Potential Energy Surfaces for the Gas-Phase Interaction between α-Alanine and Alkali Metal Ions (Li^+, Na^+, K^+). A Density Functional Study

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The conformations of α-alanine and those of the eight most stable adducts that it forms with lithium, sodium, and potassium cations were studied by means of the density functional theory using the hybrid B3LYP exchange correlation potential and the 6−311++G** basis set. Minima and transition states characterizing the energetic paths for the interaction of the three metal ions with the free amino acid were explored in detail, and the results show that they are almost the same in the cases of lithium and sodium ions. At the absolute minimum, the Li^+ and Na^+ cations appear to be contemporaneously linked to the carbonyl oxygen and nitrogen atoms of α-alanine. Slight differences were found for potassium; the most stable adduct has the cation coordinated to both oxygen atoms of carboxyl function. The influence of the low-energy conformers in the determination of the gas-phase absolute affinities is demonstrated.

Introduction

Sodium and potassium, and to a lesser extent lithium, cations have important effects from the biological point of view.[1,2] They participate in several functions of living systems such as enzyme regulation, stabilization of structural elements, transport of glucides and amino acids to proteins through transmembrane channels,[1,3,4] and so on. The understanding of the details of the local interactions between metal ions and amino acids is therefore a matter of much interest.

For these reasons, the study of metal binding affinities to biologically relevant molecules has attracted considerable attention at both experimental[1,5−10] and theoretical[3,6,7,10−16] levels.

The most common techniques for studying alkali-cationized adducts in the gas-phase are high-pressure mass spectrometry,[17,18] ion cyclotron mass spectrometry,[19,20] guided ion beam mass spectrometry,[21] and the kinetic method,[5,22] but often, only information regarding relative values of metal affinities can be obtained from these experiments. Theoretical investigations overcome the problems connected with the experimental techniques allowing the determination not only of the minima but also a means of following their formation mechanism through transition state characterization. In addition, the theoretical study of the mechanisms of the gas-phase reactions between metal ions and organic molecules provides useful insights into the properties of both absence of solvation and counterion effects.

The presence of much experimental work on the small amino acid−metal ion interactions and the availability of a good amount of comparison data have stimulated the computations for these systems, but until now, few theoretical studies have focused on α-alanine[11,14,16] or α-alanine containing peptides.[6] In view of our previous reliable results on the metalation of glycine[11,23] obtained by the B3LYP method, we considered it of interest to extend the study to the interaction of the alkali metal ions (Li^+, Na^+, K^+) with alanine.

The conformational space of free α-alanine, however, has been investigated at various levels of theory.[24−27]

Starting from a selection of several low-lying conformations for this amino acid, our investigation concerned its interaction

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(1) Hoyau, S.; Norrman, K.; McMahon, T. B.; Ohanessian, G. *J. Am. Chem. Soc.* **1999**, *121*, 8864.
(2) Lippard, S. J.; Berg, J. M. *Principles of Bioinorganic Chemistry*: University Science Books: Mill Valley, CA, 1994.
(3) Hoyau, S.; Ohanessian, G. *Chem. Eur. J.* **1998**, *4*, 1561.
(4) Stryer, L. *Biochemistry* W. H. Freeman: New York, 1988.
(5) Bojesen, C.; Breindahl, G. T.; Andersen, U. N. *Org. Mass Spectrom.* **1993**, *28*, 1448.
(6) Tomlinson, M. J.; Scott, J. R.; Wilkins, C. L.; Wright, J. B.; White, W. E. *J. Mass Spectrom.* **1999**, *34*, 958.
(7) Bouchonnet, S.; Flament, J. P.; Hoppilliard, Y. *Rapid Commun. Mass Spectrom.* **1993**, *7*, 470.
(8) Feng, W. Y.; Gronert, S.; Lebrilla, C. B. *J. Am. Chem. Soc.* **1999**, *121*, 1365.
(9) Cerda, B. A.; Wesdemiotis, C. *J. Am. Chem. Soc.* **1995**, *117*, 9734.
(10) Cerda, B. A.; Hoyau, S.; Ohanessian, G.; Wesdemiotis, C. *J. Am. Chem. Soc.* **1998**, *120*, 2437.
(11) Marino, T.; Russo, N.; Toscano, M. *J. Inorg. Biochem.* **2000**, *79*, 179.
(12) Bouchonnet, S.; Hoppilliard, Y. *Org. Mass Spectrom.* **1992**, *27*, 71.
(13) Bertran, J.; Rodriguez-Santiago, L.; Sodupe, M. *J. Phys. Chem. B* **1999**, *103*, 2310.
(14) Hoyau, S.; Ohanessian, G. *J. Am. Chem. Soc.* **1997**, *119*, 2016.
(15) Jensen, F. *J. Am. Chem. Soc.* **1992**, *114*, 9533.
(16) Wyttenbach, T.; Witt, M.; Bowers, M. T. *J. Am. Chem. Soc.* **2000**, *122*, 3458.
(17) Kebarle, P.; Hogg, A. M. *J. Chem. Phys.* **1965**, *42*, 668.
(18) Hogg, A. M.; Kebarle, P. *J. Chem. Phys.* **1965**, *43*, 449.
(19) Staley, R. H.; Beauchamp, J. L. *J. Am. Chem. Soc.* **1975**, *97*, 5920.
(20) Woodin, R. L.; Beauchamp, J. L. *J. Am. Chem. Soc.* **1978**, *100*, 501.
(21) Rodgers, M. T.; Armentrout, P. B. *J. Am. Chem. Soc.* **2000**, *122*, 8548.
(22) Cerda, B. A.; Wesdemiotis, C. *J. Am. Chem. Soc.* **1996**, *118*, 11884.
(23) Marino, T.; Mineva, T.; Russo, N.; Sicilia, E.; Toscano, M. In *Metal Ligand Interaction in Chemistry, Physics and Biology*; Russo, N., and Salahub, D. R., Eds.; Kluwer Academic Publishers: Dordrecht, 2000.
(24) Godfrey, P. D.; Firth, S.; Hatherley, L. D.; Brown, R. D.; Pierlot, A. P. *J. Am. Chem. Soc.* **1993**, *115*, 9687.
(25) Csaszar, A. G. *J. Phys. Chem.* **1996**, *100*, 3541.
(26) Stepanian, S. G.; Reva, I. D.; Radchenko, E. D.; Adamowicz, L. *J. Phys. Chem. A* **1998**, *102*, 4623.
(27) Kaschner, R.; Hohl, D. *J. Phys. Chem. A* **1998**, *102*, 5111.

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mechanism with lithium, sodium, and potassium cations. Another goal of the present study was the evaluation of the absolute value of metal ion affinities of α-alanine that have never been determined previously at a theoretical level.

Computational Methods

Density functional theory (DFT) was applied to the study of the interaction of α-alanine with metal cations (Li^+, Na^+, and K^+). The hybrid Hartree−Fock DFT method known as B3LYP which includes Becke's three[28] parameter exchange functional with Lee, Yang, and Parr[29] type nonlocal correlation functionals and the all-electron 6−311++G** internal basis set as implemented in Gaussian 98[30] were used for the computations. This choice was made due to the fact that the B3LYP/6−311++G** scheme has previously provided accurate results in the description of metal−ligand interactions.[3,11,13,23] All geometries were optimized without any symmetry constraints, and the zero point vibrational energies were computed in order to characterize all the stationary points on the three potential energy surfaces as local minima or transition states.

Metal ion affinity (MIA) was computed as the negative of the enthalpy variation (ΔH) for the reactions

$$\text{alanine} + M^+ \rightarrow \text{alanineM}^+$$

Entropic contributions to the free energy (ΔG) were obtained by thermochemical analysis at 298 K using computed vibrational frequencies.

The basis set superposition error (BSSE) was calculated following the counterpoise method[31,32] for only one of the possible complexes for each metal cation and then used to correct the MIA values of 2.0, 1.4, and 0.9 kcal/mol for Li^+, Na^+, and K^+, respectively.

Results and Discussion

Although there is no direct relationship between the lowest energy conformations of free α-alanine and those of the alanine−M^+ adducts, the former were used, in our analysis, as a set of starting structures for geometrical optimization of the complexes.

From previous theoretical investigations[24−27] a total of 13 minimum conformations have been proposed for α-alanine, but in our study only seven low-lying structures, depicted in Figure 1, were considered. The remaining conformations were neglected essentially because they were characterized by a disposition of the $-NH_2$ group not suitable for the approach of a metal cation.

All the examined structures of α-alanine were found to be in an energy range of about 7 kcal/mol (see absolute energy values in Figure 1).

The electrostatic nature of the interaction between M^+ and the neutral molecule was the discriminating element in the choice of the possible attachment sites. The selection criterion of the starting structures of complexes was based on the maximization of the two most relevant aspects from the energetic point of view. The number of complexation modes, in principle quite high, was reduced considering the greater attractive effects of the most negatively charged centers and the importance of preserving the intramolecular hydrogen bonds because of their stabilizing role. Thus, the most favorable coordination sites for the cations involve the pair of carbonyl oxygens or the amine nitrogen atom and one of the oxygens.

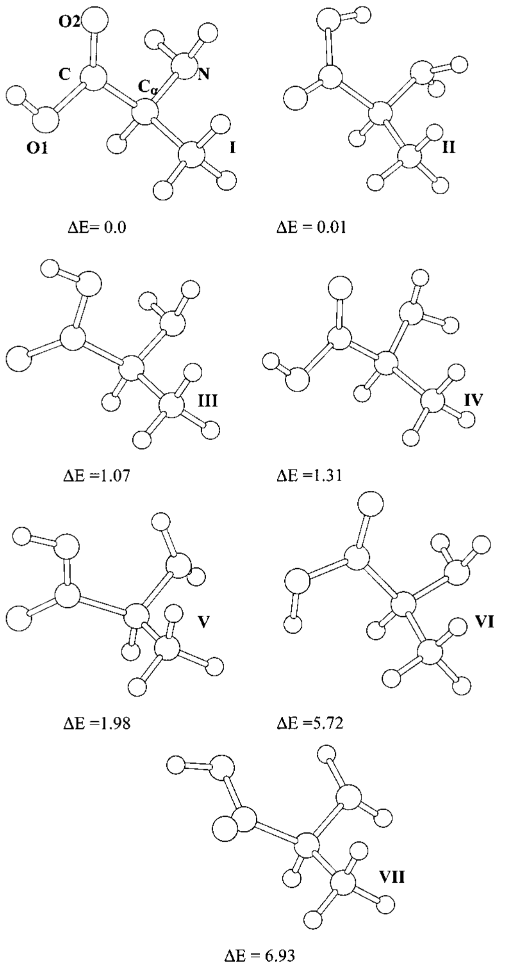

Figure 1. B3LYP/6−311++G** minimum conformations of α-alanine. Relative energies (ΔE) are in kcal/mol. The absolute energy for the most stable conformer is −323.856078 au.

Monocoordination to α-alanine, unlikely for the lithium cation, becomes slightly more favorable in the case of sodium and potassium ions. Figure 2 collects all the considered α-alanine−M^+ isomers. In the diagram, the complexes formed by the α-alanine zwitterion with the metal cations (structures **2** of Figure 2) are also included. The corresponding free zwitterion was not reported in Figure 1 because it is not a minimum on the potential energy surface (PES) of α-alanine. Although it is well-known that this conformer does not exist in gas-phase, our computations indicated its metal complexes as the most stable ones after those obtained from conformer **I** of α-alanine with lithium and sodium and from conformers **I** and **II** with potassium.

The geometrical parameters in Figure 2 show that the cation-binding site distances increase according to the dimension of the ion. In the case in which the metal bicoordination involves two different elements, the bond length with the oxygen is always shorter than that with nitrogen. This is essentially due

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(28) Becke, A. D. *J. Chem. Phys.* **1993**, *98*, 5648.

(29) Lee, C.; Yang, W.; Parr, R. G. *Phys. Rev. B* **1988**, 37, 785.

(30) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Zakrzewski, V. G.; Montgomery, J. A.; Stratmann, R. E.; Burant, J. C.; Dapprich, S.; Millan, J. M.; Daniels, A. D.; Kudin, K. N.; Strain, M. C.; Farkas, O.; Tomasi, J.; Barone, V.; Cossi, M.; Cammi, R.; Mennucci, B.; Pomelli, C.; Adamo, C.; Clifford, S.; Ochterski, J.; Petersson, G. A.; Ayala, P. Y.; Cui, Q.; Morokuma, K.; Malick, D. K.; Rabuck, A. D.; Raghavachari, K.; Foresman, J. B.; Cioslowski, J.; Ortiz, J. V.; Baboul, A. G.; Stefanov, B. B.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Gomperts, R.; Martin, R. L.; Fox, D. J.; Keith, T.; Al-Laham, M. A.; Peng, C. Y.; Nanayakkara, A.; Gonzales, C.; Challacombe, M.; Gill, P. M. W.; Johnson, B.; Chen, W.; Wong, M. W.; Andreas, J. L.; Head-Gordon, M.; Replogle, E. S.; Pople, J. A. *Gaussian 98*; Gaussian Inc.: Pittsburgh, PA, 1998.

(31) Frisch, M. J.; Del Bene, J. E.; Binkley, J. S.; Schaefer, H. F., III. *J. Chem. Phys.* **1986**, *84*, 2279.

(32) Schwenke, D. W.; Truhlar, D. G. *J. Chem. Phys.* **1985**, *82*, 2418.

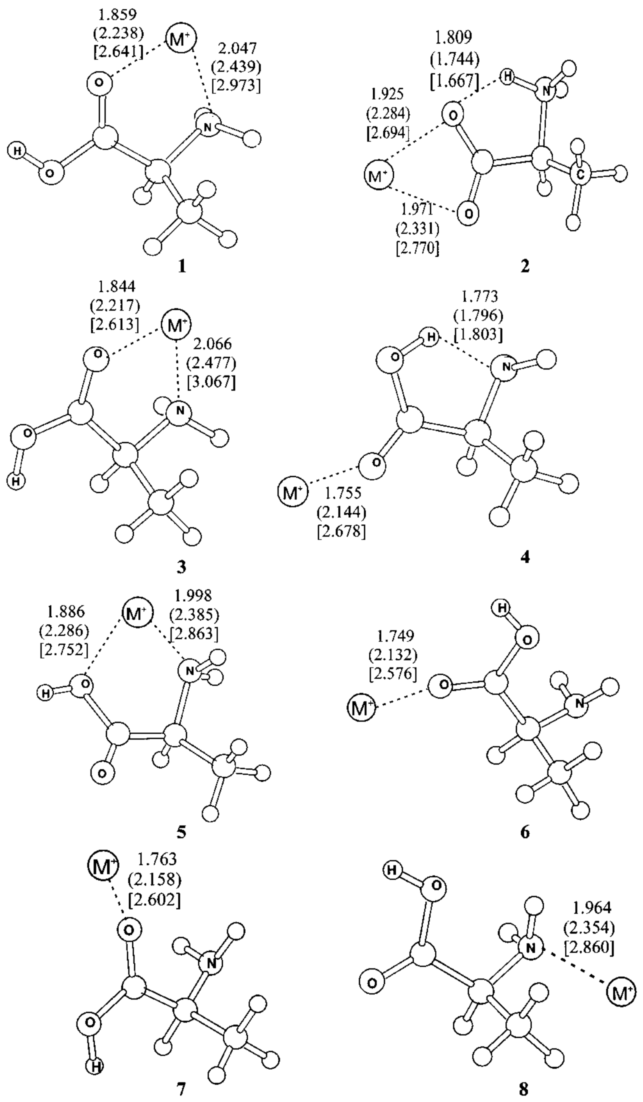

Figure 2. Structures and main geometrical parameters of the complexes obtained by metalation of α-alanine with M^+ ($M^+ = Li^+, Na^+, K^+$). Distances (Å) for Li, Na, and K ions are reported without enclosures, within parentheses, and within square brackets, respectively.

to the fact that a hard species such as M^+ prefers to interact with a harder site.

The optimization procedure for the isomer **4**, starting from a situation of bicoordination for the metal ions, yields monocoordinated systems especially in the cases of lithium and sodium cations. For potassium, the distance of 3.087 Å with the O1 atom (for the atom numbering see Figure 1), although longer than that with O2 (2.678 Å), and the M^+O2C and M^+O1C valence angle values underline the tendency of this element to bind favorably with the carboxyl moiety as previously found in other theoretical work.[6]

In Table 1 the total (E) and relative energies (ΔE) (including the zero point energy corrections) are reported for all the complexes arising from the metalation of the different α-alanine isomers. In Table 2 dipole moments and net charges on the metal, oxygen, and nitrogen atoms are given for the most stable complexes.

Table 1. B3LYP/6−311++G** Absolute (E in au) and Relative Energies (ΔE in kcal/mol) at 0 K, for the Complexes Alanine−M^+

M^+	structure	E	ΔE
Li^+	**1**	−331.128705	0.0
	2	−331.122470	3.9
	3	−331.118662	6.3
	4	−331.116216	7.8
	5	−331.114077	9.2
	6	−331.098825	18.7
	7	−331.097440	19.6
	8	−331.090485	23.9
Na^+	**1**	−485.901918	0.0
	2	−485.899930	1.2
	4	−485.897034	3.0
	3	−485.892101	6.2
	5	−485.888231	8.6
	6	−485.880679	13.3
	7	−485.879564	14.0
	8	−485.873841	17.6
K^+	**4**	−923.555890	−0.7
	1	−923.554790	0.0
	2	−923.553902	0.6
	3	−923.545554	5.8
	5	−923.541905	8.1
	6	−923.541058	8.6
	7	−923.539271	9.7
	8	−923.532450	14.0

Table 2. B3LYP/6−311++G** Dipole Moments (μ in Debye) and Net Charges on the Metal, Two Oxygens, and Nitrogen Atoms (q in $|e|$) for the Most Stable Alanine−M^+Complex

M^+	q_{M^+}	q_{O2}	q_{O1}	q_N	μ
Li^+	0.617	−0.269	−0.112	−0.384	5.25
Na^+	0.850	−0.386	−0.115	−0.515	5.81
K^+	0.977	−0.403	−0.125	−0.523	6.25

Lithium and sodium systems show similar behavior. In both cases, structure **1** (see Figure 2) appears to be the most stable resulting from the interaction between the cations and the absolute minimum of α-alanine (structure **I** of Figure 1). This structure is stabilized by the presence of the five-membered ring and by the attractive electrostatic interactions between the cations and the negative charges on the carbonyl oxygen and the amine nitrogen (see Table 2). All lithium and sodium bicoordinated systems (**1**−**5**) fall within a range of about 9 kcal/mol. The ΔE trend is the same except for structure **4**, which is stabilized with respect to **3** upon the interaction with sodium cation. Although the relative energies of the monocoordinated systems (**6**−**8**) follow the same order, in the case of sodium complexes they appear markedly reduced (by about 5 kcal/mol). For potassium, the absolute minimum is structure **4** (see Figure 2), which is really a monocoordinated complex as is evident from the M^+O2 and M^+O1 distances (see Figure 2) and from the comparison of these values with those obtained in complexes **6**−**8** in which K^+ is certainly monocoordinated. The bicoordinated structure **1** is almost degenerate, lying at only 0.69 kcal/mol above **4**. The first five compounds are included within a range of energy values, which, as in the previous cases, is about 9 kcal/mol. A further stabilization (of about 5 kcal/mol), with respect to sodium complexes, occurs for the monocoordinated structures **6**−**8**. In the absolute minimum the positive charge is entirely retained on potassium (see Table 2).

The results for lithium and sodium are in agreement with the B3LYP computations of Wyttenboch, Witt, and Bowers[16] performed using the DZVP basis set and selecting only four α-alanine isomers.

The potential energy surfaces for the Li^+−, Na^+−, and K^+−α-alanine interactions are reported in Figure 3. Table 3 collects together the relative energies of the transition states (TS) shown in Figure 4. The isomerization sequence reported in the PESs involves only one-step processes; thus, the various minima can be obtained only from a particular starting complex. The general shape of the three paths is very similar except for the height of the energetic barriers between the different minima that

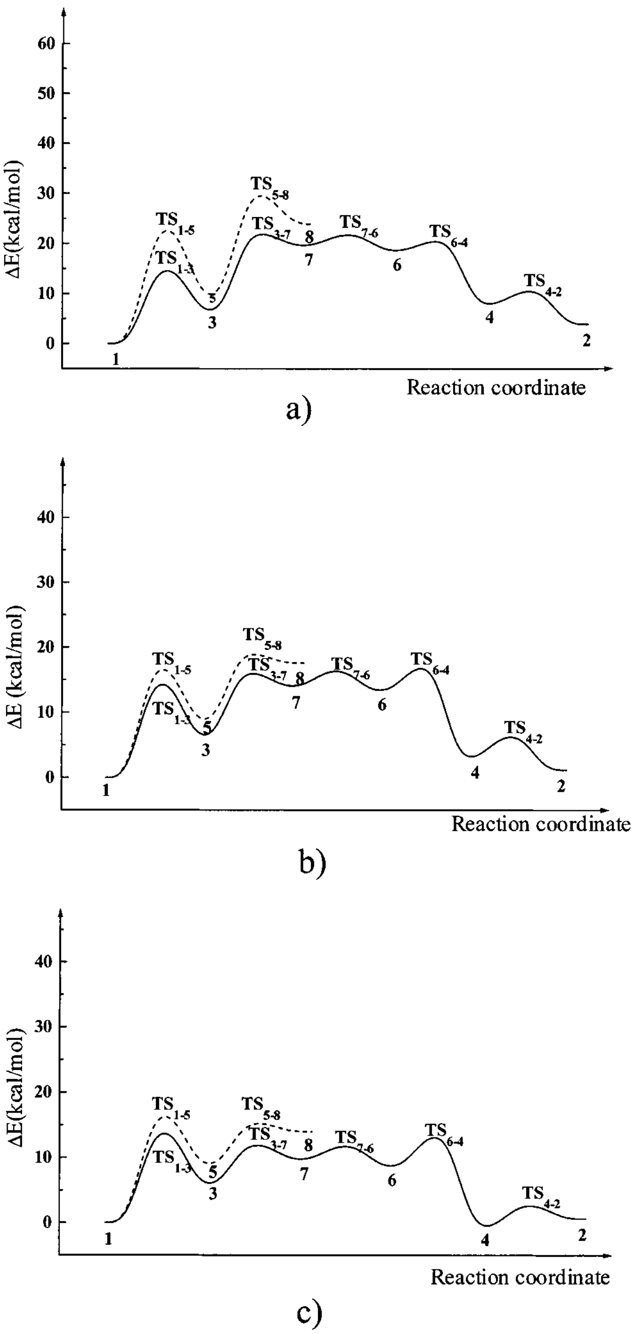

Figure 3. Energetic paths for the metalation process of α-alanine with (a) Li^+, (b) Na^+, and (c) K^+ cations.

Table 3. Relative Energies (in kcal/mol) for the Transition States on the Alanine−M^+ Potential Energy Surface at the B3LYP/6−311++G** Level

species	Ala−Li^+	Ala−Na^+	Ala−K^+
TS_{1-3}	14.9	14.7	14.5
TS_{3-7}	22.1	16.1	12.0
TS_{7-6}	21.8	16.4	11.8
TS_{6-4}	20.6	17.0	13.4
TS_{4-2}	10.6	6.4	2.7
TS_{1-5}	23.3	17.0	16.7
TS_{5-8}	29.9	19.0	15.3

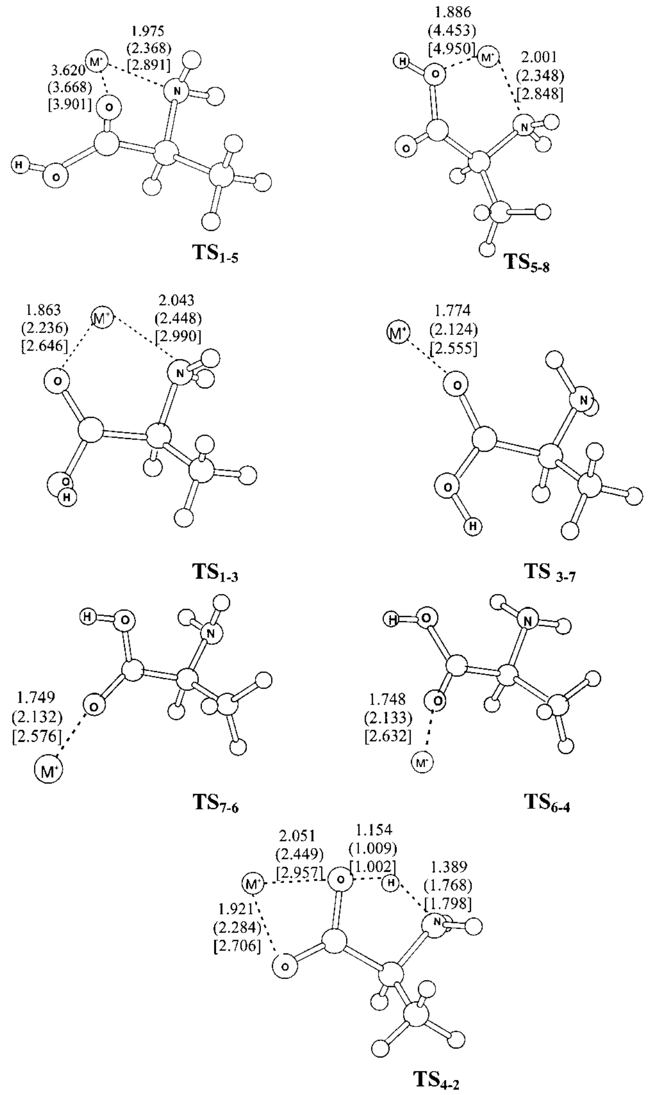

Figure 4. Structures and main geometrical parameters of the transition states (TS) connecting the minima on the energetic path for the α-alanine−M^+ interaction. Distances (Å) for Li, Na, and K ions are reported without enclosures, within parentheses, and within square brackets, respectively.

decreases in going from lithium to potassium complexes (see Table 3). The three PESs are characterized by two separate interconversion channels connecting two different sets of minima. The first channel (dashed lines in Figure 3) starts from minimum **1**, which via TS_{1-5} should arrive at minimum **5**. The isomerization processes include the cleavage of the O2−M^+ bond in **1** to form a monodentate complex and the rotation about the C−Cα bond with the formation of a new bond between O1 and M^+ as in structure **5**. Transition states TS_{1-5} are characterized by the following relative energy values: 23.3 (for Li^+), 17.0 (for Na^+), and 16.7 kcal/mol (for K^+). The alternative path, in which the in-plane H-shift could be hypothesized, is energetically unfavorable (in fact the transition states TS_{1-5} lie, in this case, at 51.2, 41.3, and 40.5 kcal/mol for Li^+, Na^+, and K^+, respectively). The barrier for the next transformation **5**−**8**, consisting of a simple migration of M^+ toward the amine nitrogen, is slightly higher with respect to the previous one, for lithium and sodium, but becomes lower for potassium cation. It is worth noting that the Li^+−O is essentially the same as in compound **5**, while the Na^+−O and K^+−O bond lengths become more similar to the corresponding ones in species **8**. The formation of complex **8** starting from minimum **1** via species **5** is, in all three cases, an endothermic process. In the second channel (solid lines in Figure 3), step **1**−**3** is a cis−trans conversion that involves the hydrogen atom of the carboxyl group. In TS_{1-3} the hydrogen is placed at 90° with respect to the plane defined by O1−C−O2. This isomerization occurs without particular problems as indicated by the values of the energetic barriers that fall in the range 14.2−14.9 kcal/mol for the three alkali metals. In complex **3,** the shift of the bicoordinated metal toward the O2 that yields the monocoordinated species **7** implies TS_{3-7} formation in which the cation appears to be out of the plane containing the O2, C, Cα, and N atoms. This transition state presents a slightly different geometry depending on the particular metal ion. The conversion **3−7** requires 15.8, 9.9, and 6.2 kcal/mol for Li^+, Na^+, and K^+, respectively. The height of the barriers for the **7−6** isomerization is practically the same for the three metal species (about 2 kcal/mol). The transition state is obtained as a result of a torsion around the C−Cα axis and a rearrangement of the hydroxyl hydrogen that occur simultaneously. TS_{7-6} are productlike species, so the very low energetic cost can be easily justified. The **7−6** isomerization is the first exothermic process along the three paths. From complex **6** it is possible to arrive at minimum **4** through TS_{6-4}. The **6−4** energetic barriers are of 1.9, 4.0, and 4.8 kcal/mol for Li^+, Na^+, and K^+, respectively. Notwithstanding the fact that these values are very similar, they nevertheless reflect that the transition from an unfavored structure (**6**) to another that becomes gradually more stable (**4**) in going from lithium to potassium is ever more expensive from the energetic point of view. The proton transfer from the O1 to the N atom occurs through TS_{4-2} giving rise to zwitterion **2** starting from complex **4**. The energetic cost for the 1,4 hydrogen shift is modest (2.8 kcal/mol for Li^+, and 3.4 kcal/mol for both Na^+ and K^+).

In summary, the interconversion between the lowest energetic minima **1**, **2**, and **4** requires many steps and the processes are kinetically controlled by the **3−7**, **6−4**, and **1−3** barriers for Li^+, Na^+, and K^+, respectively.

The experimental values of lithium and sodium affinities for α-alanine come from the kinetic method.[5] In this method metal affinities are obtained by generating a cationized dimer with a reference molecule of known affinity for the cation. The binding characteristics of the two ligands in the dimer should be sufficiently similar in order to neglect relative dissociation entropy. These assumptions can be a source of errors, and for this reason many authors[21,22,33] believe it necessary to revise the experimental data. The reference molecule used by Bojesen et al.[5] is *N*,*N*-dimethylformamide (DMF) for which an indirect measurement of MIA has been taken into account. Recently, Ohanessian and Hoyau,[3] on the basis of their MP2 computations on Li^+− and Na^+−glycine, have corrected the experimental values of Bojesen et al.[5] obtaining MIA values of 55.5 and 42.4 kcal/mol for Li^+− and Na^+−α-alanine complexes, respectively. Our MIA and free energy values corrected by BSSE are reported in Table 4. At 298 K, B3LYP/6−311++G** values for species **1** match better the original experimental result than the revised one. For the K^+−α-alanine system, for which no experimental MIA value exists, we predict a binding energy of 28.5 kcal/mol for the most stable complex **4**. Because it was demonstrated, theoretically and experimentally,[26] that free α-alanine exists in two practically degenerate forms (**I** and **II** of Figure 1), we have also considered, in the MIA determination, the complexes **4** arising from the free conformer **II**. The MIAs obtained for lithium and sodium are smaller than those corresponding to systems **1** and close to the experimental values.[3,5] For potassium complexes, the two binding energy values are very similar, reflecting the very low energy separation between **1** and **4**.

Table 4. Metal Ion Affinities (Δ*H*) and Free Energies (Δ*G*) at 298 K for Alanine−M$^+$ Most Stable Complexes[a]

	Ala−Li$^+$		Ala−Na$^+$		Ala−K$^+$	
	1	**4**	**1**	**4**	**1**	**4**
ΔH^{298}	58.8	51.0	40.5	37.5	27.8	28.5
ΔG^{298}	50.1	42.3	32.2	29.2	20.2	20.8
ΔH_{exp}[b]	52.6		39.4		−	
ΔH_{exp}[c]	55.5		42.4		−	

[a] All values are in kcal/mol. [b] Reference 5. [c] Reference 3.

The observation that more than one isomer can give MIA values in agreement with the experimental data suggests another factor that must be taken into account in the theoretical evaluation of MIA and in the interpretation of the experimental measure.

Conclusions

Computations at the B3LYP/6−311++G** level have shown that eight minima are present on the potential energy surfaces for gas-phase metalated (Li^+, Na^+, K^+) α-alanine. The lowest energy structure corresponds to a N,O-bicoordinated system arising from the most stable α-alanine conformer, in the cases of lithium and sodium, and to an O-monocoordinated complex for potassium arising from the lowest lying relative minimum of α-alanine. A total of five Li^+ and Na^+ and seven K^+ complexes fall in a range of about 10 kcal/mol above the global miminum. The three potential energy surfaces are very similar although the interconversion barriers and the isomeric energy separations decrease on going from lithium to potassium. The computed lithium and sodium affinities agree with the experimental data, allowing a reliable prediction for that of potassium. The possibility that the relative rather than the absolute minima can influence the measured values of metal affinity has been demonstrated especially in the case of potassium.

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(33) Armentrout, P. B. *J. Am. Soc. Mass Spectrom.* **2000**, *11*, 371.