex II mass spectrometer (Bremen, Germany) operating in reflection mode. The instrument was equipped with a nitrogen laser (wavelength 337 nm) and the MALDI spectra were recorded with a pulse rate of 3 Hz. The ions were accelerated by a voltage of 20 kV and detected using a micro-channel plate detector. Mass calibration was carried out before each measurement by using fullerite.

**MALDI sample preparations**

**Solvent-free method (general procedure)**

The analytes were milled together with the TCNQ matrix using a MM 2000 ball mill from F. Kurt Retsch and Co. KG (Haan, Germany). The powder was then spread on a polished stainless steel sample holder with a flat spatula, while moving the sample position. For each sample a composite MALDI spectrum was acquired by summing 500 single shots. All the MALDI spectra were recorded with the same laser power. The best conditions for the quantitative characterization were found by adjusting the laser power to well above the desorption-ionization threshold of the analytes.

**Set 1:** Quantitative MALDI analysis for the system HPB-C12/HBC-C12

In order to perform quantitative MALDI analyses on the binary mixture HPB-C12/HBC-C12, the amount of HPB-C12 was varied, whereas the amounts of HBC-C12 and TCNQ were kept constant. HPB-C12 and HBC-C12 were mixed in the ratios listed in Table 1. MALDI spectra were measured and the intensity ratios were calculated and expressed as Int.(HPB-C12)/Int.(HBC-C12) as listed in Table 2.

For the sake of simplicity, during quantitative analyses, the signal intensities of the peaks were evaluated instead of their relative areas, once the non-variance of the results had been established.

**Solvent-based method**

In order to reach the best crystallization conditions, THF was selected as a common solvent for samples and matrix to perform quantitative MALDI analyses on the mixture HPB-C12/HBC-C12. Both components were dissolved in THF at a concentration of 0.061 mg/mL (4.00 × 10⁻⁵ M) and 0.070 mg/mL (4.5 × 10⁻⁵ M), respectively, while TCNQ was dissolved in THF at a concentration of 2.5 mg/mL (1.2 × 10⁻⁴ M). Seven mixtures of the two components were prepared with different molar ratios as listed in Table 3. The quantities of HPB-C12 were varied, whereas the concentration of HBC-C12 in the mixtures was kept constant.

The responses for the different molar ratios, obtained from the MALDI measurements and expressed as Int.(HPB-C12)/Int.(HBC-C12), are reported in Table 4.

**Set 2:** Quantitative MALDI analysis for the system C70/C60

**Solvent-free method**

To quantitatively characterize binary mixtures of C70/C60 fullerenes via MALDI-TOFMS, the amount of C70 was varied, whereas the amounts of C60 and TCNQ were kept constant, analogous to the procedure in Set 1. Table 5 reports the composition of the different mixtures. The fullerite powder with unknown composition was mixed with the TCNQ matrix in a mass ratio of 1:407.

The responses for the different molar ratios, obtained from the MALDI measurements and expressed as Int. C70/Int. C60, are reported in Table 6.

**HPLC characterization of C70/C60 mixtures**

High-pressure liquid chromatography (HPLC) analyses of the C70/C60 mixtures were performed using a C18 reversed-phase column coupled with a diode-array detector working...
at a fixed wavelength of 330.2 nm, which was selected because of the strong absorption of the analytes at this wavelength. A solvent mixture composed of toluene and acetonitrile (CHROMASOLV® Plus, for HPLC, ≥99.9%) was chosen as eluent for the chromatographic analyses. Solutions of C60 and C70 were prepared by dissolving C60 and C70, respectively, in a 4:1 solvent mixture of 1,2,4-trichlorobenzene/toluene (0.20 mg/mL). To establish the calibration curve for the system C70/C60, solutions of C70/C60 mixtures were prepared with the molar ratios reported in Table 7. Three injections were performed for each sample, and the resulting data expressed as C70/C60 peak area ratios were then averaged and plotted against the respective C70/C60 molar ratios (Table 8).

The commercially available fullerene mixture, known as fullerite, was dissolved in a 4:1 solvent mixture of 1,2,4-trichlorobenzene/toluene (0.20 mg/mL) and analyzed by HPLC.

### RESULTS AND DISCUSSION

#### The standard-addition method

Our approach to the quantification rests upon applying the standard-addition method in MALDI analyses with the aim of quantitatively characterizing binary mixtures of unknown compositions. This work proceeds through the following steps:

1. A model system composed of a binary mixture is analyzed by MALDI;
2. One of the two analytes (analyte 1) is subsequently added to the initial mixture and for each new mixture a MALDI spectrum is recorded;
3. The amount of analyte 1 in the initial binary mixture is determined by evaluation of the calibration curve;
4. The amount of the second analyte (analyte 2) is calculated from the weight of the initial mixture.

### MALDI quantification of HBCs

For the quantitative characterization of a cyclodehydrogenation reaction, we have chosen HBC-C12 and its precursor molecule HPB-C12 as a model system. An HPB-C12/HBC-C12 mixture (1:1 in mass ratio), which simulates the combination of components of an incomplete cyclodehydrogenation reaction, was prepared. A MALDI analysis was then carried out by employing solvent-free sample preparation and the corresponding spectrum is reported in Fig. 1(a). Subsequently, defined amounts of HPB-C12 molecules were added stepwise to the initial mixture (Table 1), while recording the resulting MALDI spectra (Figs. 1(b)–1(g)). For each measurement the laser intensity was much higher than the desorption-ionization threshold of the analytes, which was recognized as a necessary requirement for a reliable MALDI-based quantification.

Only two abundant ions are present in the spectra, for HBC-C12 at $m/z$ 1532.3 and for HPB-C12 at $m/z$ 1544.0. No adducts due to protonation or cationization of the analytes are detected. Thus, any potential interference to the quantitative MALDI analyses can be ruled out. The spectrum of the HPB-C12/HBC-C12 mixture of mass ratio 1:1 displays strong underestimation of the precursor species.16 Quantification was achieved by calculating the intensity ratios from the MALDI spectra, expressed as Int.(HPB-C12)/Int.(HBC-C12), and these are listed in Table 2. The Int.(HPB-C12)/Int.(HBC-C12) values obtained for the other mixtures (mixture 2 to mixture 7) were then plotted versus the added weight of HPB-C12 standard molecules.

### Table 4. Measured intensity ratios of the system HPB-C12/HBC-C12 for different molar ratios

<table>
<thead>
<tr>
<th>Mixture</th>
<th>Int.(HPB-C12)/Int.(HBC-C12) SD</th>
<th>Molar ratio HPB-C12/HBC-C12</th>
<th>μL of HPB-C12 solution</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.254 0.048</td>
<td>1.12</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>0.370 0.116</td>
<td>1.41</td>
<td>1</td>
</tr>
<tr>
<td>3</td>
<td>0.422 0.086</td>
<td>1.7</td>
<td>2</td>
</tr>
<tr>
<td>4</td>
<td>0.473 0.080</td>
<td>2.0</td>
<td>3</td>
</tr>
<tr>
<td>5</td>
<td>0.483 0.086</td>
<td>2.25</td>
<td>4</td>
</tr>
<tr>
<td>6</td>
<td>0.621 0.093</td>
<td>2.53</td>
<td>5</td>
</tr>
<tr>
<td>7</td>
<td>0.665 0.199</td>
<td>2.81</td>
<td>6</td>
</tr>
</tbody>
</table>

SD: standard deviation.

### Table 5. Composition of the mixtures for the system C60/C70 prepared via solvent-free sample preparation

<table>
<thead>
<tr>
<th>Mixture</th>
<th>C60/mg</th>
<th>C70/mg</th>
<th>TCNQ/mg</th>
<th>A/M molar ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.40</td>
<td>0.17</td>
<td>200.29</td>
<td>1:1294</td>
</tr>
<tr>
<td>2</td>
<td>0.38</td>
<td>0.40</td>
<td>199.31</td>
<td>1:973</td>
</tr>
<tr>
<td>3</td>
<td>0.37</td>
<td>0.77</td>
<td>202.65</td>
<td>1:693</td>
</tr>
<tr>
<td>fullerite</td>
<td>0.50</td>
<td>0.20</td>
<td>203.55</td>
<td>Mass ratio: 1:407</td>
</tr>
</tbody>
</table>

### Table 6. Measured intensity ratios of the system C70/C60 for different molar ratios

<table>
<thead>
<tr>
<th>Mixture</th>
<th>Molar ratio C70/C60</th>
<th>Int. C70/Int. C60</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.36</td>
<td>0.680</td>
<td>0.037</td>
</tr>
<tr>
<td>2</td>
<td>0.90</td>
<td>1.625</td>
<td>0.084</td>
</tr>
<tr>
<td>3</td>
<td>1.78</td>
<td>2.432</td>
<td>0.067</td>
</tr>
<tr>
<td>fullerite</td>
<td>x</td>
<td>0.203</td>
<td>0.010</td>
</tr>
</tbody>
</table>

SD: standard deviation.
The obtained plot shown in Fig. 2 clearly displays two regimes. For small amounts of added standard molecules, the MALDI response increases linearly. For HPB-C_{12}/HBC-C_{12} mass ratios larger than 4.6, however, the instrument response deviates significantly from linearity. The experimental data points belonging to the linear regime can be interpolated by a straight line of equation \( y = 0.3789x + 0.1126 \) with a correlation coefficient \( R^2 \) of 0.9833. According to the standard-addition method, the absolute value of the x-intercept is the concentration of HPB-C_{12} in the initial mixture (mixture 1). Therefore, through extrapolation of the calibration line \( y = 0.383x + 0.1069 \) to \( y = 0 \), the amount (mg) of standard molecules in the mixture 1 is calculated to be 0.28 mg. This mass value differs from the true one by less than 4%.

The deviation from linearity of the calibration curve for HPB-C_{12}/HBC-C_{12} mass ratios larger than 4.6 can be explained by considering the total analyte-to-matrix (A/M) ratio, which increases significantly during the addition of the HPB-C_{12} standard molecules. This strongly affects the MALDI response for sample/matrix molar ratios lower than 1:850. The matrix presumably plays the role of disturbing the strong interactions between HBC molecules and it works optimally only when a certain sample/matrix threshold has been surpassed. Furthermore, low A/M ratios may favor suppression phenomena, which are emphasized for the system HPB-C_{12}/HBC-C_{12} by the different desorption-photoionization efficiencies of the analytes. According to previous investigations, HBC-C_{12} molecules create radical cations much more easily than their HPB-C_{12} precursors. For low A/M ratios, the higher contact probability between the analytes may enhance charge transfer phenomena between HPB-C_{12} and HBC-C_{12} molecules with an increased formation of HBC-C_{12} species. The importance of matrix dilution during the desorption-photoionization process has already been proven for similar systems. Within this work it was recognized that not only has a suitable matrix to be employed to avoid fragmentation and laser-induced reactions among the analyte molecules, but also that the amount of matrix has to exceed a threshold to avoid strong deviation from the linearity. Further experiments have also shown that the deviation from the linearity can be corrected by increasing the analyte-to-matrix dilution up to the threshold value of 1:850.

Comparative quantitative analyses for the system HPB-C_{12}/HBC-C_{12} were performed using solvent-based sample preparation. An equimolar mixture of HBC-C_{12} and HPB-C_{12} was prepared and analyzed via solvent-based MALDI (Fig. 3). Defined volumes of the HPB-C_{12} solution in THF

![Figure 1](image_url)

**Figure 1.** (a) MALDI spectrum of an HPB-C_{12}/HBC-C_{12} mixture 1:1 in mass ratio. (b–g) MALDI spectra of HPB-C_{12}/HBC-C_{12} mixtures with a fixed amount of HBC-C_{12} molecules and increasing amounts of HPB-C_{12}, prepared by the solvent-free sample preparation method.

<table>
<thead>
<tr>
<th>Mixture</th>
<th>Molar ratio C70/C60</th>
<th>Area ratio C70/C60</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>0.951</td>
<td>0.054</td>
</tr>
<tr>
<td>2</td>
<td>0.66</td>
<td>0.742</td>
<td>0.044</td>
</tr>
<tr>
<td>3</td>
<td>0.5</td>
<td>0.632</td>
<td>0.065</td>
</tr>
</tbody>
</table>

SD: standard deviation.

Table 8. Measured signal area ratios of the system C70/C60 for different molar ratios by HPLC

were subsequently added. The MALDI spectra of HPB-C12 and HBC-C12 mixtures with varying molar ratios are reported in Figs. 3(b)–3(g). As opposed to the solvent-free sample preparation, two abundant ions and a weak one are visible. The ions at $m/z$ 1532.3 and 1544.5 are related to the radical cations of HBC-C12 and HPB-C12 molecules, respectively, whereas the ion at $m/z$ 1567.6 is due to sodiated HPB-C12 ($\text{HPB-C12}^+ + \text{Na}^+$). It is interesting to point out that, by employing the solvent-based sample preparation, HPB-C12 molecules undergo photoionization and formation of radical cations and supplementary cation attachment to some extent. This suggests the presence of additional species, which could represent an obstacle to a reliable quantitative investigation. The presence of sodiated adducts was not recorded at all when employing the solvent-free preparation procedure. Analogously to the quantitative work, carried out via solvent-free MALDI-MS, the standard-addition method was applied in solvent-based MALDI analyses, by plotting the responses (expressed in Table 4 as intensity ratios $\text{Int.}(\text{HPB-C12})/\text{Int.}(\text{HBC-C12})$) against the increasing amount of HPB-C12 expressed in $\mu$L of added solution (Fig. 4). The sodiated HPB-C12 species were neglected.

The data points obtained via solvent-based sample preparation could be interpolated by a straight line. In contrast to the solvent-free sample preparation, no strong deviations from linearity were observed by increasing the

Figure 2. Plot of the intensity ratios $\text{Int.}(\text{HPB-C12})/\text{Int.}(\text{HBC-C12})$ versus added amount of HPB-C12.

Figure 4. Intensity ratios $\text{Int.}(\text{HPB-C12})/\text{Int.}(\text{HBC-C12})$ versus added $\mu$L of HPB-C12.

Figure 3. (a–g) MALDI spectra of HPB-C12/HBC-C12 mixtures with varying molar ratios, obtained via solvent-based sample preparation.
analyte-to-matrix concentration (Fig. 4). This phenomenon could be related to use of the solvent during the sample preparation procedure, which weakens the intermolecular $\pi-\pi$ interactions among the HBC cores. From evaluation of the data points and extrapolation of the straight line in Fig. 3, the volume of HPB-C$_{12}$ solution (µL) present in the mixture 1 (Table 4) was calculated to be 4.3 µL. This value differs from the true amount of 4.0 µL by 7.5%. However, it has to be pointed out that quantitative solvent-based MALDI analyses could be carried out only after overcoming the problems related to the presence of hot-spots. In fact, via performing solvent-based MALDI analyses, cocryotallization and segregation phenomena, which caused large variations in signal intensities, were encountered. As a result, the data points, reported in Fig. 4, possess a large error bar. The interferences due to the presence of hot-spots could be overcome by increasing the number of measurements for each sample mixture via dropping solutions with the same concentration of analytes in three different sites on the sample holder and summing 500 single shots for each site to give an average spectrum. Such homogeneity problems were not encountered during solvent-free MALDI analysis, which required many fewer measurements and which was therefore recognized to be the most favorable preparation procedure for quantitative analysis.

**Quantitative analysis of fullerenes**

Quantitative MALDI analyses of fullerenes were conducted as further proof for the method with the additional opportunity to couple the mass spectrometric results with an independent quantitative procedure. C$_{60}$ and C$_{70}$ buckyballs undergo photoionization with formation of radical cations, but, due to their spherical shape, should not lead to complications due to strong aggregation. Thus,

**Figure 5.** (a) MALDI spectra of the mixtures C$_{70}$/C$_{60}$ with different molar ratios. (b) MALDI calibration curve for the system C$_{70}$/C$_{60}$. (c) HPLC calibration curve for the system C$_{70}$/C$_{60}$. (d) MALDI spectrum of commercial fullerite mixture. (e) HPLC chromatogram of the commercial fullerite mixture.
they were chosen as another model system for this study. Mixtures of C60 and C70 fullerenes were prepared as described in Table 5 and MALDI spectra were recorded analogously to the previous measurements (Fig. 5(a)). Two abundant ions at m/z 720.3 and 840.6 are present corresponding to the C60 and C70 radical cations, respectively. A small amount of impurities, due to higher-mass fullerenes, is also present. In contrast to the previous system HPB-C12/HBC-C12, C60 and C70 molecules possess comparable photoionization-desorption efficiencies. This minimizes problems related to suppression phenomena, which have been encountered in the characterization of disc-shaped molecules, and thus simplifies the quantitative characterization. As shown in Fig. 5(a), the MALDI measurement responds linearly to the increase of one analyte (C70) and the data points expressed as Int.C70/Int.C60 versus the molar ratio of the C70/C60 mixtures (Table 6) can be interpolated by a straight line. It is interesting to point out that, for this system, no strong deviations from linearity were observed by increasing the analyte-to-matrix concentration. Probably, spherical-shaped C60 and C70 fullerenes aggregate less strongly than large and planar aromatic systems, which can assemble into columnar structures. Consequently, their desorption behavior is less influenced by variation in the dilution of the analytes with the matrix.

The measured calibration curve of the equation (y = 1.3554x) has a satisfactory R² value of 0.9749. Therefore, it was utilized to measure the C70/C60 amount in a commercially available mixture of fullerenes (fullerite). The MALDI spectrum for the fullerite mixture of unknown molar ratio is reported in Fig. 5(d). From the MALDI spectrum the intensity ratio C70/C60 is calculated to be 0.203 ± 0.010, which corresponds to a molar ratio of 0.150 ± 0.060, calculated by using the calibration curve of Fig. 5(b).

To check the accuracy of the quantitative MALDI analyses by an independent method, the fullerite mixture was also characterized via HPLC. A calibration curve for the HPLC analyses is obtained by mixing fullerenes C60 and C70 in known ratios (Table 7). The corresponding plot is reported in Fig. 5(c). The data points can be interpolated by a straight line of equation y = 0.8747x and R² of 0.9939. The HPLC response for the fullerite mixture is plotted in the calibration curve and the molar ratio C70/C60 is estimated to be 0.170 ± 0.041. Within experimental error the two values are consistent. This result therefore strengthens the feasibility of quantitative analysis via MALDI-TOFMS.

CONCLUSIONS
This work describes a novel method for the quantitative analysis of PAHs and fullerenes via MALDI-TOFMS. A laser power well above the desorption threshold, high matrix dilution of the analytes and maintaining a constant A/M ratio have been identified as prerequisites for a reliable quantitative response. During this study we recognized that the strong intermolecular interactions among large planar aromatic π systems are the largest obstacle to achieving reliable quantification. Charge transfer phenomena between the analyte molecules are in fact enhanced by high concentrations of the analyte-to-matrix ratios, especially when the analytes in the mixtures possess significantly different desorption and photoionization efficiencies, such as the disc-type HBC-C12 and its dendritic precursor HPB-C12 in the cyclodehydrogenation reaction. The homogeneity problems, encountered via solvent-based sample preparation, could be avoided by using the solvent-free sample preparation, which simplifies quantification and results in improved accuracy.

Within this investigation we explored the possibility of using the standard-addition method in MALDI analyses to determine the yield of reactions from binary mixtures when only one of the two components involved is available as a pure sample. The obtained results pave the way to quantifying an insoluble component in a binary mixture and therefore represent a new method for the quantification of intractable analytes. Its applicability to the quantitative characterization of natural PAH mixtures, such as pitches, heavy oils and asphaltenes, for which our samples represent suitable model systems, is the topic of further investigations.

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