Conformational influence on the hopping conductivity in pig insulin

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The ac conductivity of pig insulin has been reported previously [Y.-J. Ye and J. Ladik, Phys. Rev. B **48**, 5120 (1993); Int. J. Quantum Chem. **52**, 491 (1994)]. Now we have calculated in the *ab initio* scheme using Clementi's minimal basis set and the random-walk theory of Lax and co-workers the ac conductivity in another conformation that occurs in the same crystal. The results confirm the conclusions of the previous papers, that is, native proteins can be good amorphous semiconductors if they are doped. The comparison of the results of the two conformations of pig insulin shows that the ac conductivity changes two orders of magnitude in the frequency range that corresponds to the time period of the elementary steps of chemical reactions ($\omega > 10^{10}$ sec^{-1}) when the three-dimensional structure changes. However, it does not change significantly in the low-frequency range ($\omega < 10^{4}$ sec^{-1}). The conclusion is that in the high-frequency range insulin would change both the ac conductivity of itself and its receptor if it binds to a receptor. Thus insulin might change the electron transport in the receptor when it expresses its biological activity.

I. INTRODUCTION

In the preceding papers,[1,2] we have reported the hopping conductivity of pig insulin and have drawn the conclusion that polypeptide chains could be good amorphous semiconductors along their main chains if they are doped. The calculations were done on one of the molecules in a dimer, which is the asymmetric unit in the single crystal. It was necessary to compute also the hopping conductivity of another molecule in the same dimer because it has a somewhat different three-dimensional structure. Chothia *et al.*[3] and Chang *et al.*[4] compared the differences between the two molecules in the dimer of pig insulin, respectively. They have come to the conclusions that the conformations of the two molecules are similar to each other in their main chains, and almost all side chains changed only slightly; but, in the case of a few side chains, the conformations were altered significantly.

In this paper, we shall report the hopping conductivity of the other molecule in the dimer using the same approximations and methods as in the preceding papers.[1,2,5] The results show that conformational changes strongly influence the hopping conductivity of pig insulin in the high-frequency range, especially in the frequency range that corresponds to the time period of elementary chemical reactions steps.

II. THE PRIMARY HOPPING FREQUENCIES

The three-dimensional structure of native pig insulin is known. The data set of atomic coordinates that was determined from X-ray diffraction on single crystal was used in the calculations. The resolution of the data is 1.5 Å. The electronic energy levels and wave functions of pig insulin were treated in the computations as those of an isolated macromolecule in aqueous solution.[5] The same was done in the calculation of random-walk process in which we only considered the hoppings among amino acid residues.[1]

In this paper, the geometry of pig insulin was also taken from the Brookhaven Protein Data Bank.[6] The chains

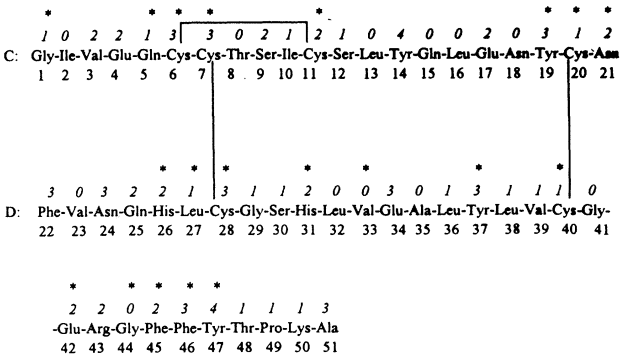

FIG. 1. The primary sequence of pig insulin (molecule II) and the distribution of the hopping centers, which are taken into account in the calculation of its ac conductivity. The numbers given above the different residues indicate the number of centers of the different orbitals localized mainly on the residue; see Eq. (2). Asterisks indicate the residues involved in the expression of biological activity of insulin.

A and B in the data set were considered as molecule I in the preceding papers,[1,2,5] so that chains C and D are identified as molecule II in this paper. There are 782 atoms in the molecule including the hydrogen atoms. 2418 basis functions were used in the calculation when Clementi's minimal basis set was applied to obtain the energy levels and wave functions of the entire molecular system. We apply the same approximations, numerical methods,[7,8] definitions and formulas, as in the previous papers.[1,2,5] For the sake of better understanding, we repeat the definitions and the expressions of hopping frequencies as well as of main residues here. The rest of the used rather involved formalism can be found in Refs. 1 and 7.

The hopping frequencies (primary jump rates) were calculated in the following way:[9,10]

$$h_{\mathbf{X}(n,i)\to\mathbf{X}(n',j)}=\nu_{\text{phonon}}\left[\sum_{r\in n}\sum_{s\in n'}C_r^{(i)}C_s^{(j)}\langle\chi_r(n)|\chi_s(n')\rangle\right]^2\exp\left[\frac{-\Delta E_{ij}}{k_BT}\right],\quad \Delta E_{ij}>0\,, \tag{1a}$$

$$h_{\mathbf{X}(n,i)\to\mathbf{X}(n',j)}=\nu_{\text{phonon}}\left[\sum_{r\in n}\sum_{s\in n'}C_r^{(i)}C_s^{(j)}\langle\chi_r(n)|\chi_s(n')\rangle\right]^2,\quad \Delta E_{ij}\le 0\,. \tag{1b}$$

Here $h_{\mathbf{X}(n,i)\to\mathbf{X}(n',j)}$, stands for the hopping frequency of a charge carrier hopping from the center of the ith orbital in the nth residue to the center of the jth orbital in the n'th residue. $\Delta E_{ij}=E_j-E_i$ is the energy difference between the final state and the primary state in the hopping. $C_r^{(i)}$ and $C_s^{(j)}$ are the linear combination of atomic orbitals coefficients of the $\chi_r(n)$ and $\chi_s(n')$ basis functions belonging to the orbitals $\Psi^{(i)}$ and $\Psi^{(j)}$ in the nth and n'th residues, respectively, k_B is the Boltzmann constant, and T the absolute temperature. The phonon frequency ν_{phonon} is taken as $\nu_{\text{phonon}}=10^{12}\,\text{s}^{-1}$ as it has been done in Refs. 9 and 10 (acoustic-phonon frequency) and in the previous papers.[1,2]

The so-called main residue, which will be only taken into account in these calculations, is defined as that residue which has a value of $a(n,i)\ge 0.4$ for its ith molecular orbital, where

$$a(n,i)=\frac{\sum_{r=1}^{m_n}C_r^2(n,i)}{\sum_{n=1}^{N}\sum_{r=1}^{m_n}C_r^2(n,i)}\,. \tag{2}$$

Here N is the total number of the residues in a protein molecule and m_n is the number of the basis functions in the nth residue.

As we have done in the preceding papers, pig insulin is considered as a quasi-one-dimensional system. Its primary sequence and the distribution of hopping centers that are taken into account in the calculations of its ac conductivity are shown in Fig. 1.

Similarly to molecule I (Refs. 1 and 2) the hopping centers that have to be taken into account (the number above the different residues in Fig. 1) are not evenly distributed along the sequence of insulin. Residues that have aromatic side chains have more than one hopping centers. This means that aromatic side chains have important contributions to the ac conductivity of proteins. Up to now, it has been experimentally found that 20 residues are involved in the expression of biological activity of insulin.[11–15] Thirteen of them have more than one hopping center. Only two of them (Val^{33} and Gly^{44}) possess no hopping centers. The residues Phe^{45}, Phe^{46}, and Tyr^{47}, one of the most important active areas of insulin, have large numbers of hopping centers. There are four tyrosine side chains in pig insulin. Three of them, Tyr^{19}, Tyr^{37}, and Tyr^{47}, have been found to be important in the expression of its biological activity; Tyr^{14} seems not to be important experimentally. All tyrosines have more than one hopping center but there is no hopping center at the nearest residues to the Tyr^{14} and only one center at its second neighbors. That is, the environment of the residues Tyr^{14} is different from the other tyrosines. It is well known that hopping conductivity depends on the values and distributions of hopping frequencies. Therefore, from the above results, we believe that there exists a relationship between the ac conductivity and biological activity of insulin: The reaction of insulin with its receptor might involve electron transport.

Chothia *et al.*[3] reported that there are three residues that have significantly different conformations in the two molecules of the dimer. These are Tyr^{A19}, His^{B5}, and Phe^{B25}. In our notation, they correspond to Tyr^{19} and His^{26} and Phe^{46}, respectively. All of them are important both from the point of view of hopping conductivity and of the biological activity.[11,12] Comparing Fig. 1 with the Fig. 1 of Ref. 1, we find that the number of hopping centers is smaller in molecule II, and the environments of them are different as in molecule I. This shows that the conformational changes of the biologically important residues strongly influence the electron transport in insulin.

In Table I, we present the largest hopping frequencies of different types (hopping between different orbitals localized on the same residues, between nearest and second nearest neighbors, hopping through disulfur bridges). Comparing them to those of molecule I, which are presented in Table II of Ref. 1, we find that both of them have qualitatively the same behavior. However, the hopping frequencies between second neighbors through disulfur bridges and between the two chains have somewhat different properties. The one from Leu^{27} to Cys^{7} (hopping to the second neighbor through the disulfur bridge Cys^{7}-Cys^{28}), which falls into the valence-bands region, is larger than the hopping frequencies to the second

TABLE I. Some primary hopping frequencies of pig insulin (Mol. II). The asterisk denotes an unfilled level.

i	j	n	n'	ΔE_{ij} (eV)	$h_{\mathbf{X}(n,i)\to\mathbf{X}(n',j)}$	$h_{\mathbf{X}(n',j)\to(n,i)}$
22[a]	18	4	4	0.047 23	5.20×10^{7}	3.05×10^{8}
38	25	14	14	0.197 75	4.41×10^{6}	7.28×10^{9}
3*[b]	5*	46	46	0.087 70	4.44×10^{5}	1.19×10^{7}
5	2	51	51	0.213 42	3.03×10^{5}	8.99×10^{8}
45	17	34	34	0.347 75	4.92×10^{4}	2.4×10^{10}
46*	49*	25	24	0.059 66	1.83×10^{7}	1.71×10^{8}
33	18	5	4	0.232 62	2.46×10^{3}	1.50×10^{7}
35	19	30	31	0.248 17	1.63×10^{3}	1.78×10^{7}
43	28	24	25	0.182 15	1.23×10^{3}	1.14×10^{6}
33	22	5	4	0.185 39	1.05×10^{3}	1.09×10^{6}
24*	28*	31	29	0.240 74	4.45×10^{3}	3.67×10^{7}
34	28	27	25	0.090 02	4.37×10^{3}	1.27×10^{5}
26	24	38	36	0.034 49	1.29×10^{3}	4.71×10^{3}
40	32	49	47	0.097 48	2.65×10^{2}	1.02×10^{4}
27	23	7	9	0.042 03	1.06×10^{2}	5.12×10^{2}
34	27	27	7	0.118 67	3.22×10^{4}	2.74×10^{6}
29*	32*	6	12	0.044 86	0.381	2.05
13*	18*	19	40	0.359 10	3.35×10^{-2}	2.34×10^{4}
10	3	10	6	0.349 01	3.08×10^{-2}	1.47×10^{4}
29*	40*	6	28	0.299 26	4.17×10^{-3}	3.08×10^{2}
40*	45*	28	7	0.069 63	3.17×10^{5}	4.30×10^{6}
27	6	7	28	0.572 96	0.125	2.62×10^{8}
19*	40*	7	28	1.293 37	4.95×10^{-12}	5.48×10^{9}
19*	45*	28	7	1.363 00	4.33×10^{-14}	6.52×10^{8}
15*	29*	11	6	1.373 66	9.85×10^{-15}	2.21×10^{8}

[a]The numbering of filled levels starts from the highest occupied one.
[b]The numbering of unfilled levels starts from the lowest unoccupied one.

neighbors within a chain. Further, the other primary jump rates through disulfur bridges to the second-neighbor residues are much smaller than those within a chain. Again, the hopping frequencies through the disulfur bridges (except the one in the conduction-bands region through Cys^{7}-Cys^{28}) become negligibly small if we go from lower energy levels to higher ones. On the other hand, they are larger than those between other nearest neighbors when the hopping events happen from higher energy levels to lower ones. This confirms the conclusion in Ref. 1 that the transport of electrons between the two chains in their ground states is generally difficult. It becomes, however, easy when external electrons are pumped to the conduction-bands region so that the disulfur bridges in an insulin molecule can have their special function in biochemical reactions. Moreover, the disulfur bridge Cys^{7}-Cys^{28} might have different features than the others. The experimental fact is that any mutation at this disulfur bridge hinders the activity of insulin even if its original three-dimensional structure remains unchanged.[12]

Comparing Table I with Table II of Ref. 1, we can see that the change of the three-dimensional conformation strongly influences the values of the hopping frequencies. The values of the largest hopping frequencies along the main chains, presented in Table I, are smaller than those in Table II of Ref. 1 by about one order of magnitude. This means that the electron transport along the main chains in molecule II is less efficient than in molecule I. Furthermore, those hopping centers that have the largest hopping frequencies belong to quite different residues in the two different molecules. For example, the largest nearest-neighbor frequency belongs to $Asn^{24}\to Val^{23}$ in molecule I and to $Gln^{25}\to Asn^{24}$ in molecule II. For further details, see Table II in Ref. 1 and Table I in this paper. These facts mean that the paths of electron transport in insulin alter by the changes of its three-dimensional conformation. We can expect that these differences would strongly influence the ac conductivities of the same molecule system in the two conformations.

III. THE HOPPING CONDUCTIVITY OF MOLECULE II OF PIG INSULIN

Hopping conductivity in disordered lattices and quasi-one-dimensional systems of inorganic materials has been investigated by random-walking theory in the last two decades.[16–20] In the preceding paper,[1] we have applied the formulas of Odagaki and Lax[16] to calculate the hopping conductivity of pig insulin, which is one of the smallest

native proteins.

In this paper, we use the same approximations and methods as in the preceding paper[1] to calculate the ac conductivity of the other conformation in the dimer of pig insulin. It is treated as a quasi-one-dimensional disordered finite system for which the three-dimensional coordinates of the atoms of pig insulin were used in the calculation, that is no periodic boundary conditions were applied (for details see Ref. 1 and Ref. 2). The formalism of Odagaki and Lax[16] has been applied again to calculate numerically its complex ac hopping conductivity as that has been done in Refs. 1 and 2. In this, and in the preceding papers, only the ac conductivity has been calculated numerically. (The dc conductivity cannot be determined analytically with the help of the expressions applied for *random* primary jump rates.[1,2] Numerical estimates for them based on the frequency dependent ac results will be given below.) It should be mentioned, however, that the value of the dc conductivity is not very important for the biological activity of insulin, because the series of biochemical reactions which are triggered by insulin binding to a receptor at the external part of the cell membrane have for their elementary steps a time scale of 10^{-10} sec.

As in the previous papers, we have taken the dimensionality, $d=1$, and the approximation that only those hoppings were taken into account that happen between first and second neighbor main residues. Further, we have considered only the 50 highest filled and 50 lowest unfilled orbitals in the calculation. In this way, the number of charge carriers is 100. The molecular volume of pig insulin is 22 250 Å^3 (Refs. 1 and 2) also for molecule II.

The calculated ac conductivity of pig insulin II is shown in Fig. 2. The previous results[2] for the ac conductivity of molecule I are also shown in Fig. 2 for comparison.

From Fig. 2, we can see that the ac conductivity of molecule II of insulin increases also with the frequency ω, like in the case of disordered inorganic solids (chalcogenides).[9]

The ac conductivity curves of pig insulin II are qualitatively similar to those of molecule I (Refs. 1 and 2). In the frequency range of $10^3<\omega<10^8$ sec^{-1}, the $|\sigma(\omega)|$ curve of the ac conductivity of the molecule II also lies between the curves of Te_2AsSi and As_2Se_3, and has the same order of magnitude as $Te_{48}As_{30}Si_{12}Ge_{10}$ (see Fig. 7.15 of Ref. 9). All these results confirm the conclusion that native proteins are good amorphous conductors if they are doped. Putting together these results with those obtained for pig insulin I (Refs. 1 and 2) and for lysozyme,[2] we believe that the above described behavior is a common property of ac conductivity in proteins.

Comparing the curves in Fig. 2, we find that there are certain differences between the ac conductivities $[\sigma(\omega)]$ of the two molecules. The largest differences occur in the high-frequency range, $\omega>10^{10}$ sec^{-1}. The real part of $\sigma(\omega)$ is larger in insulin I than that of molecule II in the frequency range of $10^4<\omega<10^7$ sec^{-1} and $\omega>10^{10}$ sec^{-1}. The imaginary part of molecule I is also larger than that of molecule II but only from $\omega>10^7$ sec^{-1} and its max-

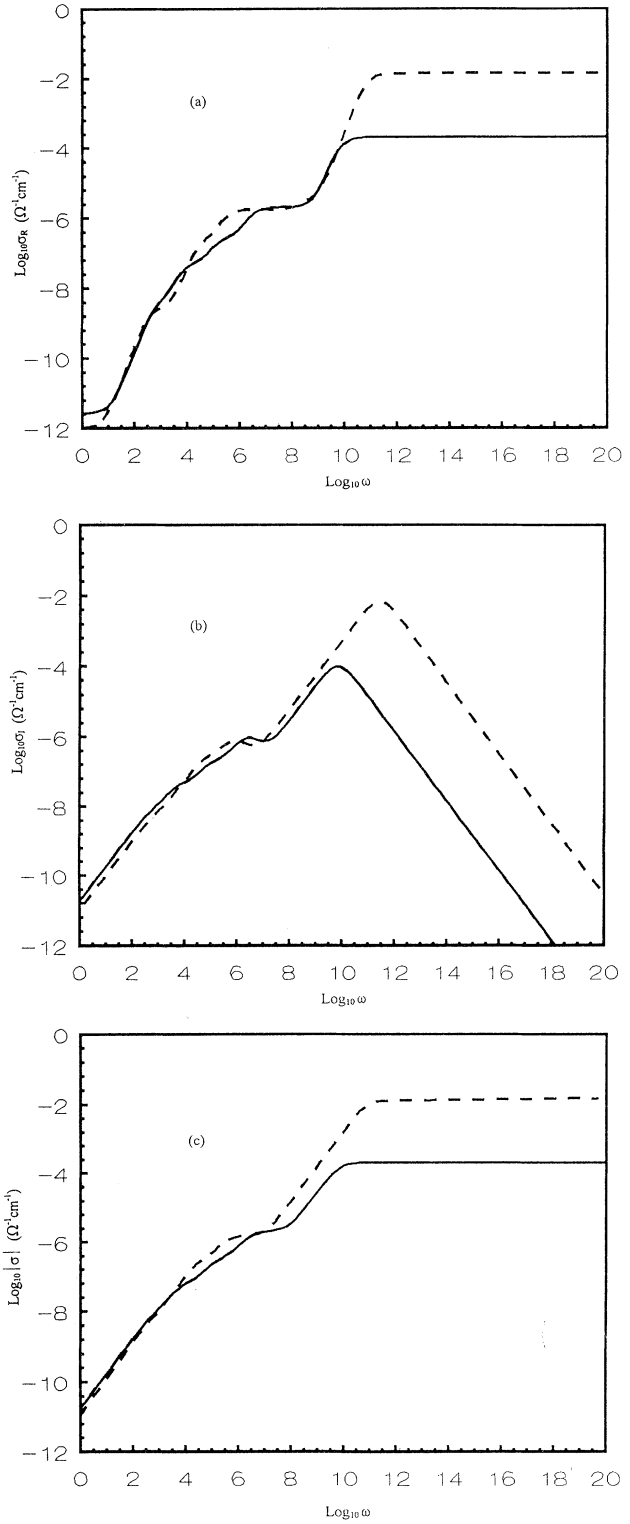

FIG. 2. The ac conductivity of pig insulin. The full line indicates the results of molecule II and dashed line indicates the results of molecule I.[2] [Molecule II consists of chains *C* and *D* (which differ not in their sequence, but only in their conformations from chains *A* and *B*, in molecule I) (Ref. 12)] (a) the real parts of the conductivity; (b) their imaginary parts; (c) their absolute values.

imum lies at $\omega \approx 10^{11}$ sec^{-1}. $|\sigma(\omega)|$ of insulin I is also larger than that of II in the frequency range of $10^4 < \omega < 10^6$ sec^{-1} and $\omega > 10^7$ sec^{-1}. The conductivity of molecule I is much higher than that of molecule II in the range of $\omega > 10^{10}$ sec^{-1}, which corresponds to the time period of an elementary step of a biochemical reaction. $|\sigma(\omega)|$ of molecule I still increases at $\omega \approx 10^{10}$ sec^{-1} while in the molecule II it becomes a constant at the same ω value; it is nearly two orders of magnitude larger in insulin I than in II when the curve of molecule I reaches its constant value at $\omega \approx 10^{12}$ sec^{-1} [see Fig. 2(c)]. This fact means that electron transport is easier in insulin I than in II when the external electric field changes its direction in a short time period. Therefore, insulin I should have more activity than II if the reaction involves electron transport.

From these results we can conclude that the ac conductivities of proteins are sensitive to conformational changes in the high-frequency range, especially if ω corresponds to the time period of an elementary step of a chemical reaction. Therefore, we believe that not only fitting of the reactants to proteins is needed, if they show biological activities, but most probably also their ac conductivity is important in reactions of proteins with their reactants when electron transport is involved in them. This property of proteins could be strongly influenced by conformational changes caused by the reactions.

It should be mentioned that by redrawing the curves of Fig. 2 in nonlogarithmic scales and enlarging their very low-frequency ranges, one could estimate the dc conductivity by extrapolation of pig insulin I and II, respectively. According to the results obtained $\lim_{\omega \to 0} |\sigma(\omega)| \sim 3 \times 10^{-12}$ $\Omega^{-1}\,\mathrm{cm}^{-1}$ for insulin I and $\sim 7 \times 10^{-12}$ $\Omega^{-1}\,\mathrm{cm}^{-1}$ for insulin II. [This is caused first of all by the inequality $\lim_{\omega \to 0} \sigma^{\mathrm{II}}_{\mathrm{Re}}(\omega) > \lim_{\omega \to 0} \sigma^{\mathrm{I}}_{\mathrm{Re}}(\omega)$ as one can see from Fig. 2(a).] Finally, it should be mentioned that Refs. 19 and 20 contain different partially quite sophisticated model calculations applying the random-walk theory for disordered one-dimensional chains. In this way, the authors were able to obtain also analytical expressions for the dc conductivity of these chains. Since, however, all these models contain some restrictions (the ratio of different hopping frequencies, periodic boundary conditions etc.), they are not applicable to real protein chains where no restrictions exist neither in the values for the different hopping frequencies, nor the conformation and relative orientation of the different subunits (different amino acid residues).

IV. DISCUSSION AND CONCLUSIONS

The ac conductivity calculated for molecule II of pig insulin that forms a dimer with molecule I in the single crystal was presented. The results confirm the conclusions of our previous papers that native proteins are good amorphous conductors along their main chains if they possess external electrons due to doping because the absolute value of the conductivity at a given frequency lies in the range of some typical good inorganic amorphous semiconductors. It has been found that the conformational differences between the two molecules of pig insulin strongly influence their ac conductivity in the high-frequency range, especially if the frequency corresponds to the time period of chemical reactions. This change is caused by the alterations in the distribution of hopping centers and of hopping frequencies, which are sensitive to conformational changes.

The biological action of insulin is somewhat complicated.[11,12] Many details remain unknown. Up to now, it is certain that pig insulin binds to a special receptor that is in the cell membrane and consists of three parts (external side of membrane, transmembrane, and internal membrane side parts, see Refs. 11 and 12). The insulin molecule binds to the external part of the receptor and then the internal part of the receptor stimulates a series of biochemical reactions. These reactions are stopped, if the insulin molecule is removed from the external part of the receptor.[11,12]

The atoms that are involved in the biochemical reactions change their positions in a very short time period. This time period corresponds to the high-frequency range of conductivity ($\approx 10^{-10}$ sec). The change of the positions of the atoms is, however, irreversible. Therefore, the treatment of a high-frequency electric current can be used as an approximation to describe such processes. This is the reason why our interests are focused on the high-frequency range of ac conductivity.

It has been found also that the conformation of molecule II is more close to the conformation of pig insulin in aqueous solution.[21] Further, it has been shown that pig insulin in this conformation has a lower ac conductivity by nearly two orders of magnitude in the frequency range that corresponds to the time period of an elementary step of the occurring biochemical reactions. This means that pig insulin has a lower ac conductivity when it is in aqueous solution. Until now the conformation of pig insulin bound to its receptor is unknown; therefore, its ac conductivity in its reactive state cannot be computed. However, we can consider the dimer of insulin as two molecules bound to each other. One molecule can be considered as a receptor of the other. It is known, at least, that insulin changes its three-dimensional conformation when it binds to its receptor.[12,22] This implies that pig insulin in aqueous solution has a lower ac conductivity in the high-frequency range before it binds to another molecule, which might be its receptor. To summarize the results of this paper and the previous ones,[1,2] we can conclude that insulin would change the ac conductivity of itself and of its receptor in the frequency range corresponding to the time period of chemical reactions when it binds to its receptor. In this way, the electron transport of the insulin receptor would change at the time when it expresses its biological activity.

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[1]Y.-J. Ye and J. Ladik, Phys. Rev. B **48**, 5120 (1993).
[2]Y.-J. Ye and J. Ladik, Int. J. Quantum Chem. **52**, 491 (1994).
[3]C. Chothia, A. M. Lesk, G. G. Dodson, and D. C. Hodgkin, Nature **302**, 500 (1983).
[4]W.-R. Chang, J.-B. Dai, J.-P. Zhang, B. Kuang, D.-L. Xie, D. Stuart, R. Todd, and D.-C. Liang, Chin. Biochem. J. **2**, 221 (1986).
[5]Y.-J. Ye and J. Ladik, J. Math. Chem. **14**, 141 (1993).
[6]J. Bordas, G. G. Dodson, H. Grewe, M. H. J. Koch, B. Krebs, and J. Randall, Proc. R. Soc. London. Ser. B **219**, 21 (1983).
[7]Y.-J. Ye, J. Math. Chem. **14**, 121 (1993).
[8]J. H. Wilkinson, *The Algebraic Eigenvalue Problem* (Clarendon, Oxford, 1965).
[9]N. F. Mott and E. A. Davis, *Electronic Processes in Non-Crystalline Materials* (Clarendon, Oxford, 1971), p. 215.
[10]J. Ladik, M. Seel, P. Otto, and A. K. Bakhshi, Chem. Phys. **108**, 203 (1986).
[11]S. Gammeltoft, Physiol. Rev. **64**, 1321 (1984).
[12]E. N. Baker, T. L. Blundell, J. F. Cutfield, S. M. Cutfield, E. J. Dodson, G. G. Dodson, D. M. C. Hodgkin, R. E. Hubbard, N. W. Isaacs, C. D. Reynolds, K. Sakabe, N. Sakabe, and N. M. Vijayan, Philos. Trans. R. Soc. London **319**, 369 (1988).
[13]G. P. Schwartz, G. T. Burke, and P. G. Katsoyannis, Proc. Natl. Acad. Sci. U.S.A. **84**, 6408 (1987).
[14]G. P. Schwartz, G. T. Burke, and P. G. Katsoyannis, Proc. Natl. Acad. Sci. U.S.A. **86**, 458 (1989).
[15]S. H. Nakagawa and H. S. Tager, J. Biol. Chem. **266**, 11 502 (1991).
[16]T. Odagaki and M. Lax, Phys. Rev. B **26**, 6480 (1982).
[17]H. Scher and M. Lax, Phys. Rev. B **7**, 4491 (1973).
[18]T. Odagaki and M. Lax, Phys. Rev. B **24**, 5284 (1981).
[19]J. W. Haus and K. W. Kehr, Phys. Rep. **150**, 263 (1987).
[20]T. Odagaki, M. Lax, and R. S. Day, Phys. Rev. B **30**, 6911 (1984).
[21]P. Kruger, W. Strassburger, A. Wollmer, W. F. van Gunsteren, and G. G. Dodson, Eur. Biophys. J **14**, 449 (1987).
[22]D.-C. Liang, W.-R. Chang, J.-P. Zhang, and Z.-L. Wan, Sci. China B **35**, 547 (1992).