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Identifying the source of a strong fullerene envelope arising from laser desorption mass spectrometric analysis of meteoritic insoluble organic matter

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Abstract

Insoluble organic matter (IOM) has been obtained from two carbonaceous chondrite meteorites and subjected to analysis by laser desorption mass spectrometry (LDMS) using standard operating conditions that were optimized for fullerene detection (3–6 μJ pulses at 337 nm focused to a spot size of approximately 100 μm in diameter). The preparation process yields no free C_{60} in the IOM, and other experiments suggest that this material does not contain appreciable amounts of fullerenes. Nevertheless, a pronounced high-mass envelope is observed in LDMS, extending from 720 amu to about 4000 amu, with peaks spaced apart every 24 amu (corresponding to the gain or loss of C_2 units). We attribute this high-mass envelope to the existence of various fullerene molecules. The present work demonstrates that these fullerene molecules are created by the laser desorption laser ionization process under typical laser conditions used for studying free fullerenes in organic solvent extracts of natural samples (toluene and 1,2,4-trichlorobenzene). The implications of this false positive detection of fullerene molecules on the reports of fullerenes in other meteoritic samples have been investigated by introducing IOM into typical fullerene extraction procedures and examining the LDMS results. We found that IOM is capable of producing false positive signals in these experiments. The effect of ambient laboratory contamination producing fullerene signals is also described. It is found that extensive centrifugation of the meteoritic extracts is able to reduce the observed fullerene envelope, which points to an association of this envelope with IOM particulates that have passed through the filtering steps. We suggest the exercise of extreme caution in interpreting fullerene data from LDMS experiments.

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1. INTRODUCTION

Ever since the first mass spectrometric detection of C_{60} from laser ablation of graphite by Kroto et al. (1985), there has been keen interest in determining where fullerene molecules might occur naturally and what role they play in geochemical and cosmochemical phenomena. The first observation of naturally occurring fullerenes was made in 1992 (Buseck et al., 1992), and much work has been done since. Excellent reviews have been presented by Buseck (2002) and by Heymann et al. (2003).

It is reported that fullerenes occur in a variety of natural materials, especially in materials where evidence has been recorded of high energy events. In terrestrial samples, fullerenes are found in metamorphosed carbon-rich rocks, sedimentary rocks from formations at the Cretaceous-Tertiary (K/T) boundary and the Permo-Triassic (P/T) boundary layers, and breccias taken from large-body impact structures. In extraterrestrial materials, results are less consistent. However, fullerenes have been detected in the residue of one of the impact craters on the Long Duration Exposure Facility interplanetary particle collectors (di Brozolo et al., 1994) and in a series of experiments performed by Becker and co-workers in meteorites (Becker et al., 1994; Becker and Bunch, 1997; Becker et al., 1999, 2000; Pizzarello et al., 2001).

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The search for fullerenes in meteorites began immediately after the discovery of fullerenes, when it was suggested that these highly stable carbon allotropes may be abundant in the interstellar medium (Kroto, 1988). However, all of the initial investigations failed to detect fullerenes, and several searches subsequent to Becker's positive results have also failed to corroborate this evidence (Ash et al., 1991; de Vries et al., 1991; Gilmour et al., 1991; de Vries et al., 1993; Heymann, 1995, 1997). Buseck (2002) summarized these contradictory results as follows: "In spite of all of the published work, many unresolved questions remain about the identity and formation of fullerenes in geological environments... [specifically] why all research groups (ours included) other than that of Becker have been unsuccessful in locating fullerenes in meteorites."

Differences in the results of these experiments can be attributed to many factors associated with the challenging analytical aspects of targeted fullerene searches, including the low concentration at which natural fullerenes tend to appear, the apparent heterogeneity of fullerenes present in natural samples, and the strengths and weaknesses of the different analytical techniques used to perform the searches. A typical investigation for fullerenes requires much careful sample preparation, including crushing the material into an extractable powder, removing minerals by treatments with strong acids, extracting fullerenes with organic solvents, and analyzing the concentrated extract. The papers by Becker and co-workers rely chiefly on laser desorption mass spectrometry (LDMS) to characterize the presence of fullerenes. When using high-boiling solvents like 1,2,4-trichlorobenzene (TCB) or tetramethylbenzene, they found high-mass carbon envelopes in the LDMS spectra, which they attributed to fullerenes in a variety of samples including three carbonaceous chondrite meteorites—Tagish Lake, Murchison, and Allende. Mass spectra from extracts of Tagish

Lake and Murchison meteorites are shown in Fig. 1a and b, respectively. Clearly evident are mass peaks separated by 24 amu, which can be attributed to the loss or gain of C_2 fragments in the structures of these compounds. This pattern is diagnostic of the presence of fullerenes.

The strength of LDMS analysis is that it is able to detect fullerenes sensitively and report mass information that helps to identify specifically the analyte as fullerene. Also, it is capable of simultaneously reporting information about a series of fullerene peaks up to very large mass (C_{400} and above). The biggest weakness of LDMS is that under certain conditions, fullerenes can be generated by the laser desorption process when carbonaceous materials are present. This limitation has been observed in laser ablation studies of coals (Greenwood et al., 1990), polymers (Campbell et al., 1990), kerogen (Rose et al., 1993), and graphite (Kroto et al., 1985). Becker et al. relied on this information in their 1997 study and showed that, even at laser energies that fragment C_{60} in their system, no fullerenes were detected in desorption studies including coal, hydrothermal sediments, Green River Oil Shale, soot, K/T boundary sediments, the EETA79001 Martian meteorites, several Antarctic carbonaceous chondrites, and the Murchison meteorite (Becker and Bunch, 1997).

Interest in the fullerene content of natural samples received additional attention when Becker et al. reported that trace amounts of noble gases were encapsulated in fullerenes extracted from rocks at the Sudbury impact crater (Becker et al., 1996). The isotope ratio of these noble gases differed from the solar average, establishing a likely extraterrestrial origin for the fullerenes. The samples were analyzed by noble gas mass spectrometry (stepwise heating of the sample and ionization by electron bombardment). Further investigations found nonterrestrial isotope ratios in many of the samples where previously fullerenes had been

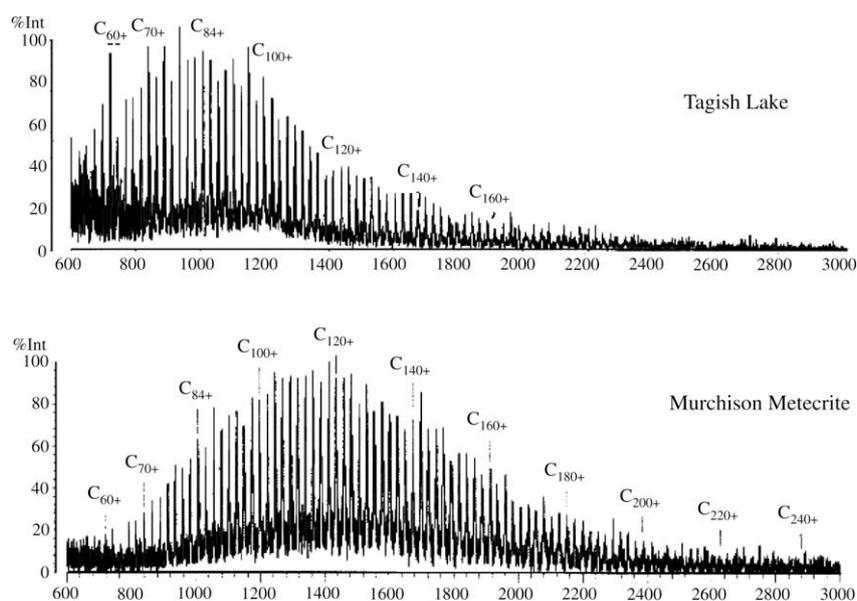


Fig. 1. LDMS Spectra reported for extraction analyses of fullerenes from carbonaceous chondrite meteorites. Originally published by Becker et al. (2000).

reported: sediments taken at mass extinction boundary layers (Becker et al., 2000, 2001) and in the carbonaceous chondrites (Becker et al., 2000; Pizzarello et al., 2001; Becker et al., 2006).

These results suffer, however, from many of the same deficiencies as their initial reports of fullerenes, including failures to reproduce the results on similar samples (Mukhopadhyay et al., 1998; Farley and Mukhopadhyay, 2001; Koeberl et al., 2004; Farley et al., 2005), questions regarding sample identity (Braun et al., 2001; Isozaki, 2001), and quantitative concerns about planetary gas contributions to the recorded fullerene gas signals (Koeberl et al., 2004; Heymann et al., 2006). The identity of fullerenes as the carrier of the gas isotope anomaly is also circumstantial, although the ability of C₆₀ to encapsulate helium-3 is well known and has become a powerful tool in studying C₆₀ chemistry (DiCamillo et al., 1996; Saunders et al., 1996; Cooper et al., 2002). Buseck sums up the controversy thusly: "Although the occurrence of the noble gases in the fullerene products of solvent extraction is presumably diagnostic, it is also possible that minor amounts of other gas carriers might have survived extraction. It would be more satisfying if the same mass spectra that show the natural fullerenes would also provide evidence of the encapsulated noble gases (Buseck, 2002)."

We have undertaken a study similar to previously reported investigations of the fullerene content of meteorite samples, and we find that a high-mass envelope of comparable appearance to the positive results for carbonaceous chondrites can be generated from samples that are themselves expected to be free of fullerenes. Specifically, we investigated the insoluble organic matter (IOM) from the Allende meteorite. Thus, we believe that the LDMS technique is capable of producing false positive fullerene signals at laser energies relevant to LDMS investigations of fullerene content. These false positives can hide the possible presence of the sought-after fullerene molecules of interest. Moreover, they also can confound the interpretation of the data.

In order to understand the ability of IOM to interfere with the search for fullerenes, we mimicked fullerene extraction experiments performed by Becker and co-workers by replacing powdered meteorite material with the equivalent amount of IOM. For the IOM to interfere with the fullerene analysis, it must become deposited on the chip substrate surface, requiring the IOM to pass through the physical barriers associated with the different preparation steps: the cellulose thimble used in the Soxhlet extraction step, and the 0.2 μm polytetrafluoroethylene (PTFE) filter used after the extraction step to remove particulates. Results from our experiment demonstrate that IOM can pass through these barriers, and that the IOM particulates do emit a strong enough fullerene envelope signal to interfere with the search for fullerenes. Further support for this conclusion is provided by centrifugations studies of the extracts, which were able to reduce the intensity of the fullerene envelope.

2. EXPERIMENTAL

We purified IOM from 10 g of the Allende meteorite according to the procedure reported by Gardinier et al.

(2000) with slight modification to aid in the removal of potential soluble fullerenes. A fresh, inner part of the meteorite was pulverized finer than 100 μm using an agate mortar. The powder was stirred in 500-mL HPLC-grade toluene (Fischer) for 24 h. Fluid and solid were then separated by centrifugation (Sorvall RC-5B, DuPont, Wilmington, DE), and the volume of the fluid was reduced to about 10 mL by vacuum evaporation. The fluid extract was then passed through a 0.2 μm PTFE filter (Whatman), reduced in volume by vacuum evaporation to ~500 μL, and stored for later analysis. We call this Extract E. The meteorite powder was then extracted with a mixture of dichloromethane and methanol (CH₂Cl₂/MeOH, 2/1, v/v) by refluxing for 24 h to remove soluble organic materials and elemental sulfur. After centrifugation and supernatant elimination, the residue was treated with 6 N HCl (sonication, 24 h) and then digested with a mixture of 6 N HCl/concentrated HF (1/1, v/v, 24 h). The residue was washed three times with water. The residue was treated by 6 N HCl followed by extraction with the CH₂Cl₂/MeOH mixture three times. Much care was taken to collect the floating black materials at each step, though some loss was inevitable. The final yield of the IOM was 2.3% wt, indicating that some mineral material remains (Pearson et al., 2006). A second Allende rock sample (5.6 g) was also prepared according to the above procedure with two modifications: one additional round of digestion with 6 N HCl/concentrated HF plus two additional rounds of HCl followed by CH₂Cl₂/MeOH extraction were performed. The final yield for this sample was 0.64% wt. Dichloromethane and methanol were distilled once before use; hydrochloric acid, hydrofluoric acid, and toluene of high purity grade reagents were used without further purification. Glassware used in this process was baked at 550 °C for 12 h before use to avoid contamination. Tagish Lake meteorite used in this study was prepared by Sandra Pizzarello at Arizona State University, (according to the procedure developed by Cronin et al. (1987).

The prepared IOM was divided in half and subjected to two identical extractions that follow the meteorite fullerene extraction procedures described in the literature (Becker and Bunch, 1997; Heymann, 1997). One fraction of the IOM powder was placed in the thimble of a Soxhlet extractor and extracted for 24 h with 200 mL of HPLC-grade 1,2,4-trichlorobenzene (TCB), which was purchased from OmniSolv, Wingate, NC. The liquid in the distillation flask was then reduced to a volume of ~30 mL by evaporation at reduced pressure at a temperature of 90 °C. We call this Extract A. The second fraction was placed in the thimble of a Soxhlet extractor and extracted for 24 h with 200 mL of toluene. The liquid in the distillation flask was then reduced to a volume of ~20 mL by evaporation at reduced pressure at a temperature of 50 °C. We call this Extract C. Each extraction thimble had been refluxed three times (24 h) in its respective solvent prior to the IOM extraction. An aliquot (100 μL) of each extract was taken and saved for later analysis, while the remaining liquid was filtered through a 0.2 μm PTFE filter into a vial. The volume of the filtered extract was again reduced by evaporation under reduced pressure. For the TCB extract, the final volume was ~10 mL,

whereas for the toluene extract the final volume was ~ 5 mL. We call the former Extract B and the latter Extract D. These filtered extracts and the extracts collected above were then analyzed by laser desorption mass spectrometry (LDMS).

Several other samples were prepared to provide additional information for the analysis done above. Suspensions of either ~ 700 μg untreated Allende meteorite powder or ~ 10 μg of IOM powder (Allende and Tagish Lake) were prepared by sonication in 100 μL of HPLC grade toluene, which are designated Control A, Control B, and Control C respectively. A control solution containing IOM powder was prepared by sonicating 90 μg of IOM powder in 200 mL of TCB. This sample was reduced by vacuum filtration to a volume of 30 mL, passed through a 0.2 μm PTFE filter, and further reduced to a final volume of 15 mL. This solution is designated Control D. A second control solution was also prepared by sonicating 678 μg of Allende IOM in 500 mL of TCB and is designated Control F. Several aliquots of 10 mL of this solution were collected and subjected to 1 h of increasingly high-speed centrifugation. The sample centrifuged at 12,000 rpm is designated Control G, and the sample run at 18,000 rpm is called Control H. After the removal of a 2 μL aliquot for analysis, Extract B was further treated with centrifugation at 4000 rpm for 60 min and subjected to a second round of filtration with the PTFE filters. We label this resulting solution Control E. Finally, the second sample of prepared IOM powder (5.6 g) was subjected to a modified extraction procedure. The procedure was as follows: the IOM powder was placed in the thimble of a Soxhlet extractor that been refluxed three times in TCB and then three times in toluene (12 h each). The IOM was extracted for 24 h with 200 mL of HPLC-grade toluene. The liquid in the distillation flask was then reduced to a volume of ~ 15 mL by evaporation at reduced pressure at a temperature of 50 $^{\circ}\text{C}$ and then passed through a 0.2 μm PTFE filter. The liquid was again reduced in volume by vacuum evaporation, this time to a final volume of ~ 100 μL . This reduced solution is called Extract F. For clarity, a list of all of the different extracts and control samples analyzed is provided (Table 1).

LDMS analysis was carried out on the Bio-Rad ProteinChip SELDI System PCS4000 Enterprise Model (Bio-Rad Laboratories, Hercules, CA). For each LDMS experiment, analyte was deposited in 2 μL aliquots onto a general-purpose NP20 ProteinChip Array (with silicate functionality) and dried in ambient conditions. Further, an apparatus blank (solvent passed through the apparatus before addition of IOM and treated according to the same analytical procedure) was spotted alongside each experimental extract. The spotted chip array was then loaded into the vacuum chamber. This instrument uses a nitrogen laser (337 nm wavelength, >5 ns pulse duration) to desorb and ionize molecules from the sample spot (100 μm diameter spot size), with subsequent detection of the ions in a linear time-of-flight mass spectrometer. The laser is scanned over the sample surface and a summed average is collected of all laser shots (typically 1000 shots for these experiments). The mass spectra of individual laser pulses can also be collected and stored. Laser energies between 1 and 6 μJ per pulse

were used, though results are typically reported for laser energies at 3 μJ . Because even trace amounts of carbon material on the chip surface can create a fullerene-containing envelope of mass peaks, the chip surfaces were scrupulously cleaned by sequential sonication in distilled methanol (20 min) followed by cleaning in a UV/Ozone oven (T16x16/OES model, UVOCS, Montgomeryville, PA) for 15 min until the chip surface showed no signs of fullerene signal even at laser energies two times stronger (6 μJ per pulse) than those used for the various IOM extract analyses. Further, a chip surface blank from each ProteinChip array was analyzed during each experiment to ensure that the surface remained free of carbon contamination. In all cases, chip surface blanks and apparatus blanks were free of contaminating signal. Although the Bio-Rad instrumentation permits modification of the sample substrate to selectively enhance desorption of certain compounds, we did not use such modification in this work. Thus, the LDMS experiments run in this study are similar to the LDMS technique used to detect fullerenes in other studies (e.g., (Becker et al., 1994, 1996, 2000; Pizzarello et al., 2001)), using the same combination of substrate, laser wavelength, one-step desorption/ionization, and time-of-flight mass spectrometry.

3. RESULTS AND DISCUSSION

Fig. 2 presents the results from LDMS investigations of Allende powder (Fig. 2a), Allende IOM, and Tagish Lake IOM. The high mass region of the two IOM spectra (Fig. 2b and c) are dominated by even-number carbon clusters, whereas no such peaks are seen in the unextracted meteorite powder spectrum (Fig. 2a). Laser ablation of graphite has long been known to produce envelopes of high-mass, even-numbered carbon clusters that include fullerenes (Kroto et al., 1985; Bowers et al., 1993). Creation of fullerenes from other carbonaceous materials, including coals, kerogens, polymers, and aromatic hydrocarbons has been reported. Zhang et al. summarize this situation by the statement: "These studies reveal that the laser energy required for fullerene formation from such materials is much lower than that required for graphite and, in general, scales with the aromaticity of the materials (Zhang et al., 1999)." Meteorite IOM is similar to type-III coal kerogen in that it is an amorphous matrix of small, polycyclic aromatic moieties held together with short, highly branched aliphatic chains, including many heteroatoms (Gardinier et al., 2000; Binet et al., 2002). Tagish Lake IOM is high in aromatic character, as is the Allende meteorite (Cody and Alexander, 2005). From this perspective, it is not unexpected that meteoritic IOM would generate a fullerene envelope when analyzed by a laser, and Fig. 2 demonstrates that IOM taken from Tagish Lake and Allende meteorites all produce strong fullerene signals. Given the high aromatic character of the Tagish Lake IOM, it is also not surprising that the signal from equivalent amounts of IOM is stronger for the Tagish Lake meteorite than for Allende. It is noteworthy that the Allende meteorite powder did not produce a fullerene signal, despite having roughly the same total amount of carbon deposited on the surface.

Table 1
Designation and description of extracts and controls

Designation	Description
Extract A	Soxhlet extract of Allende IOM (234 mg) with 200 mL TCB; collected and reduced to ~30 mL final volume
Extract B	Extract A filtered through 0.2 μm PTFE filter and reduced to 10 mL final volume
Extract C	Soxhlet extract of Allende IOM (226 mg) with 200 mL toluene; collected and reduced to ~20 mL final volume
Extract D	Extract C filtered through 0.2 μm PTFE filter and reduced to 5 mL final volume
Extract E	Extract (sonication) of 10 g Allende powder extracted with 500 mL toluene; filtered (0.2 μm PTFE) and reduced to ~500 μL final volume
Control A	700 μg crushed Allende powder in 100 μL toluene
Control B	10 μg Allende IOM in 100 μL toluene
Control C	10 μg Tagish Lake IOM in 100 μL toluene
Control D	Colloidal solution of 90 μg Allende IOM in 200 mL TCB; filtered (0.2 μm PTFE) and reduced in volume to 15 mL
Control E	Extract B centrifuged (4000 rpm, 1 h) and filtered a second time (0.2 μm PTFE)
Extract F	36 mg Allende IOM (0.64% wt) Soxhlet extracted with toluene, filtered, and reduced to a final volume of ~100 μL
Control F	678 μg of Allende IOM in 500 mL TCB
Control G	Supernatant of 10 mL of control F centrifuged for 1 h at 12,000 rpm
Control H	Supernatant of 10 mL of control F centrifuged for 1 h at 18,000 rpm
Blank A	Supernatant 10 mL of IOM-free TCB centrifuged for 1 h at 18,000 rpm

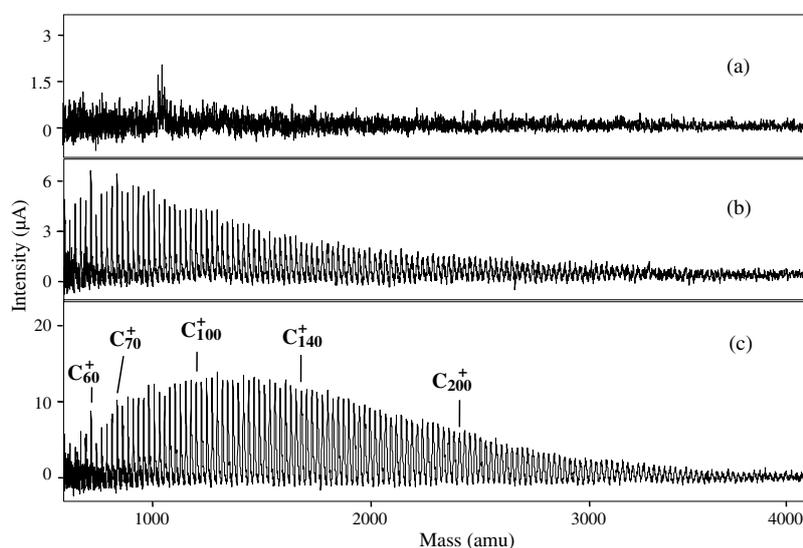


Fig. 2. LDMS spectrum (1000-shot average, 3000 nJ per pulse) of a 2 μL aliquot of (a) 700 μg of crushed Allende meteorite powder suspended in 100 μL toluene (Control A), (b) 10 μg of Allende IOM suspended in 100 μL toluene (Control B), and (c) 10 μg of Tagish Lake IOM suspended in 100 μL toluene (Control C).

Gu et al. pointed out that the creation of fullerenes from carbonaceous matrices is highly dependent on the power of the desorption laser and that ablation power densities are typically orders of magnitude higher than desorption powers (Gu et al., 1995). This observation indicates that LDMS experiments of free molecular material in extracts can be safely performed, even in the presence of background carbonaceous matrices, provided that the ablation threshold of the laser energy is much higher in intensity than that needed for desorption/ionization of the material of interest. For a typical investigation of fullerenes in natural materials, where the fullerene concentrations are low, the laser intensity is adjusted empirically by finding the maximum signal intensity for the C_{60} ion before fragmentation through C_2 loss is apparent. In our experimental setup, we deposited 2 μL aliquots of 27 μM C_{60} in toluene on the sample chip and optimized the laser intensity. The operating condition

was found to be near 3000 nJ per pulse and is in the same range as laser intensities used in previous successful analyses of C_{60} in natural samples in our laboratory (Elsila et al., 2005). The results presented in Figs. 2–4 were recorded with this same laser energy, and similar envelopes with diminished intensities were recorded at lower laser energies (2000 and 1000 nJ). This result shows that meteoritic IOM is capable of producing a strong false-positive signal that can easily mask the presence of low-concentration fullerenes in extraction experiments under typical investigation conditions.

Fig. 3 presents results from extraction experiments that simulate previous work except that the extractions are being done with IOM rather than meteorite powder. Fig. 3A and C show that for both solvent extractions (toluene and TCB) contamination by IOM in the collected extract produces a fullerene envelope. However, the two

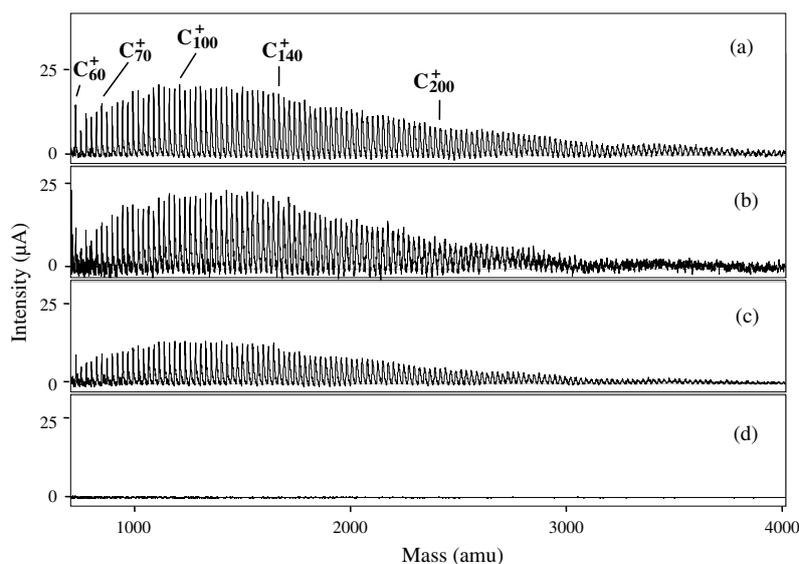


Fig. 3. LDMS spectra (1000-shot average, 3000 nJ per pulse) of a 2 μL aliquot of extract from the Soxhlet extraction of Allende IOM (a) with TCB before (Extract A) and (b) after filtration (Extract B), and with toluene (c) before (Extract C) and (d) after filtration (Extract D).

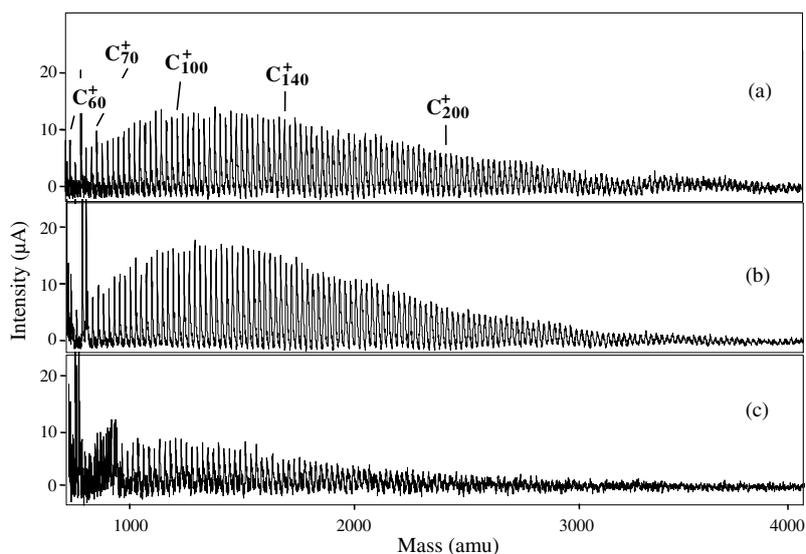


Fig. 4. LDMS spectra (1000-shot average, 3000 nJ per pulse) of a 2 μL aliquot of (a) 90 μg colloid solution in 100 mL TCB after filtration and volume reduction (Control D); (b) TCB extract of Allende IOM after centrifugation and double-filtration (Control E); and a 10 μL aliquot of (c) toluene extract from the modified Soxhlet extraction of 36 mg of Allende IOM (Extract F).

solvent extracts behave differently during the filtration process. Fig. 3B shows that a fullerene envelope is seen in the TCB extract even after it has been passed through the PTFE filter (Extract B); however, the PTFE filter effectively removes IOM below the limit of detection for this procedure when toluene is used as the extraction solvent (Fig. 3D, Extract D). We attribute this difference to the behavior of IOM suspended in the two different solvents. In toluene, the IOM behaves as a suspension, with visible aggregates settling to the bottom of the flask within minutes after sonication. Whereas, in TCB, the IOM behaves as a colloidal solution, turning the solution a uniform brown color and no visible signs of settling even after several

weeks. The lack of aggregation in TCB allows the IOM to pass through the filter more easily than in toluene, and the IOM material was observed to be less likely to condense on the plastic areas of the filters in TCB.

One of the challenges of this study is the removal of all fullerenes from the IOM. While Heymann demonstrated that up to 80% of C_{60} fullerene doped onto Allende meteorite powder can be recovered (Heymann, 1995), Jehlicka et al. reported that the recovery of fullerenes from carbonaceous materials can be heavily dependent on extraction technique and fullerene concentration, with recovery efficiencies consistently less than 5% when fullerenes were doped at the ppm level or lower (Jehlicka et al., 2005). Their

results indicate that the small recovery yields for low-level fullerenes are caused by destruction of the fullerenes by some as-yet-unknown mechanism. Laser desorption laser ionization mass spectrometry (L^2MS) analysis of the Allende and Tagish Lake IOM revealed them to be free from large polycyclic aromatic hydrocarbons (Plows et al., 2003), although L^2MS lacks the ability to validate that all of the extractable fullerenes had been removed from the IOM (Elsila et al., 2005). L^2MS , which is a gentle desorption process without the formation of a plasma, is less prone to generating complex species than LDMS, as recently demonstrated in studies on asphaltenes (Pomerantz et al., 2008).

In order to confirm that the fullerene signals observed in our extraction experiments arise from the presence of IOM and not residual extractable fullerenes, several control experiments were performed (see Fig. 4). In the first experiment, a colloid of 90 μg of IOM powder was prepared in TCB (Control D). The IOM was removed from the Soxhlet extraction thimble of the TCB extraction apparatus. This mass is the estimated amount of IOM that was collected in the extraction flask for the TCB extract, though the measurement was imprecise owing to the difficulty of scraping the IOM out of the collection flask and the potential for residue left behind by the TCB. The mass of 90 μg was chosen as a conservative estimate. If there are residual fullerenes present in the IOM, even assuming their presence at the 100 ppb level, only 0.00090 ng of fullerene are present for analysis, which is well below the detection limit for this instrument. Therefore, the presence of a strong fullerene envelope (Fig. 4A) certainly originates from the presence of the IOM. Fig. 4B shows what happens when we make additional efforts to remove IOM from Extract B (Control E). First, this extract was subjected to centrifugation (60 min, 4000 rpm). Second, the supernatant was passed a second time through the PTFE filter. The mass spectrum presented in Fig. 4B demonstrates that the fullerene envelope persists after attempts to eliminate particulate matter by this means. We followed the same procedure for Extract

F, this time with efforts to increase the likelihood that IOM would pass through the filter. To this end, several modifications were made. First, the IOM was treated with acid until the mass yield dropped below 1%. Second, the Soxhlet extraction thimble was washed both by TCB and toluene, which is expected to soften it and allow it to become more porous. Third, the filtered extract was reduced to 100 μL , in comparison to 10 mL, providing a much more concentrated sample for analysis (Extract F). In addition, 10 μL of this extract were deposited on the LDMS substrate for analysis. Because high-mass fullerenes are sparingly soluble in toluene (Becker et al., 1999), the fullerene envelope seen in Fig. 4C is indicative of a false-positive signal resulting from the presence of IOM.

It was suggested to us by Pizzarello (private communication) that if particulates are the source of the fullerene envelope, then high-speed centrifugation might demonstrate this fact. It should be recalled that centrifugation by us at 4000 rpm was not effective, and it is important to note that Becker and co-workers do not report using centrifugation as a purification step for their TCB extracts (Becker et al., 1999, 2000; Pizzarello et al., 2001). Consequently, we investigated the effect of centrifugation at higher rotor speeds. Fig. 5A shows the fullerene envelope for a colloidal suspension of 678 μg in 500 mL TCB (Control F). Aliquots of this solution were subjected to 1 h of increasingly high-speed centrifuging (12,000 rpm, Fig. 5B, Control G); and 18,000 rpm, Fig. 5C, Control H). The fullerene signals are seen to decrease in intensity with increasing centrifuge speed, up to 4 h of centrifugation at 18000 rpm (not shown) beyond which point the centrifuge tubes used for this experiment cracked or became distorted. Fig. 5D presents the result of IOM-free TCB that had been run through the centrifuge. This figure demonstrates that the signals appearing at 685 and 701 amu are background contaminations encountered during sample handling or storage. Above 16000 rpm, a small amount of black residue was visible at the bottom of the centrifuge tube, and we estimate from the LDMS results that 80–90% of the IOM is removed by

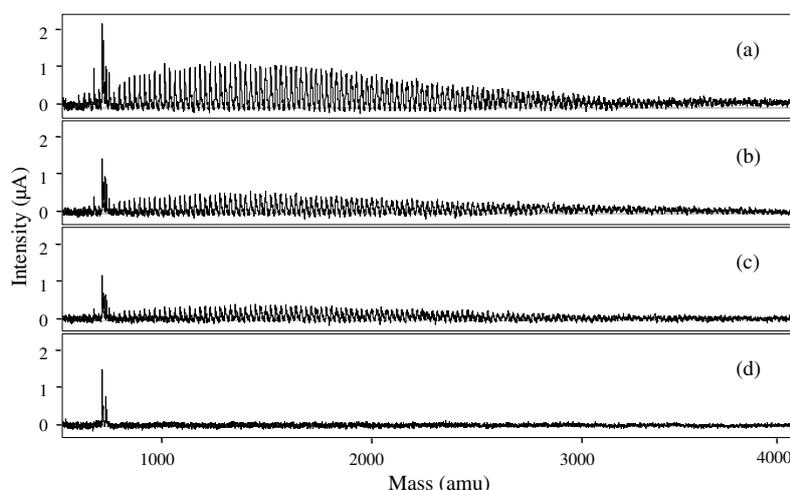


Fig. 5. LDMS spectra (1000-shot average, 3000 nJ per pulse) of a 2 μL aliquot of a colloidal solution of 678 μg of Allende IOM in 500 mL of TCB (a) before (Control F) and (b) after centrifugation for 1 h at 12,000 rpm (Control G) and (c) 1 h at 18,000 rpm (Control H). A blank aliquot of TCB was also centrifuged at each speed and the blank at 18000 rpm is shown in (d) (Blank A).

this centrifugation protocol. The decrease of the fullerene signal intensity with centrifugation indicates a strong correlation between the fullerene signal and the presence of IOM particulates, as we had suspected from the more indirect evidence we had already found.

LDMS data have been heavily relied upon for understanding the formation conditions of fullerenes. Irle et al. (2006) used a variety of simulation methods to elucidate the mechanism of fullerene formation from carbon vapor in high-temperature, non-equilibrium conditions. They found that this process results in the creation of giant fullerene molecules of differing sizes, and that factors like vapor density, temperature, and heating duration affect the average size of the generated fullerene envelope. They specifically mentioned the fullerene content reported by Becker and co-workers as being examples of fullerenes produced by non-equilibrium hot-carbon vapors. Their interpretation contrasts sharply with the expectations that Allende fullerenes have reached or approached equilibrium during their formation or in post-formation processing. Allende has been exposed both to thermal metamorphism (Rouzaud et al., 2005; Bonal et al., 2006, 2007) and to high temperatures during Rouzaud atmospheric flight and collision with earth (Heymann et al., 2006; Braun et al., 2001). Thus, it might be expected that the fullerene envelope would have a more uneven distribution (Slanina et al., 2000). The LDMS signal generated by meteoritic IOM is also indicative of non-equilibrium formation conditions, and this signal could easily interfere with or mask information about the formation conditions of native fullerenes in meteorites. Other factors that are likely to affect the distribution envelope include the presence of other light-absorbing/ionizing molecules in the extract. In the case of IOM, the free organic matter has been removed by iterative extractions, but UV-absorbers like PAHs are expected to be present in fullerene extraction experiments. Their presence might also change the distribution by providing additional carbon sources or altering local heating.

Our results cannot prove that no fullerenes occur in meteorites, though we note that neither C_{60}^+ nor C_{70}^+ were observed in our LDMS analysis of the toluene extraction of 10 g of Allende powder above the 600 ppt level (data not shown). The fact that high-mass fullerene peaks appear in LDMS spectra taken of TCB extracts more easily than in toluene extracts mirror the observation that high-order fullerenes are observed only for TCB extracts of acid-digested meteorites (Becker et al., 2000). Given the persistence of IOM in TCB extracts despite efforts to remove particulates by filtration, the results presented here suggest that the meteoritic fullerene signals previously reported by Becker and co-workers suffer from contamination and represent a LDMS detection method applied to samples that inadvertently contain IOM. The results of the present work raise serious doubts about the interpretation of previous meteoritic studies searching for fullerenes. In our opinion, no experiment to date has established the existence of fullerenes in meteorites, although we know that naturally occurring processes can generate fullerenes.

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