Nanocrystalline Aggregation of Serine Detected by Electrospray Ionization Mass Spectrometry: Origin of the Stable Homochiral Gas-Phase Serine Octamer

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Introduction

Did homochirality precede life or was homochirality a consequence of life? This issue1 is complex enough to warrant study from all avenues. This paper addresses a novel mechanism for the spontaneous generation of homochirality, or homochirogenesis. Homochirogenesis may be achieved by at least three fundamental mechanisms: (1) selective synthesis of only one enantiomer of a chiral molecule,2 (2) the preferential destruction of one enantiomer of a heterochiral mixture,3 and (3) separation of a racemic mixture into distinct homochiral parts.4

The spontaneous breaking of symmetry for a racemic mixture of chiral molecules has only been achieved by a handful of methods. Certain molecules will spontaneously separate from a racemic solution into homochiral crystals.4 The assembly of these macroscopic homochiral crystals must be orchestrated at the molecular level. Intermediate to the formation of macroscopic crystals from single molecules, noncovalently bound molecular clusters may be formed. The study of these intermediate molecular clusters is likely to offer insight into the most fundamental requirements for spontaneous symmetry breaking. Molecular clusters may share structural similarities with their parent crystal structures.5 Furthermore, homochiral self-assembly into small molecular clusters offers a possible target of opportunity for conversion of the aggregates into a homochiral polymer or macromolecule with well-defined stereochemistry.

Electrospray ionization mass spectrometry (ESI-MS) is an excellent method for observing molecular clusters in the gas phase.6 A variety of noncovalent clusters have been studied by ESI-MS, ranging from the molecular recognition of organic molecules7 to the structures of salt clusters.8 The flexibility and sensitivity of ESI-MS allows for the examination of solutions over a wide range of concentrations.

By itself, mass spectrometry provides only the molecular weight of a detected species and no direct information relating to molecular structure in general or to chirality in particular.

This has led to the development of a variety of methods to circumvent this deficiency. These experiments, involving chiral amino acids,9 amino acid derivatives,10 tartrates,11 metals and amino acids,12 and host/guest chemistry involving crown ethers,13 cyclodextrins,14 cyclofructans,15 and monosaccharides16 have all utilized mass spectrometry and taken advantage of the different physical properties of diastereomers to successfully discriminate between enantiomeric pairs. Experimental techniques include both kinetic and equilibrium methods. The preferred method uses one of the enantiomers isotopically labeled to distinguish it from the other.8–16

Alternatively, mass spectrometry can be coupled with another technique such as ion mobility spectrometry (IMS).17 IMS gives direct structural information about the gas-phase conformation of a molecule or cluster in the form of a collisional cross section. The experimental cross section can then be compared to the theoretical cross section determined computationally for likely structures of a particular molecule or cluster.

We have recently developed a new method to determine the extent to which spontaneous chiral separation occurs in small molecular clusters.5,18 Our previous work18 demonstrated the successful application of this new technique on the serine octamer. The serine octamer was originally reported by Cooks and co-workers,19 using a different experimental methodology, to have a homochiral preference.

In the current work, we report detailed experiments that elucidate the structure of the serine octamer and offer an explanation for its unusual abundance. Examination of a mixture of d-serine and labeled L-serine using ESI-MS indicates that the octamer strongly prefers a homochiral composition. IMS experiments indicate that the structure of the octamer is consistent with a cubic arrangement of serine. Blockage of the N-terminus or C-terminus of serine leads to no octamer formation, suggesting that amino and carboxylate functionalities play a critical role in the bonding. The spontaneous symmetry breaking and likely involvement of zwitterionic serine aggregates in solution suggest that nanocrystals of serine precede...
the formation of the gas-phase octamer. DFT calculations reveal several low-energy structures that are related to the crystal structure of serine. The analogues threonine and homoserine may also form similar structures. The experimental results for these molecules are compared to the results found for serine.

Experimental

Mass spectra were obtained using a Finnigan LCQ ion trap quadrupole mass spectrometer without modification. The signal was optimized using the automatic tuning capabilities of the LCQ on the protonated serine octamer. For serine and analogues, the settings used were source voltage 4.15 kV, capillary voltage voltage of between 0.98 and 2.45 V for a period of 30 ms. Collision-induced dissociation (CID) was performed on isolated parent ions by applying a resonance excitation RF gate with a 556 V cm$^{-1}$ field against a 2000 sccm flow of helium. The signal may also form similar structures. The experimental results for the formation of the gas-phase octamer. DFT calculations reveal the structure of serine. The analogues threonine and homoserine may also form similar structures. The experimental results for these molecules are compared to the results found for serine.

**TABLE 1: Progressive Stabilization of Structure 4 at Various Chain Lengths**

<table>
<thead>
<tr>
<th>Structure</th>
<th>zwitterion$^a$ (kcal/mol)</th>
<th>neutral$^b$ (kcal/mol)</th>
<th>$\Delta$ (kcal/mol)</th>
<th>$\Delta_{\text{ser}}$ (kcal/mol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2mer</td>
<td>-500335.09</td>
<td>-500341.65</td>
<td>6.56</td>
<td>3.28</td>
</tr>
<tr>
<td>4mer</td>
<td>-1000716.07</td>
<td>-1000726.37</td>
<td>10.3</td>
<td>2.58</td>
</tr>
<tr>
<td>6mer</td>
<td>-1501103.56</td>
<td>-1501094.01</td>
<td>-9.54</td>
<td>-1.59</td>
</tr>
<tr>
<td>8mer</td>
<td>-2001491.09</td>
<td>-2001468.19</td>
<td>-22.9</td>
<td>-2.86</td>
</tr>
<tr>
<td>10mer</td>
<td>-2501870.68</td>
<td>-2501825.12</td>
<td>-45.5</td>
<td>-6.16</td>
</tr>
</tbody>
</table>

$^a$ Total energy as determined by single point calculations on the PM3 minimized neutral structures at the 6-31G/B3LYP level, corrected for zero-point energies.

**TABLE 2: Calculated Energies and Cross Sections for Structures 4–6**

<table>
<thead>
<tr>
<th>Structure</th>
<th>relative gas-phase energy (kcal/mol)$^a$</th>
<th>computed cross section ($\AA^2$)</th>
<th>relative gas-phase binding energy (kcal/mol)$^b$</th>
<th>relative solution phase binding energy (kcal/mol)$^c$</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
<td>76</td>
<td>224$^b$</td>
<td>112</td>
<td>-55</td>
</tr>
<tr>
<td>5</td>
<td>0</td>
<td>196$^c$</td>
<td>-</td>
<td>-16</td>
</tr>
<tr>
<td>6</td>
<td>0</td>
<td>189$^b$</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

$^a$ Energies listed relative to the energy of 6 for protonated octamers. $^b$ Cross sections for protonated structures to be compared with an experimental value of 187 Å$^2$. $^c$ Cross section calculated for neutral structure (the neutral cross sections for 4 and 6 are 227 and 189 Å$^2$, respectively). $^d$ Binding energy calculated relative to 8 separated zwitterionic serines. $^e$ Energies calculated for neutral octamers in water with a dielectric constant of 80.37 and a probe radius of 1.4. This structure is not well behaved and does not fully minimize.

Stability of the cluster is sensitive to the chirality of the individual molecules, the observed distribution will deviate from the binomial distribution. A preference for homochirality will be indicated by an increase in the relative intensities of the pure L and pure D clusters. A preference for heterochirality might also be observed, with the mixed species being more abundant than predicted by the statistical distribution.

The experimental cross sections of the serine clusters were measured using a high-resolution ion mobility apparatus. A solution was prepared by dissolving five milligrams of L-serine in 1 mL H$_2$O, 0.2 mL CH$_3$COOH, and 0.1 mL CH$_3$CN. The solution was electrosprayed using a 5 kV potential across a 0.5 cm gap. The ions were guided into a drift tube through an ion gate with a 556 V cm$^{-1}$ field against a 2000 sccm flow of helium. The 63 cm long drift tube contains 46 guard rings and was operated at 10,000 V with ~760 Torr of helium buffer gas.

Ions exit the drift tube through a 0.125 mm aperture and are focused into a differentially pumped region where they are mass selected by a quadrupole mass spectrometer and detected by an off-axis collision dynode and dual microchannel plates. A multichannel scaler that is synchronized with an electostatic shutter located between the ion gate and the drift tube records arrival-time distributions. Drift times are determined by correcting the arrival times for the time that the ions spend traveling from the drift tube to the detector. Collision cross sections are calculated using eq 1

$$
\Omega_{\text{avg}}^{(1,1)} = \frac{(18\pi)^2}{16} \left[ \frac{1}{m} + \frac{1}{m_B} \right] \frac{z_e^2}{(k_B T)^2} \frac{L}{\rho} E
$$

In this expression, $m$ and $m_B$ are the masses of the ion and buffer gas, $z_e$ is the charge on the ion, $\rho$ is the buffer gas number density, $L$ is the length of the drift tube, and $E$ is the drift field. Ions with the same $m/\rho$ but different structures have different collisional cross sections and can be distinguished. This is particularly useful in distinguishing “multimers” where, for example, the peak for the singly charged monomer is overlapped by the doubly charged dimer.

**Results**

**Serine.** Serine is the simplest amino acid with a polar side chain, the hydroxymethyl group. The chiral center is marked with an asterisk in structure 1. In separate ESI-MS experiments, the concentration of serine was varied by factors of 10 from 10 μM to 0.1 M. Maximum clustering was observed at 0.01 M concentration. The positive ion mass spectrum obtained with 0.01 M L-serine is presented in Figure 1. The protonated
charged serine clusters as evidenced by $^{13}$C distribution. The
In this case, collisional heating eliminated all of the multiply
higher than that of $[8\text{Ser}^+\text{H}]^+$. The theoretical cross section for a proposed
structure should agree to within approximately $1156$. This series could only have resulted
from a larger cluster with more than three charges on it, most
likely $[44\text{Ser}^+\text{H}]^{4+}$. The relative intensities of the two series indicate that the quadruply charged species comprises a
significant portion of the total intensity for the peak at $m/z$ 1156. However, in this case there is an
additional series of peaks separated by 35 mass units with $m/z$ ratios higher than 1156. This series could only have resulted
from a larger cluster with more than three charges on it, most
likely $[44\text{Ser}^+\text{H}]^{4+}$. The relative intensities of the two series indicate that the quadruply charged species comprises a
significant portion of the total intensity for the peak at $m/z$ 1156.

Figure 4a shows the distribution of serine octamer clusters observed in a 54:46 mixture of D-serine with L-serine, respectively. The L-serine is labeled with deuterium in the three C–H bonds. In these experiments, collisional heating of all ions by mild excitation of the entire mass range was used to eliminate the multiply charged clusters, yielding only singly charged octamer peaks. The resulting experimental distribution of mixed clusters differs significantly from the predicted statistical distribution, included in Figure 4a for comparison. These results are in stark contrast to those for the serine dimer. Figure 4b illustrates the results of the same experiment for the protonated serine dimer in which no preference for homochirality is observed.

At first glance, the data in Figure 4a suggests that octamers containing a mixture of 6D and 2l. or 2D and 6D serines are favored. A more detailed analysis is shown in Figure 5. Dividing the observed intensities by the statistical prediction (Figure 5a) yields relative intensities for the octamers which better reflect their energetic stabilities. This is more evident in the semilog plots in panels b and c of Figure 5, where starting with either the 8D or 8l. octamers, respectively, there is a sequential replacement of serines of one enantiomer with the other. In both instances, the limiting case of a single replacement indicates an energetic cost of $2.9 \pm 0.3$ kJ/mol, calculated from the slope of the semilog plot assuming a Boltzmann analysis is valid, for incorporating the incorrect enantiomer in the homochiral cluster.

Serine Derivatives. The spectra for L-serine methyl ester and L-N-tertbutoxycarbonylserine (L-tboc serine) are shown in panels a and b, respectively, of Figure 6. As seen in Figure 6a, the...
Figure 3. (a) Collision-induced dissociation spectrum for the singly charged serine octamer. The octamer preferentially loses a neutral serine dimer. (b) Collision-induced dissociation spectrum for m/z 1051. Nearly all of the fragments are separated by 35 m/z, indicating that the parent ion is composed primarily of [30Ser+3H]^{3+}. (c) Collision-induced dissociation spectrum for m/z 1156. The emergence of a cluster distribution with a separation of 35 m/z at a higher m/z suggests that a significant portion of this cluster is composed of [44Ser+4H]^{4+}.
C-terminal methyl ester group eliminates the pattern of clustering that is observed for serine. The dimer is the only prominent peak in the spectrum. The small distribution of clusters at higher masses contains multiply charged clusters of L-serine methyl ester with sodium (present as a contaminant). Similarly in Figure 6b, the dimer of L-tboc serine is the dominant peak, followed by some higher-order clusters. The trimer and higher mass clusters are primarily sodiated. The distribution of clusters is again quite different than that observed for serine itself (Figure 1).

Homoserine and Threonine. Two amino acids closely related to serine are threonine (structure 2) and homoserine (structure 3). Homoserine contains an additional methylene group, which extends the side chain relative to serine. The mass spectrum for L-homoserine is presented in Figure 7a. The monomer and dimer are both abundant, but the singly charged octamer does not exhibit unusual abundance (in contrast with serine). The base peak corresponds to 477 $m/z$, which might suggest that homoserine forms an unusually abundant tetramer. However, upon closer inspection of the carbon-13 peaks, it becomes clear that the peak at 447 $m/z$ is primarily composed of $[8Hser + 2H]^+$ (where Hser = L-homoserine). Therefore, homoserine also forms an unusually abundant octamer. The peak corresponding to $[11Hser + 2H]^+$ is also unusually abundant with respect to the surrounding distribution.

Figure 4. (a) Comparison between the predicted statistical distribution and observed distribution for the serine octamer for a 54:46 mixture of d-serine and isotopically labeled L-serine. Full spectrum CID was used to break up the multiply charged aggregates of the octamer. A clear preference for homochirality is indicated by these data. (b) Comparison between the predicted statistical distribution and observed distribution for the serine dimer. No clear preference for chirality is indicated.

Figure 5. (a) Ratio of the observed to statistical intensities for the serine cluster distribution shown in Figure 4a. (b) Plot of the ln of the ratios in panel a vs the number of L-serines switched for D-serine starting with the D-serine octamer. (c) Plot of the ln of the ratios in panel a vs the number of D-serines switched for L-serine starting with the homochiral L-serine octamer.
Several experiments were performed with mixtures of L-homoserine and L-serine. A 50/50 mixture of L-serine and L-homoserine will yield mixed clusters, but the mixed clusters are not unusually abundant. A 6/2 mixture of L-serine/L-homoserine yields abundant mixed serine octamers with the incorporation of 1 or 2 homoserine molecules into the cluster (Figure 7b). Isolation of [6Ser+2Hser+H]+, followed by CID, yields the spectrum shown in Figure 7c. The mixed cluster preferentially loses two neutral serines. Homoserine is always retained.

The mass spectrum for D-threonine is shown in Figure 8 (allo-threonine isomers gave very similar results). This spectrum shares many similarities with the spectrum of homoserine. As in the case with homoserine, [8Thr+H]+ and [8Thr+2H]2+ are both present. [7Thr+H]+ is only prominent in the threonine spectrum. The peak corresponding to [11Thr+2H]2+ is unusually abundant. However, the combined intensity of the octameric peaks is not as prominent as it is in the case of serine or homoserine.

A 50/50 mixture of D-threonine and D-serine was electro-sprayed and is shown in Figure 9a. The singly and doubly charged mixed octamers are labeled in Figure 9a, where it is observed that they exhibit unusual abundance when compared to the other clusters. Figure 9b shows the distribution for the singly charged mixed threonine-serine octamer. Mixed clusters ranging from [6Ser+2Thr+H]+ to [2Ser+6Thr+H]+ are easily observed. The abundance of the two remaining mixed clusters and the two pure clusters cannot be discerned from noise. A similar distribution is observed for the doubly charged mixed octamers ([6Ser+2Thr+2H]2+ to [2Ser+6Thr+2H]2+) in Figure 9c. The different distribution, particularly the extra prominence of [4Ser+4Thr+2H]2+, is likely due to the overlap with mixed tetramers such as [2Ser+2Thr+H]+.
Structure 4 is formed from zwitterionic serine, which is the predominant form of serine in both solution and the solid phase. Would a zwitterionic structure also be stable in the gas phase? Table 1 demonstrates the progressive stabilization of zwitterionic serine with increased cluster size for a model structure similar to 4, in the absence of any net charge. The structures are stabilized in the zwitterionic state by the Coulombic attraction that is derived from the two rows of salt bridges. This Coulombic attraction compensates for the energy required to generate zwitterionic serine, which is not the lowest energy structure for the isolated serine molecule in the gas phase. If the Coulombic attraction overcomes the "zwitterion penalty," the zwitterionic state of the cluster will be favored. A net charge has been shown previously to enhance the stability of zwitterions in the gas phase. Our calculations indicate that structures similar to 4, with varying numbers of serines in the chain, would be stable as zwitterions with 8 or more serine molecules, even with errors of 5–10 kcal/mol in the calculations.

Thus 4 is a reasonable candidate structure for the serine octamer because it can exist both in solution and the gas phase. Furthermore, 4 is a derivative of the crystal structure of L-serine and, as a result, displays a strong preference for homochirality. It is not possible to change the chirality of four of the serines and generate a cluster composed of 4 D-serines with 4 L-serines (a 4D/4L cluster). The highly ordered bonds of 4 suggest that the structure may be very stable. This is confirmed in Table 2, which lists the calculated gas-phase cross sections with the accompanying solution and gas-phase energetics of several possible structures for the serine octamer. In solution, 4 has the best binding energy of the three structures. However, the calculated cross section for the protonated structure is 224 Å², which is much greater than the experimental value of 187 Å². Therefore 4 must be ruled out as the gas-phase structure for the serine octamer.

Structure 5 is a cubic arrangement of 8 serines that is closely related to 4. Structure 5 is formed by taking 4 and essentially folding it in half to form a cube-like structure. The structure has four rows of salt bridges, two interior and two exterior. The structure shown is the minimized unprotonated structure. The cross section calculated for the neutral structure 5 is in reasonable agreement with the experimental results (Table 2).

However, structure 5 undergoes significant reorganization in the gas phase when protonated. The protonated structure will not properly minimize, suggesting that it is significantly unstable in the gas phase. Furthermore, the two interior rows of zwitterions are not well exposed, leading to unfavorable solvation energetics which substantially decrease the solution-phase binding energy (Table 2). Additionally, a similar heterochiral 4T/4T structure exists. Therefore, it is unlikely that 5 is the structure for the serine octamer, but it may be an important intermediate structure, as will be explained below.

Reorganization of structure 5 in the gas phase leads to structure 6. The minimized gas-phase protonated structure of
6 is shown, but it is virtually identical to the neutral structure. All eight serines are bound via a central zwitterionic core. The resulting cubic structure is further stabilized by hydrogen bonding between facing pairs of serines. The computed cross section for 6, 189 Å², is in excellent agreement with the experimental value of 187 Å² (Table 2). The gas-phase binding energy of this structure is the best of any of the candidates that we have tested (Table 2). Given that the zwitterionic core of structure 6 would be essentially buried from any solvent, it has the worst solution-phase binding energy of the three structures. Furthermore, a 4L/4L cluster can be assembled with a zwitterionic core and binding energy similar to that for structure 6. For these reasons, 6 is not likely to be the solution-phase structure for the serine octamer. However, 6 represents the most likely gas-phase structure, particularly in light of the excellent gas-phase energy and agreement with the experimental cross section.

Of the experimental constraints noted above for the structure of the serine octamer, 2 and 3 are the most difficult to simultaneously satisfy. A three-point interaction between each serine and the rest of the cluster would constitute the minimal requirement for chiral recognition. It is easy to generate a variety of clusters that satisfy this requirement, but such clusters do not necessarily satisfy requirement 2. Given that eight serines must be arranged in an unusually stable structure with a cross section of 187 Å², cubic motifs (such as 5 and 6) seem the most consistent with the ion mobility data. However, the symmetry of a cube generates additional problems that must be taken into account. For example, a cubic octamer connected at four points through the neutral carboxylic acids (a structure actually proposed in a concurrent work by Cooks and co-workers during the preparation of this manuscript) can be assembled from homochiral serine or from 4D and 4L serines. Although each serine is held in a three-point interaction in both clusters and the energetics are nearly identical, requirement 2 is clearly not satisfied by such a structure. Furthermore, the data presented in Figure 4 clearly demonstrates the extremely low abundance of 4D/4L clusters. In fact, it is entirely likely that all cubic structures (the only reasonable geometry that satisfies the experimental cross section) will have an accompanying 4D/4L cluster.

The combination of requirements 2 and 3 suggest that a noncubic structure, such as 4, exists in solution which becomes cubic in the gas phase. This is depicted in cartoon fashion in Scheme 1. In general terms, A depicts various noncubic
nanocrystals of serine forming in bulk solution. In B, a charged serine octamer is driven to the surface of a highly charged droplet (other charges not shown for clarity). In C and D, as the number of solvent molecules is reduced (either through ion evaporation or evaporation of the solvent), the octamer begins to collapse on itself. The fully desolvated protonated serine octamer with a cubic structure is shown in E. In this manner, the requirements for homochirality and a cubic structure can be satisfied simultaneously.

Singly protonated structures similar to 4 (i.e., 6mers and 10mers) may also form in solution; however, their individual transfer to the gas phase is not as favorable because only the octamer can form a cube. Aggregation of these singly charged species into multiply charged multimers (such as the multimers of the octamer shown in Figure 2) allows for enhanced stabilization in the gas phase. Thus larger, multiply charged clusters such as those shown in Figure 1 and Figure 3 are also present in the gas phase. However, the stability of these multimers is derived from their size, not from their structure. Of the singly charged species, the octamer is king. In the case of the octamer, the unusual abundance is attributed to the special stability of the cubic structure.

Both homoserine and threonine can form structures analogous to 4. The crystal structure of homoserine allows for a homoserine analogue of 4 with very little rearrangement. However, in the case of threonine, substantial rearrangement from the crystal structure is necessary. This may account for the reduced relative abundance of the octamer for threonine (Figure 8). Notwithstanding, the facile incorporation of threonine into the octamer as shown in Figure 9b,c is compatible with the solution phase structure given in 4 and the gas-phase structure shown 6. It is anticipated that the clusters producing the spectra for homoserine and perhaps threonine are largely analogous to the structures that lead to the serine spectrum. The somewhat higher proton affinity of threonine and homoserine may be related to the observation that the cluster distributions of the analogues sometimes exhibit higher charge states than those observed for serine itself.

Conclusion

We have conducted a number of experiments designed to elucidate the structure of the serine octamer, and explain its abundance in electrospray ionization mass spectrometry. The octamer demonstrates a strong preference for homochirality. Experiments using L-serine methyl ester and L-N-tertbutoxycarbonyl-serine (t-boc serine) show no cluster formation, indicating the amino and carboxylate functionalities are necessary for formation of the cluster. Ion mobility data indicates the cross section of the octamer to be 187 Å² (±2%). DFT calculations and the experimental data lead to a structure that is derived from solution aggregates that precede formation of crystalline serine (structure 4). Structure 4 provides the necessary enantiomeric discrimination observed in the serine octamer and the folding of 4 into 5, which relaxes to 6, enables the octamer to conform to the measured cross-section with an energetically favored structure. Both threonine and homoserine may form structures analogous to the serine octamer. Threonine may incorporate freely into serine clusters because the additional methyl group does not interfere with the bonding of the cluster.

The combination of the solution and gas-phase properties of the serine octamer lead to its unusual abundance when sampled by mass spectrometry. We have taken advantage of this unusual abundance to demonstrate clearly that small molecular clusters can demonstrate a preference for homochirality. This offers a molecular cluster parallel to the macroscopic observation of chiral symmetry breaking through crystal formation, and a possible pathway for the establishment of prebiotic homochirality. In appropriate energetic or chemical environments, covalent coupling of the components of the clusters could occur, creating homochiral polymers or macromolecules with well-defined stereochemistry. Such stereoregular molecules could then serve as templates in reproductive chemical systems. No inherent preference is given to a particular enantiomer in the formation of these clusters, and therefore an additional mechanism would be required to explain the eventual elimination of one enantiomer.

On a different note, ESI may offer a new experimental technique for investigating the early stages of homogeneous crystal nucleation in solution. A universal theory for the explanation of homogeneous nucleation is still lacking. Several spectroscopic methods have been employed to study crystal nucleation, but mass spectrometry offers the additional ability to sample small clusters and study them in the gas phase. As appears to be the case in the present study, the solution-phase structures of the clusters may not be retained in the gas phase. The difference between the solution and gas-phase cluster distributions will depend on the detailed mechanism by which the are transferred from solution to the gas phase in the ESI process. The ESI process may increase the monomer concentration through evaporation prior to droplet fissioning. This could conceivably induce greater cluster formation in a super saturated and cooled droplet. If ESI does enhance the formation of nanocrystals, then one could conceivably devise a crystal-seeding source designed to initiate crystallization.

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References and Notes


