constant. A higher rotational temperature results in a decrease in the probability of the dipole of the molecule "locking in" on the charge of the ion (a rear attack). Consequently, a lower reaction probability results. The pure temperature dependences of the rate constant data are given by the slopes of the lines through those data points obtained with no electric drift field. For the CH_3Cl, CH_3Br, and CH_3I reactions, they are $T^{-0.8}$, $T^{-0.7}$, and $T^{-0.3}$, respectively. The theoretical results are $T^{-0.48}$, $T^{-0.45}$, and $T^{-0.41}$, respectively.

It should be noted that the trend in both the kinetic energy and temperature dependences of the rate constants for the reactions studied is $CH_3Cl > CH_3Br > CH_3I$, both experimentally and theoretically. The exothermicities of the reactions are 34.3, 41.4, and 48.2 kcal mol^{-1} for CH_3Cl, CH_3Br, and CH_3I, respectively. Therefore the less exothermic the reaction, the more negative the energy/temperature dependence. This may reflect a more severe orientation requirement for the less exothermic reactions or that the central barrier becomes more important as the exothermicity decreases.

The disagreements between theory and experiment in the present work may well be due to the approximate nature of the potential used. Especially critical is the assumption that the geometry of the neutral and the charge distributions of the ion and neutral are fixed at their separated positions even when the reactants approach to a hard sphere collision diameter. Allowing for changes in geometry and charge distribution could easily allow for a more favorable orientation during a collision. In addition, no attempt was made here to include explicitly any steric hindrance. A further error is the neglect of the central barrier. Neglect of the central barrier may be more important for the lighter halides, since the reactions are less exothermic. However, it is worth noting that the model is a phenomenological description of the orientation dependence, rather than a causal mechanism.

In spite of the simplicity of the theoretical calculations, there are substantial similarities between the results of the theoretical model and the results of the experiments, indicating that the approach taken here is a useful first approximation to including orientation effects. The similarities are such that it appears that the collision angle plays an important role in influencing S_N2 reaction rates, as expected, and that the reactions being studied do occur via a rear attack in the gas phase. Additionally, the changes in slope of the curves showing rate constants vs $\langle KE_{cm} \rangle$ appear to be explained simply by the fact that the slope changes occur at the energies at which the hard sphere collision diameter is equal to the capture radius. This has the effect of not allowing much orientation during an encounter at higher $\langle KE_{cm} \rangle$.

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Electron-Transfer Reactions in Proteins: A Calculation of Electronic Coupling

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The distance and medium dependences of the electronic coupling for electron-transfer reactions in proteins (particularly the ruthenated myoglobins studied by Gray et al.) are calculated. An extended-Hückel method is used to treat individually the donor, the acceptor, and the intervening protein. A search method is used to select a subset (~15–20) of the approximately 150 amino acid residues as being the most relevant for the electron transfer. Approximate agreement is found between ratios of experimental matrix elements (approximated as ratios of square roots of rate constants) and of calculated electronic matrix elements. No adjustable parameters were introduced in the diagonalization. Only parameters available in the literature from other (non-electron-transfer) sources were employed.

I. Introduction

Long-range electron transfer plays a major role in many biological processes, as in respiration and photosynthesis. Biological electron-transfer reactions have been shown to occur rapidly over large molecular distances. In recent years, there has been considerable experimental research in the field of intramolecular electron-transfer reactions in native and modified proteins.[1-4] There have also been many studies of long-range intramolecular electron transfer in synthetic organic and metal complex systems, in which investigations were made of the effects of driving force, reorganization energy, distance, and the intervening material on the rate of long-range electron-transfer reactions.[5-8]

In electron-transfer theory, the rate constant for transfer of an electron from a donor to acceptor can be expressed in terms of a golden-rule type expression, namely, as the product of the square of an electronic coupling matrix element (H_{DA}) and a nuclear Franck-Condon factor (FC):[9-11]

$$k_{ET} = (2\pi/\hbar)|H_{DA}|^2(FC) \qquad (1.1)$$

Expressions for FC are given, for example, in a recent review.[9]

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FC depends on reorganizational (λ) and driving force ($-\Delta G°$) factors.[9]

For small systems, the electronic matrix element, H_{DA}, has been calculated by using all-electron SCF models.[12-14] This approach has been shown to give detailed information on the effect of ligand orientation on H_{DA}, for example. However, at the present time, the application of such models to biological systems, such as proteins, is a formidable problem. It is, therefore, desirable to employ a simpler model to understand and estimate the electronic coupling between a donor and an acceptor separated by 15–20 Å of protein sidechains and backbone.

In our recent study of long-range intramolecular electron transfer in synthetic model systems[15] the one-electron extended Hückel theory[16,17] was used to calculate the electronic coupling between the donor and acceptor. Experimental results were available for four different series, two of them purely organic and two of them involving metal ions connected by an organic bridge. In a theoretical treatment of the four series,[15] it was found that though the absolute values of H_{DA} agreed with the experimentally determined values only approximately in some cases, the agreement of the distance dependence of H_{DA} between the theoretical and experimental values was very good. In the present paper, we extend the method used in ref 15 to study electron-transfer reactions in proteins.

Previous theoretical studies of electron transfer within proteins[17-22] have employed different approaches, such as a frontier orbital analysis of the groups on the tunneling pathway,[21] pseudopotential-based path integral calculations,[18] and a modified tight-binding method.[22] Even a relatively small protein molecule can contain up to 800 heavy atoms, and therefore even a valence-electron calculation would require diagonalization of an approximately 3200 × 3200 matrix. Hence, it is clear that further simplification of the problem is desirable. We have made this simplification in the following way:

(i) We have used a searching method to select the "important" amino acid residues for electron transfer between the donor and acceptor sites.

(ii) We have then made use of a partitioning technique[17,23,24] to calculate the electronic coupling.

The present method avoids in this way an alternative method of calculating H_{DA}, in which the latter is obtained as an extremely small number, in the case of electron transfer in proteins, a small difference between two large eigenvalues. It has been shown in ref 15 that both the latter direct diagonalization method and the more approximate method of partitioning lead to comparable values of H_{DA} for the systems studied there.

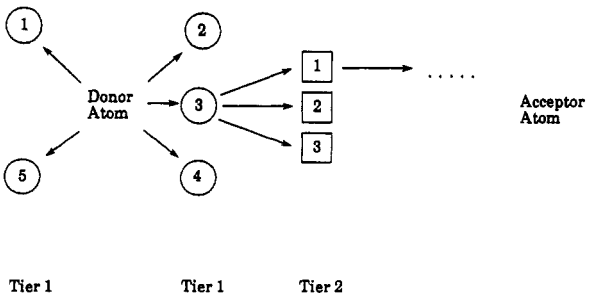

Figure 1. Schematic illustration of the search procedure, in which the circled atoms 1–5 are those with the largest matrix elements for electronic coupling to atoms of the donor species. The constitute tier 1 of the search. Each is coupled to another set of atoms, those with the largest matrix elements being called tier 2. Part of tier 2 are atoms 1–3 in squares; they having the largest couplings to the circled atom 3. Also part of tier 2 (not shown) are the atoms with the largest couplings to circled atoms 1, 2, 4, and 5.

II. Theory. Protein Structure and Searching Procedure

In the case of an electron-transfer reaction in a protein molecule, it is clear from the experimental data (e.g., ref 4) that the reaction cannot proceed by a purely through-bond pathway, which would involve an unusually long path. It is also equally clear that it cannot be a purely donor-to-acceptor "through-space" electron transfer, not making use of molecular orbitals of the intervening material, since the rate then would be considerably slower than that experimentally determined. It is necessary, instead, to consider a combination of through-bond and, occasionally between residues, through-space links between the donor and acceptor.

We consider proteins whose crystal structure is known. To formulate a search for the "important" amino acids or atoms of amino acids, it is necessary to have a measure of the electronic coupling matrix element V_{AB} between atoms. We use the following plausible expression[25]:

$$V_{AB} = K\sum_{a}^{A}\sum_{b}^{B} S_{ab}(\epsilon_a + \epsilon_b/2) \qquad (2.1)$$

where a denotes an atomic orbital on atom A, b denotes an atomic orbital on atom B, S_{ab} is the overlap integral between a and b, ϵ_a and ϵ_b are the orbital energies, and K is a constant. This expression for V_{AB}, but not summed over the a and b orbitals, is the well-known Wolfsberg–Helmholtz formula for the resonance integral between two atomic orbitals.[26] Expression 2.1 has the advantage of employing well-tested parameters. (K is usually taken to be 1.75.[16]) Further, V_{AB}, as calculated above, depends directly on the overlap between atomic orbitals and thus takes into account the effect of the mutual orientation of the amino acids. In the present study, to keep the model as parameter-free as possible, both the through-bond and through-space couplings are

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calculated by using the above expression.

Given a protein crystal structure, it is possible to establish covalent and hydrogen-bonded connections between atoms in the protein.[27] As a result, one obtains the through-bond connections between the atoms in the protein. Then, for each bonded pair of atoms A and B, V_{AB} is calculated. The through-space links are obtained as follows: Through-space couplings between each atom A and any other atom B within a given radius of the atom A, but not bonded to A, are calculated.

Once both the through-bond and through-space connections and their individual electronic couplings have been determined, a tree-like search algorithm is implemented, starting from the donor atom and reaching the acceptor site (Figure 1). For the search to yield more than just a few amino acids between the donor and acceptor, it was found important to consider a reasonable number (given below) of the amino acid (or other) atoms coupled to the donor in the first step in the selection in Figure 1. These atoms are chosen on the basis of the magnitude of their coupling to the donor atom, i.e., the atoms with the largest couplings V_{AB} to the donor atom are selected and constitute the first tier of the search. In the next selection the atoms with the largest V_{AB}'s on going from the first to a new set of atoms were retained and constitute the second tier of this search.

At this stage, after these two tiers, an angle restriction (given below) was imposed so that the search would ultimately point from the donor toward the acceptor. Then from each of these tier-2 atoms (minus duplicates, if any), useful paths to the acceptor group were sought by an iterative procedure: once a path (i.e., a combination of through-bond and through-space links) was found from any one of the tier-2 atoms to the acceptor group, a net measure of electronic coupling, calculated as the product of the V_{AB}'s of the links in that path, was assigned to it. That is, a "net coupling" for a particular path of $n + 1$ atoms is given by

$$\text{net coupling} = \prod_{i=1}^{n} V_{i,i+1} \tag{2.2}$$

where $V_{i,i+1}$ is the V_{AB} for successive atoms along the path. Then, other paths are sought from the same tier-2 atom to the acceptor, and finally only that path that has the greatest net electronic coupling is retained. There are a number of such paths, since there are a number of tier-2 atoms. A preset threshold value for the net electronic coupling for any path is used in order to exclude particularly unimportant paths from the search. All amino acids that contain atoms of the accepted path are then included in a later diagonalization. It can perhaps be stressed that more than one path from the donor to the acceptor is included.

Since we are using a treelike search algorithm, it is desirable to perform searches in both directions, i.e., starting from the donor and reaching the acceptor as well as starting from the acceptor and reaching the donor. Since the number of atoms chosen for expansion in the first and second tiers of the search can be varied, this method increases the likelihood that the important amino acids will be selected in the search. It may also be noted here that to shorten the time taken for the search, it is sometimes advisable not to treat the whole protein but a subset of it, chosen by including in the search all the amino acid residues within, say, 10 Å of the donor and the acceptor sites, in addition to others within a cylinder of radius 10 Å, the axis of the cylinder being the line of centers of the donor and acceptor.

In this way a set of atoms that lie on the paths from the donor to the acceptor was selected. The amino acids to which these atoms belong are regarded as the important amino acids for electron transfer in the protein. These residues, collectively, can then be considered to constitute the important part of the molecular bridge that mediates the electron transfer in the protein. By selection of only a subset of the amino acids, there are "dangling atoms", which link these amino acids to those not selected. In each case an H atom was added to complete the valency of the dangling atom. In summary, we determine the more important residues using the path concept but then use in the final calculations all of the amino acids associated with the selected paths.

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III. Theory. Calculation of Bridge Orbitals and H_{DA}

Once the particular amino acids were selected, in the search, as the bridge, the calculation of the properties of the bridge proceeded as in ref 15. The eigenvalues and eigenvectors for the bridge orbitals were calculated via a diagonalization technique, using the various atomic orbitals of this bridge as a basis set.[15]

We now recall the results of using the partitioning technique in a one-electron description of the system containing a donor D, a bridge B, and an acceptor A.[15,17] For the case where the bridge B is linked to the donor by only one atomic orbital of each and where B is similarly linked to the acceptor, the electron-transfer matrix element H_{DA} is given by[15,17]

$$H_{DA} = \eta_D \eta_A \sum_{\nu} \frac{C_{D\nu} C_{A\nu}}{b_\nu - a} \tag{3.1}$$

where a is the energy of the localized molecular orbital of D, equal to the energy of A in the transition state for the electron transfer, b_ν is the energy of the νth molecular orbital of the bridge B, $C_{D\nu}$ is the coefficient of that bridge orbital at the point of contact of B with D, $C_{A\nu}$ is the corresponding quantity at the point of contact of B with A, η_D is the matrix element for the interaction between D and the adjacent atomic orbital of B, and η_A is the corresponding quantity for A.

When D and A are each linked to more than one atomic orbital of B, eq 3.1 is replaced by[15,17] where $\lambda_j(\mu_k)$ is the matrix element

$$H_{DA} = \sum_{\nu} \left(\sum_{j} \lambda_j C_{j\nu}\right)\left(\sum_{k} \mu_k C_{k\nu}\right)/(b_\nu - a) \tag{3.2}$$

for the interaction of the D(A) orbital and the jth (kth) adjacent atomic orbital of B.

When the donor D and the acceptor A have a number of atomic orbitals of their own linked to the various bridge orbitals, eq 3.2 is replaced by[15,17]

$$H_{DA} = \sum_{\nu} \left(\sum_{j}\sum_{l} C^{D}_{l\chi} \lambda_{jl} C_{j\nu}\right)\left(\sum_{k}\sum_{m} C^{A}_{m\rho} \mu_{km} C_{k\nu}\right)/(b_\nu - a) \tag{3.3}$$

where χ and ρ denote the molecular orbitals on D and A that are involved in the electron transfer, l and m denote the atomic orbitals of D and A, and j and k denote the atomic orbitals on B connected to D and A, respectively. C^D and C^A are the MO coefficients of D and A. the λ_{jl} are the interaction matrix elements of the donor and bridge orbitals, and μ_{km} are the corresponding quantities for the bridge and acceptor orbitals. Here a is the energy of the molecular orbital χ of D and is matched to that of the molecular orbital ρ of A in the transition state, "matched" by replacing each such energy by the mean of these two energies.

IV. Results and Discussion

The proteins considered here are the ruthenium-labeled myoglobin systems studied by Gray and co-workers.[28] Sperm-whale myoglobin contains four surface-accessible histidine residues, which can be individually and singly ruthenated, giving four new compounds. Further, the heme was replaced by Zn or Mg mesoporphyrin.[28] The four resulting $(NH_3)_5Ru(His)Mb$ derivatives (with Zn or with Mg) now contain identical donor and acceptor species but at different fixed sites. In these systems, the free energy of reaction and reorganization energy of the reaction are regarded as held fixed (they may have, in principle, some distance dependence), allowing only the electronic distance and medium effects to be examined for the protein.

Thus in the present calculational procedure, the donor is the ruthenated histidine group, the acceptor is the metal porphyrin,

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TABLE I: Amino Acid Residues Determined by the Search Procedure for the Four Ruthenated Systems

His-48 deriv	His-81 deriv	His-116 deriv	His-12 deriv
		Search 1	
Thr-39	Glu-83	Val-13	Val-13
Leu-40	His-82	Trp-14	Trp-14
Glu-41	Ala-84	Val-17	Ala-15
Lys-42	Leu-86	His-24	Lys-16
Phe-43	Lys-87	Ile-28	Val-17
Asp-44	Ala-90	Leu-29	Ile-28
Arg-45	His-93	Val-66	Leu-29
Phe-46	Phe-138	Thr-67	Leu-32
Lys-47	Asp-141	Val-68	Leu-69
Leu-49	Ile-142	Leu-69	Leu-72
Lys-50	Trp-146	Thr-70	Phe-106
Glu-54		Ile-111	Ile-107
Ala-57		Val-114	Ile-111
Ser-58		Leu-115	Leu-115
Asp-60		His-119	Asp-122
		Phe-123	Phe-123
	Extended Search: Additional Amino Acids		
Val-68	Ser-92	His-64	His-64
Thr-67	His-97	Ser-117	
Val-66	Lys-98	Ile-112	
Gly-65	Lys-42		
	Phe-43		

and the bridge consists of the collection of amino acids determined by the search. The protein coordinates were obtained from the Brookhaven data bank.[29] Of the structures of the modified myoglobins, modified by adding $(NH_3)_5Ru$ to one of the four surface histidines, His-48, His-81, His-116, and His-12, only the His-48 one has been determined experimentally.[30] For consistency, all were "determined" by using BIOGRAF,[31] by minimizing the conformational energy of each structure. This particular part of the calculation included only the contribution of those atoms within 8.5 Å of the ruthenated histidine and resulted in only minor modifications of the myoglobin structure.[30] The resulting structure files were used in the search algorithm.

In the first tier of the search, five atoms were retained, and for each of them three atoms were retained in the second tier (Figure 1). Then, from each of the 15 atoms in the second tier, a path to the acceptor was found, as described earlier. The angle between the donor, any atom found by the search after the first and second tiers, and the acceptor was constrained to be less than or equal to 120°.[32] The search was then performed in the reverse direction, starting from the acceptor and seeking the donor. In Table I, we give the amino acid residues that were determined by the search for each of the four ruthenated myoglobin structures. It is seen that search 1 provided 11–16 important amino acids for the electron transfer. Myoglobin itself has about 150 amino acids. It is clear that the needed reduction in the number of amino acids to be considered has been achieved by the searching technique, thus constituting a major simplification of the problem.

Once the important amino acids were determined for each of the four ruthenated myoglobin systems, an extended-Hückel calculation was performed for the donor, bridge, and acceptor separately, and eq 3.3 was used to calculate the electron-transfer matrix element.[33] The results obtained for the four systems are given in Table II. The value of H_{DA} extracted from the experimental data for the His-48 derivative is 0.006 cm^{-1}.[28] Of course, it is important to note that the value of H_{DA} that was derived experimentally is itself model-dependent and might change if a different analysis of the experimental data were used. Further, the extended Hückel calculations as judged from the results in ref 15 are more reliable for relative values of H_{DA}. A comparison of the relative values is also given in Table II. It should perhaps be stressed that because of the empirical nature of the extended Hückel approach, it is preferable to use the present results for comparisons among the four derivatives rather than for the absolute values of the electronic coupling elements themselves.

To assess the importance of amino acids not found by the present search for electron transfer, the search was again performed for all the four ruthenated myoglobins but with more tolerant cutoffs (10^{-12} for the His-48 derivative and 10^{-28} for the others) than those used[32] in the first search. This extended search then yielded, in addition to the paths in search 1, additional paths. Again, once the amino acids were determined, eq 3.3 was used to calculate H_{DA}. It was found that the magnitude of the electronic coupling did not change much from what was found before, essentially no change for the large H_{DA} (His-48) and changes within a factor of 2–3 for the small H_{DA}'s (Table II). This result provides further evidence that the search performed does, in fact, within such limits, select the important amino acids.

Experimentally, it has been found that the rate constant of electron transfer in the His-48 modified myoglobin (7×10^4 s^{-1}) is much larger than that in the other three modified myoglobins, for which the rate constants are more or less the same, of the order of 100–85 s^{-1}. Thus, the rate constant for the His-48 derivative is about 700–800 times larger than for the others. This factor of 700–800 in the rate constant would appear as a factor of 26–30 in the electron-transfer matrix element H_{DA}, when the free energy and reorganization energy effects are assumed constant. For the calculated values also, it can be seen from Table II that H_{DA} for the His-48 derivative is considerably larger than the matrix elements for the other three derivatives. The theoretical ratio of the electron-transfer matrix element in the His-48 to that in the His-81 derivative, for example, is 14 in search 1, thus showing that the calculated values are at least reasonable and what would be expected on the basis of the available experimental data. Additional experiments on the temperature effects and on measurements of $\Delta G°$, if possible, in all four cases would be helpful in seeing whether the FC in eq 1.1 is approximately the same for the four compounds. The present model neglects any effects due to conformational fluctuation of the protein coordinates on H_{DA}. An interesting observation from the results is that there is an exponential decrease, $|H_{DA}|^2 \propto \exp(-\beta R)$, where R is the edge-to-edge separation distance between donor and acceptor and where the calculated $\beta \simeq 0.8$–0.9 $Å^{-1}$. This β is in the same range as is found for electron transfer across saturated bridges, for example.[5]

Further, with the model, some assessment can be made of the role played by specific amino acid residues in the protein electron-transfer reactions. For example, it has been speculated[28] that since the rate of electron transfer in the His-12 derivative is slightly higher than would be expected from the distance correlation, the aromatic residue, Trp 14, which lies in a parallel orientation directly between the donor and the acceptor, may enhance the electronic coupling and thus increase the rate of electron transfer. With the present model, it is possible to make some a priori estimate of such an effect. By replacing the Trp 14 residue in the myoglobin structure by another similarly nonpolar but saturated residue such as leucine or proline,[34] we found that there is

(29) Brookhaven Protein Databank: Bernstein, F. C.; Koetzle, T. F.; Williams, G. J. B.; Meyer, Jr., E. F.; Brice, M. D.; Rodgers, J. R.; Kennard, O.; Shimanouchi, T.; Tasumi, M. *J. Mol. Biol.* **1977**, *112*, 535.

(30) Mottonen, J.; Ringe, D.; Petsko, G., unpublished results, cited in: Lieber, C. M.; Karas, J. L.; Mayo, S. L.; Albin, M.; Gray, H. B. *Proceedings of the Robert A. Welch Foundation, Conference on Chemical Research XXXI. Design of Enzymes and Enzyme Models*; 1987; Vol. 31, p 9. These authors also give a computer graphic projection comparing the His-48 ruthenated and the unruthenated structures.

(31) BIOGRAF/III: BIOGRAF was designed and written by S. L. Mayo, B. D. Olafson, and W. A. Goddard III. It is a product of Biodesign Inc., Pasadena, CA.

(32) It was found that the presetting of a threshold value for the net electronic coupling for any path was, by itself, sufficient to stop the search from proceeding in directions not leading to the acceptor. For the His-48 derivative, a cutoff value of 10^{-10} was used; for the other three derivatives, the cutoff value used was 10^{-16}.

(33) The extended Hückel program and the basis sets were obtained from the Caltech MQM files. See also: Huzinaga, S. *Gaussian Basis Sets for Molecular Calculations*; Physical Sciences Data 16; Elsevier: New York, 1984. Since the molecular orbitals of the donor, bridge, and acceptor are determined separately, there is a possibility that the energy differences in eq 3.3 may become very small. In the present case, none of the eigenvalues of the bridge are very close to the mean energy of the donor and acceptor orbitals, and hence the above problem did not arise.

TABLE II: Experimental and Calculated Ratios of Electron-Transfer Rate Constants or Matrix Elements for Four Ruthenated Myoglobin Systems

		H_{DA}(calcd), cm^{-1}		$H_{DA,i}/H_{DA,1}$ (calcd)		
i (deriv)	R (edge-to-edge), Å	search 1	extended search	search 1[b]	extended search	$(k_i/k_1)^{1/2}$[a] (expt)
1 (His-48)	12.7	0.11[c]	0.12[c]	1	1	1
2 (His-81)	19.3	0.008	0.003	0.07	0.03	0.04
3 (His-116)	20.1	0.004	0.01	0.04	0.08	0.04
4 (His-12)	22.0	0.002	0.001	0.02	0.01	0.04

[a] An experimental H_{DA} for only the His-48 derivative has been determined. Use of $(k_i/k_1)^{1/2}$ for the ratio $H_{DA,i}/H_{DA,1}$ omits any correction of the ratio for any R-dependent effect that may occur in FC, i.e., in the solvation effect on $\Delta G°$ and on λ. [b] The results in ref 27 for these four ruthenated histidines are 1, 0.02, 0.07, and 0.001, respectively. [c] $H_{DA,1}$(expt) $\simeq$ 0.006 cm^{-1}.

only a minor decrease in the value of H_{DA} from 0.0016 to 0.001 cm^{-1} in the case of the leucine-modified His-12 and to 0.0008 cm^{-1} in the case of the proline-modified His-12. These minor differences are within the uncertainties in the present calculations and may even be due to small geometrical changes. The present calculations thus suggest that a dramatic increase in the electronic coupling may not be expected due to the presence of a single aromatic residue. It is, of course, possible that an extended conjugated subsystem in the protein may give rise to a noticeably enhanced coupling. Pseudopotential calculations of Kuki and Wolynes[19] also did not indicate any special role for the intervening tryptophan residue in the His-12 derivative.

It is useful to compare the present approach and that of Betts and co-workers.[27] Betts et al.[27] considered all bonds to be equivalent and assigned a uniform value for what was called there "the decay factor" across any bond. Their through-space decay factors were calculated by an exponential-type formula with adjustable parameters. In our method the electronic coupling between any pair of atoms is explicitly calculated, instead, by means of eq 2.1, and is therefore dependent on the nature of the atoms themselves. The search procedures of the two methods also differ, the present one making use of a broader base of atoms to start from, while that of Betts et al. finds a path between the donor and acceptor and then searches the region near the acceptor for alternate routes. A third difference is that we use the search only as a means to find the amino acids that are important in the electron transfer and then proceed to make a quantum mechanical calculation (matrix diagonalization) using all of these residues, in order to obtain H_{DA}. Betts et al. determine, instead, the electron-transfer matrix element itself by making use of their parametrized (uniform) through-bond and through-space decay factors. It is perhaps worthwhile stressing that in the present study no adjustable parameters have been employed to obtain the matrix elements used in the diagonalization. The parameters used were those available in the literature from non-electron-transfer sources.

Brooks et al.[21] have used a frontier orbital analysis of groups in an extended-Hückel type formalism to calculte electronic coupling in model polypeptide systems. In that work, the orientational dependence of the interaction between groups was included, though only a pair of orbitals was used for each group.

V. Conclusions

A theoretical model is proposed for calculating the dependence of electron-transfer matrix elements on the amino acid residues in the protein. This method employs a search technique that yields the important amino acids for the mediation of electrons between the donor and the acceptor. Once the search is accomplished, a quantum mechanical method tested earlier for model rigid-bridge experimentally studied systems was applied to the ruthenated protein systems and was shown to give relative values of electron-transfer matrix elements consistent with experimental data.

The present model, by making use of a search algorithm, drastically reduces the number of amino acids to be considered in the quantum mechanical calculation and hence is suitable for large biological systems. The method considers all potential pathways rather than just a limited number of them and takes into account the nature and orientations of the intervening amino acid residues. Accordingly, it will be useful to investigate the dependence of electron-transfer rates on changes in the structure of proteins, which can be achieved by site-directed mutagenesis, as well as to treat electron transfer in other proteins. It is planned to investigate also the sensitivity of the calculated electron-transfer matrix element to the individual electronic properties.

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(34) BIOGRAF was used (see footnote 31). A local energy minimization (including all atoms within 8.5 Å of the replaced residue) was also performed in order to obtain a favorable conformation for the replaced residue.