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Short communication

Gas-phase Cu^+– and Ag^+–glycine complexes produced with a new source

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Abstract

A combination of the laser ablation of a metal target with an effusive amino acid oven and the free-jet expansion of helium is employed in order to produce gas-phase metal cation–amino acid complexes in a molecular beam. With this source, Cu^+–glycine and Ag^+–glycine complexes are formed through gas-phase reactions and are analyzed using a time-of-flight mass spectrometer. From the mass spectra we conclude that copper is dicoordinated while silver can attract up to four glycine molecules.

Keywords: Metal–amino acid complexes; Mass spectrometer; Glycine

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1. Introduction

The binding of metal ions to amino acids, peptides and proteins plays a very important role in biology. Metal ions in solutions with biomolecules cause a series of chemical processes such as catalysis, electron transfer, O_2 transport, stabilization, etc. [1]. Furthermore, metal ions play a very crucial role in organizing three dimensional biological structures [2]. These phenomena originate from the ability of metal ions to bind preferentially to several bioligands, thus affecting strongly the structure of nucleic acids. In solution, there are many factors (e.g., pH) that influence the structural properties of the nucleic acids and, therefore, investigations of these properties are very complicated.

In contrast, studies on metal ion–biomolecule complexes in the gas phase provide valuable information, as the complexes are free molecules and therefore are not affected by local environment (solvent) that can stabilize or destabilize them. Thus, such gas-phase studies compared with the solution-phase ones, are very useful for understanding the role of metal ions in initiating biochemical processes.

In the last few years, there has been an intensive effort in developing sources able to produce free complexes between metal ions and biomolecules. Fast atom bombardment (FAB) sources have been employed to produce metal ion–amino acid complexes [3]. In recent years, the matrix-assisted laser

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desorption/ionization (MALDI) [4] and the electrospray ionization (ESI) [5] are the most widely used methods in producing readily gas-phase biomolecular ions. However, by these techniques it is not very clear how far the products formed are the result of gas-phase reactions, or are directly desorbed from the bulk phase (matrix or solution) [6].

In this communication, we present another approach that produces metal–amino acid ions via gas-phase reactions of preionized metal ions and gas-phase volatile amino acids. We have produced Cu^+–glycine and Ag^+–glycine complexes using a combination of laser ablation-ionization of the metal and an effusion oven for the glycine (G) neutral molecule. The obtained mass spectra, which reflect the stability of the complexes formed and the coordination of the two metals, are analyzed and the results are compared with theoretical calculations from other groups.

2. Experimental

The experiments have been performed in a molecular beam apparatus that contains a reflectron time-of-flight (TOF) mass spectrometer and is already described in detail elsewhere [7,8]. The metal ion–glycine source is displayed in Fig. 1. The production of metal ions (Cu or Ag) process through the vaporization of a high purity (>99.95%) Cu or Ag targets in form of rods by laser ablation using the fundamental (1064 nm) of a Nd–YAG laser. The produced plasma is mixed with the evaporated glycine from an effusion oven placed below the ablation point and the final complexes are "pushed" towards the spectrometer by a pulsed beam of He gas (1 bar) from a home-built nozzle. The effusion oven is simply a Cu block where glycine is placed in a hole and is covered with a conical cap. The whole block is heated with the aid of a 250 W halogen lamp to a temperature of 120 °C. At this temperature, which is measured with iron/constantan thermocouples, the vapor pressure of glycine is 10^{-4} mbar [9].

This combination of laser ablation, effusion of neutral glycine molecules and thermalization with the helium supersonic expansion, produces a molecular beam of metal ion–glycine complexes, which can then be mass analyzed with the TOF mass spectrometer.

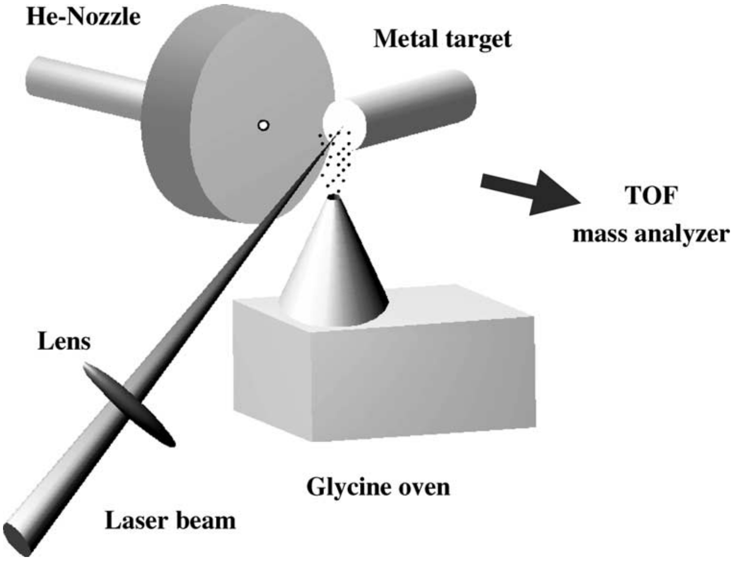

Fig. 1. A schematic diagram of the source employed to produce metal cation–glycine complexes.

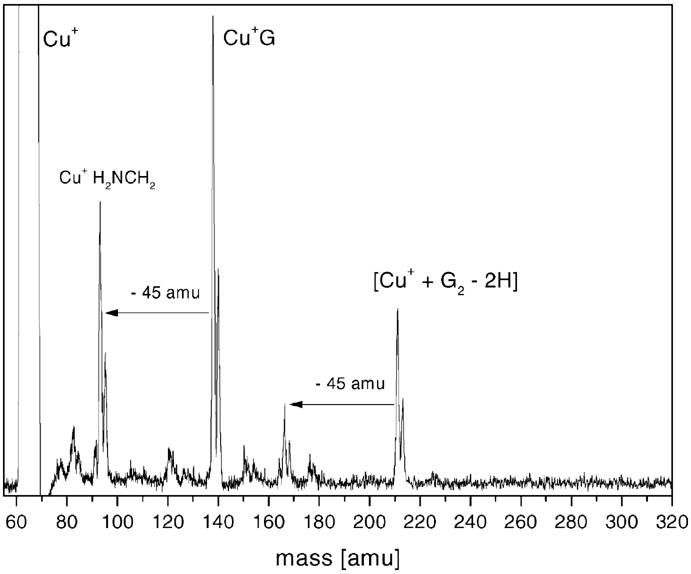

Fig. 2. Mass spectrum that results from the gas-phase reaction of laser ablated Cu^+ and the effusion of glycine (G) in a He nozzle beam.

3. Results and discussion

The products from the gas-phase reactions between cations of Cu and Ag with G are shown in the TOF mass spectra of Figs. 2 and 3 respectively. It is generally accepted that the intensity of the peaks in a mass spectrum reflects the stability of the corresponding ion. In the case of Cu the high intensity peak (out of scale in Fig. 2) corresponds to Cu^+ at $m/z = 63$ and 65 amu (due to two Cu-isotopes with abundance ratio

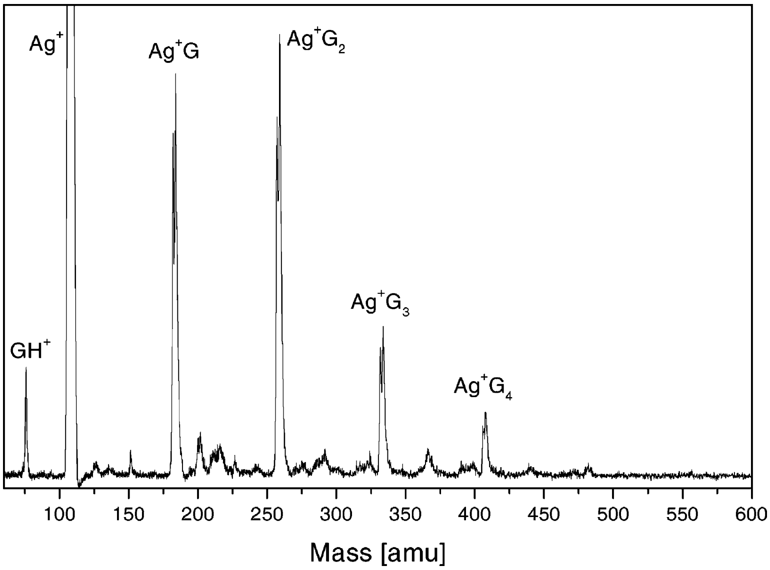

Fig. 3. Mass spectrum that results from the gas-phase reaction of laser ablated Ag^+ and the effusion of glycine (G) in a He nozzle beam.

$^{63}Cu/^{65}Cu \approx 7/3$). There is no intensity at the expected mass of G (H_2NCH_2COOH) at 75 amu or its protonated form GH^+ at 76 amu. Doublets, which correlate with the Cu-isotopes with significant intensities, appear also at masses 93/95, 138/140, 166/168, and 211/213 amu. The peaks at 138/140 and 211/213 amu correspond to Cu^+G and $[Cu+G_2–2H]^+$ respectively. The ions with masses 93/95 and 166/168 amu differ from the previous series at 45 amu. There is no indication for the formation of double charged species.

Cu–G complexes have been produced recently by gas-phase reactions [10], by electrospraying aqueous solutions of Cu sulfate and G [6,11] and by fast atom bombardment (FAB) of sample matrices [3]. In the most cases the mass spectra obtained indicate the formation of Cu^+G (at 138/140 amu) and $[Cu^{2+}G(G–H)^-]^+$ (at 212/214 amu) complexes. Hence, with the exception of the Cu^+G complex, the most ions appearing in the spectrum of Fig. 2 have not been observed before.

The structure of the ion $[Cu+G_2–2H]^+$ (mass 211/213 amu) can not be inferred from the mass spectrum of Fig. 2. This complex can have different forms: $Cu^+(G–2H)G$ or $Cu^+(G–H)_2$, i.e., one or both glycine molecules have undergone dehydrogenation.

The ion with mass 93/95 amu in Fig. 2 can be attributed to $[CuH_2NCH_2]^+$. In our experimental arrangement we assume that it is produced trough two different ways. First, it can be the result of the insertion reaction:

$$\begin{aligned} &Cu^+ + H_2NCH_2COOH \\ &\quad \rightarrow [H_2NCH_2–Cu^+–COOH] \\ &\quad \rightarrow Cu^+H_2NCH_2 + COOH, \end{aligned}$$

where the intermediate is activated via (multi)photon absorption from the laser (∼10 ns pulse duration). This fragmentation channel (removal of a neutral carboxyl radical or loss of 45 amu) has not been observed before, but it is discussed by Polce et al. [3], where they suggest that Cu catalyzes the cleavage of C–COOH bond by insertion. The fragmentation of Cu^+G complex has been investigated in several experiments [3,10,12,13] and the loss of 46 amu is observed in contrast to our results.

A second possibility is that the $[CuH_2NCH_2]^+$ ion is the result from the reaction of immonium ion $H_2NCH_2{}^+$ with a neutral Cu atom. In that case, the immonium ion is the decarboxylated ion fragment of the multiphoton ionization of neutral glycine molecule. This process has been observed in recent experiments of Vorsa et al. [14] concerning the photoionization of amino acids. The immonium ion can also be formed from the loss of neutral CuCOOH from CuG^+ as this has been observed by Wen et al. [13].

In contrast to Cu^+–G complexes where the heaviest species observed is $[Cu+G_2–2H]^+$, in the case of Ag^+–G the development of the Ag^+G_n series with $n = 1–4$ is clearly observed in the spectrum of Fig. 3. The small peaks at masses 483 and 558 amu, which are just visible in the spectrum, correspond to Ag^+G_5 and Ag^+G_6 respectively. Each doublet is due to Ag stable isotopes 107/109 which appear naturally with almost the same abundance. For Ag^+–G complexes, to our knowledge no other experimental data are available.

Comparing now the two transition metals Cu and Ag, it is evident from the spectra that whereas Cu is dicoordinated, Ag can attract up to four ligands. Although the experimental conditions are more or less identical, i.e., the glycine-oven temperature is in both cases the same, the MG_n complexes are formed in the gas phase and reflect the different reactivity/coordination of the two metals. Furthermore, for both transition metal cations no peptide bonds (e.g., glycilglycine with mass = 132 amu) are formed.

Theoretical calculations concerning the binding energies and the geometrical structure of Cu^+–G and Ag^+–G have been performed by several groups [15,16]. These calculations show stable metal–glycine complexes and the lowest energy structure is that, where the metal ion interacts with both the carbonyl oxygen and the amino nitrogen. Furthermore, the interaction of Cu^+ with G is stronger than that of Ag^+ [16].

The different coordination between Cu^+ and Ag^+ with glycine, evident from Figs. 2 and 3, resembles the different binding behavior observed with several other molecules [16,17]. Bauschlicher et al. [18] and Armentrout and coworkers [19,20] have shown that Cu cation tends to bind strongly two ligand molecules (H_2O or NH_3) in a linear arrangement. The binding energies for larger ligands drop substantially down. This is due to the 4s-3dσ hybridization of Cu^+ orbitals and also to ligand–ligand repulsion [18]. This mechanism reduces the electron density along the *z*-axis (ligand–ligand axis) allowing thus the closer approach of the two ligands to the Cu^+ core. Additional ligands placed at out-of-axis positions experience higher repulsion and thus the binding energies are much weaker than the first two ligands. This leads to unstable complexes whose formation is less favorable, as the absence of tricoordinated Cu–G clusters in our spectra of Fig. 2 indicates. Thus, from our data the above discussed mechanism is likely only in the case where Cu^+ binds with only one atom from glycine molecule. In order to infer the structure of such a molecule further experimental and theoretical work is necessary.

For Cu cation the separation energy for a fully promoted electron from the $3d^{10}$ ground state to the first excited $3d^9 4s^1$ state is 2.81 eV, while for Ag cation the $4d^{10}$–$4d^9 5s^1$ promotion energy is 5.04 eV [21]. Thus, the hybridization is much weaker in the case of Ag^+ and therefore this ion behaves like a closed shell alkali ion, where the coordination is dictated from the geometrical size of the ion [22,23]. The size of Ag^+ is larger than that of Cu^+ and the ligand–ligand repulsion is reduced allowing the coordination of more ligands in the case of Ag^+.

Experimental and theoretical studies of Ag^+ with NH_3, [24], HCN and $NCCH_3$ [25] indicate that maximal four ligands can be directly coordinated to Ag cation. The geometrical conformation of Ag^+L_2 is linear, Ag^+L_3 is an equilateral triangle and Ag^+L_4 is a tetrahedron. Additional ligands are less tightly bound to Ag^+. This is also evident in our spectra in Fig. 3 where the intensity for Ag^+G_5 and Ag^+G_6 is very low. These weak complexes can constitute the second solvation shell over the first tetrahedral one.

In summary, we have shown that a simple source based on a combination of laser ablation and effusion of volatile biomolecules is able to produce gas-phase metal–ligand complexes. This is demonstrated in the cases of Cu^+–G and Ag^+–G, where a series of metal–glycine ionic complexes have been observed. From the produced ions the different coordination of the two coinage metal ions is deduced in agreement with previous theoretical expectations.

Acknowledgements

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