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Relative Silver(I) Ion Binding Energies of α-Amino Acids: A Determination by Means of the Kinetic Method

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The relative silver(I) ion binding energies of 19 α-amino acids have been measured by means of the kinetic method. In general, they are similar to the relative copper(I) ion binding energies of corresponding amino acids although there are differences that can be accounted for by differences in silver(I) and copper(I) chemistry. The correlation with proton basicities is comparatively poorer. Again, the differences between silver(I) and proton binding can be attributed to differences in silver(I) and proton chemistry. The relative silver(I) binding energies measured are best described as relative basicities or $\Delta\Delta G^{\circ}_{Ag}$'s. The observed internal consistency during construction of a silver(I) ion basicity ladder implies that $\Delta\Delta S^{\circ}_{Ag}$ is approximately zero except when histidine and lysine are involved. For 16 α-amino acids, their relative silver(I) ion basicities ≈ relative silver(I) ion affinities or $\Delta\Delta G^{\circ}_{Ag} \approx \Delta\Delta H^{\circ}_{Ag}$. (J Am Soc Mass Spectrom 1998, 9, 760–766) © 1998 American Society for Mass Spectrometry

The bio-inorganic chemistry of the silver(I) ion is rich and fascinating. The silver(I) ion has long been used as a bactericide in the form of eye drops for newborns [1, 2]. Some of its complexes display remarkable antimicrobial activities [3, 4]. The metallothioneins, a class of small proteins believed to be responsible for heavy-metal detoxification in mammals, exhibit very high affinity for Ag(I) [5–7]. The silver(I) ion binds relatively strongly to peptides and proteins; collision-induced dissociation of these complexes in the gas phase yields abundant Ag(I)-bound product ions [8].

We report in this article the first measurement of the relative gas-phase silver(I) ion binding energies of 19 essential α-amino acids. Measurements of gas-phase binding energies, especially affinities, of biological ligands for small ions, including protons [9–23], alkali-metal ions [24], and recently Cu(I) [25], are often made with the desire to know the fundamental, intrinsic binding in the absence of water—an environment which approximates that in the interior of proteins. The kinetic method, developed by Cooks and co-workers [26–30], is a particularly effective method for measuring relative ion binding energies of amino acids and peptides because it does not require generating a population of nonvolatile neutral ligands in the gas phase. It is based on measuring the logarithm of the relative abundance of the product ions arising from the dissociation of an ion-bound heterodimer of the ligands, which is proportional to the logarithm of the relative rate of dissociation of the two reaction channels, to estimate the relative binding energies of the two ligands for the ion:

$$[B_i\text{---}M\text{---}B]^+ \rightarrow B + B_iM^+ \text{ (rate constant} = k_i)$$

$$\rightarrow B_i + BM^+ \text{ (rate constant} = k)$$

where M is the central ion, B_i is a reference base whose binding energy to M is known, and B is the unknown base whose binding energy is being measured. Accord-

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ing to the transition state theory [31], the rate constants may be expressed as

$$k_i = (RT_{\text{eff}}Q_i^*/hQ_i) \exp [-\varepsilon_0(i)/RT_{\text{eff}}]$$

$$k = (RT_{\text{eff}}Q^*/hQ) \exp [-\varepsilon_0/RT_{\text{eff}}]$$

R is the gas constant, T_{eff} is a parameter in temperature units that reflects the internal energy of the dissociating heterodimer, Q^* is the partition function of the activated complex, Q is the partition function of the heterodimer, ε_0 is the activation energy, and h is the Planck constant. Because the same ion-bound heterodimer is involved, $Q_i = Q$

$$\ln(k_i/k) = \ln(Q_i^*/Q^*) + [\varepsilon_0 - \varepsilon_0(i)]/RT_{\text{eff}}$$

Assuming that abundances reflect rate constants and that no reverse activation barriers exist [22, 23, 26, 30],

$$\ln([B_iM]^+/[BM]^+) = \ln(Q_i^*/Q^*) - \Delta H^\circ_M/RT_{\text{eff}} + \Delta H^\circ_M(i)/RT_{\text{eff}}$$

where ΔH°_M is the ΔH of the dissociation reaction $BM^+ \rightarrow B+M^+$ or the metal ion affinity of B. If B_i and B are structurally similar, $Q_i^* \approx Q^*$

$$\ln([B_iM]^+/[BM]^+) \approx [\Delta H^\circ_M(i) - \Delta H^\circ_M]/RT_{\text{eff}}$$

A plot of $\ln([B_iM]^+/[BM]^+)$ versus $\Delta H^\circ_M(i)$ for a series of reference bases, B_i's, would be linear with a slope $= 1/RT_{\text{eff}}$ and an intercept/slope $= -\Delta H^\circ_M$. This approach forms the basis of many proton affinity measurements and their results are typically in good agreement with those measured using more accurate, conventional methods [14, 15, 30].

It is not always possible to find reference bases that are structurally similar to the unknown. If the bases are not similar, i.e., $Q_i^* \neq Q^*$, but no reverse activation barriers exist, $\ln(Q_i^*/Q^*) = -\Delta\Delta S^\circ_M/R$ [15, 22, 23], where ΔS°_M is the entropy change of the reaction $BM^+ \rightarrow B+M^+$.

$$\ln([B_iM]^+/[BM]^+) \approx -\Delta\Delta S^\circ_M/R + \Delta\Delta H^\circ_M/RT_{\text{eff}}$$

$$\ln([B_iM]^+/[BM]^+) \approx \Delta\Delta G^\circ_M/RT_{\text{eff}}$$

That is to say, a plot of $\ln([B_iM]^+/[BM]^+)$ versus $\Delta G^\circ_M(i)$ for a series of B_i's would be linear with a slope $= 1/RT_{\text{eff}}$ and an intercept/slope $= -\Delta G^\circ_M$ [15, 29, 32].

Fenselau and co-workers [15, 22], and Cerda and Wesdemiotis [23] provided a different perspective: if the reference bases are not structurally similar to the unknown, but are structurally similar among themselves, and no reverse activation barriers exist, a plot of $\ln([B_iM]^+/[BM]^+)$ versus $\Delta H^\circ_M(i)$ for the B_i's would also be linear with a slope $= 1/RT_{\text{eff}}$, the intercept, however, now equals $\ln(Q_i^*/Q^*) - \Delta H^\circ_M/RT_{\text{eff}}$. If measurements are made at several collision energies (different T_{eff}'s), a plot of the intercepts [i.e., $\ln(Q_i^*/Q^*) - \Delta H^\circ_M/RT_{\text{eff}}$] versus $1/RT_{\text{eff}}$ would be linear with a slope $= -\Delta H^\circ_M$ and an intercept $= \ln(Q_i^*/Q^*)$. Because

$$\ln(Q_i^*/Q^*) = -\Delta\Delta S^\circ_M/R = [\Delta S^\circ_M - \Delta S^\circ_M(i)]/R$$

Not only can the ΔH°_M be measured, but the ΔS°_M, the entropy change of the demetallation reaction, can also be calculated from known $\Delta S^\circ_M(i)$'s.

For the measurement of silver(I) binding energies, very few ΔH°_{Ag} values are known and they are all for simple ligands [33, 34]. The kinetic method, however, can still be used to estimate the relative Ag^+ binding strength of α-amino acids by measuring the dissociation of a silver(I)-containing heterodimer of the α-amino acids. [This is identical to the approach of Cerda and Wesdemiotis [25] for the determination of Cu(I) binding energies of α-amino acids.] Since the α-amino acids all contain different sidechains, most of which bear functional groups that can participate in metal ion binding, their metal ion-binding chemistry is rich and it is debatable if they are structurally and chemically similar for the purpose of the kinetic method [22, 23, 30]. For this reason, the relative silver(I) binding energies measured are more accurately described as relative silver(I) basicities or $\Delta\Delta G^\circ_{Ag}$'s. We will show later on that the $\Delta\Delta S^\circ_M$'s for the majority of the α-amino acids are ≈ 0. That is to say, for the majority of the α-amino acids, $\Delta\Delta G^\circ_{Ag} \approx \Delta\Delta H^\circ_{Ag}$.

In this study, we produced gas-phase Ag(I)-bound heterodimers of α-amino acids by electrospraying a solution containing Ag(I) and the two constituent α-amino acids. The relative abundances of the silver(I)-bound monomer fragment ions were then measured to give a relative silver(I) binding energy scale of the α-amino acids.

Experimental

Experiments were conducted on an atmospheric pressure ionization mass spectrometer of triple quadrupole (QqQ) design (TAGA 6000E, SCIEX, Concord, Ontario, Canada). The electrospray probe was fabricated from an approximately 3-cm-long, 33-gauge stainless steel tube (Hamilton, $\sim$ 100 μm i.d.) that had been attached to a length of 1/16-in.-o.d. stainless steel tube with epoxy glue. The probe tip was electropolished prior to use. The optimum probe position was established from time to time, but was typically with the tip about 1–2 cm from the interface plate and with the spray off-axis from the orifice. Biasing of the probe tip was achieved via a 50 MΩ current-limiting resistor in series with a high-voltage power supply (Tennelec, Model TC 950) set typically between 2.5 and 3.5 kV. The electrospray current was monitored via a custom-built microammeter that could be floated above ground.

Table 1. Reference bases (B_i's) for leucine, histidine, and lysine

B_i (Leu)	Ala	Val	Ser	Thr	Met
PA[a] (kcal/mol)	215.5	217.6	218.6	220.6	223.2
GB[a] (kcal/mol)	207.4	209.5	210.5	212.5	215.5
ΔS°_H (cal/K mol)	27.18	27.18	27.18	27.18	25.84
B_i (His and Lys)	$N(Me)_3$	$N(Et)_3$	$N(Pr)_3$	$N(Bu)_3$	
PA[a] (kcal/mol)	226.8	234.7	237.1	238.6	
GB[a] (kcal/mol)	219.4	227.3	229.7	231.1	
ΔS°_H (cal/K mol)	24.83	24.83	24.83	25.17	

[a][37] and [38].

Gas-phase silver(I)-bound heterodimers of α-amino acids were generated by means of electrospraying 50/50 water/methanol solutions containing a binary mixture of α-amino acids, 1 mM each, and 2 mM of silver(I) nitrate. Altogether, 52 different Ag(I)-bound dimers of 19 constituent α-amino acids were examined; the only common α-amino acid that was omitted was cysteine, which precipitates with Ag^+ [35]. To measure the relative abundance of the Ag(I)-bound monomer fragment ions, the Ag(I)-bound heterodimer ion was mass selected in the first quadrupole (Q1), fragmented in q2 via collision with Ar ($E_{\text{center-of-mass}}$ (E_{cm}) = 3.57 eV; collision gas thickness = 0.5×10^{14} atoms cm^{-2} [36]), and the product ions mass analyzed in Q3 with a dwell time of 10–50 ms per mass-to-charge ratio unit. The T_{eff} of the dissociating silver(I)-bound heterodimers was estimated from that of dissociating proton-bound heterodimers measured at the same time and under identical conditions.

Results and Discussion

Because the kinetic method is mostly practiced on sector instruments, a preliminary investigation was carried out to ascertain that it may be used on our triple quadrupole mass spectrometer. The exercise involved measuring the proton affinities and basicities of leucine, histidine, and lysine using both the original Cooks' procedure [26, 29, 30] as well as Fenselau's modification [22, 23]. The reference bases chosen for leucine were alanine, valine, serine, threonine, and methionine, whereas those for histidine and lysine were trimethylamine, triethylamine, tripropylamine, and tributylamine. Table 1 lists their recently evaluated proton affinities (PAs), basicities (GBs) and ΔS°_H's [37, 38]. The fact that the reference bases are all amino acids or tertiary amines and they have similar ΔS°_H's satisfied the requirement of structural similarity among the reference bases for an application of Fenselau's modification. The measurements were carried out under five collision energies: E_{cm} = 1.80, 2.50, 3.00, 3.75, and 4.20 eV. For leucine, the proton affinities and basicities measured using the original Cooks' procedure were independent of collision energy. For histidine and lysine, these values were found to decrease with increasing collision energy. This effect had been attributed to differences in entropies between the binding of the reference base and that of lysine or histidine [15]. With Fenselau's modification, accurate proton affinities and basicities were obtained; these are tabulated in Table 2, along with those of the Cooks' method and the evaluated values [37, 38]. It is evident that accurate measurements of thermochemical data may be performed using the kinetic method with our triple quadrupole instrument.

Having satisfied ourselves that the method performed satisfactorily, we then proceeded to the measurement of silver(I) binding energies of the α-amino acids. Six representative product ion spectra are shown in Figure 1. The product ion spectra are simple and comprise silver(I)-bound amino acids as fragment ions. This condition was almost always met throughout this

Table 2. PAs, GBs, and ΔS°_H's of leucine, histidine, and lysine[a]

	Cooks'		Fenselau's			Reference	
B	PA[b]	GB[b]	PA	ΔS°_H[c]	GB[d]	PA[e]	GB[e]
Leucine	219.2 ± 0.1	211.1 ± 0.1	219.2	27.20	211.1	218.6	210.5
Histidine	228.2 ± 0.8	220.8 ± 0.8	236.6	34.27	226.4	236.1	226.8
Lysine	227.4 ± 0.9	220.0 ± 0.9	238.1	38.77	226.6	238.0	227.3

[a]All PAs and GBs in kcal/mol; all ΔS°_H's in cal/K mol.
[b]Average of 5 E_{cm}'s; uncertainty of one standard deviation.
[c]$\Delta S^\circ_H = \Delta S^\circ_H(i) - \Delta\Delta S^\circ_H$.
[d]$GB = PA - T\Delta S^\circ_H$.
[e][37] and [38].

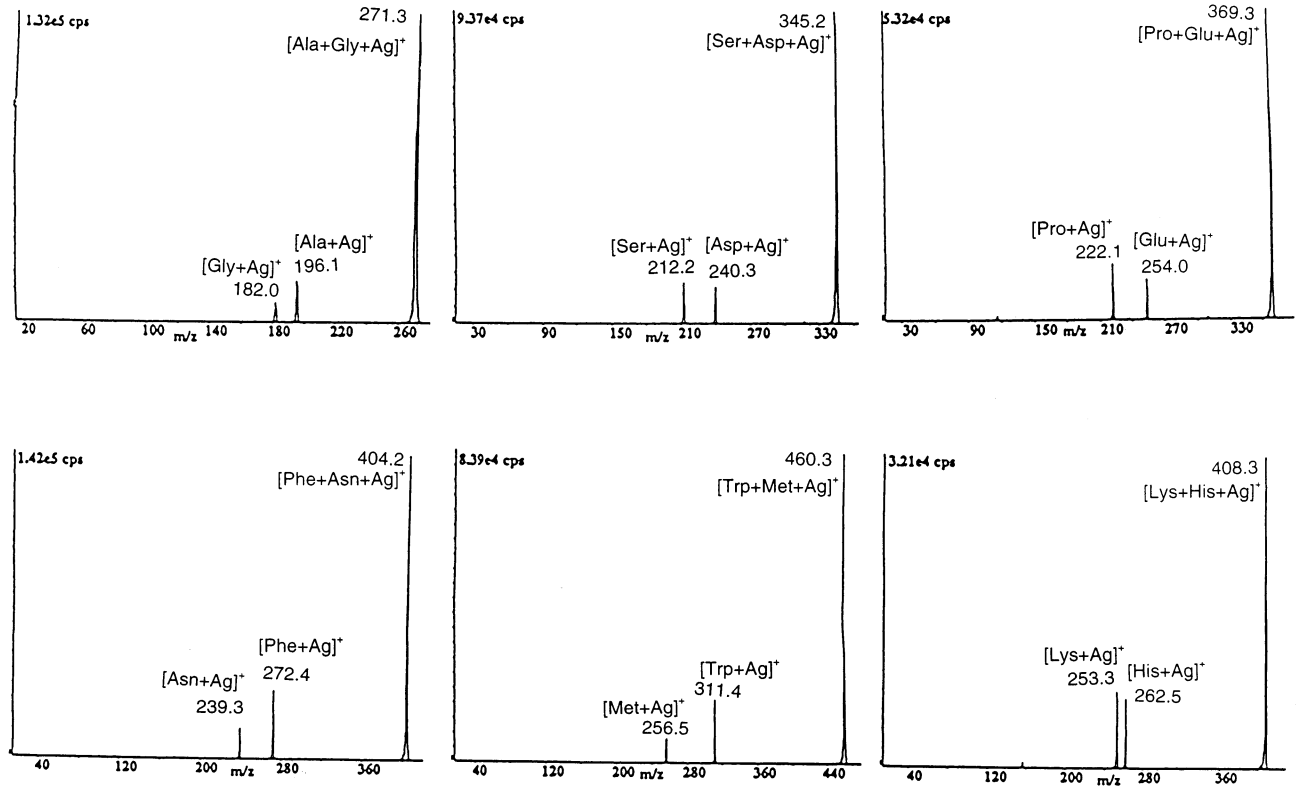

Figure 1. Product ion spectra of Ag(I)-bound heterodimers of α-amino acids.

study, which satisfied the requirement of relatively weak ion–ligand bond. When other fragment ions were present, their total abundance was always <5% of the total abundance of the two Ag(I)-amino acid product ions (the only exception being the dissociation of the dimer ion, [lysine–Ag–arginine]$^+$). It is apparent by means of simple inspection that the silver(I) ion binding energy of alanine is higher than that of glycine, serine higher than aspartic acid, proline higher than glutamic acid, etc.

Figure 2 shows a silver(I) ion-binding ladder of the α-amino acids constructed from results of the 52 pairs of amino acids that were examined. It is evident that the $\ln(k_1/k_2)$ values measured are internally consistent for all pairs with the exception of those involving histidine and lysine. For example, (a) $\ln(k_1/k_2)$ for the tyrosine/serine pair is 3.40, which is comparable to the value obtained by summing the values of six amino acid pairs, i.e., 0.76+0.05+0.25+1.67+0.76+0.04 = 3.53 or (b) that for the serine/alanine pair is 0.98, a value comparable to that obtained by summing five amino acid pairs, 0.63+0.05+0.15+0.11+0.04 = 0.98. Most differences are no larger than 0.2 with the exception of those involving histidine and lysine. For example, (a) $\ln(k_1/k_2)$ for the glutamine/histidine pair is found to be 3.08, a value that is very different from summing three sequential values 1.43+0.90+2.39 = 4.72, and (b) $\ln(k_1/k_2)$ for the lysine/tryptophan pair is 3.63, whereas that from the sum of two amino acid pairs, 2.39+0.32 = 2.71. As a result of these inconsistency, all measurements involving histidine and lysine were judged to be less reliable and they are indicated as such by means of dashed lines. As others have pointed out [12, 15–19], the discrepancies indicate that the dissociation of Ag–histidine and Ag–lysine complex may be associated with entropic changes that are different from those of the other amino acids. Arginine has a silver(I) ion binding energy much higher than that of the next higher amino acid, lysine; in this article, we are reporting only a lower limit for arginine.

To report relative silver(I) ion binding energies of the α-amino acids in units of kcal/mol, the effective temperature, T_{eff}, of the dissociating silver(I) ion-bound dimers needs to be estimated. Here, as in previous studies [24, 25, 39, 40], we assume that the effective temperature of dissociating proton-bound dimers measured along with the silver(I) ion-bound dimers to be a good estimate of the effective temperature of the silver-containing complexes. Table 3 shows the relative silver(I) ion binding energies measured for the 19 α-amino acids; the uncertainties show one standard deviation of five measurements with T_{eff} = 902 ± 26 K. It is apparent that quite a few pairs of amino acids have comparable silver(I) ion binding energies, e.g., valine and leucine, aspartic acid and serine, threonine and glutamic acid, and phenylalanine and tyrosine.

Table 3 also lists the relative copper(I) ion affinities [25] and relative proton basicities of the α-amino acids

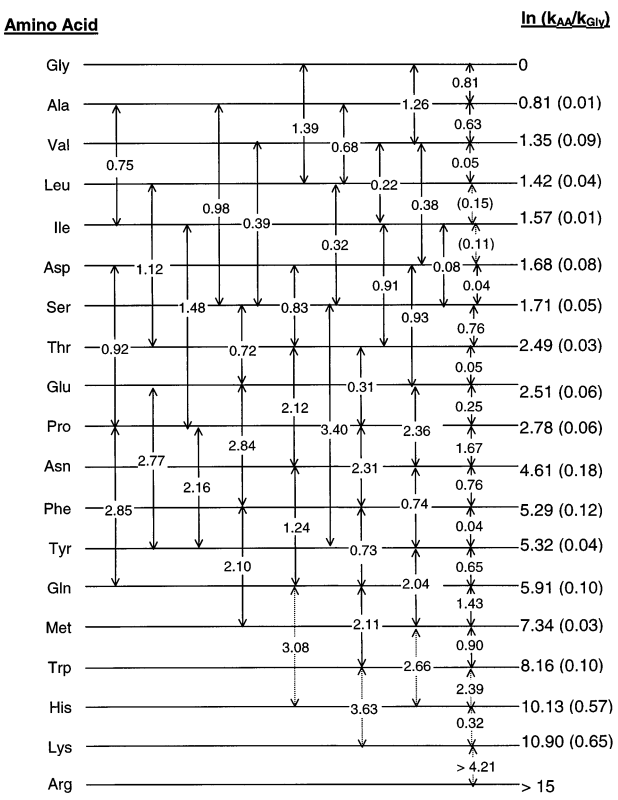

Figure 2. Silver(I) ion-binding ladder of the α-amino acids.

[37, 38]. Overall, the ranges of energies decrease from the proton to silver(I) and to copper(I). The correlation between Ag(I) ion binding and Cu(I) ion binding is shown in Figure 3. The full line is the best fit among the data points (r^2 = 0.930), whereas the dashed line shows a one-to-one correspondence of the energies. For α-amino acids that contain alkyl sidechains, such as valine, leucine and isoleucine, their relative silver(I) ion binding energies are smaller than their corresponding copper(I) ion binding energies. This observation is in accordance with the longer Ag(I)–N bond relative to the Cu(I)–N bond, which makes the inductive effect of the alkyl sidechain less effective for the silver-containing complexes [41–44]. For the two amino acids that contain carboxylate sidechains, aspartic and glutamic acid, their relative silver(I) ion binding energies are also smaller than their corresponding copper(I) ion binding energies; the silver(I) ion is a softer Lewis acid than the copper(I) ion, and hence will bind less strongly to hard bases, such as the oxygen sites on the sidechain carboxylic group [45–47]. For amino acids that contain nitrogen-bearing sidechains, i.e., histidine, tryptophan, glutamine, and asparagine; aromatic sidechains, i.e., tryptophan, tyrosine, and phenylalanine; and sulfur-bearing sidechain, i.e., methionine; their relative silver(I) ion binding energies are larger than their corresponding copper(I) ion binding energies. This effect is likely due to more favorable binding because of Ag(I)'s larger size [41–44] and softer properties relative to Cu(I) [45–47].

Table 3. Relative binding energies (kcal/mol) of α-amino acids to silver(I), copper(I), and the proton

Amino acid	$\Delta\Delta G^\circ_{Ag}$ [a]	$\Delta\Delta H^\circ_{Cu}$ [b]	ΔGB [c]
Gly (G)	0.0	0.0	0.0
Ala (A)	1.4 ± 0.0	1.7	3.7
Val (V)	2.4 ± 0.2	3.7	5.8
Leu (L)	2.5 ± 0.1	4.1	6.8
Ile (I)	2.8 ± 0.0	4.3	7.6
Asp (D)	3.0 ± 0.2	5.0	5.4
Ser (S)	3.2 ± 0.4	3.1	6.8
Thr (T)	4.6 ± 0.1	4.6	8.8
Glu (E)	4.6 ± 0.1	7.2	6.4
Pro (P)	5.0 ± 0.1	4.8	8.1
Asn (N)	8.3 ± 0.3	6.7	9.4
Phe (F)	9.5 ± 0.3	8.0	8.8
Tyr (Y)	9.6 ± 0.1	8.3	9.6
Gln (Q)	10.7 ± 0.2	9.8	11.4
Met (M)	13.1 ± 0.1	10.4	11.8
Trp (W)	14.5 ± 0.2	11.5	15.0
His (H)	18.0 ± 1.0	13.3	23.1
Lys (K)	19.8 ± 1.2	>13.3	23.6
Arg (R)	>26.8	>13.3	36.9

[a] $\Delta\Delta G^\circ_{Ag} \approx \Delta\Delta H^\circ_{Ag}$ except for His, Lys, and Arg; uncertainties show standard deviations of 5 measurements; T_{eff} = 902 ± 26 K.
[b] [25].
[c] [37] and [38].

Figure 4 shows the correlation between relative silver(I) ion binding energies and relative proton basicities. Relative to that in Figure 3, the degree of correlation exhibited in Figure 4 (r^2 = 0.897) is poorer. The amino acids that show disproportionally large relative Ag(I) binding versus relative proton binding are tryptophan, methionine, phenylalanine, tyrosine, asparagine, and glutamine; the sidechains of these amino acids interact much more strongly with the silver(I) ion than with the proton [34, 49–52]. Those amino acids that exhibit disproportionally small relative Ag(I) binding in comparison to relative proton binding are valine, leucine, isoleucine, serine, and threonine; valine, leucine, and isoleucine have alkyl sidechains that better stabilize the shorter proton–nitrogen bond than the longer silver–nitrogen bond; serine and threonine have hydroxyl-containing sidechains that interact more strongly with the harder proton than with the softer silver(I) ion.

In spite of the assumptions and potential inaccuracy of the kinetic method, the internal consistency displayed in the laddering of the amino acids for silver(I) binding, and the good correlations exhibited in the comparisons with copper(I) ion and proton binding, show that the method provides, at the very least, useful approximations of binding energies, especially for nonvolatile bases or ligands where no alternative method exists. A recent high-level ab initio study showed that the amino acids glycine, serine, and cysteine bind the copper(I) ion via di- and even tricoordination [48]. The copper(I) ion is bound to the amino nitrogen, the carbonyl oxygen, and for serine and cysteine, the oxygen and sulfur on the sidechain. In other words,

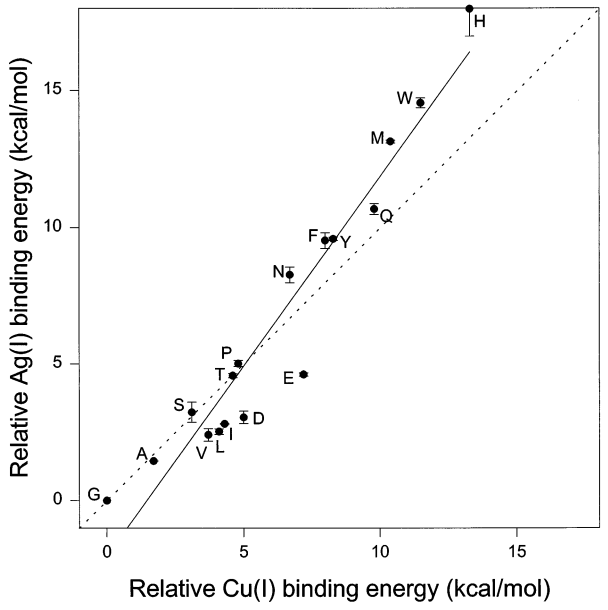

Figure 3. Relative Ag(I) binding energies vs. relative Cu(I) binding energies of the α-amino acids: full line, best fit with r^2 = 0.930; dashed line, 1:1 correspondence.

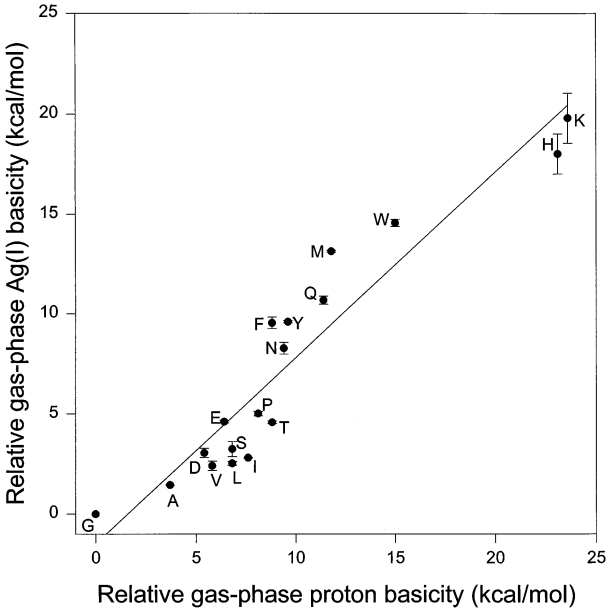

Figure 4. Relative gas-phase Ag(I) basicities vs. relative gas-phase proton basicities of the α-amino acids: r^2 = 0.897.

the metal ion interacts with the two termini as well as the sidechain of the amino acid. Assuming first that the binding of amino acids to silver(I) is similar to that to copper(I), and second that the nature of the binding is the same irrespective of whether one or two amino acids are involved, it may be speculated that the binding of silver(I) to the amino acids in the heterodimer may not be all that different from the copper binding described above. It may be reasoned that, given the structural differences among the amino acids (the sidechains are different), the relative entropy changes of demetallation ($\Delta\Delta S^\circ_{Ag}$) among the amino acids may be significant for certain pairs. This is certainly correct for pairs involving histidine and lysine (vide supra).

As pointed out in an earlier section, the relative silver(I) binding energies measured are more accurately described as relative silver(I) ion basicities rather than affinities because of the structural differences among the amino acids. The relative silver(I) ion basicities measured would be those at an ion temperature that is unknown and, therefore, undefined. As it turns out, the internal consistency displayed in the laddering of the silver(I) ion basicities of 16 out of 18 amino acids (the exceptions being histidine and lysine) strongly suggest that $\Delta\Delta S^\circ_{Ag}$ among the 16 amino acids is approximately zero. Because $\Delta\Delta G^\circ_{Ag} = \Delta\Delta H^\circ_{Ag} + T\,\Delta\Delta S^\circ_{Ag}$, $\Delta\Delta G^\circ_{Ag} \approx \Delta\Delta H^\circ_{Ag}$ for 16 α-amino acids. That is to say, the relative silver(I) binding energies displayed in Table 1 may be defined as either basicities or affinities for the first 16 amino acids (from glycine to tryptophan). For histidine and lysine, the values are more appropriately described as relative silver(I) basicities of an undefined temperature.

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References

1. Petering, H. G. *Pharmac. Ther. A* **1976,** *1*, 127–130.
2. Wigley, R. A.; Brooks, R. R. In *Noble Metals and Biological Systems*; Brooks, R. R., Ed.; CRC: Boca Raton, 1992; pp 277–297.
3. Nomiya, K.; Kondoh, Y.; Nagano, H.; Oda, M. *J. Chem. Soc. Chem. Commun.* **1995,** 1679–1680.
4. Nomiya, K.; Onoue, K. I.; Kondoh, Y.; Kasuga, N. C.; Nagano, H.; Oda, M.; Sakuma, S. *Polyhedron* **1995,** *14*, 1359–1367.
5. Narula, S. S.; Mehra, R. K.; Winge, D. R.; Armitage, I. M. *J. Am. Chem. Soc.* **1991,** *113*, 9354–9358.
6. Stillman, M. J.; Presta, A.; Gui, Z.; Jiang, D.-T. In *Metal-Based Drugs Vol. 1*. Gielen, M., Ed.; Freund: London, 1994; pp 375–393.
7. Gui, Z.; Green, A. R.; Kasrai, M.; Bancroft, G. M.; Stillman, M. J. *Inorg. Chem.* **1996,** *35*, 6520–6529.
8. Li, H.; Siu, K. W. M.; Guevremont, R.; Le Blanc, J. C. Y. *J. Am. Soc. Mass Spectrom.* **1997,** *8*, 781–792.
9. Moet-Ner, M.; Hunter, E. P.; Field, F. H. *J. Am. Chem. Soc.* **1979,** *101*, 686–689.
10. Locke, M. J.; McIver, R. T., Jr. *J. Am. Chem. Soc.* **1983,** *105*, 4226–4232.
11. Bojesen, G. *J. Am. Chem. Soc.* **1987,** *109*, 5557–5558.
12. Bojesen, G.; Breindahl, T. *J. Chem. Soc. Perkin Trans. 2* **1994**, 1029–1037.
13. Isa, K.; Omote, T.; Amaya, M. *Org. Mass Spectrom.* **1990,** *25*, 620–628.
14. Wu, Z.; Fenselau. C. *Rapid Commun. Mass Spectrom.* **1992,** *6*, 403–405.
15. Wu, Z.; Fenselau. C. *Rapid Commun. Mass Spectrom.* **1994,** *8*, 777–780.

16. Gorman, G. S.; Spier, J. P.; Turner, C. A.; Amster, I. J. *J. Am. Chem. Soc.* **1992,** *114*, 3986–3988.
17. Bliznyuk, A. A.; Schaefer, H. F. III; Amster, I. J. *J. Am. Chem. Soc.* **1993,** *115*, 5149–5154.
18. Campbell, S.; Beauchamp, J. L.; Rempe, M.; Lichtenberger, D. L. *Int. J. Mass Spectrom. Ion Processes* **1992,** *117*, 83–99.
19. Li, X.; Harrison, A. G. *Org. Mass Spectrom.* **1993,** *28*, 366–371.
20. Lias, S. G.; Liebman, J. F.; Levin, R. D. *J. Phys. Chem. Ref. Data* **1984,** *13*, 695–808.
21. Lias, S. G.; Bartmess, J. E.; Liebman, J. F.; Holmes, J. L.; Levin, R. D.; Mallard, W. G. *J. Phys. Chem. Ref. Data,* **1988,** *17*, Suppl 1, 1–861.
22. Cheng, X.; Wu, Z.; Fenselau, C. *J. Am. Chem. Soc.* **1993,** *115*, 4844–4848.
23. Cerda, B.; Wesdemiotis, C. *J. Am. Chem. Soc.* **1996,** *118*, 11884–11892.
24. Bojesen, G.; Breindahl, T.; Andersen, U. N. *Org. Mass Spectrom.* **1993,** *28*, 1448–1452.
25. Cerda, B. A.; Wesdemiotis, C. *J. Am. Chem. Soc.* **1995,** *117*, 9734–9739.
26. Cooks, R. G.; Kruger, T. L. *J. Am. Chem. Soc.* **1977,** *99*, 1279–1281.
27. McLuckey, S. A.; Cameron, D.; Cooks, R. G. *J. Am. Chem. Soc.* **1981,** *103*, 1313–1317.
28. McLuckey, S. A.; Schoen, A. E.; Cooks, R. G. *J. Am. Chem. Soc.* **1982,** *104*, 848–850.
29. Majumdar, T. K.; Clairet, F.; Tabet, J.-C.; Cooks, R. G. *J. Am. Chem. Soc.* **1992,** *114*, 2897–2903.
30. Cooks, R. G.; Patrick, J. S.; Kotiaho, T.; McLuckey, S. A. *Mass Spectrom. Rev.* **1994,** *13*, 287–339.
31. Robinson, P. J.; Holbrook, K. A. *Unimolecular Reactions*; Wiley Interscience: London, 1972.
32. Wu, Z.; Fenselau, C. *Tetrahedron* **1993,** *49*, 9197–9206.
33. Holland, P. M.; Castleman, A. W., Jr. *J. Phys. Chem.* **1982,** *76*, 4195–4205.
34. Deng, H.; Kebarle, P. *Proceedings of the 45th ASMS Conference*, Palm Springs, CA, June 1–5, 1997.
35. Greenstein, J. P.; Winitz, M *Chemistry of the Amino Acids Vol 1*; Wiley: New York, 1961; pp 646–647.
36. Dawson, P. H.; French, J. B.; Buckley, J. A.; Douglas, D. J.; Simmons, D. *Org. Mass Spectrom.* **1982,** *17*, 205–211.
37. Hunter, E. P.; Lias, S. G. *J. Phys. Chem. Ref. Data*, in press.
38. NIST Chemistry WebBook, http://webbook.nist.gov/chemistry.
39. Chen, L.-Z.; Miller, J. M. *Org. Mass Spectrom.* **1992,** *27*, 883–890.
40. Chen, L.-Z.; Miller, J. M. *Organomet. Chem.* **1993,** *448*, 225–232.
41. Cromer, D. T.; Larson, A. C.; Roof, R. B., Jr. *Acta Crystallogr.* **1965,** *19*, 192–197.
42. Yamaguchi, T.; Lindqvist, O. *Acta Chem. Scand.* **1983,** *A37*, 685–689.
43. Lewin, A. H.; Michel, R. J.; Ganis, P.; Lepore, U.; Avitabile, G. *J. Chem. Soc. Chem. Commun.* **1971**, 1400–1401.
44. Nilsson, K.; Oskarsson, A. *Acta Chem. Scand.* **1982,** *A36*, 605–610.
45. Pearson, R. G. *J. Am. Chem. Soc.* **1963,** *85*, 3533–3539.
46. Fleming, I. *Frontier Orbitals and Organic Chemical Reactions*; Wiley: New York, 1976; pp 34–85.
47. Glusker, J. P. *Adv. Protein Chem.* **1991,** *42*, 1–76.
48. Hoyau, S.; Ohanessian, G. *J. Am. Chem. Soc.* **1997,** *119*, 2016–2024.
49. Winstein, S.; Lucas, H. J. *J. Am. Chem. Soc.* **1938,** *60*, 836–847.
50. Andrews, L. J.; Keefer, R. M. *J. Am. Chem. Soc.* **1949,** *71*, 3644–3647.
51. Featherstone, W.; Sorrie, A. J. S. *J. Chem. Soc.* **1964,** 5235–5242.
52. Thompson, N. R. *The Chemistry of Copper, Silver and Gold*; Pergamon: Oxford, 1973; pp 79–128.