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Charge Transfer

Distal Charge Transport in Peptides

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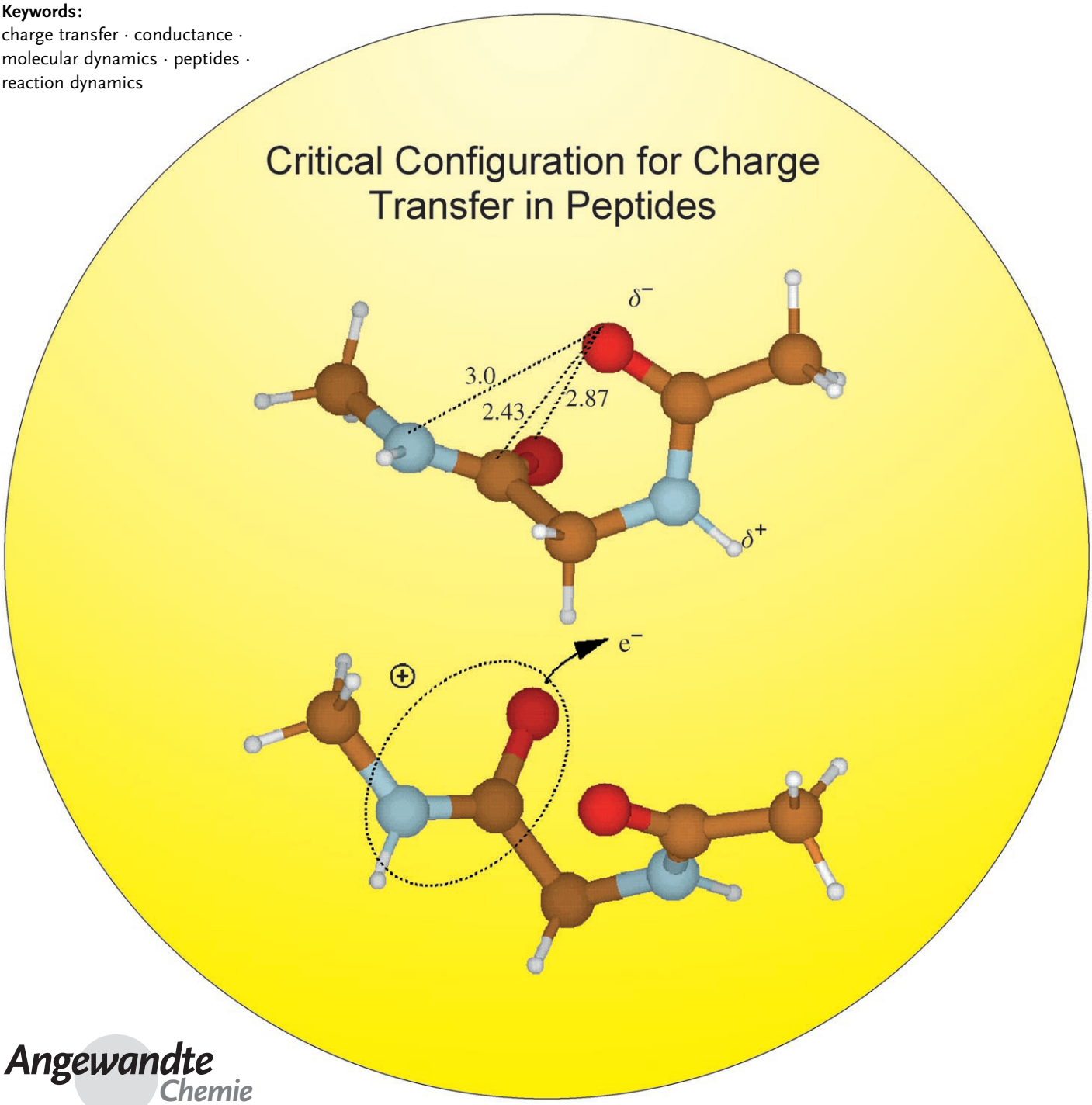

Biological systems often transport charges and reactive processes over substantial distances. Traditional models of chemical kinetics generally do not describe such extreme distal processes. In this Review, an atomistic model for a distal transport of information, which was specifically developed for peptides, is considered. Chemical reactivity is taken as the result of distal effects based on two-step bifunctional kinetics involving unique, very rapid motional properties of peptides in the subpicosecond regime. The bifunctional model suggests highly efficient transport of charge and reactivity in an isolated peptide over a substantial distance; conversely, a very low efficiency in a water environment was found. The model suggests ultrafast transport of charge and reactivity over substantial molecular distances in a peptide environment. Many such domains can be active in a protein.

From the Contents

1. Introduction

One of the many important functions occurring in proteins is the conveying of information with the help of a charge or some chemical change over substantial molecular distances in the protein chain, for instance, the transport of charge across a cell wall to activate intracellular chemistry. Chemistry at a point distant from the origin of excitation is a common observation for proteins and as such is of fundamental importance through, nevertheless, not yet well understood on an atomistic basis.[1–17] Conventional theories of chemical kinetics do not directly apply to action over substantial distances and as such chemical transport often involves charge migration as well. Hence, it is of some interest to evolve models for distal action, to show how action at one site generates molecular motions at a distant removed site. Such action can be considered as a chemical reaction that is carried out far away from the point at which the signal/charge has originated. We refer herein to this process of reactivity (R) and charge (C) transfer as RC transduction, a process of importance in more-complex signal transduction. A problem of such a model is the central issue that possible dissipative processes involving the many vibrational degrees of freedom along the way often prevent chemical reactivity at the distant site and thus prevent any substantial transport.[18,19] Herein, we wish to consider RC transduction for simple peptides that are shown to display ultrafast long-range transduction in spite of the many degrees of freedom present.

Long-range electron transfer (ET) is a fundamental mechanism in a variety of biological systems.[20] Electron transfer is involved directly or indirectly in many biological reactions,[21] such as oxidative phosphorylation, photosynthesis,[22–24] conductivity of the DNA helix,[25] and aerobic respiration.[26] Usually these involve a metal ion–biomolecule–metal ion system so that the metal ions are separated by a long distance, in some cases, this range may be longer than 10 Å. In 1980, Isied and co-workers[27] showed the electron transfer between two redox centers mediated by a polypeptide bridge. Gray and Winkler[26] were able to attach the redox centers to protein systems such as myoglobin, cytochrome *c*, etc. This modified donor–protein–acceptor system enables one to investigate the distance and structure dependence for the ET processes.[21,28–34]

What would a pure charge placed on a peptide, such as in localized photoionization at the C terminus, do to the chemistry and the charge transmission to the N terminus in the peptide? One might test such a model for distal processes on several levels. First, we tested the transport of charge and reactivity for the case of the intrinsic, isolated molecule. Second, we could then investigate the understanding of the perhaps much changed behavior of the process in a medium, such as water. Here the question would be: How does the medium influence information transport with respect to rate or yield? Experiments on simple model peptides have now revealed that the process in isolated molecules can be extremely efficient, whereas the same process in water is now predicted to typically be approximately two orders of magnitude less efficient.[35–38] One desiderata would be to find a simple model that presents us with an understanding of such extremes, at least for the case of peptides.

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We have performed a series of experiments on such isolated model peptides in which we have observed experimentally highly efficient charge transduction along the backbone of the peptide that was accompanied by ensuing long-range chemistry.[1–4] Although these peptides are of modest sizes, they represent an enormous number of microscopic states owing to their substantial number of degrees of freedom and as such predict very little reactivity in conventional models. Furthermore, experiments have revealed that such transduction was severely controlled and even switched on or off by the introduction of special amino acids in the chain. An atomic mechanism by which such long-range transduction occurs is of central interest. It would be desirable to understand the atomic mechanism of how transduction occurs efficiently in nonconjugated special organic systems to such distal sites through the use of atomic motions. Long-range transduction might have important model character for mechanisms of biological reactions.

Chemical reactions are traditionally located locally or proximally (the immediate neighborhood of the position of the excitation).[39,40] The customary mechanistic theory in chemistry explaining reactivity only relates to local excitations and an eventual bond rupture or other bond change as a result of energy being coupled and funneled into this bond by a statistical process. This lies at the heart of the transition-state formulation or statistical unimolecular theory of chemical kinetics[41] and has been the mainstay in one form or another of most theoretical interpretations of chemical kinetics since the first application of the Rice, Ramsperger, Kassel, and Marcus (RRKM) theory[42] by Rabinovitch and co-workers approximately 50 years ago.[43–45] These statistical processes typically couple to all the degrees of freedom of the reactant species. Such a model of coupling cannot be applied to long-range biological RC transduction as the number of eigenstates in such large systems and thus the associated phase space becomes astronomical even for peptides. Any local energy would be dissipated prior to the arrival at the distant site or would require astronomical time scales for the chemical reaction of systems involving more than two or three residues.[46]

In view of the fact that such distal processes have been proven experimentally for many biological systems, and that

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they occur with some efficiency, some other model must be sought to explain such processes. Levinthal[18,19] recognized this problem of protein folding[47] in which too many degrees of freedom would again prevent the process from occurring in any finite time scale. Many possible suggestions have been made to circumvent this problem, but the issue is still a subject of intense discussion.[48–50] These many degrees of freedom for peptides, however, do not couple at the low energies of about 200 cm^{-1} involved here. Thresholds for intramolecular vibrational redistribution (IVR) in kinetics are typically at least 1200 cm^{-1}[51] or 2200 cm^{-1}[52] and at low energies are even higher,[53,54] although the picture for very large molecules with soft modes may be different. Hence, the coupling suggested herein takes place prior to communication of all modes by IVR and thus in a much-reduced phase space.

First we want to review recent experimental data for model peptides. These data already provide strong evidence that such efficient action at a distance, even for an isolated molecule, exists, indicating this to be a pure molecular property. We present a model that attempts to encompass the new data that are now available and suggest special molecular conditions under which such distant actions can or cannot occur. Furthermore, we must explain the observed strong environmental influence on this process as the efficiency in water is much reduced.

A typical theory to understand conductivity in proteins might calculate the matrix elements for the coupling of the amino acids positioned as some averaged conformation. Such calculations were performed for a model peptide (Figure 1)

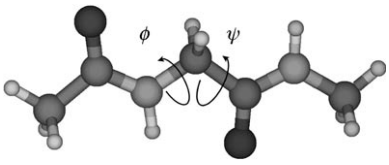

Figure 1. The model dipeptide used for the calculation of the interaction between two neighboring amino acids for charge transfer across a peptide backbone.

and show that the energetics between neighboring sites typically involves surmounting a barrier of some 0.4 eV (Figure 2). For biological systems at typical temperatures, this is a very large barrier to cross and is difficult to attain. This leads to some inefficiency in the process as was in fact observed in studies in aqueous media. It does not, however, explain the highly facile charge transport observed in the gas phase.

The Marcus theory shows that an ET rate is proportional to the product of the square of the electronic coupling constant and the Franck–Condon factor, which relies on the driving force and solvent reorganization energy. A distance-dependent factor was not at first contained in the Marcus theory. Later, the Marcus theory was extended[55] to include a distance-dependent factor $e^{-\beta(R-R_0)}$, where β is the distance-decay constant, R the distance between redox sites, and R_0 the distance of closest approach between donor and acceptor. A typical experimental β value for DNA is approximately

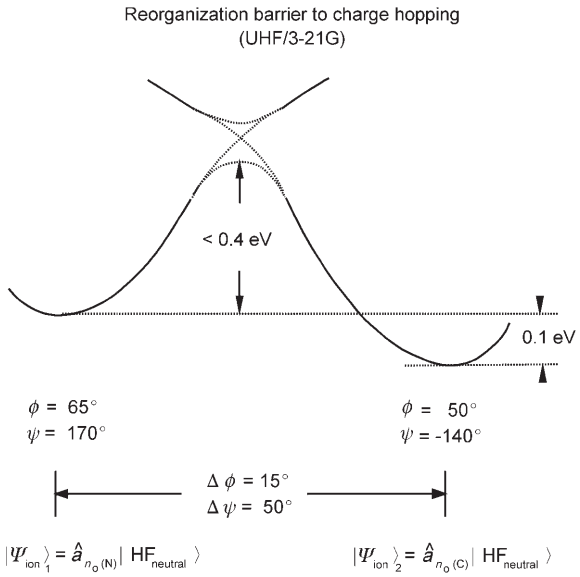

Figure 2. Lowering of the barrier for charge transport between neighboring amino acids (identical residues) as a function of the Ramachandran angles ψ and ϕ shown in Figure 1. The charge hops from ionic state 1 to ionic state 2 through a non-adiabatic transition. After diabatic transformation, the barrier for the charge transfer is about 0.4 eV. The charge transfer is actually dependent on the Ramachandran angles. Some more-detailed notations are shown in Figure 3. Ab initio calculation of the energy of the states is performed at the UHF/3-21G level. $|\psi_{ion}\rangle_1$ = electronic wavefunction with the charge located at the N side, $|\psi_{ion}\rangle_2$ = electronic wavefunction with the charge located on the C side.[6]

0.77 $Å^{-1}$;[56,57] for an α helix it is approximately 1.26 $Å^{-1}$, and for a β sheet it is about 1.00 $Å^{-1}$.[56,57] Theoretical investigation has been done by many groups such as Beratan, Onuchic, and co-workers by using a tunneling-pathway model to explore the distance-dependent factor.[58–86] A superexchange model based on nonadiabatic electron tunneling has also been proposed by many groups.[87–89] These models are often based on an averaged background chain structure.

In earlier work,[11] we suggested that the key may be that peptides have a very unique molecular property derived from the individual amino acid sites undergoing quite facile rotations over very large angles (the so-called Ramachandran angles) of the polypepide. These motions are characterized by a nearly flat potential energy surface inside the Ramachandran plot. This is particularly seen for the free molecule. We then observed the further surprising result that the energetics between typical model amino acid sites are not constant, but rather vary strongly with the Ramachandran angles of the two sites with respect to one another; hence the average behavior is not the behavior at the average angle. Baranov and Schlag[6] found that there is, in fact, a favored angle between two neighboring amino acids at which even the barrier between sites becomes negligible, as compared with the 0.4-eV barrier at the average angle. The motion can be seen in Figure 3 in which the ionic state is twisted until it reaches a near-degenerate state where the curves cross and the charge on that site returns to the ground state. From this point, it rotates

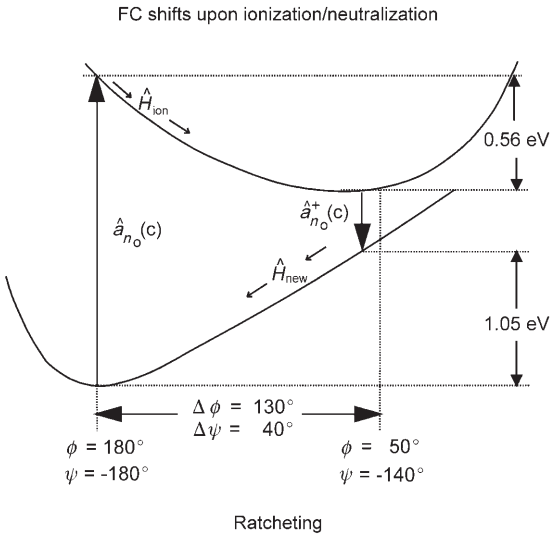

Figure 3. Potential surface for charge transport as a result of an angular twist between neighboring amino acids; note the high efficiency of transport when the neighboring CO groups are close together; FC = Franck–Condon, $\hat{H}_{ion}$ = ionic energy, $\hat{H}_{new}$ = neutral energy. The vertical lines represent the transition energies between the relevant electronic states.[6] The upper curve represents the ionic state and the lower curve the neutral state. The charge is excited from the ground state following a Franck–Condon transition. The twisted motion of the Ramachandran angles shifts the ionic state toward the curve crossing point and charge is transferred back to its ground neutral state. This process can be described as a type of ratcheting.

back to the equilibrium conformation. This again points to motion being an important condition for reactivity.[90]

Suppose we asked about the transduction between identical neighboring amino acid groups. Performing ab initio calculations for the charge being transported away from the α carbon atom, we find that one direction proceeds to the N side and the other direction proceeds to the C side. Our calculations show that there is a 0.4 eV discrepancy between the two directions, thus making the transport of charge at moderate energies quite inefficient.[6] Such poor couplings in amino acids are well known.[91] The calculations carried out on a simplified model by Baranov and Schlag showed that at the extreme deflection of the dihedral angle when two neighboring carbonyl groups are separated by only approximately 2.8 Å, this barrier becomes negligible, producing a type of hybrid state between the two residues.[6] Two amino acids could thus couple strongly, but only at the extremes of the Ramachandran angles. For strong coupling, we need to examine the extreme rotational deflection of the relative angles of rotation. Hence, in this sense we refer to this as a two-step model, first rotation and then charge transfer, that is, a bifunctional model. Detailed calculations on pentaglycine have recently confirmed that the probability of hopping of a charge is strongly correlated with the alignment of the CO groups in a full quantum mechanical calculation.[6,92]

We undertook molecular dynamics (MD) simulations of the motions of these angles to determine mean first-passage times for reaching the low-energy near-degenerate conformation by rotation of the Ramachandran angles. We then presumed charge transfer (CT) to the next-lower energy site to occur when the angles reach this critical conformation in which neighboring carbonyl groups are approximately 2.8 Å apart. The calculation employed a variant of the typical MD calculation. Usually in MD calculations, all the motions are initiated at one time, however, in our variant, we want to activate energy at a defined site, usually the C terminus, and observe a time evolution of this specific local excitation. For this we had to modify the code of the CHARMM 24 program[93,11] The time of interest is taken here as the first time for two neighboring CO groups at a common C_α atom of an individual amino acid in the peptide to rotate through their Ramachandran angles to a critical distance ($\approx$2.8 Å) between the oxygen atoms, a point at which we find the strong coupling leading to RC transduction[6,8] (see Figure 4)—the mean first-passage time. We suggested, in

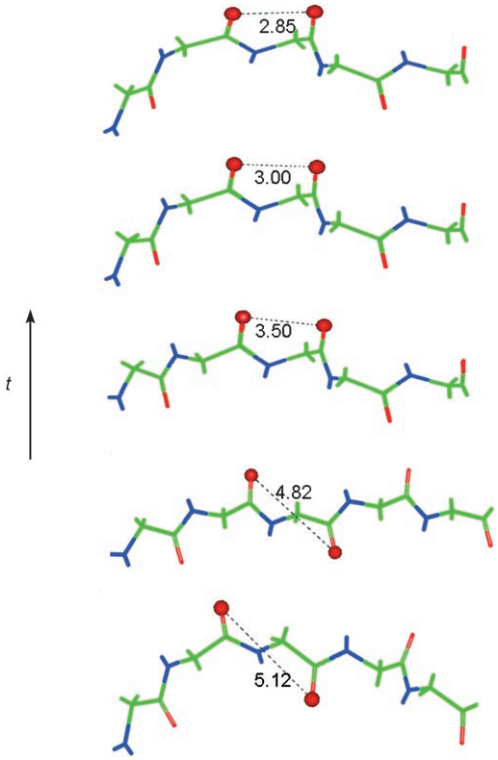

Figure 4. A snapshot of the firing process (reaching a critical distance and RC transfer) of Gly_3 (green C, $H_{(C)}$; blue N, $H_{(N)}$; red O). An MD simulation for the short peptide is done at 1667 K, that is, 150 meV.

first-order calculations, that other angles are inefficient for transduction. Hence, the key to this mechanism is that little transport occurs at typical angles of the peptide, but facile mechanical rotation of the peptide chain along the Ramachandran angles until it reaches a firing point results in highly efficient transfer of the charge at an energetically nearly degenerate state.

The interesting result from these MD calculations is that the mean first-passage time for peptide rotations of a single step for simple peptides occurs at a precise very fast time scale of approximately 100–150 fs and, as such, presents a basic

rapid subprocess for protein motion in general (Figure 5). Such a fast time scale at first appears surprising, but recent work by Hamm and co-workers in 2001 has experimentally directly determined a very similar time scale of 120 fs for a tripeptide.[61] This agreement of the experiment with our MD calculations is encouraging. This time scale may vary for more-complex systems but still appears to be much faster than typical IVR times. This was first shown for chemically reacting systems by Rabinovitch and co-workers to be approximately 1–2 ps[94,95] and is longer still at low excitation energies.[51] Although anharmonically coupled IVR can be as fast as 300 fs,[96] it is still slower than the rotations considered herein. This early and as yet weak coupling to the vibrations of the peptide may be another means for avoiding the large number of degrees of freedom, at least in segments of the peptide chain, that would lead to dissipation of the initial excitation. This is a unique case in which only a few degrees of freedom are coupled on the subpicosecond time scale. In such a greatly reduced phase space there is highly efficient motion with little energy dissipation. We suggest that such a highly efficient motion in peptides on the very short subpicosecond timescale constitutes an important part of the very early dynamics of protein motions that contribute to very long-range reactivity. Such a very fast time scale in a reduced phase space might be of interest for finding stable, even near-native conformations in subsections on the very early time scale of an evolving protein system. These conformations could interact with each other as sections or domains and subsequently induce protein macroscopic motions.

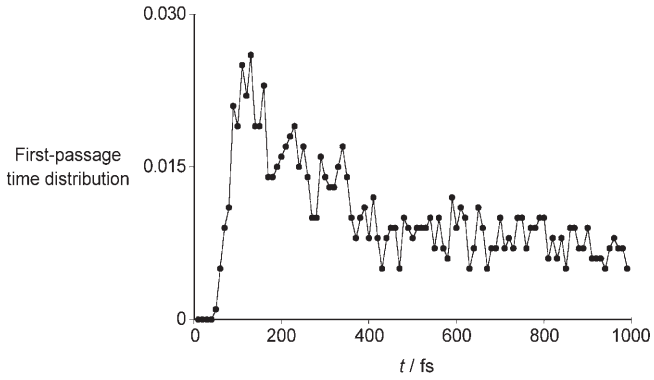

Figure 5. Typical mean first-passage-time distribution for reaching the firing position of the charge between neighboring residues at 100–150 fs. The sharp onset of the local heating is followed by a tail, which shows the thermal noise owing to the vibrational modes starting to set in. Here we adopt a short polypeptide chain of 10 Gly residues and perform a MD simulation after global heating of the molecule.[8] We then determine the first-passage-time distribution versus time of the collision of CO groups around the third C_α atom of the polypeptide chain.

This very rapid time scale for rotation in the peptide to the point of efficient coupling to the next amino acid is taken here as the mechanistic precursor for the efficient charge transmission. We suggest that it is the rotation to this critical conformation that makes for facile charge mobility. Conversely, we must then also conclude that if such transduction is impeded, there should be no charge transmission. In the rigid environment of the peptide, no transduction should occur by this mechanism.[97] This could lead to some interesting speculation on which environments might be detrimental for charge transport in biological systems (see Section 6).

2. Energetics

When residues are not identical, one must consider further energetic considerations because even the correct rotation must find an energetically favorable path for transduction. The simplest view might be to consider each residue to have a characteristic energy, which is only loosely coupled to the other residues. This weak coupling regime appears reasonable as a first-order approach. To estimate these individual energies, we might consider the ionization energies of each of the amino acids. This, however, cannot be exact even for the isolated molecule in the gas phase as each residue is attached to a chemical environment that is quite different from that of an isolated amino acid. In particular, the neighboring groups are typically not acid groups, etc., thus even simple-model calculations have to consider a neutralized environment (Figure 6).

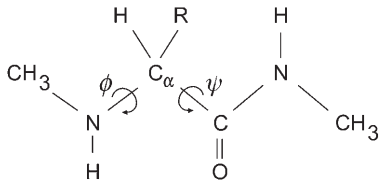

Figure 6. Model compound used for calculation of the IP of an amino acid residue within a peptide. ψ and ϕ are the Ramachandran angles.

Furthermore, one has to consider that the ionization of a particular residue in the peptide environment is unlike the ionization of the bare amino acid in which the electron is removed to an infinite distance. The down-chain ionization proceeds to CT states in which the charge is not removed to infinity. These states are typically around 1 eV below the standard ionization energies for the bare amino acids. Nevertheless, the model described here is of transport being substantially controlled by varying local energies, which appears to broadly explain our results. This presents a useful first-order model for charge transport in peptide ions and could also have possible applications for large systems observed, for example, in a mass spectrometer.

Hence, we now have a bifunctional model that describes the CT along the polypeptide chain from the C terminus to the N terminus. On each C_α atom there is an N side as well as a C side. The torsion angles around the C_α hinge are confined in certain domains inside the phase space (ψ, ϕ) that are depicted within the Ramachandran plot. Different pairs of amino acids for the α helix and β sheet occupy different subregions inside the Ramachandran plot. When the charge is locally excited from the C side of one C_α atom, it jumps to the nearby N side of the C_α atom through the adjacent O–O atom collision, which is located near the C_α hinge. This jumping

process itself may take less than 10 fs. However, the torsion motions of the carbonyl groups for the ψ and ϕ angles are on a 150-fs time scale, although these times vary according to the various starting conformations and are thus not correlated with one another. Hence, these rotational motions are taken to be a virtual Brownian particle moving inside the Ramachandran plot domain with nearly free motion.[8] As the O–O atoms collide at a certain distance, for example, 2.8 Å, and the virtual Brownian particle reaches a fixed point in the Ramachandran plot domain, a gate for the Brownian particle to escape is defined. This gate condition is reached by the Brownian rotational motion. This creates the condition of a quasidegenerate state that then leads to the occurrence of rapid charge transfer. This charge transfer occurs as soon as the CO distance is reached and is essentially electronically instantaneous on the time scale of the Brownian motion. Hence, the sequential propagation of the charge along a polypeptide chain is a sequential combination of 1) escape of the Brownian particle and 2) jumping of the charge.

As an example comparing the experimental results for Leu-Leu-Leu-Trp with Gly-Gly-Gly-Trp, we find that the former conducts charge completely from an ionized Trp to the N-terminal Leu, whereas in the latter case, no transport at all is observed.[1,2] This can be attributed to a small difference in CT energies between Leu and Gly, the former being about 0.2 eV lower. As the energy of Trp lies in between, replacing Trp with Tyr in both cases above enables complete transport for both systems as Tyr lies at a higher energy. These are but two examples of a considerable number of peptides that have been investigated.[1,2]

The facile transport of charge then also results in transduction in the form of a chemical reaction proceeding at a site quite distant from the point of ionization. In other words, both transduction of reactivity and charge are taking place. This so-called RC transduction is the result of two conditions: first, that ultrafast motion of the dihedral angles to the state where charge transfer occurs is not impeded and, second, that the energetics are favorable at that site. This state is now quite different from typical proximal chemical kinetics. The excitation in our system is at one end and the chemical reaction proceeds at the other end as a result of charge transport.

One might ask where this leaves normal unimolecular processes in peptides? The answer is, of course, that such processes are still operative, but are on a much longer time scale. This was shown for smaller peptides in the gas phase by Lifshitz and co-workers,[46,98] who have elegantly demonstrated that Leu-Tyr and Leu-Leu-Tyr are both capable of undergoing normal unimolecular chemical reactions, after IVR, albeit far more slowly. The rate constants are then in the range predicted by the RRKM theory.[42] Extrapolating these times to our larger systems above would lead to reaction times in the range of 0.1 s, which would not be observable in our apparatus—indeed it would compete with radiative decay. We propose that we are dealing with two different regimes with a very much reduced phase space at short times and a normal larger phase space after IVR. For peptides with low energies of excitation, such as is typical for biological systems, we have an ultrafast RC transduction occurring to a low-energy residue down the chain before IVR occurs.[53,99] This process proceeds with utmost efficiency owing to the very small phase space involved.

Suppose we now consider an energetic surface in the peptide that is not even but contains some substantial energetic contours. Consider, as an example, a mixed case of Leu-Gly-Leu-Trp. We again observe extremely efficient RC transduction from the C terminus, but interestingly only to the Gly residue. This high-energy local site now serves as a bottleneck to RC transduction. Note that although the N terminus is still the lowest energy in the chain as before, this lowest energy site is not observed here as the final site for RC transduction. Hence, this mechanism does not seek out the lowest energy final state as in standard statistical kinetics; the bottleneck often leads to stoppage in the transduction process. The bottleneck experiment also demonstrates that RC transduction proceeds along the chain of the peptide and not by some other process.

This activity at a distance, which involves such a greatly reduced phase space, is a process that may be quite unique to polypeptides and is made possible by the ultrafast large-amplitude motions of the Ramachandran angles outstripping the IVR as well as local electron sites of the individual amino acid residues along the chain.

As an aside, if we apply this thinking to some of the photoionization data from the mass spectrometry of peptides, we find that here too reactivity in the ion is transported over substantial distances to produce the many fragments observed. A statistical process would not have enough time to produce such extensive fragmentation of large ions located so far from the site of excitation on the time scale of a typical mass spectrometer.[100]

3. Calculations

Although many energizing steps for peptides, such as redox potentials, could be envisaged, here we considered the simple process of direct photoionization, and this only at the C-terminal end of the peptide. For the determination of the local energies, we proceeded to determine the ionization energies of the 20 natural amino acids. Only fourteen values have experimentally been determined and for the remaining six amino acids, we proceeded to an ab initio calculation as seen in Figure 7. For these calculations, we employed a DFT program[101] that uses the B3LYP level of theory and a 6-31 + G* basis set. It was determined that Gly is indeed 0.2 eV above Leu, as is postulated by the model.

The initially photoionized amino acid, here Trp, is a special case as the ionization at this site removes the electron to an infinite distance, whereas all further local ionizations are CT states. This was already demonstrated with the dimer Leu-Trp, in which the charge travels readily to the Leu. Furthermore, this transfer is stable and apparently irreversible even after the cessation of the laser and on a 10-ms time scale.[46]

The ionization of Trp is uniquely evoked by the optical transition in the benzene moiety at 260 nm and is close to the ionization energy of the bare amino acid. Such ionization energy is the energy required to place an initial positive charge on the molecule and at the same time remove the

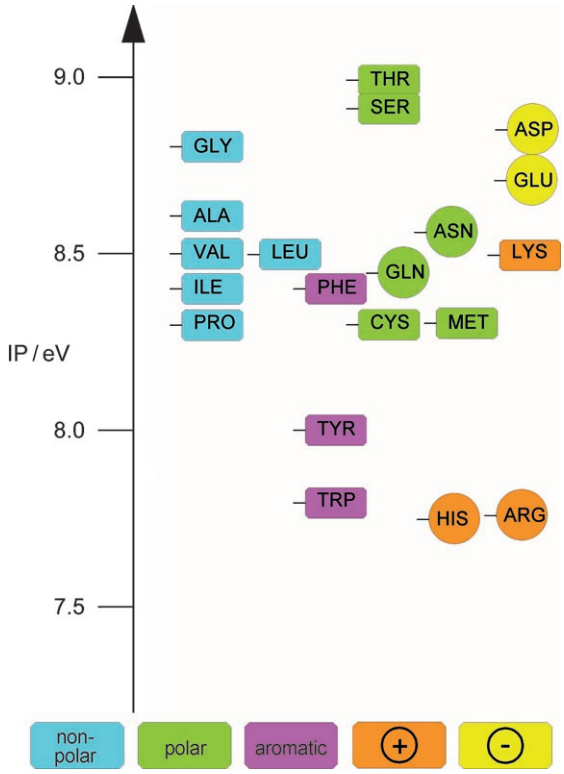

Figure 7. Ionization potentials of amino acids. □ Experimental value, ○ calculated value (B3LYP/G-31 + G*) with optimized geometry. We labeled the polar, aromatic, cation, and anion species along the *x* axis.

electron to infinity. If we consider this to simply be the first-order ionization energy of the bare amino acid, then the energetics for these processes are given in Figure 7.

For the Leu-Trp dimer, this would place the charge as Leu-Trp$^+$. We could now imagine the charge subsequently hopping from this first site to the second site at the Leu. The energy expended is different for the removal of the electron from these two sites. In the latter case, the removal of this electron does not require being moved to infinity, but rather only back to the first site. From a simple Coulomb model, one can estimate the energy saving to be approximately 1 eV; any further sites in the chain after the first site are then all CT sites. This means that we are now in the Leu$^+$-Trp state.

If we now perform a DFT calculation on the Leu$_2$-Trp tripeptide, we find that the vertical ionization energy is near 7.8 eV, whereas the adiabatic energy is near 7.0 eV. Hence, for purposes of modeling, we need to calculate prototypical ionization energies of the amino acids embedded in a pseudo peptide environment (Figure 6). These energies are indeed all about 1 eV lower than those of the bare amino acids. This simply means that the lowest energy state is an adiabatic state that, although lower in energy, is not readily accessible by the vertical process.

As a rough simplification of the above argument, we take the vertical ionization energy from the bare amino acid for the first site, but the CT energies for all successive sites. To show the application, we consider a dimer of Leu and Trp (LT), that is, let us suppose that we choose the laser wavelength and intensity in such a way that Trp in LT is ionized [Eq. (1)].

$$\mathrm{LT} \xrightarrow{\hbar\omega} \mathrm{LT}^+(v) \qquad (1)$$

$\mathrm{LT}^+(v)$ represents the vertical ionization state, which is determined by the Franck–Condon transition. From Figure 7, we can estimate that the minimum energy will be around 7.8 eV for $\mathrm{LT}^+(v)$, whereas from Figure 8, we can see that the

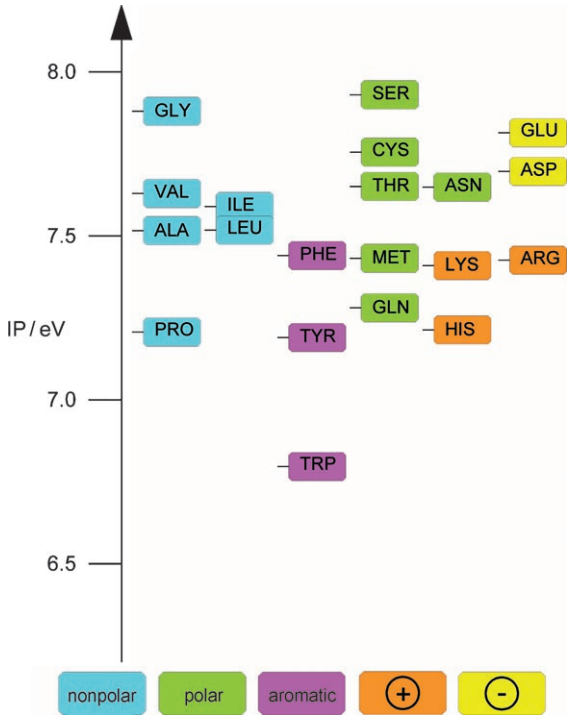

Figure 8. Ionization potential of amino acid residues within a peptide calculated from the model compound in which the amino acid is in a pseudo peptide environment. The basis sets and the species are the same as for Figure 7.

adiabatic ionization energy is only 6.8 eV for $\mathrm{LT}^+(a)$ (a = adiabatic). As $\mathrm{LT}^+(a)$ only has an energy of 7.5 eV, the CT shown in Equation (2) can occur.

$$\mathrm{LT}^+(v) \rightarrow \mathrm{LT}^+(a) \qquad (2)$$

To calculate these adiabatic energies, we put the amino acid in a typical environment (Figure 6). In this way, we calculated a set of adiabatic energies for the 20 amino acids as is shown in Figure 8. Note that these energies are all typically about 1 eV less than the vertical ionization energies in Figure 7. If we now take the ionization potential (IP) of Trp from Figure 7 and compare it with the adiabatic energies of Figure 8, we see that the value for Trp lies just between the values of Gly and Leu. This predicts charge transport to Leu, but not to Gly, which conforms with our observation. The absorption to the adiabatic state of Trp (Figure 8) is the lowest energy state of the system, but has simply too little oscillator strength for a typical absorption experiment. Nevertheless, it is predicted that some small cross section will also reach this state of Trp in the peptide and this is then the lowest state of the overall system. For this reason, there will be no CT possible from this state.

It is important, as noted above, that the electron transfer (ET) in Equation (2) above takes place before IVR. Thus, the ET in Equation (2) should be described by the single-level ET rate constant W_{iv}. For the case in which ET is much slower than IVR and thus vibrational equilibrium is established rather than ET, we have the so-called microcanonical ET rate constant $W(E)$ [Eq. (3)], where E represents the excitation energy of the system and $P_{iv}(E)$ denotes the microcanonical distribution [Eq. (4)]. $\delta(E-E_{iv})$ is the delta function and $\rho_i(E)$ represents the density of states with energy E [Eq. (5)]. Equation (3) indicates that IVR spreads the distribution of excitation energy over all the vibrational states of equal energy. Each state is equally probable and is described by $P_{iv}(E)$. The observed rate $W(E)$ now has to include all the possible W_{iv} weighted by their $P_{iv}(E)$. In other words, IVR will decrease the ET rate.

$$W(E) = \sum_{v} P_{iv}(E) W_{iv} \tag{3}$$

$$P_{iv}(E) = \frac{\delta(E-E_{iv})}{\rho_i(E)} \tag{4}$$

$$\rho_i(E) = \sum_{v} \delta(E-E_{iv}) \tag{5}$$

It should be mentioned that although these are general conclusions for RC transduction, there will be subtle differences when the appropriate excitation energy is derived from processes other than the photoionization process considered here. Notably, after transfer of the charge to the N-terminal Leu, the reverse transfer back to the adiabatic state of the C-terminal Trp is too slow and hence not favored as the preferred process is now local dissociation.

The reverse charged state, which lies approximately 1 eV below the state of the Leu^+-Trp state results in a considerable energy gap. As these are all radiationless processes within electronic transitions, we can deduce from the theory of radiationless transitions that the lifetime of the Leu^+-Trp state approximately increases with the energy gap[65,102] according to the Equation (6), where ω_{if} denotes the energy gap, s the Huang–Rhys factor, T_{fi} the electronic coupling matrix element, ω the excitation frequency, and $\bar{\omega}$ the average frequency. A gap of 1 eV leads to a lifetime of some 100 ns as a lower limit. On this time scale, the Leu^+-Trp state has time to fragment. Experiments on Leu-Tyr show the charge to remain on the Leu site in the dimer.[46]

$$W_{i0} = \frac{|T_{fi}|^2}{\hbar^2}\left(\frac{2\pi}{\omega_{if}\omega}\right)^{1/2} \exp\left[-\frac{\omega_{if}}{\omega}\left(\ln\frac{\omega_{if}}{s\bar{\omega}}-1\right)-s\right] \tag{6}$$

A more detailed DFT computation of Leu_2-Trp system exhibits a torsion-angle-dependent CT process that supports our bifunctional model. In Figure 9, the torsion angle between the Leu2–CO group and the Leu1–CO group is fixed. Changing the torsion angle between the Leu2–CO group and the Trp–CO group induces a variation in the charge distribution. The medium residue acts as a switch. Only at a certain torsion angle is the charge allowed to transfer.

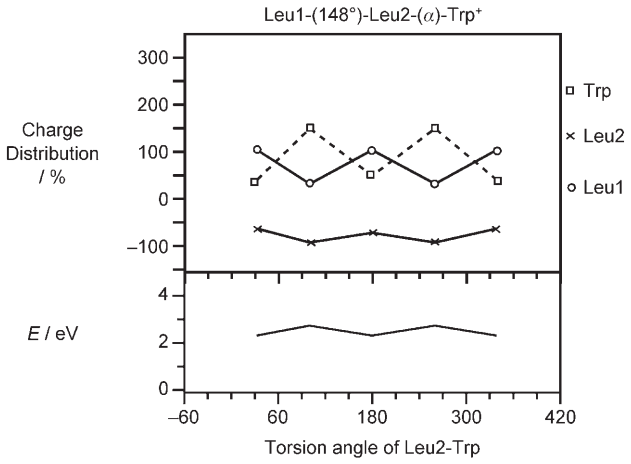

Figure 9. Charge distribution in the tripeptide Leu-Leu-Trp as a function of the torsion angle α(Leu 2-Trp). The lower portion of the figure shows the relative energy of the system. Here we calculate the Mulliken population of the Leu-Leu-Trp cation. The angle between Leu 1 CO group and the Leu 2 CO group is fixed at 148°. Moreover, the angle between the Leu 2 CO group and the Trp CO group is plotted for a variable α. These angles are fixed during the energy minimization of the peptide geometry. Note the alternation in the charge distribution as the angle is twisted. This shows the charge transfer based on the torsional motion of the Ramachandran angles.

A gas-phase DFT computation encourages us to propose a more universal model that contains intrinsic polypeptide back-bond dynamics. This model should cover gas- and liquid-phase behavior. The bifunctional model has the advantages that it not only covers bond dynamics but also the environment effects, such as hindrances to bond dynamics. On such a very short time scale, water becomes a severe obstruction and hence leads to a severe reduction in CT efficiency. Interestingly this is not the case for lipids; preliminary calculations for a lipid environment yielded much higher efficiencies.[103]

4. MD Simulation Method and Some Results of Mean First-Passage-Time Distribution

Many transmembrane proteins carry the CT process with an energy gradient in the order of 0.4 eV. This energy requires a substantial driving force for the propagation of charge from one amino acid to its adjacent amino acid. In our bifunctional model, this energy is provided to the carbonyl group of the N side of the C_α atom, which drives the torsional motion and thus allows the virtual Brownian particle to move inside the Ramachandran plot. Only at special locations is this energy barrier reduced to near zero.

Our bifunctional model is a wait and release process. The main physics here is the injection of the driving energy into the rotation degrees of freedom, that is, to drive the rotational motion of the carbonyl group before the vibration motion of the background polypeptide chain sets in. The O–O atoms collision should be effective. As one knows, the vibrational motion involved in the process is dissipated by IVR after several picoseconds. Hence, after several sequential O–O

collisions, for example, approximately 5 collisions, the rotational energy here too is dissipated.

As we mentioned in the Section 2, the firing and escaping times of the Brownian particle are on different time scales. The background chain motion is the rate-determining step. This bifunctional model is computed by a classical MD simulation method. We initiate the process by rotating the $\vec{C_\alpha C}$ axis (Figure 10) with a driving energy of approximately 150 meV. The basic question we ask is what the efficiency is for a successful O–O collision. Furthermore, we now suggest the following definition for efficiency.

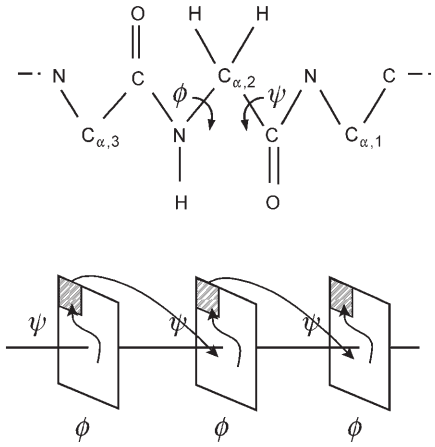

Figure 10. Hopping mechanism as a function of the dihedral angles of the residues. We can imagine that the charge is first transferred starting from the C terminus and stays at the C side of the C_α atom. The C-side carbonyl group waits for the torsion motion until the carbonyl group from the N side approaches a given angle upon which the charge starts to jump. This process is depicted in the lower portion of the figure. For the individual amino acids, the Ramachandran plot is shown in the lower portion of the figure. The torsion motion of each amino acid is similar to a virtual Brownian particle moving in a phase space of the Ramachandran angles. Once this Brownian particle reaches the gate part (shaded area), transfer of the charge occurs at this place and time. This figure shows a sequential escaping process for our bifunctional model.

Efficiency = successful conformations/total conformations.

We performed the O–O collision simulation with approximately 3000 conformations and found that there are less successful conformations with the critical distance than there are total conformations. The ratio suggests an efficiency.

With this efficiency, we can produce a relationship with the distance-decaying factor through a simple argument. First, we define the rate constant for CT, k_t, and a rate constant for loss to the bath k_b. The fraction of charge that survives after n linked amino acids is described in Equation (7).

$$a^n = |\frac{k_t}{k_t + k_b}|^n \tag{7}$$

This power form can be transformed to an exponential form as $e^{-\beta n}$, where β is typically 0.8–1.4 $Å^{-1}$. As each amino acid length is approximately 3.7 Å, the distance-dependent decaying factor is expressed as $e^{-\beta R} = e^{-3.7\beta n}$. We can apply $a^n = e^{n \ln a}$, where $a = k_t/(k_t + k_b)$, to obtain Equation (8).

$$\beta = -\frac{\ln a}{3.7} \tag{8}$$

As we can see from Equation (8), a is the efficiency and we obtain the distance-decaying factor β value. Although the β value is small, the CT efficiency is large. A very efficient transfer occurs in DNA where the β value is approximately 0.2 $Å^{-1}$. The ratio of k_b/k_t is 1:1. Now both processes are equally fast. For $\beta = 1.4$ $Å^{-1}$, one obtains $k_b/k_t = 177$. This means that less than 1% CT at each step corresponds to this typical value of β.

In our simulation, a single-site modified CHARMM 24 program[93] is introduced, implementing the local-heating method described in Section 1 (Figure 5). For the native initial conformation, the rotational direction affects the mean free path and thus the mean first-passage time. The model provides instantaneous driving energy to a single local site and this energy is transferred along the polypeptide chain down to the next nearby C_α hinge. Subsequently, MD simulations covering the CT from an isolated gas phase to a hydrated system were performed.

5. Isolated Systems

Our mass spectrum of short polypeptide chains $(Leu)_n$-Trp ($n = 1$–4) shows only the peak of the molecular ion and the N termini with total efficiency of approximately 0.47.[1–3] There are no other bond breaks at the intermediate residues. Hence the efficiency 0.47 is for the first Leu residue and the other residues have unit efficiency of almost one. This unit efficiency warrants the conclusion that the energy is transferred with high efficiency. In Figure 10, as there is no energy dissipation, the rotation energy is totally transferred to the next nearby carbonyl group owing to conservation of momentum. By using the local-heating method described in this work, the remaining phonons are seen not to influence the subsequent motion leading to the charge-transport process. The results in Table 1 confirm a near unit efficiency and a ballistic motion in the bifunctional model.

Table 1: Efficiency at each site away from the local-heating site.[a]

Residue number[b]	Efficiency	Residue number[b]	Efficiency
2	0.18	12	0.90
3	0.0033	13	0.46
4	0.41	14	0.22
5	0.29	15	0.22
6	0.25	16	0.084
7	0.40	17	0.11
8	0.69	18	0.013
9	0.85	19	0.025
10	1.0	20	0.26
11	0.78		

[a] Position of the local-heating site = Val_{10}. Local-heating temperature = 2667 K. Background temperature = 300 K. [b] Mb_{20} = (N terminus)Glu 1-Asp 2-Leu 3-Lysn 4-Lysn 5-Hsd 6-Gly 7-Val 8-Thr 9-Val 10-Leu 11-Thr 12-Ala 13-Leu 14-Gly 15-Ala 16-Ile 17-Leu 18-Lysn 19-Lysn 20(C terminus). Lysn = neutral lysine.

In Table 1, we excite the C side of the C_α hinge at the Val_{10} site of a polypeptide chain 20 mer Mb_{20} with an excitation energy of 150 meV. This 20 mer was cut from an α helix of the myoglobin molecule. The efficiency is high even at the fifth site away from the local-heating site (residue 15). At residue 16, the efficiency decreases abruptly. At Val_{10} (the local-heating site), the mean first-passage (mfp) peak is very strong (Figure 11).

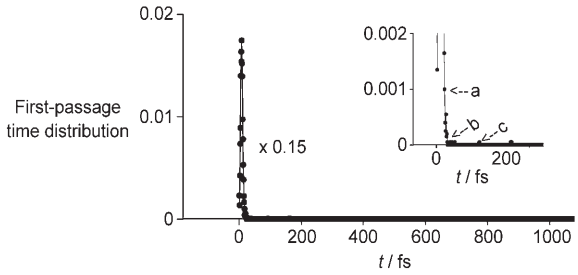

Figure 11. Mean first-passage-time distribution versus time at the local-heating site, Val 10, for the polypeptide 20 mer Mb_{20}. This is the same peptide chain as in Table 1. Here, we show a strong peak corresponding to the local-heating site. As there are two directions for the rotation of the CO group, one is the forward motion from low amino acid number to higher amino acid number and the other is the backward rotation from higher amino acid number to lower amino acid number. The inset shows three small peaks that correspond to the forward motion immediate after locally heated (a), the second peak (b) denotes a backward motion, and the third peak (c) is a recursive motion from the C side towards the N side.

6. The Water Environment

The described model now appears to be quite successful in interpreting the highly efficient transport of a charge that is placed by ionization on the C terminus of the isolated peptide molecule and travels down the chain from there. How does this compare with the observed smaller conductivity in the water medium? In such experiments, the charge is typically introduced via the end of a redox donor–acceptor complex[26,35,36] and removed at the other end. The charge decays exponentially with distance, in fact, decreases in efficiency by approximately two orders of magnitude. Considering the great importance of water as a medium for biological processes, this must be addressed for our case as well.

The conductivity of proteins in a water medium has been measured by a number of groups and is characterized by the decay of the charge with distance.[35,36] This is given as an exponential function [Eq. (9)], where β is typically observed to be 1 $Å^{-1}$ (Y = yield, A = molecular parameter, $R = n$ times spacing between amino acids, n = number of amino acids). The implication is that the signal/charge decays to 1/e after a distance of only 1 Å. If we now cast this into a hopping model between amino acid sites, this decay translates into a hopping efficiency of 3% for a typical spacing of $R = 3.7$ $Å^{-1}$ of amino acids, as in angiotensine. This implies that only 3% of the energy is transported to the next site for each hopping, clearly not a high efficiency, particularly when compared with the near 100% efficiency in the isolated molecule observed in the gas phase. It is of some interest to ask to what extent the isolated molecule of our studies would be influenced by the medium. Again MD calculations were performed as in the gas phase, but instead in a medium of approximately 500 individual water molecules. The water environment was observed to facilitate protein mobility by softening the hydrogen-bond network,[12] however, water was found to produce a tight cavity around the peptide. Such a cavity is too sluggish to respond to the ultra-fast angular motions of the peptide. Water thus tends to make a hydrophobic cage. This creates a barrel around the peptide structure with an opening of only about 6 Å (Figure 12).

$$Y = A\, e^{-\beta R} \tag{9}$$

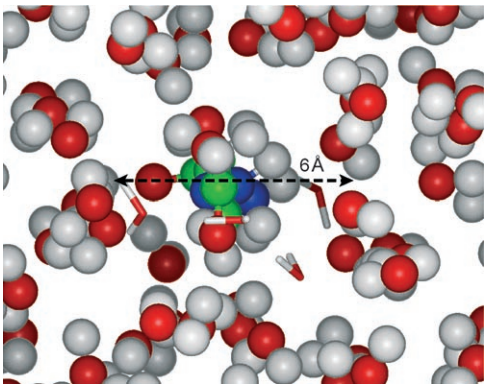

Figure 12. Water barrel around a peptide Gly_3 with an inner diameter of about 6 Å. Here we clearly show a water cage wrapped around the peptide chain (red O, white H, blue N, green C). These water molecules surround the peptide chain and prohibit the rotation motion of the Ramachandran angles. This lowered the charge transfer efficiency. Note that the solvent-dynamics effect is present after about 100 fs.

Performing MD calculations on our simple peptides in water as a medium, one still observes a similar mean first-passage time of approximately 150 fs. This is indeed surprising because it states that the constraint of water, though severe, does not significantly alter the mean first-passage time of the Ramachandran rotations in the peptide. (Figure 13) However, the observation of the MD results in Figure 13 clearly indicates considerable noise in the many trajectories studied. This is due to the fact that even though the time scale is not changed, only very few of the many trajectories are successful in producing the large amplitude required for charge transport to the next site. Thus the first-passage time to rotation is hardly affected, but instead the efficiency falls off drastically in water.

We can attempt to use the MD program to estimate the number of successful hits compared with the number of trajectories at the start of each run. Here, we observe that only about 2.8% of the initial trajectories actually undergo a "collision", which explains the noise in Figure 13. Apparently, the hydrophobic barrel observed in the MD structure interferes with the large-amplitude internal rotations of the

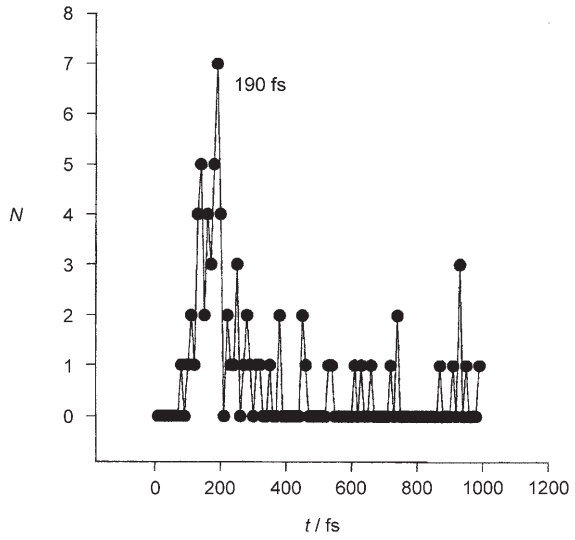

Figure 13. Typical distribution of mean first-passage times for firing between adjacent residues for the water medium for the example of a Gly_3 peptide chain dissolved in water; N = number of O–O distances up to 3 Å. The peak position is shifted to longer time values owing to the water cage prohibited motion. Note that the mean first-passage time is similar to the gas-phase value but with greatly reduced efficiency.

Ramachandran motions. This eventually results in IVR dissipation for most initial trajectories but does not effect the timing. Only a few trajectories survive to the next site in these MD calculations. This produces an overall efficiency here that is in the same range as previous work for charge transport in water. The inefficiency of charge transport in water is, however, the direct result of the rotational motions of the dihedral angles being impeded by a 6-Å hydrophobic barrel of assembled water on the outside of the peptide. Again it should be noted that the motions of the dihedral angle are in this subpicosecond regime: on this time scale the water molecules act as a rigid water barrel. The water movements are on a longer time scale and simply act rigidly on the subpicosecond time scale of the dihedral motions.

Our results indicate that water, although normally helpful, can be a problematic environment for charge transmission here. For other processes in protein dynamics, the water environment is clearly helpful, even essential as it strongly influences the H-bonding network and its dynamics.[15] However, for the very rapid RC transduction in water, the theoretically predicted value for transmission from site to site is 3%. Interestingly, this is in the range of other data where the value of β is around 1 $Å^{-1}$, which is a well-known experimental result.[35,36] The work here constitutes an alternate explanation for the extreme drop in RC transduction efficiency in water as observed when compared with our highly efficient gas-phase results. Biological processes of conductivity may depend on more-protected and friendly environments, like membranes. RC transduction may be better realized in a more flexible environment, as, for example, found in lipids.[103]

7. Secondary Structure

If we translate experimental β values into efficiencies, the β sheet is about three times more efficient than the α helix. Interestingly though, the efficiencies are different for the native α helix when compared with the β sheet. Only very small motions are needed in the Ramachandran plot to lead to charge transport. This is facilitated by the close proximity of the groups in the α helix to the firing position.

To explain the high efficiency of the β sheet, we have studied the first-passage-time distribution of rigid β-sheet structures in azurin. We found that the solvated β sheet has a weaker hydrogen bond than an isolated one. Solvation breaks the strong interaction between chains inside the β sheet and those bound through the H bond observed in the isolated system. The β value obtained through our local-heating method is 1.3 $Å^{-1}$, which is in exact agreement with the experimental results for azurin.[31]

8. Conclusion

We reviewed experiments together with a theoretical model for action at a distance, in which excitation takes place at one end of the peptide and the charge is transferred intact to react at a distal point at the other end of the peptide or to the point of blockage in the chain. The process makes use of very fast facile molecular motions that are unique to peptides. In fact, such structures could even lead to an interesting new class of electronic devices.[63,104–106]

We suggest that one of the principal motions for peptides reacting and transferring charge on a subpicosecond ultrafast time scale are the dihedral motions between neighboring amino acid sites. These unique motions could be of fundamental importance for the very early processes in protein dynamics. On this time scale, vibrational coupling has not yet taken place so that such long-range dynamics proceeds with atypical facility as a result of a greatly reduced phase space. Hence, the vibrational couplings are highly efficient and not very dissipative. The very rapid large-amplitude dihedral motions between the amino acid sites are a result of negligible rotational potential-energy barriers; this is a special feature of the motions of the Ramachandran angles in proteins. At the external positions of such large-amplitude motions, the coupling of neighboring sites becomes very efficient. In this case, the distorted structure is energetically preferred, much as is in an sp^3 hybrid. We postulate that it is this external position, and not the average position, that is responsible for RC transduction as the barriers at the average intersite angles are too high for efficient transport.

Furthermore, of course, the energy profile along the peptide chain must be favorable. For the purpose of a model, we suggest, as a first approximation, an interaction of locally independent sites with their own inherent energies. These can be determined with an approximate model and are tabulated herein (Table 1). We introduce the energy along with the charge at the C terminus of the peptide, as seen in Figure 7, and then transfers the energy to CT states along the chain (Figure 8). Additionally, other types of charge and energy

introduction, such as in a redox system, could also be envisaged here and could also produce the modest energy of 150 meV required.

Furthermore, we suggest that environmental factors can seriously interfere with these large-amplitude motions and that this interference can lead to a gross reduction in charge and reactivity transport; However, this apparently does not significantly affect the mean first-passage time of the fundamental motions even though water is present. Bulk water is interestingly not seen as a good medium for this very fast process even though it is essential for other slower processes. It is predicted in this model to reduce transport efficiency by about two orders of magnitude, a result that now brings the calculations of this highly efficient gas-phase model in line with other experimental observations in water. This model predicts quite well a change in signal transport over approximately two orders of magnitude, depending on the environment.

The converse conclusion is also formed, that a peptide, which cannot rotate, cannot produce a good RC transduction—any such transduction in a nonflexible medium will become very inefficient. One might then ask the question if there exists an environment that permits facile charge transport in peptides. Recent work appears to show that lipids constitute such an environment.[103] A reduction of signal/charge transport in proteins in a strongly constraining biological environment might lead to the quite important suppression of biological functions.

The model presented herein, together with the special MD calculations, describes an integrated overall picture for the long-range chemistry of simple peptides, a subject of some interest for any understanding of far-range reactions in which conventional models of reaction kinetics appear to be unsuccessful. The basic, extremely rapid RC transduction can be seen in experiments that determine the isolated molecular yield[1,3] and its time scale[107] and also in the femtosecond motions of simple peptides. The model explains the highly efficient results observed in the isolated gas phase as well as the highly reduced efficiency in water where the efficiency values obtained were similar to those seen for electron transport of peptides in water. This is a first-order model of RC transduction leading to transport of charge and reactivity over long distances in peptides, a process commonly observed in biological systems. Such a process can not be immediately reconciled with small-molecule rate theories.

9. Epilogue

One could ask the question if this rapid charge transport might manifest itself in further protein motions. As charges are important in ribosomal proteins, one might suppose that this facilitation takes place here as well. The presence of the charge would thus facilitate the subpicosecond energy transfer in a domain of limited size to some binding point, such as a hydrogen bond. The adjustment of equilibrium within this domain would thus be very rapid up to this first possible hydrogen bond. This process would be favored for a very small number of residues, such as an α helix; thus, the formation of the α helix is preferred, even as a precursor to further motions and larger domains with new hydrogen bonds. The hydrogen bond is known not to be stationary, but opens up with a gate time of some 10 ps,[15] which then permits coupling to the next domain and in turn allows relaxation to a new state. Hence, we have a system of domains of varying size that are in internal equilibrium and couple at random intervals through 10-ps gates that open and close. These domains can then undergo large-amplitude structural rearrangements. The optimal long-range motions of such domains are much more facile than the motions of all individual atoms,[90,108–111] which leads to rapid equilibria of local structures that then are incorporated into the slower skeletal motions.

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