Theory of ^{14}N and ^{17}O Nuclear Quadrupole Interactions in the Single Amino Acids Occurring in the Protein Chain of Cytochrome c

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The understanding of electron transport in proteins based on a novel technique involving muon spin rotation (μSR) measurements is a topic of great current interest. The technique, which involves study of spin relaxation of a positive muon (μ^+) trapped in amino acids in protein chains due to the fluctuating magnetic field that the moving electron produces, is based on the premise that the electron is generated by ionization of a muonium (Mu) which was trapped at the same site as the μ^+ left behind. In attempting to test this premise from first-principles for the Cytochrome c (Cyt c) system in which recent μSR measurements have been made, we have carried out Hartree-Fock investigations of the electronic structures of the bare amino acids and amino acids with μ^+ and Mu trapped at the oxygen of the C=O group common to all amino acids. With the aim that the comparison of theoretically predicted experimental nuclear quadrupole interaction (NQI) parameters will provide a useful test of the electron distribution in the amino acids of Cyt c, we present results for the nuclear quadrupole coupling constants (e^2qQ) and asymmetry parameters (η) for the bare amino acids and the amino acids with trapped μ^+ and Mu. The trends in e^2qQ and η for ^{14}N and ^{17}O between the various amino acids, as well as the changes in these parameters in the presence of μ^+ and Mu are being analyzed. It would be helpful to have experimental data for e^2qQ and η to compare with our predictions for the amino acids as they occur in vitro in polycrystalline Cyt c in which the μSR measurements have been carried out. It is also hoped that the μSR technique will be able to provide experimental data on e^2qQ and η for the ^{14}N and ^{17}O nuclei to compare with our predictions.

Key words: Hartree-Fock Calculations; Amino Acid Molecules; Muon and Muonium Trapping; Nuclear Quadrupole Interactions.

1. Introduction

Electron transport in protein chains is a phenomenon of great interest due to its occurrence in many biological processes, such as photosynthesis [1]. The electron transfer macromolecule Cyt c has recently been studied [2] through the μSR technique with the goal to reveal the microscopic details of the path an electron takes during its motion through the protein chain. An incoming μ^+ from a 100% spin-polarized muon beam picks up an electron, thereby becoming Mu, which then slows down and eventually gets trapped somewhere along the protein chain of Cyt c. It then loses the electron brought in, changing back to μ^+ which is assumed to stay at the same trapping site. From there it can probe the departing electron through spin-lattice interaction caused by the fluctuating magnetic field of the moving electron. By measuring [2] the nature of the dependence [3, 4] of the relaxation rate of the muon spin

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on the magnitude of the external magnetic field one can find the dimensionality of the path the electron is taking.

One of the assumptions the μSR experiment makes is that there exist sites in the protein chain of Cyt c which are able to trap both Mu and μ^+. Using the first-principles Hartree-Fock Cluster procedure, we have investigated potential trapping sites for their hospitality to Mu and μ^+. In the course of this, we also calculated e^2qQ and η for ^{14}N and ^{17}O in a set of amino acids with and without Mu or μ^+ attached, with the motivation that comparison between experimental results and theoretical predictions can provide a test of the accuracy of the calculated electronic charge distributions. We also report on the trapping properties of Mu and μ^+, and how they affect e^2qQ and η as compared to the bare molecules.

2. Procedure

In order to make the ab initio calculations feasible, we had to divide the protein chain, which consists of a sequence of 104 amino acid molecules [5], into smaller clusters. The straightforward approach is to use individual amino acids as the basic units. We have chosen to investigate four different types of amino acid molecules, namely Glycine, Cysteine, Alanine and Lysine. Each of them occurs at multiple locations in the protein chain of Cyt c.

Regarding the structure of the amino acids under investigation, we decided that the normal structure (Fig. 1) [6] in which neither the amino group is protonated nor the carboxyl group deionized would

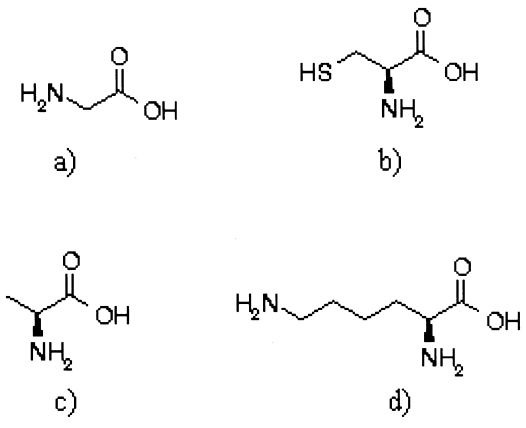

Fig. 1. Structure of the four amino acids investigated. a) Glycine, b) Cysteine, c) Alanine and d) Lysine.

be appropriate. This is because the Cyt c samples used in the μSR experiment were extracted from horseheart and prepared in crystalline form. Therefore no exchange of H^+ ions is expected to take place.

For each of the four amino acids we carried out three Hartree-Fock calculations [7]: one for the bare molecule with the geometry taken from the structure found experimentally [6], one for the case when Mu is placed near a potential trapping site, and one for the case when μ^+ is placed near the same site. In the latter two cases, the positions of Mu and μ^+ as well as the positions of nearest and next nearest neighboring atoms were optimized for minimum energy. In the Hartree-Fock calculations we simulated Mu by a hydrogen atom and μ^+ by an H^+ ion, the mass difference not influencing our results, since we were not concerned about vibrational effects in this study. The binding energy of Mu and μ^+ was then determined by subtracting the total energy found for the case where either Mu or μ^+ was attached, from the total energy of the bare molecule. For Mu one needs to subtract also the total energy of Mu, which is essentially equal to the ground state energy of a hydrogen atom (–13.6 eV). If the binding energy for a particular site is positive for both Mu and μ^+, then one can conclude that this site is indeed able to first trap Mu and then subsequently hold on to the μ^+ when the electron leaves the Mu.

The electronic wave functions obtained by the Hartree-Fock calculations were used to evaluate the EFGs at the ^{14}N nucleus in the amino group (NH_2) and at the two ^{17}O nuclei in the carboxyl group (COOH) of each amino acid, and in the case of Lysine the ^{14}N nucleus in the residual part. The quadrupole coupling constants (e^2qQ) can be obtained from the calculated values of the largest principal component of the EFG $V_{zz} = -eq$ for the bare form of the amino acids and with trapped μ^+ or Mu using the values of Q available in the literature, either [8] 0.0193 and 0.0258 barn for ^{14}N and ^{17}O or the slightly different values also available in the literature [9] 0.0201 and 0.02558 barn, respectively. The other pertinent parameter for nuclear quadrupole interaction is the asymmetry parameter $\eta = (V_{xx} - V_{yy})/V_{zz}$, with $|V_{xx}| \leq |V_{yy}|$. It is hoped that these predictions can be tested by NQR experiments on the bare individual amino acid molecules and in the trapped μ^+ or Mu systems by some special techniques associated with μSR measurements.

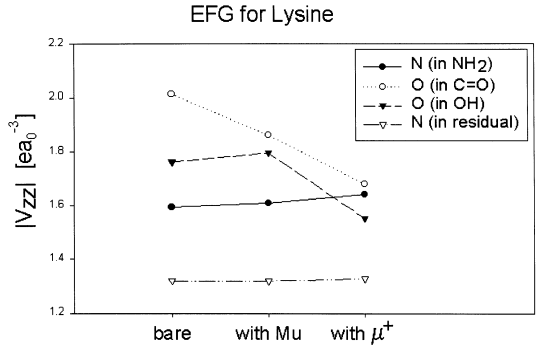

Fig. 2. The absolute values of the maximum principal component V_{zz} of the EFG tensor for two ^{14}N nuclei and two ^{17}O nuclei in Lysine. Three cases are shown: bare molecule, muonium attached and positive muon attached (at the double bond oxygen). The EFG is expressed in atomic units (ea_0^{-3}), where e is the electronic charge and a_0 the Bohr radius.

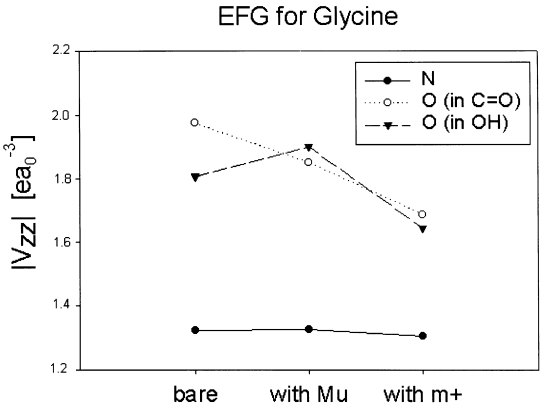

Fig. 3. The absolute values of the maximum principal component V_{zz} of the EFG tensor for one ^{14}N nucleus and two ^{17}O nuclei in the amino acid Glycine.

3. Results and Discussion

Through extensive testing [10], we have found that Mu can only be trapped in any of the four amino acids by breaking the double bond in the carboxyl group between C and O (Fig. 1), changing it into a single bond. The unpaired electron at either C or O can then form a covalent bond with the Mu. The positively charged μ^+, on the other hand, is only stable at sites which carry an effective negative charge. Among C and O, the two possible trapping sites for Mu, only O is partially negatively charged. Therefore, it is only the double bonded O which can trap both Mu and μ^+. This result was found first for Cysteine and was subsequently verified [10] for Glycine, Alanine and Lysine. This trapping site for both μ^+ and Mu involving the O in the C=O group in COOH is therefore of main interest for the interpretation of μSR data [2] and is the one for which we have investigated ^{14}N and ^{17}O NQI parameters in the trapped systems.

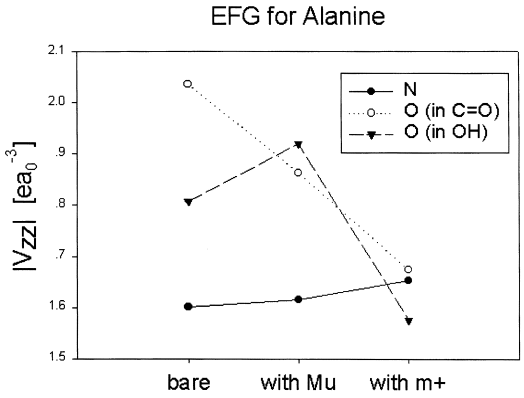

Fig. 4. The absolute values of the maximum principal component V_{zz} of the EFG tensor for one ^{14}N nucleus and two ^{17}O nuclei in the amino acid Alanine.

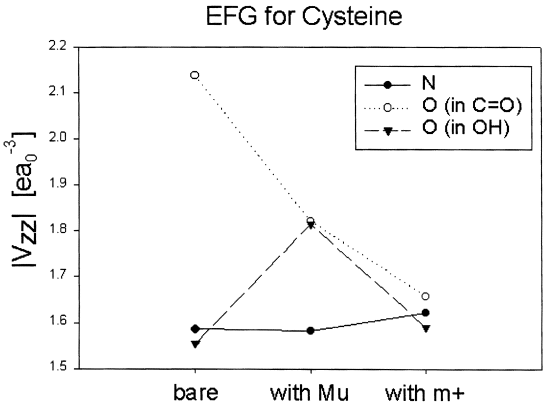

Fig. 5. The absolute values of the maximum principal component V_{zz} of the EFG tensor for one ^{14}N nucleus and two ^{17}O nuclei in the amino acid Cysteine.

From the optimization procedure used in our work, the length of the bond between Mu and O was found to range from 0.947 Å to 0.952 Å for the four amino acids investigated, and the bond angle C-O-Mu from 113.0° to 114.2°. For μ^+ the corresponding intervals are 0.963 Å to 0.968 Å and 119.8° to 120.8°.

We turn next to the determination of the EFG for ^{14}N and ^{17}O for the purpose of predicting e^2qQ and η, and to examine what effect the presence of Mu and μ^+ has on the electronic distribution. We present

our results in Figs. 2-5 and discuss details only for Lysine, since it acts as a representative example for the other three amino acids because it was found that the EFG at ^{14}N and ^{17}O does not change very much for different amino acids and the qualitative behavior is essentially the same too.

Considering the bare systems, we notice from Figs. 2-5 that the largest principal component $|V_{zz}|$ in all four amino acids seems to have the trend of having the largest value for ^{17}O in the C=O group, next largest for ^{17}O in the OH group and smallest for the ^{14}N in the NH_2 group. This trend could be understood from physical considerations, associated with the number of neighbor atoms to which the atom containing the nucleus in question is bonded, the greater spherical symmetry being expected to hold for the N atom with three neighbors, with increasing departures from such symmetry for the O of the OH group with two neighbors and the O of the C=O group with only one neighbor. For Lysine, where there is a nitrogen in the residual part of the amino acid, the $|V_{zz}|$ at ^{14}N is even weaker than for the nitrogen in the amino group nearer to the C=O group. This trend could be associated with the smaller departure from spherical symmetry expected for the N in NH_2 in the residual region because of lesser double bond character as compared to the N of the NH_2 group nearer to C=O, where there is greater departure from pyramidal behavior with a tendency towards planarity.

We see in Fig. 2 that q (where q refers to the maximum principal component V_{zz} of the EFG tensor) at ^{14}N in the amino group (NH_2) is much less influenced by the presence of Mu and μ^+ than those at the two ^{17}O. This is of course expected since ^{14}N is located relatively far away from the trapping site of Mu and μ^+. This is even more true for the ^{14}N in the residual part, where the dependence of q on the presence of Mu and μ^+ is almost negligible. In any case (with the exception of O in OH of Cysteine) μ^+ affects the q more strongly than Mu does. This result was not anticipated, since Mu bonds covalently, which one would intuitively have expected to have a strong effect on the electron distribution, while μ^+ as a point charge could electrostatically polarize the electron cloud which would appear to correspond to a perturbation type effect.

The magnitude of q at O in C=O is seen from Fig. 2 to get reduced when Mu is attached and is further reduced by about the same amount when μ^+ takes the place of Mu. For the O in OH, Mu slightly increases q while μ^+ reduces it with respect to the bare system. Cysteine is the only one of the four amino acids studied which represents an exception to this behavior for O in OH, with the trapped μ^+ seen from Fig. 5 to increase q with respect to the bare molecule but staying below the value for trapped Mu. The trends reported are of course the same for both the q and the nuclear quadrupole coupling constant e^2qQ.

From the three principal components V_{xx}, V_{yy} and V_{zz} of the EFG tensor we calculated the asymmetry parameters η for the four different amino acids with and without μ^+ or Mu trapped. The results for these are shown in Tables 1 - 4. As it can be seen, η for the bare molecules is relatively smaller for N and O (C=O) as compared to O (OH), except for Cysteine. A possible explanation for this behavior can again be given in terms of the symmetry of the different sites, the important feature here being the departure from axial symmetry. For N the symmetry is close to three-fold pyramidal and would be completely three-fold if the adjacent C atom were replaced by H; O in C=O

Table 1. Asymmetry parameter η for Lysine.

	N (NH_2)	O (C=O)	O (OH)	N (residual)
bare	0.130	0.083	0.808	0.138
with μ^+	0.207	0.052	0.397	0.198
with Mu	0.079	0.825	0.987	0.128

Table 2. Asymmetry parameter η for Glycine.

	N	O (C=O)	O (OH)
bare	0.199	0.145	0.565
with μ^+	0.367	0.090	0.212
with Mu	0.167	0.856	0.796

Table 3. Asymmetry parameter η for Alanine.

	N	O (C=O)	O (OH)
bare	0.133	0.053	0.898
with μ^+	0.215	0.088	0.557
with Mu	0.082	0.829	0.920

Table 4. Asymmetry parameter η for Cysteine.

	N	O (C=O)	O (OH)
bare	0.072	0.986	0.298
with μ^+	0.217	0.099	0.228
with Mu	0.106	0.901	0.999

has linear geometry and η would have been zero if the double bond did not exist; O in OH is closer to two-fold symmetry than in H_2O, with a strong difference expected in V_{xx} and V_{yy} associated with the x- and y-directions. For Cysteine the value of η is large for O (C=O) showing that V_{yy} and V_{zz} are similar to each other in magnitude but with opposite signs. This difference in nature compared to the other three amino acids perhaps comes from the residual group, particularly the sulfur atom influencing O.

The effects of μ^+ and Mu are seen to be in general quite important but with no particular trend as to which is stronger in influencing η as compared to the bare molecules. This feature could perhaps be explained by noting that μ^+ may not be taking a mainly polarizing role as one would intuitively expect as pointed out earlier. In fact it is likely that the O next to the trapped μ^+ makes a donation of electrons to the μ^+ making it partially resemble a Mu, leading to some covalent bonding as in the trapping of Mu itself.

As a concluding remark, we would like to mention that measurements of NQI, for both bare molecules by NQR or NMR technique and molecules with μ^+ and Mu, with some sophisticated techniques associated with μSR, would be very helpful in testing the predictions of theory.

[1] G. Feher and M. Y. Okamura, in: R. K. Clayton, W. R. Sistrom (Eds.), The Photosynthetic Bacteria, Plenum Press, New York 1978, Chapter 19.
[2] K. Nagamine, F. L. Pratt, S. Ohira, I. Watanabe, K. Ishida, S. N. Nakamura, and T. Matsuzaki, RIKEN Rev. **20**, 51 (1999); K. Nagamine, F. L. Pratt, S. Ohira, I. Watanabe, K. Ishida, S. N. Nakamura, and T. Matsuzaki, Physica B**289-290**, 631 (2000).
[3] M. A. Butler, L. R. Walker, and Z. G. Soos, J. Chem. Phys. **64**, 3592 (1976).
[4] R. Risch and K. W. Kehr, Phys. Rev. B **46**, 5246 (1992).
[5] L. Banci, I. Bertini, H. B. Gray, C. Luchinat, T. Reddig, A. Rosato, and P. Turano, Biochem. **36**, 9867 (1997).
[6] Webpage of the Institute of Chemistry, Freie Universität Berlin. http://www.chemie.fu-berlin.de/chemistry/bio/amino-acids_en.html
[7] See for example: papers on the Hartree-Fock Cluster procedure in "Electronic Properties of Solids Using Cluster Methods", T. A. Kaplan and S. D. Mahanti ed., Plenum Press, New York 1995.
[8] Pramila Raghavan, Atomic Data and Nuclear Data Tables **42**, 189 (1989).
[9] P. Pyykkö, Z. Naturforsch. **47a**, 189 (1992).
[10] D. Cammarere, R. H. Scheicher, N. Sahoo, T. P. Das, and K. Nagamine, Physica B **289-290**, 636 (2000).