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Short communication

Advantages and limitations of laser desorption/ionization mass spectrometric techniques in the chemical characterization of complex carbonaceous materials

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ABSTRACT

Laser desorption/ionization techniques coupled with mass spectrometry analyzers have evolved rapidly in the recent years and are currently capable of providing valuable information about the chemical composition and structure of very high molecular weight species, mainly biopolymers or synthetic polymers.

In view of this rapidly increasing interest a thorough understanding of the desorption/ionization process is not only of scientific interest, but also important for a correct spectra interpretation and for further improvements of the technique.

In the present paper, the effect of main experimental parameters on mass range detectable by laser desorption/ionization techniques has been investigated for standard aromatic molecules, like polycyclic aromatic hydrocarbons (PAH), fullerenes, polyacenaphthylene (PACE) and for complex carbonaceous materials like heavy fractions of fuel oils and combustion-formed particulate. In particular, it has been shown that laser power (or more specifically, the surface power density of the laser spot, named laser fluence) as well as the surface concentration of samples are crucial parameters controlling the highest detected molecular weight range. However, neither of these parameters is easily well-controlled and therefore more work is necessary for the standardization of laser/desorption techniques in the analysis of complex samples.

1. Introduction

Laser desorption/ionization (LDI) techniques were firstly developed in the early seventies [1] but only in the late eighties [2,3], with the introduction of matrix-assisted laser desorption/ionization (MALDI), they became an established method for the mass spectrometry of macromolecular compounds. The role of matrix is to absorb UV laser radiation and to give the energy to the analyte, often not absorbing in UV region, for ionizing it in a softer way. However, many polycondensed systems, especially with aromatic moieties and, therefore, strong UV absorption, have the so-called "been self-matrix" property [4,5] that means the capability of the sample (or a part of it) to act as matrix by itself. In this case, their direct photoionization takes place by laser irradiation without the necessity of an external matrix addition.

The physicochemical nature of the desorption/ionization process is still not fully understood as several parameters have a strong influence on it, such as, for example, the laser wavelength and pulse width, the laser fluence and its profile on the sample, the properties of analyte, etc. However, a thorough understanding of desorption/ionization processes is crucial for exploiting the enormous potentiality of the technique in the analysis of high molecular weight and structurally complex samples.

In the present paper, the effect of the main experimental parameters on the mass range detectable by LDI techniques has been investigated for standard aromatic molecules, like polycyclic aromatic hydrocarbons (PAH), polyacenaphthylene, fullerenes and for complex carbonaceous materials like heavy fractions of fuel oils and combustion-formed particulate.

The advantages and limitations of such powerful techniques in the analysis of polydisperse and chemically heterogeneous samples have been critically examined. In particular, the spectra reported in the present study, acquired in different operative conditions, put in evidence the artifacts that can lead to a misleading interpretation of the signals obtained by LDI techniques.

Therefore, a critical evaluation of the LDI data, taking into account for the parameters used, has been suggested based on the analysis of standard molecules and extended to ill-structurally defined complex mixtures.

2. Experimental

2.1. Materials

Standard compounds: PAH mixture is from Supelco (EPA 610). Fullerenes C60 is from Sigma Aldrich. Polyacenaphthylene (Sigma

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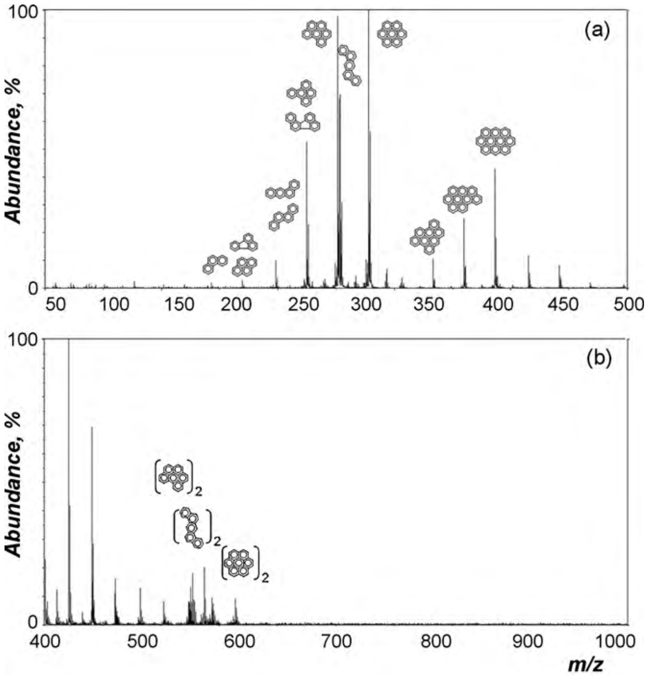

Fig. 1. Linear LDI-MS spectra of a synthetic mixture of PAH in two m/z ranges: (a) 50–500; (b) 400–1000.

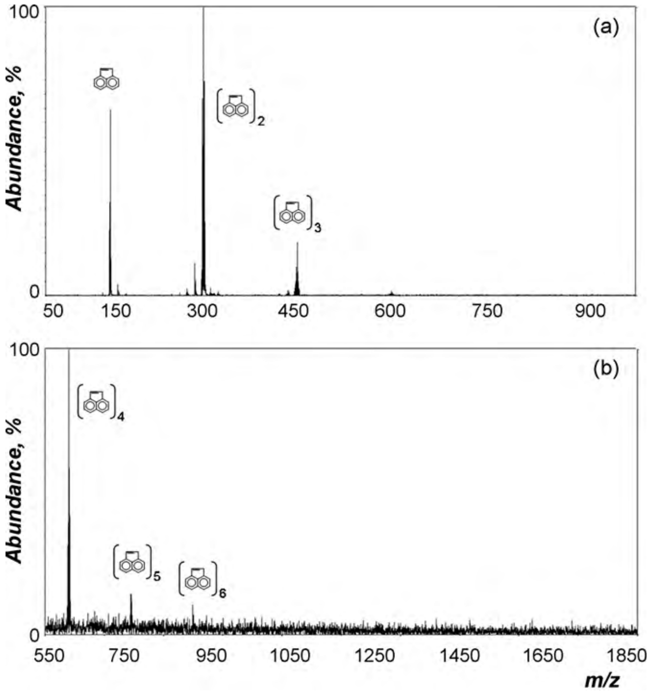

Fig. 2. Linear LDI-MS spectra of PACE in two m/z ranges: (a) 50–1000; (b) 550–1900.

Aldrich), called PACE in this work, is a yellow solid material synthesized by radical polymerization of acenaphthylene (MW 5000–10,000 u) [6].

Samples: Asphaltenes are the alkane insoluble/aromatic soluble fraction of a #6 commercial heavy fuel oil separated by ASTM (D200-75) standard procedure.

The carbonaceous particulate was sampled in a fuel-rich premixed laminar ethylene/oxygen flame (equivalence ratio $\Phi = 3.03$), produced on a commercial McKenna burner. Solid and condensed material collected on the probe walls, on the Teflon filter and in an ice-cooled trap placed in the sampling line was extracted by dichloromethane (DCM) to separate the DCM soluble organic material (named soot extract), from the insoluble solid carbonaceous material (soot). More details are reported in previous works [7,8].

2.2. Techniques

LDI-TOF-MS. Positive linear laser desorption ionization-time-of-flight mass spectrometry spectra were recorded on a Voyager DE STR Pro instrument (Applied Biosystems, Framingham, MA). The target was prepared by depositing a volume variable from 1 and 10 μl of a solution of the sample, dissolved in DCM, on the metallic sample plate. Acceleration and linear voltages set up were: target voltage 25 kV, first grid at 96% of target voltage, delayed extraction at 100–150 ns. More details are given in a previous paper [4].

AP-LDI-MS. The atmospheric pressure laser desorption ionization mass spectra were obtained with an Agilent 1100 Series MSD Trap (Agilent Technologies, Palo Alto, CA, USA). More details are given in a previous paper [9].

3. Results and discussion

The LDI-MS spectra presented in this work are collected in linear mode, in order to get the maximum possible sensitivity in the larger molecular weight range detectable, even though with lower resolution in respect to reflector mode.

The LDI-MS spectrum of a synthetic mixture of PAH from naphthalene to coronene (128–300 u) is reported in Fig. 1. The spectrum was measured using 60% of maximum laser power. In Fig. 1a, the spectrum is reported in the range m/z 50–500, covering the mass range of the mixture, extending up to 300 u, and showing some clusters in the range m/z 300–500. In Fig. 1b the spectrum is reported in a larger range, up to 1000 u, in order to put in evidence the formation of PAH clusters.

It is known that PAH readily aggregate upon desorption under favorable conditions of density and laser power and that the intensity and size of clusters detected increase with the number of fused rings [10]. However, in Fig. 1 it is possible to easily distinguish above 300 u, peaks with gaps among them at 12, 24 and 26 u, ascribable to the growth of PAH for an insertion of a CH_2 bridge followed by hydrogen elimination (12 u gap) and a net sequential addition of C_2 as an ethylene bridge (24 u gap) eventually followed by isomerization (mass increment of 26 units) [4]. The presence of dimer, trimer and bigger homologs of PAH present in the mixture is visible only in the high MW range (552, 556 and 600 u dimers of 276, 278 and 300 u respectively). Therefore, apart from clusterization, growth reactions appear to occur in the laser plume, when high concentration (5 μl of solution on the target spot) and high laser power (60% of the maximum power) are used, as in the case of spectra in Fig. 1. Decreasing the surface concentration and/or the laser power, results in the disappearance of the higher molecular weight (MW) peaks, above 300 u. However, a further increase of concentration and laser power also causes the disappearance of the higher MW peaks, probably because the higher concentration hinders the desorption of the sample, reducing the plume ionic concentration and too high laser power causes a simultaneous cluster fragmentation, globally causing no clusters detection.

On the other hand, the presence in aromatic structures of alkane substituents reduces aggregation largely because of steric effects [10] and also favors fragmentation. To verify this effect a polymer like PACE, with two rings aromatic systems linked among them by aliphatic bridges, presented a strong fragmentation, at the same laser power conditions used for the PAH mixture (60% of the maximum power). Thus, it was not possible to obtain a signal corresponding to its average MW (about 8000 u). In Fig. 2 the LDI spectrum of PACE in linear configuration is reported in two ranges:

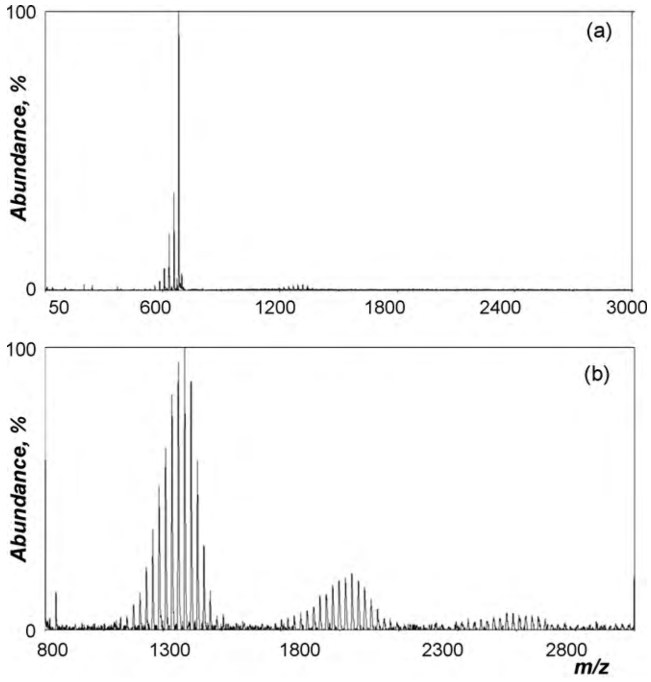

Fig. 3. Linear LDI-MS spectrum of fullerene in the *m*/*z* range 50–3000 (a) and its zoom (b).

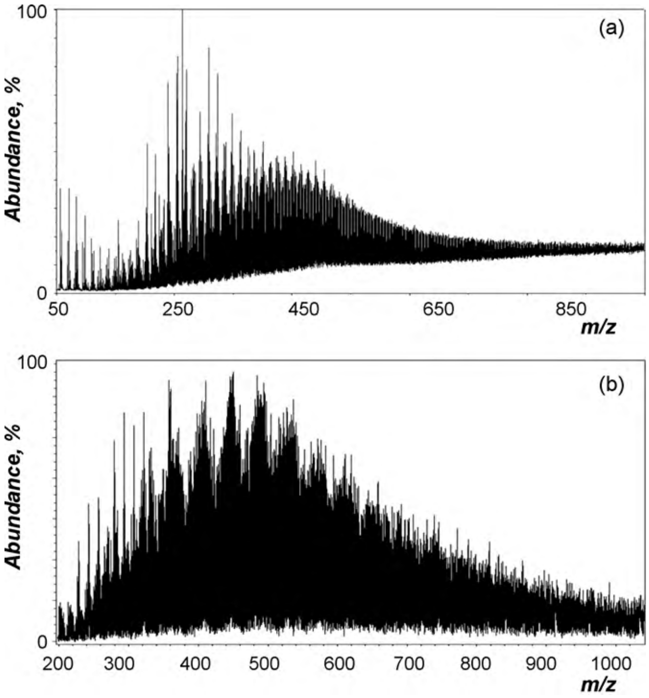

Fig. 4. Spectra of asphaltenes acquired by LDI-MS (a) and AP-LDI-MS (b).

m/*z* 50–1000 (Fig. 2a) and *m*/*z* 550–1900 (Fig. 2b). Peaks between 152 and 911 u with gaps at 152, which is the mass of the monomer (acenaphthylene) can be observed whereas no other signals beyond 2000 u was detected (not reported here).

In the case of fullerene, whose spectrum is reported in Fig. 3, parent peak at 720 u, with the typical isotopic pattern dominates the spectrum (Fig. 3a) whereas no fragmentation is observed with the same laser fluence (60% of maximum laser power) with respect to those used for the previous spectra. Fullerene clusters in the range *m*/*z* 1000–3000 are observed in Fig. 3b, with gaps of 600 u, corresponding to C_{50}.

In order to investigate the potentiality of LDI technique, also a much more complex carbonaceous sample, constituted of mixed aromatic/aliphatic moieties, as fuel oil asphaltenes, has been analyzed and the relative spectrum reported in Fig. 4.

The real molecular weight distribution of asphaltenes from fuel oils, coal or petroleum, is a widely debated subject. Briefly, mass spectrometric measurements with electrospray, chemical and field desorption ionization, along with florescence-based diffusion measurements and electron microscopy experiments that relate molecular size to molecular weight, suggest an average MW in the range 100–1000 u [11,12]. On the other hand, fast atom bombardment and plasma desorption mass spectrometry show a pronounced tail extending at times beyond 10,000 u, and size exclusion chromatography (SEC) gives a bimodal distribution containing an intense peak at >10^6 u, even though it can be overestimated due to steric effects [13,14]. Also by using LDI-MS, different groups have reported mean MW spanning 2 orders of magnitude for similar LDI setups and asphaltenes samples. However, recently, Pomerantz et al. [15] obtained data by using L^2MS–MS (two colors laser desorption MS) supporting the hypothesis that LDI asphaltene mass spectra peaking at >1000 u result from aggregation in the LDI plasma plume.

The asphaltene spectrum measured in the present work, reported in Fig. 4a, presents a mass distribution extending up to about 1000 u with a maximum around 400 u. This is in agreement with spectra reported by Traldi and co-workers [16], showing no clustering phenomena and MW distribution shifted at lower weight for asphaltenes from young fuel oils with respect to asphaltenes from aged heavy fuel oil, coal and petroleum. The low resolution obtained also in Reflectron conditions in early work [17] is strongly related to the complexity of the sample under examination. It is reasonably due to the high number of ionic components generated by laser irradiation; when present in high density, these ionic species are responsible for space charge phenomena, which lead to an enlargement of the kinetic energy distribution and the consequent peak broadening. The optimization of LDI setups has increased the quality of the spectra, but an apparent noise is still present, largely due to the partial resolution of peaks at every nominal mass in the complex mixture. Moreover, in the present work it is confirmed by using linear configuration what reported in a previous work [17]: asphaltene structure is so labile that can be subjected to fragmentation during ionization desorption, as the same samples, analyzed by atmospheric pressure photoionization mass spectrometry, APPI-MS, presented a MW distribution shifted at higher masses, with a maximum around 600 u. Alternatively, asphaltenes contain species not ionizable by laser in UV range, but easily ionizable by photoionization.

Repeating the LDI analysis by using AP-LDI/MS, a spectrum very similar to that obtained under vacuum is obtained (Fig. 4b).

In the ion source of this system the ions are produced at normal atmospheric pressure differently from the conventional MALDI/LDI ion source where ions are formed inside the vacuum system of the mass spectrometer. The AP-LDI-MS demonstrated large tolerance to the laser fluence variations and minimal fragmentation of molecular ions due to fast thermalization of the ion internal energy at atmospheric conditions. Therefore, the probable fragmentation presented by asphaltenes also in this softer condition, never underlined in the previous works in literature, suggests caution in using LDI techniques in the asphaltene analysis.

The LDI-MS spectrum of a combustion-formed soot extract sample dissolved in DCM is reported, in different *m*/*z* ranges, in Fig. 5, by using similar laser power with respect to the previous spectra (60% of the maximum power).

In Fig. 5a it is possible to observe the typical PAH trends with gaps at 12 and 24–26 u [4]. However, in Fig. 5b and its inset it is

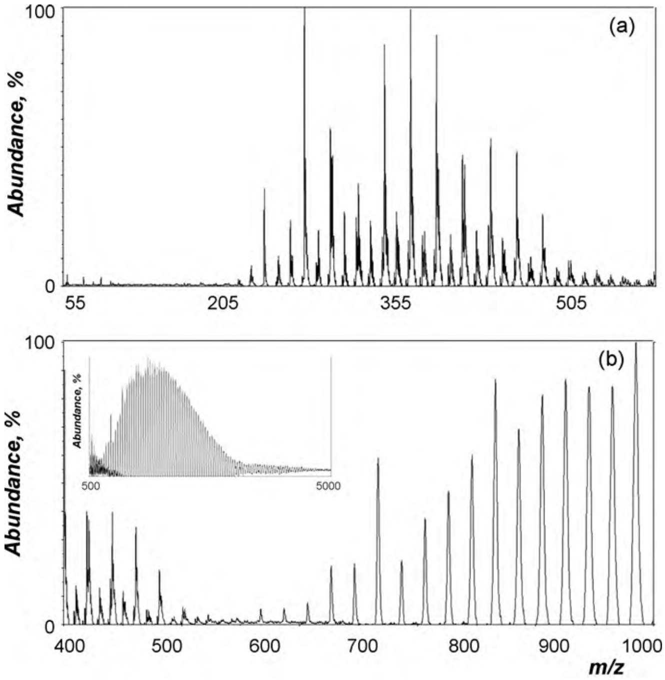

Fig. 5. Linear LDI-MS spectra of soot extract in three different *m*/*z* ranges: (a) 50–600; (b) 400–1000; (inset of figure b 500–5000).

observable that after 600 u the gap at 12 u largely decreases up to a complete disappearance, leaving place to only one sequence spaced at 24–26 u, extending up to about 3000 u. The mass difference in the detected peaks of only 24 u gap and their presence only in spectra acquired in linear configuration (for spectra of similar samples in reflectron mode see Refs. [4,17]) could give rise to the suspicion that they do not represent real ionic species, but originate by a clustering into the plume. Indeed, the high laser power used in low vacuum LDI conditions could induce clusterization of large PAH from the samples. However, the 24 u structure does not disappear even in different linear LDI setups. In particular, decreasing the delay time between the ionization and the extraction do not affect the high mass peak intensity.

Moreover, the high mass sequence is much more evident if lower sample concentration is used, in contrast with the clustering occurrence favored in higher concentration conditions.

Similar structures have been found in recent works [5,18] in asphaltene spectra, but only when the parent oil was aged and therefore, when the graphitic structure of asphaltenes was larger. In those papers, the clustering occurrence was ruled out as the spectra were acquired with different instruments with very different setups and 24 μm gaps were always present, but only in samples with extended graphitic moieties.

Soot extract samples have been analyzed also by AP-LDI-MS, after fractionation by SEC for improving sensitivity of ion trap MS, and the spectra were reported in a previous work [9].

The AP-LDI-MS spectra of the heaviest MW fractions separated by SEC (600–2000 u and 600–5000 u fraction) showed a very different pattern with respect to those reported in Fig. 5, appearing as a continuous spectrum of masses typical of polymeric structures, with gaps at 14 and 16 u.

These interesting features can be interpreted with a very intriguing hypothesis.

Two classes of species are present in soot extract and probably constitute soot precursors: the former has mainly aromatic moieties, extending up to about 3500 u (about 2 nm), with structure progressively becoming tridimensional for the insertion of pentagons that are located in the PAH periphery at the beginning (and therefore two PAH sequences are present, one attributable to even PAH and the other to odd-PAH [4]) and afterwards migrate inside the PAH skeleton [19], generating a curvature which brings the net back on itself to form a spheroidal shell which, in turn, can close to form a spherical soot particle; the second class has a polymeric structure with small aromatic moieties linked by aliphatic bridges and probably extends up to nanometric dimensions, but in AP-LDI spectra is observable just up to 1000 u, due to fragmentation occurring at high MW when aliphatic bridges are present.

The softer LDI conditions, due to atmospheric pressure in AP-LDI-MS and very lower laser power, can favour the desorption and therefore the detection of the polymeric structure, whereas the low vacuum and higher laser fluence in LDI-MS conditions hinder the polymeric structure detection for its complete fragmentation (as for PACE, Fig. 2) and allows to detect graphitic moieties, much more difficult to desorb and fragment.

This very fascinating speculation requires deeper investigations that are already in progress.

4. Conclusions

Under appropriate experimental conditions, laser ionization can be a very efficient process, and when combined with the high ion transmission of a time of flight mass spectrometer, excellent sensitivity can be achieved [20].

However, it was noteworthy that the MW distribution can be influenced by operating parameters. In particular, laser energy and sample concentration can lead, if too high, to the production of a high density plume, inside which clustering reaction can occur, with the consequent formation of ions at high *m*/*z* values, especially when condensed aromatic structures are present.

On the other hand, if the laser fluence is too high fragmentation can occur, in particular when aliphatic moieties are present and when low vacuum LDI is used.

Therefore, a critical evaluation of the LDI data has to be carried out especially when very complex mixtures are analyzed.

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