Complexation of Na^+ and K^+ to Aromatic Amino Acids: A Density Functional Computational Study of Cation-π Interactions

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Binding energies were calculated for the complexes of Na^+ and K^+ with phenylalanine (*Phe*), tyrosine (*Tyr*), and tryptophane (*Trp*), along with energies of low-energy conformers of the neutral amino acids. Structures were optimized and energies determined by density functional theory (DFT) with the B3LYP functional, using a basis set of 6-31+g(d) on all, or nearly all, heavy atoms. For all but one cation/ligand system, the most energetically favorable binding geometry was the tridentate N/O/Ring chelate. For K^+/*Trp*, however, the advantage of placing the metal ion over the phenyl region of the indole side chain was dominant, leading to a most favored bidentate O/Ring binding geometry. All of the systems, and particularly the *Trp* systems, have multiple conformers with stabilities within a few kcal mol^{-1} of the most stable. Zwitterion forms of the complexes were not unreasonable, but were less stable than the normal forms by ~5 kcal mol^{-1}. To assess the importance of cation−π interactions, conformers were examined in which the side chain was rotated out of chelation. This indicated cation−π stabilization energies of ~5 kcal mol^{-1}.

Introduction

Although the affinity of π faces for cationic centers has long been appreciated in simple molecular systems, it is more recently that this interaction has come to the forefront in considerations of biological systems.[1−17] In particular, such interactions have been discussed as structural determinants in folding and assembly of large systems,[1,2,8−11,17−20] and as playing a central role in the functioning of ionic channels in the transport of metal ions into and out of cells.[12−15,21−24] The latter idea gives special motivation to the present study, because it is the binding of Na^+ and K^+ to exposed π faces of aromatic acids associated with the ionic channel that is hypothesized to play a role in the selective transport of these metal ions through the channel.[25,26]

The cation−π binding motif has an obvious biological role in the interaction of the ubiquitous Na^+ and K^+ metal ions with the three aromatic amino acids phenylalanine (*Phe*), tyrosine (*Tyr*), and tryptophane (*Trp*), and it is toward a more secure and quantitative characterization of these interactions that the present work is directed. Histidine might in principle also be a site of cation−π interactions, but as argued by Mecozzi et al.,[5] it is actually an unlikely candidate both on electrostatic grounds and also on the grounds that it is likely to be protonated, so this possibility is not pursued here.

Full molecular understanding of these situations must begin with a detailed characterization of the binding of these metal ions to the simple, gas-phase monomeric amino acids. On the experimental side, the "kinetic method" of mass spectrometry[27−30] has been applied to binding energy measurements of Ag^+[31] and Cu^+[32] to these molecules. A forthcoming study also using the kinetic method,[33] parallel to the present work, reports apparently the first experimental binding energies of Na^+ and K^+ to these amino acids.

Complementary to such experiments, computational study of these systems is important for several reasons: (a) density functional (DFT) calculations are now feasible with sufficient basis sets to give accuracy and confidence comparable to experiment; (b) the "kinetic method" experimental approach, which is thus far the only one which has been applied to these problems, is not definitive, since it is not a rigorous binding energy measurement, but depends on various approximations and assumptions; (c) only by computation can we readily examine the different possible binding sites and geometries of a given complex, and compare their energetics; (d) computation offers the possibility of dissecting the cation−π interaction contribution to the binding out of the total binding interaction, so that we can assess its actual importance in determining structures and energies of metal−ion complexes. The present work takes the computational approach to addressing three questions about the gas-phase complexation in these systems. First, the preferred sites and geometries of metal ion binding are explored. Second, relative and absolute binding energies are assigned for the different ions and neutrals. And third, the quantitative contribution of the cation−π interaction to the binding in these systems is assessed.

As metal−amino acid interactions are considered in a variety of contexts, particularly in biological systems, the architectural constraints of various systems will favor binding modes differing from the most stable gas-phase complexation geometry. Thus one of the principal aims of the present work was to locate and characterize other low-energy conformers of the complexes besides the most stable ones. It is a daunting task to characterize all the myriad possible sites and binding geometries in systems of such complexity, but characterization of even a limited number of low-energy conformers can serve to suggest likely modes of binding which can carry over to other contexts.

Methods

Current work is active in applying a variety of quantum chemical approaches to complexes of metal cations with small-to-medium-sized molecules. For transition metals, DFT calculations are attractive, combining accuracy comparable to other approaches with computational tractability (for example, see ref

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34 for a review of some of this work). Main-group metal ions are somewhat less demanding, and various approaches are current, including DFT, MP2, CI, and G2 methods. Stöckigt[35,36] has made systematic comparisons of different methods for Al^+ binding to π ligands. Of interest to the present work, he considered that the B3LYP-DFT binding energies for these systems were systematically too low by ~5 kcal mol^{-1}.

More important to the present work are recent studies of Na^+ complexes by two groups, reported by Hoyau et al.[37] and by Armentrout and Rodgers.[38] Hoyau et al.[37] used MP2 theory to calculate an extensive series of complexes, and compared their calculations to a small set of experimental measurements by high-pressure mass spectrometry. They also reviewed the available thermochemical literature for Na^+ complexes. Armentrout and Rodgers[38] made an extensive series of measurements by threshold collision-induced dissociation, and also made a systematic comparison of calculated binding energies using a variety of computational approaches. These studies give a good picture of the variations among different computational approaches to Na^+ binding energies, and the level of agreement with the best experimental values. As a generality, the various computed values span the experimental values, and show variations of a few kcal mol^{-1}. Basis set explorations generally show that inclusion of polarization and diffuse functions is indispensable for obtaining results with reasonable confidence at a level of a few kcal mol^{-1} accuracy.

The present quantum chemical calculations were all carried out with the DFT approach, using the B3LYP hybrid functional. For the present purposes, the choice of the DFT approach was clear, because other sufficiently accurate approaches are computationally too demanding for these large systems using adequate basis sets. Within the DFT realm, the choice of the B3LYP hybrid functional was less clear-cut. This functional was chosen because it has been widely used, making it perhaps easier to correlate our results with other work, and because its demonstrated success for transition metal cationic systems will ease future extension of the present studies to transition metal complexes of these amino acids. For Na^+ complexation, Armentrout and Rodgers[38] found the B3P86 functional to be somewhat better than B3LYP in terms of agreement with more accurate theoretical approaches and with their experimental results. B3LYP results appear to be on the order of 1 kcal mol^{-1} too high, and also scatter somewhat more widely. Also relevant to the present purposes is the fact that B3P86 gives Na^+ a slightly greater phenol binding increment relative to benzene (0.7 kcal mol^{-1}, compared with 0.2 kcal mol^{-1} for B3LYP). However, their measured increment from benzene to phenol is 2.4 kcal mol^{-1}, suggesting that neither of these functionals serves to produce fully adequate comparisons of benzene vs phenol systems, a problem which presumably carries over to the present comparison of *Phe* and *Tyr* complexes. It may be interesting in the future to revisit these systems with other functionals besides B3LYP, and with other more intrinsically accurate ab initio approaches.

The GAUSSIAN 94[39] and GAUSSIAN 98[40] program suites were used. The basis sets used in energy calculations were in general as follows: 6-31g(d) (double-ζ plus polarization) on hydrogen, 6-31+g(d) (polarization and diffuse functions) on first-row atoms and on Na and K. In those calculations done with the GAUSSIAN 94 package, the polarization functions for K were taken from literature values,[41] and the diffuse functions on K were generated by us. (Results using this home-constructed set of diffuse functions did not differ significantly from those later obtained using the 6-31+g(d) basis functions contained in the GAUSSIAN 98 package, and were considered essentially equivalent to them.) In the Na^+/*Trp* calculations, the three side-chain carbons were used without diffuse functions (6-31g(d)), and in the K^+/*Trp* calculations diffuse functions were also removed from three of the ring carbons far from the metal ion. Geometries were fully optimized in all cases at the same calculational level as the energy calculations, except that for the K^+/*Trp* complexes the geometries were optimized at the lower level of 6-31+g(d) on K and 6-31g(d) on all first-row atoms.

As a test case, a calculation of Na^+/glycine was carried to a higher level of basis, 6-31g(d,p) on H, and 6-311++g(2df) on all heavy atoms. Results with various sizes of basis ranging from 6-31g up to this large basis indicated variations in binding energy of this complex of less than 1−2 kcal due to basis set incompleteness, as long as the basis was at least as large as 6-31g(d) on all atoms. In another set of model calculations on Na^+/benzene, it was found that the calculated binding energy was unchanged within ~0.2 kcal as the basis set was progressively expanded from a basis set similar to that used here (6-31+g(d) on all heavy atoms, 6-31g(d) on H) up to a large basis set of 6-311++g(3d) on Na, 6-31+g(3d) on C, and 6-31+g(d,p) on H. (In this latter case, smaller basis sets without diffuse functions on the carbons did not diverge greatly in binding energy, but the BSSE corrections of several kilocalories made the results less satisfactory.) These model studies strengthened our confidence that the basis sets used for the amino acid complexes, which included polarization functions on all atoms and diffuse functions on at least all of the heavy atoms involved in metal−ion interaction, were amply large so that basis set incompleteness was not a significant limitation on the reliability of the DFT calculations.

The experience of our group, reinforced by similar results of others with alkali ion complexes, suggests that the basis sets used here were adequate to reduce basis set superposition error (BSSE) effects for the alkali metal ion complexes to the order of 1 kcal or less. A number of the more important systems had BSSE corrections calculated explicitly using the counterpoise correction scheme described by Xantheas,[42] and these results are footnoted in the tables. It is well-known that at best such corrections provide only a very approximate amelioration of the BSSE problem, serving as much to indicate the magnitude of the possible error as to give a reliable correction, so it is fortunate that these corrections turn out to be small. The results shown confirm that this correction is of the order of 1 kcal mol^{-1} for the Na^+ complexes, and less than 0.5 kcal mol^{-1} for K^+ complexes. For those systems where a counterpoise correction was not explicitly evaluated, a generic correction was applied of 1.0 kcal for Na^+, and 0.5 kcal for K^+. (The BSSE correction always reduces the binding energy).

Zero-point energy (ZPE) effects are typically found to be around 1 kcal for metal-ion/aromatic-ligand binding (always acting to decrease the binding energy). A number of ZPE corrections were calculated here for the important complexes, as footnoted in the tables. Frequency calculations for the ZPE corrections were nearly all done using a reduced basis of 6-31+g(d) on the alkali metal and 3-21g(d) on all other atoms. One check of this level of frequency calculation against a frequency calculation using the full basis, carried out on Na^+/phenylalanine, gave the same ZPE correction within 0.1 kcal for both basis sets. For those complexes where vibrational frequencies were not calculated explicitly, it seemed appropriate to use a generic ZPE correction of 1.5 kcal mol^{-1} for the Na^+ complexes and 1.0 kcal mol^{-1} for the K^+ complexes.

It would be possible that ZPE corrections could affect the assignment of the lowest-energy neutral structures, but limited trials indicated that the zero-point energy of the neutral molecules varies little among the different conformers, and no comprehensive frequency calculations were carried out for the various neutral conformers other than the most stable ones. We can emphasize that most of the interpretations involving binding energies in the present work are based on comparisons of binding energies of similar complexes, for which the BSSE and ZPE errors should be largely similar and canceling.

A challenge in these calculations is the large number of possible conformations of both neutrals and ion complexes. For the neutral amino acids there is guidance from previous studies of non-aromatic amino acids, of which we will note the alanine study by Stepanian et al.[43] That study indicates the most stable conformation to be one (Stepanian's structure I) with both amine hydrogens interacting with the carbonyl oxygen. For the present study of the aromatic amino acids, the reasonable configurational possibilities for the neutral molecules were explored to see if the presence of the aromatic side chain changes this picture. First a broad survey of possible conformations was made using semiempirical (AM1) calculations to locate the minima on the potential surface lying within a few kilocalories of the most stable conformation. Then these candidate structures were reoptimized and their energies determined with full DFT calculations with the same basis set as was used for the corresponding complexes. It was found that the relative AM1 energies of the different conformers gave quite a faithful representation of the relative energies of the final DFT determinations, within ~1 kcal.

For the complexes, the various conformers were explored using DFT calculations with smaller basis sets. For *Phe* with its simple and symmetrical side chain, the number of possibilities is manageable, and the conformers described below probably encompass all of the low-energy structures. With *Tyr*, the ring hydroxyl was found to be essentially irrelevant, with its orientation having no effect (within one or two tenths of a kcal) on the energies. Furthermore, no stable conformations were found in which this hydroxyl was in an interacting location relative to the metal ion. Thus the conformer possibilities for *Tyr* were essentially the same as those for *Phe*. With *Trp*, however, the presence of the unsymmetrical and electrostatically complex side chain made the conformer search more daunting, and it is with only moderate confidence that we claim to have located all the low-energy conformers.

Results and Discussion

Neutral Molecules. The most favorable conformations found for neutral *Phe* are shown in Figure 1, and those for *Trp* in Figure 2. The corresponding energies are displayed in Tables1 and 2. The ring hydroxyl of *Tyr* was found to give negligible changes in the relative energetics as determined for *Phe*, so *Tyr* structures and energies are not displayed. It is evident from these figures that changing the orientation of the aromatic side chain of *Phe* has only a minor effect on the energies, so that for instance conformations *Phe*(a), *Phe*(b), and *Phe*(c) are almost the same. Changing the conformations of the amino and carboxyl groups has effects similar to those reported for alanine.[43] The existence of so many conformers of the neutrals within energies of the order of *kT* (room temperature) above the most stable conformer has obviously important implications for calculations of entropies and free energies of binding, but such calculations were not attempted here; for the present purposes, it is sufficient to be confident, as we are, that there are no unexplored neutral conformers having lower DFT energy than those described.

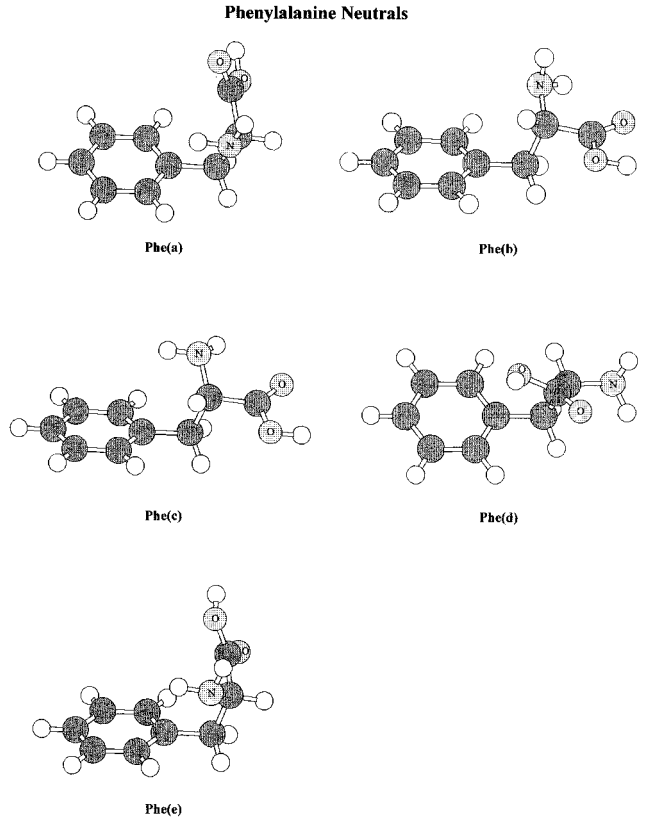

Figure 1. Low-energy conformers of neutral phenylalanine.

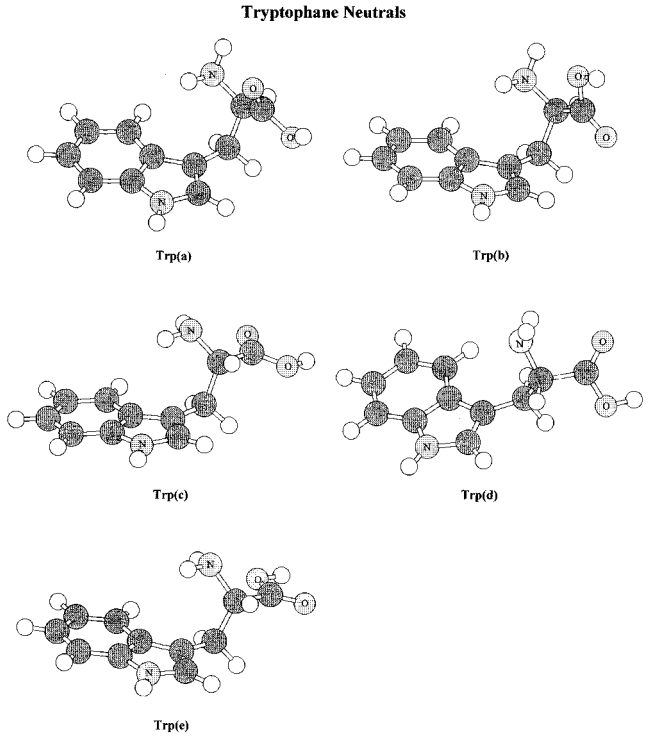

Figure 2. Low-energy conformers of neutral tryptophane.

Complexes. The most stable conformations located for each of the six complexes are displayed in Figure 3. (In looking at the figures, note that the vertical line drawn down from the metal ion toward the ring is perpendicular to the ring, and always terminates at the point where it crosses the plane of the ring.

TABLE 1: Neutral *Phe* Structures and Energy Increments Relative to the Most Stable Conformer (kcal mol^{-1})

structure	energy increment
Phe(a)	0
Phe(b)	+0.1
Phe(c)	+0.3
Phe(d)	+0.4
Phe(e)	+0.7

TABLE 2: Neutral Tryptophane Structures and Energy Increments Relative to the Most Stable Conformer (kcal mol^{-1})

structure	energy increment
Trp(a)	0
Trp(b)	+0.6
Trp(c)	+1.0
Trp(d)	+1.8
Trp(e)	+1.9

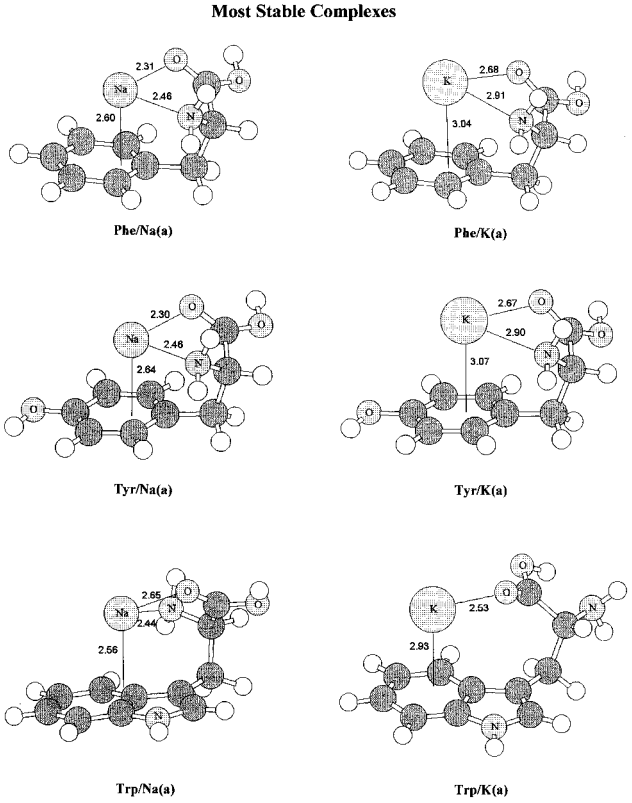

Figure 3. Most stable calculated conformers for the six metal-ion/amino acid combinations. (Distances in Å).

This allows one to picture the location of the metal ion above the π face.) The binding energies are given in Table 3. (We will use the term "binding energy" here to denote the zero-Kelvin ground-state-to-ground-state enthalpy of binding. Thermal corrections to room temperature, and entropy corrections to give free energies, are obviously of interest but are outside the scope of this study.) With only one exception, the most stable conformer is a tridentate N/O/Ring chelating structure placing the metal ion in a favorable position for cation-π interaction.

The exceptional case is K^+/*Trp*.The geometrical constraints do not allow either Na^+ or K^+ to form an N/O/Ring chelate involving the phenyl ring site of the indole side chain, which is the most favorable site for metal-ion binding. As known from model studies of the indole moiety,[44] Na^+ can reside above the pyrrole part of the indole π system, but at a cost of $\sim$3$-$4 kcal in energy relative to the phenyl site, and the situation for K^+ is presumably analogous. It is thus possible to form tridentate N/O/

TABLE 3: Calculated Energies and Binding Energies of Amino Acid Complexes (kcal mol^{-1}); Most Stable Known Conformations of Neutrals and Complexes (calculated ZPE and BSSE corrections have been applied as noted in the footnotes)

	phenylalanine		tyrosine		tryptophane	
	energy[a]	binding energy[b]	energy[a]	binding energy[b]	energy[a]	binding energy[b]
neutral	554.8061		630.0266		686.3763	
Na^+ complex	716.9687	48.0[c]	792.1896	48.3[d]	848.5432	52.0[e]
neutral	554.8061		630.0266		686.3763	
K^+ complex	1154.5891	34.7[f]	1229.8095	34.7[g]	1286.1621	37.6[h]

[a] B3LYP-DFT energy in Hartrees. (Na^+ energy is 162.0812, K^+ energy is 599.7240). [b] Binding energy in kcal mol^{-1}. [c] ZPE 1.8, BSSE 1.2. [d] ZPE 1.7, BSSE 1.2. [e] ZPE 1.8, BSSE 1.6. [f] ZPE 1.0 (est), BSSE 0.4. [g] ZPE 1.0 (est), BSSE 0.4 (est). [h] ZPE 0.7, BSSE 0.4.

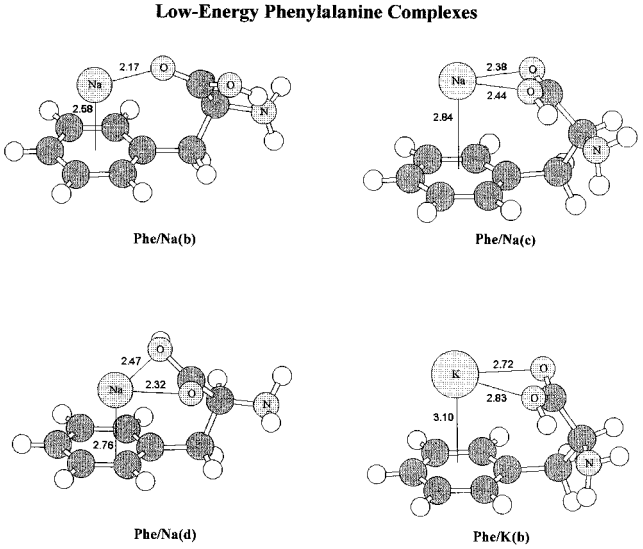

Figure 4. Various low-energy conformers of Na^+ and K^+ complexes with phenylalanine. (Distances in Å).

Ring chelates, but at substantial energetic cost. As shown in Figure 3, Na^+/*Trp* prefers to form an N/O/Ring chelate in this latter fashion. However, for K^+/*Trp*, it is slightly more favorable to adopt an alternative bidentate O/Ring chelating conformer which allows the metal ion to move over the phenyl ring of the side chain, as shown in the figure. The sacrifice of the K^+/N interaction energy involved in adopting this conformer is more than compensated by the larger cation$-\pi$ interaction available at the phenyl end of the indole group.

As expected, the binding energies to *Phe* and *Tyr* were found to be essentially equal. For both metal ions, binding to *Trp* is $\sim$3$-$4 kcal mol^{-1} stronger than to *Phe* or *Tyr*. Binding to Na^+ is $\sim$14 kcal mol^{-1} stronger than K^+, which is in accord with the larger radius and weaker electrostatic interactions for K^+ compared with Na^+.

Other Conformers and Isomers. *Phenylalanine.* The binding energies of a number of the lowest energy alternative complex structures found for *Phe*/Na^+ and *Phe*/K^+ are shown in Figure 4, and the corresponding energies are summarized in Table 4. *Phe*/Na(c) shows the slightly less favorable tridentate O/O/Ring chelation, as does *Phe*/K(b), while *Phe*/Na(d) illustrates an alternative, less favorable arrangement of the CO and NH_2 groups. Structure *Phe*/Na(b) is noteworthy, showing the possibility of bidentate O/Ring chelation. This conformation is quite favorable; its short Na^+/Ring distance indicates that it gains stability by a particularly strong cation$-\pi$ interaction,

TABLE 4: Structures of Various Isomeric *Phe* and *Tyr* Complexes and Energy Increments Relative to the Most Stable Conformer (kcal mol^{-1}) (ZPE and BSSE corrections were calculated or estimated as noted in the footnotes, while for cases not footnoted, the generic corrections described in the text were used)

	Phe		Tyr	
	structure	energy increment	structure	energy increment
Na^+				
	Phe/Na(a)	0[a]	*Tyr*/Na(a)	0
	Phe/Na(b)	+1.4[b]	*Tyr*/Na(b)	+1.8[d]
	Phe/Na(c)	+8.7[c]	*Tyr*/Na(c)	+8.9[d]
	Phe/Na(d)	+18.3		
K^+				
	Phe/K(a)	0		
	Phe/K(b)	+5.3		

[a] ZPE 1.8, BSSE 1.2. [b] ZPE 0.5, BSSE 1.0. [c] ZPE 1.4, BSSE 0.8. [d] ZPE and BSSE estimated to be the same as for phenylalanine.

which largely compensates for the loss of stability due to the absent Na^+/N interaction. For comparison, the optimal Na^+/Ring distance in the model complex Na^+/benzene is 2.40 Å (at the same level of computation), so that the *Phe*/Na(b) conformer in Figure 4, at a distance of 2.58 Å, is apparently quite close to maximizing the benefit of the cation$-\pi$ interaction. The Na^+/Ring distance in the most stable tridentate chelate *Phe*/Na(a) in Figure 3 is nearly as short, but the other conformers in Figure 4 have significantly longer distances.

The optimal K^+/Ring distance in the model complex K^+/benzene is calculated to be 2.92 Å. It can be seen that the K^+/Ring distances in both 3:*Phe*/K(a) and 4:*Phe*/K(b) are closer to the optimum than is true for the Na^+ complexes. The tridentate chelating cavity formed by phenlyalanine provides a better fit to K^+ than to Na^+. It allows the K^+ ion to achieve a position relative to the π face close to the optimum for cation$-\pi$ interaction with less distortion of its position relative to the other ligands than in the Na^+ case.

Tyrosine. Two considerations lead one to expect the binding energy to tyrosine to be very little changed from phenaylalanine. It is not geometrically feasible for the ring hydroxyl to participate in any reasonable chelating complex involving the other electronegative atoms of the amino acid group, so that it is unfavorable to move the metal ion close enough to this hydroxyl for significant interaction. Moreover, the bond dipole of the ring hydroxyl is oriented in a direction which minimizes its electrostatic effect in the vicinity of the metal ion binding over the ring.

In fact, as was found for a number of calculated structures, the binding energy differences between *Tyr* complexes and the corresponding *Phe* complexes are very small, certainly less than the uncertainty in the calculations. The structures are also very similar. Accordingly, structures of the *Tyr* complexes are not displayed other than the most stable forms shown in Figure 3. A few calculated Na^+/*Tyr* energies are shown in Table 3. The points made in discussing the *Phe* complexes can be taken as applying equally to the *Tyr* complexes.

Tryptophane. Figure 5 and Table 5 show various low-energy conformers found for *Trp* complexes. As noted above, low-energy conformations exist with the metal ion interacting either with the phenyl or the pyrrole regions of the indole π face. The indole nitrogen carries a substantial positive charge, and a common feature of all the low-energy structures is the tendency of the metal ion to move as far away from it as possible without losing the advantage of interaction with the π face.

The bidentate O/Ring chelated Na^+ complex exists (Figure 5:*Trp*/Na(b)) in which the Na^+ ion has virtually optimal cation-π interaction orientation relative to the phenyl ring region of the π face, just as in the K^+ complex of Figure 3:*Trp*/K(a). However, in contrast to the K^+ case, this conformation is slightly less favorable than the most stable N/O/Ring chelate.

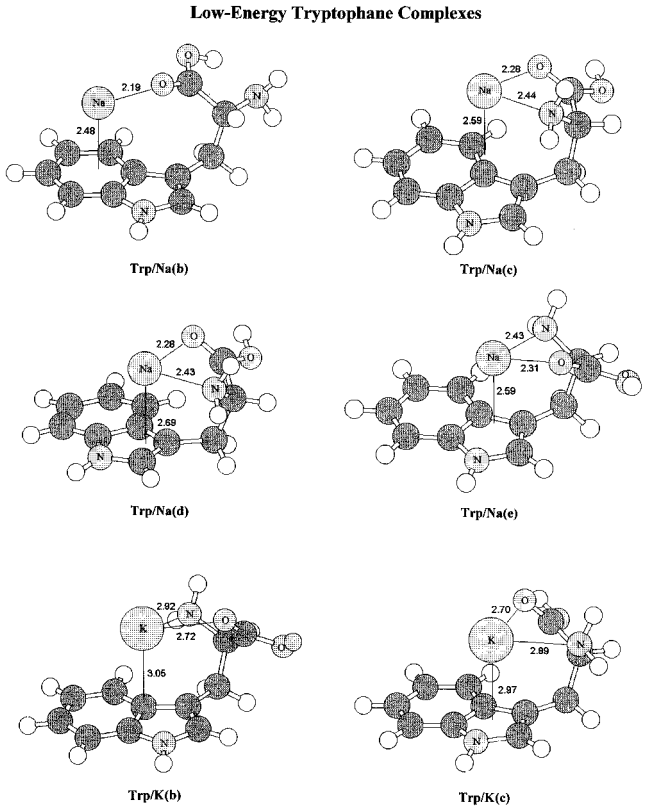

Figure 5. Various low-energy conformers of Na^+ and K^+ complexes with tryptophane. (Distances in Å).

TABLE 5: Structures of Various Isomeric *Trp* Complexes and Energy Increments Relative to the Most Stable Conformer (kcal mol^{-1}) (ZPE and BSSE corrections were calculated or estimated as noted in the footnotes, while for cases not footnoted, the generic corrections described in the text were used)

structure	energy increment
Na^+	
Trp/Na(a)	0[a]
Trp/Na(b)	+2.0[b]
Trp/Na(c)	+2.7
Trp/Na(d)	+1.8
Trp/Na(e)	+1.3
K^+	
Trp/K(a)	0[c]
Trp/K(b)	+0.8[d]
Trp/K(c)	+2.2

[a] ZPE 1.8, BSSE 1.6. [b] ZPE 1.8 (est), BSSE 0.8. [c] ZPE 0.7, BSSE 0.4. [d] ZPE 1.2, BSSE 0.4.

An interesting feature of *Trp* binding, illustrated by comparing Figures 3:*Trp*/Na(a) and 5:*Trp*/Na(e), is that the metal ion can move far off to the edge of the π face to optimize the electrostatic interaction between the metal ion and the dipole field of the indole ring. There thus exist distinct conformers, similar in other respects, in which the Na^+ either lies over the pyrrole ring (5:*Trp*/Na(e)), or lies outside the periphery of the ring framework (3:*Trp*/K(a)). Another such pair of conformers is 5:*Trp*/Na(d) and 5:*Trp*/Na(c).

As suggested by the figures, various arrangements of the amino acid's electronegative atoms are reasonably similar in

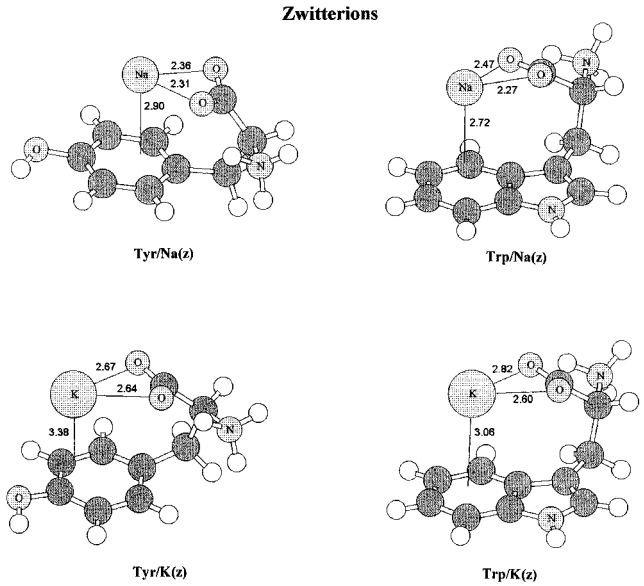

Figure 6. Most stable calculated zwitterion structures of tyrosine and tryptophane complexes. (Distances in Å).

TABLE 6: Energy Increments for Zwitterion Structures of *Tyr* and *Trp* Complexes Relative to the Most Stable Complex (kcal mol^{-1})[a]

	Na	K
Tyr	+4.1	+6.7
Trp	+4.6	+6.5

[a] The generic ZPE and BSSE corrections described in the text were applied.

energy, with the differences between the conformers presumably governed by small differences in electrostatic interactions.

Zwitterion Structures. There is evidence that the most stable cation-bound structures of some amino acids, notably arginine, are the zwitterion forms,[45] which benefit from a particularly favorable electrostatic interaction between the side chain and the ionized groups on the amino acid. Accordingly, some exploration was made of possible zwitterion structures for the present complexes. (Only structures with the proton on the amino nitrogen were considered, since other possibilities were expected to have even lower stability.) The most stable zwitterion conformers of *Tyr* and *Trp* complexes found are shown in Figure 6, with their binding energies shown in Table 6. While in all cases these zwitterions are not unreasonably high in energy, nevertheless they are less stable than the most stable conformers of the normal chelating complexes by significant amounts (4−7 kcal). Thus it seems unlikely that the zwitterions form an important fraction of the gas-phase thermal populations of any of these complexes.

Cation-π Contribution. There are two computational approaches one can pursue to assess the importance of the cation-π interaction in the binding of these complexes. One is to compare the metal-ion binding to an aromatic amino acid versus binding to an otherwise comparable amino acid lacking the aromatic side chain. This approach was followed in a forthcoming study[33] in which the non-aromatic amino acids used for comparison were glycine and alanine. The binding enhancement for the aromatic amino acids relative to alanine was substantial, amounting to 7−8 kcal mol^{-1} for *Phe* and Try, and 10−12 kcal mol^{-1} for *Trp*. This comparison is, however, not fully satisfactory, because the aliphatic amino acids are also expected to bind more weakly as a result of their lower polarizabilities and consequent lower polarization contributions to the binding. It

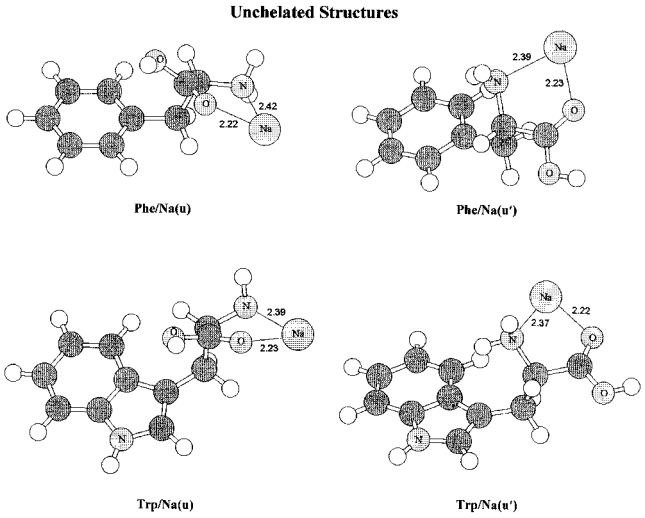

Figure 7. Conformers of Na^+ complexes with the aromatic side chain rotated out of chelation. (Distances in Å).

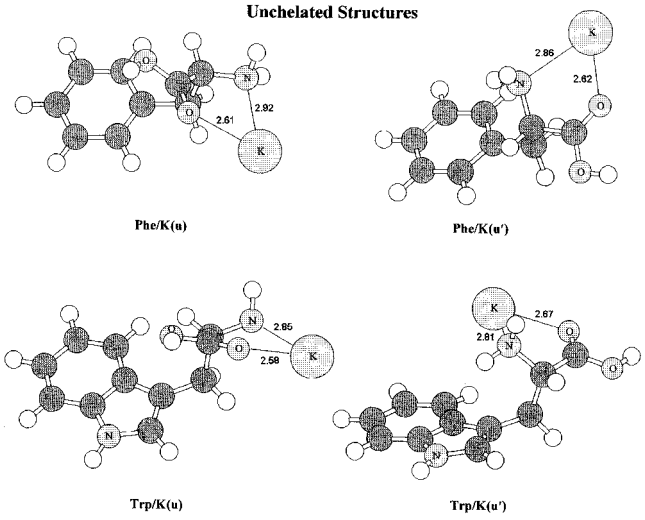

Figure 8. Conformers of K^+ complexes with the aromatic side chain rotated out of chelation. (Distances in Å).

is not clear how to separate the polarization and cation−π effects using this approach.

Another approach to assessing the cation−π contribution, which is somewhat more satisfactory in this regard, is to compare the energy of the π-chelated complex of an aromatic amino acid with the energy of the same complex in a conformation where the aromatic side chain is rotated out of chelation with the metal ion. Upon rotating the side chain around the CH_2 linkage, the present complexes were each found to have two such unchelated conformations, corresponding to rotations of 120° and 240° around the C−C bond, whose structures are shown in Figures 7 and 8 (denoted as the u and u′ conformers). (The *Tyr* complexes are similar to the *Phe* complexes, and are not displayed.) In all cases these nonchelating conformers were found to be local minima on the potential surface, and the structures were fully optimized at these minima with the same level of calculation as for the corresponding chelated conformers. Aside from the rotation of the side chain, it was not found that the bonding geometries changed radically relative to the chelating conformations.

The energies of these are summarized in Table 7. It would lead to a convenient interpretation if the two unchelated conformations had similar energies, but as the table shows, this is not quite the case. The differing electrostatic interactions of

TABLE 7: Binding Energy Comparison for Unchelated Conformers vs the Cation−π Chelated Conformer (kcal mol^{-1}) (ZPE and BSSE corrections were calculated or estimated as noted in the footnotes, while for cases not footnoted, the generic corrections described in the text were used)

	π-chelated [a]	u	u′	cation−π energy
Na^+				
Phe	48.0	41.6	44.2[b]	5.1
Tyr	48.3	41.8	44.6	5.1
Trp	52.0	45.3	47.9	5.4
K^+				
Phe	34.7	28.6	30.7	5.0
Tyr	34.7	28.9	31.4	4.6
Trp	36.8[c]	29.2	32.3	6.0[c]

[a] From Table 3 and Table 5. [b] ZPE 1.7, BSSE 0.9. [c] The comparison for K^+/*Trp* is made assuming the N/O/Ring chelated structure, Figure 5:*Trp*/K(b), as the chelated conformer.

the side chain with the electronegative groups of the amino acid are clearly important, and lead to a substantial energy difference between the (u) and (u′) unchelated conformations in each case. There is thus some ambiguity in trying to decide how much energy is gained by rotating the side chain into the conformation where the cation−π chelating interaction of the metal ion with the ring is possible. It was decided to use the average energy of the two unchelated conformations in assigning a value to this cation-π energy. The resulting energy increments are given in the last column of Table 7. The cation-π contributions gauged in this way are consistent, amounting to ∼5 kcal mol^{-1} for both Na^+ and K^+ complexes.

The interaction energies gauged this way are smaller than those gauged by the alanine comparisons in ref 33. This is reasonable, since the alanine comparison does not take into account stabilizing polarization and electrostatic interactions of the aromatic side chains that are not entirely removed when the side chain is rotated out of chelation.

Confirmation by MP2 Calculations. Ab initio calculations of these complexes at the MP2 level with the same polarization/diffuse basis sets used in the present DFT calculations are very demanding of computer time. However, it seemed worthwhile to make one such comparable calculation, with three purposes. First, the substantial cation−π interactions found here are noteworthy, and it is useful to confirm by a wholly independent approach that they are not simply an artifact of the DFT method. Second, various other work on cation−π complexes has used MP2 calculations, making it useful as a point of reference to do one of our systems by MP2. And third, we can point out the much larger BSSE correction calculated by MP2 compared with DFT using the same basis, taking this as a further justification of our preference for the DFT approach.

The smallest complex, Na^+/phenylalanine (structure Phe/Na-(a), Figure 3), was chosen for this study; at the same time a similar set of calculations was carried out on Na^+/alananine. These results are shown in Table 8. It is seen that the MP2 result for Na^+/alananine gives a binding energy (after BSSE correction) 1.3 kcal mol^{-1} lower than the DFT result, which seems like acceptable agreement. The BSSE correction using the MP2 approach is strikingly large, however, at 4.6 kcal, which is almost 4 times larger than the correction in the DFT approach. Given the uncertain quantitative validity of such counterpoise corrections, this suggests that there is a substantial degree of uncertainty in the MP2 result associated with basis-set incompleteness problems, and that this uncertainty is much less severe using the DFT approach.

Considering the comparison of alanine with phenylalanine, Table 8 shows that MP2 actually gives a somewhat larger binding energy increment (9 kcal) than does DFT (7 kcal). This difference in the two methods is probably not significant. However, the large increment obtained with MP2 does give a clear confirmation that the substantial effect calculated for introduction of the aromatic substituent is not just an artifact of the DFT method, but does actually reflect a real cation-π interaction effect.

TABLE 8: MP2 Calculations (kcal mol^{-1})

	Na^+/alanine[a]		Na^+/phenylalanine[b]	
	DFT	MP2	DFT	MP2
uncorrected binding energy	41.6	40.2	49.2	51.3
BSSE	0.9	2.6	1.2	4.6
corrected binding energy	40.7	37.6	48.0	46.7

[a] Basis 6-31g(d) on H, 6-31+g(d) on all heavy atoms. The calculated ZPE correction of 1.9 kcal mol^{-1} was subtracted from all binding energies. [b] Basis 6-31g(d) on H, 6-31+g(d) on all heavy atoms. Structures optimized with 6-31+g(d) on Na^+, 6-31g(d) on all other atoms. The calculated ZPE correction of 1.5 kcal mol^{-1} was subtracted from all binding energies.

Conclusions

The binding site formed by tridentate N/O/Ring chelation provides strong binding for both alkali ions to all three amino acids, and gives the most stable complex conformation in all except the K^+/*Trp* case. In some cases, the geometry of an alternative bidentate O/Ring chelation pattern allows the metal ion to move into a more nearly optimal cation−π interaction with the phenyl ring; such complexes are also very favorable, and in the K^+/*Trp* case this is the best binding conformation. For *Phe* and *Tyr*, the geometry of the binding cavity in the N/O/Ring chelates offers a somewhat better fit to the larger K^+ ion than to Na^+, as indicated by comparisons of the metal/ring distances. For *Trp*, neither metal ion can achieve optimal chelation to the phenyl region of the indole π surface without sacrificing some of the interaction with the N or O ligands. The resulting conformational compromises, involving either disruption of the N/O chelation, or cation−π interaction with the less favorable pyrrole region of the π face, lead to the existence of a variety of binding geometries with similar energies.

With respect to binding energies, geometries, and variety of available complex structures, no significant differences were found between *Phe* and *Tyr*. This supports the discussion of Mecozzi et al.,[5] who found no distinction between *Phe* and *Tyr* from an electrostatic viewpoint. They suggested that the preference for *Tyr* as a cation−π ligand in biological contexts must be due to effects other than the intrinsic binding, for instance the ability of the hydroxyl group to constrain a favorable orientation of the ring, or the additional interactions provided by groups hydrogen bonded to the hydroxyl.

Even though *Trp* is unable to assume conformations which simultaneously optimize the chelation with all three ligands, nevertheless it binds more strongly to either metal ion than *Phe* or *Tyr*, by about 3−4 kcal mol^{-1}. For any given amino acid, Na^+ binds more strongly than K^+ by about 14 kcal mol^{-1}.

The comparison of π-chelating vs nonchelating conformers of the same complex seems to offer a good quantitative insight into the stability gained by turning on the cation−π interaction. This analysis gave results of gratifying consistency, showing a gain of ∼5 kcal mol^{-1} for both the π−Na^+ interaction and the π−K^+ interaction.

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