Quantum effects in signal transduction biology: perspectives for 21st century nanoelectronics

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Abstract. Sizeable quantum effects in a specific biological signal transduction event have been recently discovered. Linear-scaling quantum mechanical calculations performed on the complex between the SH2 domain of a protein tyrosine kinase and a phospho-peptide indicate that upon binding the macrodipole of the protein is rotated by ~150 degrees from its initial orientation. This interesting finding suggests about the possibility of employing the SH2 domain as a molecular switch for 21st century nanoelectronics. Furthermore, additional aspects concerning both signal integration and signal amplification in biological systems are also discussed.

1. Introduction

The quest for increasing the miniaturization of electronic devices is driving both scientists and engineers towards novel and exciting avenues. The atom-by-atom approach whereby nano-objects are built by manipulating single atoms one at a time has its origins in Feyman's notorious lecture "There is plenty of room at the bottom" delivered in the late 1959 (Feynman, 2000). In 1989, Feynman's words became reality: Don Eigler and his coworkers at IBM used a liquid-helium-temperature scanning tunneling microscope (STM) to build nano-structures at the atomic level (Eigler and Schweizer, 1990). Following the excitement provoked by this breakthrough, Wada at Hitachi Ltd. suggested the realization of atom-based devices such as the atom-relay transistor (Wada, 1997). One of the limitations of atom-based electronics might be encountered in the large-scale production of nanostructures (atom-based nanocircuits) which will require the development of nanofactories (Phoenix, 2003).

Other promising approaches have also been investigated. The molecule-by-molecule approach, for example, takes its advantage from the fact that molecules are stable collections of atoms properly arranged in 3D space that can be synthesized in large quantities. The presence of specific chemical groups favor their self-assembly to supramolecular systems stabilized by intermolecular, non-covalent interactions. So far, several functional molecules have been synthesized and tested for their possible use in molecular electronics (ME). ME is concerned with synthetic molecules capable of signal processing ability. A typical example of this kind is the well known molecular rectifier firstly proposed by Aviram and Ratner in 1974 (Aviram and Ratner, 1974), which is regarded as the first molecular-scale device ever conceived. Other relevant examples include molecular wires, switches, transistors, and logic gates (Goser et al., 2004).

Biomolecular electronics (BioME), on the other hand, is concerned with the use of

biomolecules in nanoelectronics (Nicolini, 1996). The main difference between BioME and ME is that biomolecules are stabilized by and do work in an aqueous environment whereas their synthetic counterparts are generally soluble in organic solvents. It follows that the majority of biomolecules (proteins, DNA, carbohydrates, etc.) are thermodynamically stable within a limited range of pH and temperatures. This has important consequences for their correct functioning as molecular-scale devices. However, the advantage in using biomolecules rather than synthetic molecules is that important functional properties such as electron transfer and proton tunneling have already been implemented by Nature during the course of evolution. This together with the possibility of using genetic engineering as a mean to modify natural proteins to yield mutants with enhanced characteristics makes BioME very attractive a technology.

Reducing the size of electronic components at the molecular level, however, is not enough for 21st century nanoelectronics. The individual components need to communicate with one another as well as with the outside world. Hence, novel paradigms are necessary so as to achieve both signal integration and communication. In this contribution I shall make the following proposal: let us look at how information is transmitted from the outside to the inside of the cell and see whether this complex biological process might be exploited and applied in nanoeletronics. The field that we are going to explore is known as signal transduction (ST) biology. After a brief overview of the field, I will present some recent results concerning the discovery of quantum mechanical effects in a specific ST event. It will follow a discussion on possible means to implement ST phenomena and elements in nanoelectronics.

2. Signal transduction biology

ST biology studies how an extracellular signal is transformed (transduced) into an intracellular biological effect (Eyster, 1998). In general, a ST process starts with the interaction of

a hormone (generally a polypeptide) with its target receptor on the surface of the cell. Ligand-receptor binding activates a series of intracellular (molecular) processes such as receptor dimerization, phosphorylation, and conformational changes which initiate a cascade of signals inside the cell. Three types of membrane-bound receptors are known: (a) G-protein-linked receptors, (b) enzyme-linked receptors, and (c) ion channels (Gomperts et al., 2002; Helmreich, 2001).

Let us briefly explain the different characteristics of each membrane-bound receptor. G-protein receptors are seven-pass transmembrane proteins that are being activated upon ligand binding. A conformational change of the receptor favors the interaction with an intracellular linker/transducer G-protein which subsequently dissociates to interact with an effector enzyme. The signaling cascade is then started by the effector enzyme in the active (ON) state. When the effector enzyme becomes inactive (OFF state) the transmission of the signal is halted. Enzyme-linked receptors are somewhat simpler than G-protein-linked receptors for the catalytic domain is part of the receptor and, hence, they do not need the assistance of G-proteins. This characteristic makes them attractive objects for bio-inspired nanoelectronics. ST pathways involving enzyme-linked receptors start also from a ligand binding event that favors receptor dimerization. This, along with a conformational change of the intracellular domain, gives rise to the autophosphorylation of the receptor. Intracellular proteins that recognize the phosporylated residues can now interact with the receptor so as to be subsequently phosphorylated. Protein phosphorylation is a key event which starts the downstream propagation of the signal inside the cell. Finally, ion channels are membrane proteins that facilitate the transport of charged ions across the membrane. It is worth mentioning that only recently it has been possible to crystallize ion channel proteins to obtain single crystals of a quality that is sufficiently good for X-ray structure analysis. The 2003 Nobel Prize for Chemistry has been awarded to MacKinnon who

firstly determined the atomic structure of the potassium (K^+) channel.

Two intracellular processes are particularly important for ST biology. The first is the process of *signal integration* schematically shown in Figure 1. Here two membrane-linked surface receptors, R1 and R2, are independently activated by their associated ligands, L1 and L2, respectively. Two signaling cascades are then propagated towards a common target, the protein (P), which integrates the signal and propagates it downstream inside the cell.

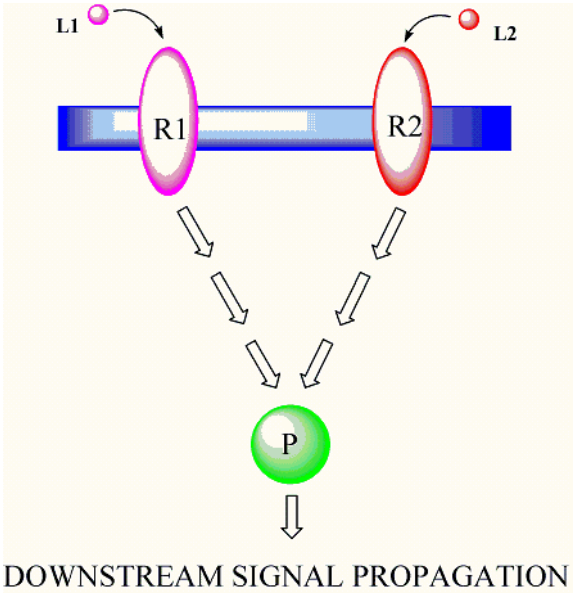

Figure 1. Schematic view of a process of signal integration. R1 and R2 are membrane-bound receptors activated by their associated ligands L1 and L2, respectively. The signal cascade converges to a common protein (P) molecule which integrates the signal and propagates it downstream inside the cell.

The second important process for ST biology is that of *signal amplification* shown in Figure 2. Here ligand binding to a membrane-linked surface receptor activates the signaling cascade inside the cell. The signal is then amplified at every step. An interesting example of

signal amplification is that occurring in the visual transduction cascade where the activation of one Rhodopsin molecule produces the hydrolysis of 10^5 cyclic GMP molecules at the end of the catalytic cascade (Alberts et al., 2002). This means that the signal is amplified 10^5 times!

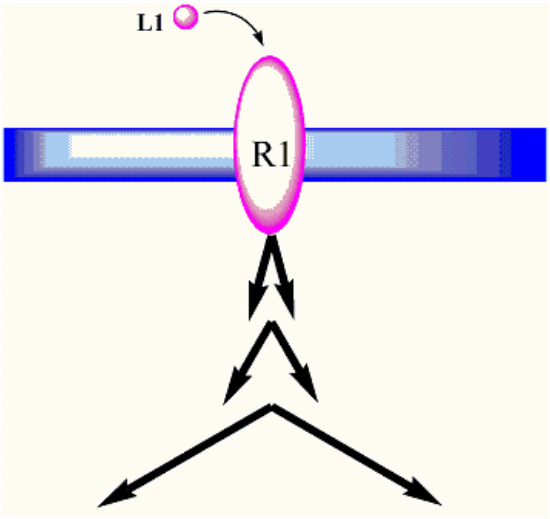

Figure 2. Schematic view of a process of signal amplification. Ligand binding to a putative membrane-bound receptor initiates a signaling cascade inside the cell that is amplified at every step.

It is important to keep in mind that the above processes of signal integration and signal amplification do require the concerted participation of many enzymes and substrates. Enzymes that participate in a specific ST pathway are being synthesized by the cell itself while many of the substrates are either transported across the membrane or synthesized *in situ*.

3. Discovery of quantum effects in ST biology

The occurrence of quantum mechanical effects in biological systems has been discussed by several authors with particular reference to important biological processes such as

enzymatic catalysis and genetic information transfer (Igamberdiev, 1993; Matsuno and Paton, 1999). So far and to the best of our knowledge, however, QM effects in ST biology have not been investigated yet. The reason behind this delay might be due to the lack of high-resolution structures of proteins that are involved in ST pathways or to the inherent complexity of such processes. The availability of a crystal structure of the Src homology 2 (SH2) domain solved at atomic resolution along with important advancements in the field of computational quantum chemistry stimulated me to perform a series of calculations on this protein domain. The SH2 domain is one of the most important recognition domains for phospho-peptides. Its structure is composed of a central anti-parallel beta-sheet flanked by two alpha-helices to form the so called alpha/beta/alpha motif. High affinity binding of phospho-peptides is achieved through the insertion of a phospho-tyrosine head into a binding pocket located at one of the two alpha/beta interfaces. This is one of the key events in the ST pathways originated from the receptors of the tyrosine kinase family (RTK).

I have selected the crystal structure of the SH2 domain of p56lck tyrosine kinase since it has been recently determined at atomic resolution (1.0 Å) (Tong et al., 1996). The atomic coordinates of this protein domain have been deposited in the Protein Data Bank (PDB) under the reference code 1LKK. The protein has been crystallized at pH 6.5 in complex with a short phospho-peptide, denoted to as pYEEI (pY=phosphotyrosine, E=glutamate, I=isoleucine) which is bound to it through eight hydrogen bonds and three salt bridges. Six hydrogen bonds do involve the phospho-tyrosine (pY) moiety which interacts with the side-chains of two arginine residues and two serine residues.

Linear-scaling semiempirical QM calculations were performed so as to assess the magnitude of the interaction energy of the protein-ligand complex, the effect of the ligand on the charges of the amino acid side-chains of the protein, and the topology of the molecular orbitals

(Pichierri, 2004a). This type of calculations can also provide an estimate of the permanent dipole moment of the protein, which can be regarded as a measure of the polarization of electronic charge in a molecule. The magnitude of the dipole moment calculated with this method has been validated for alpha-chymotrypsin (Pichierri, 2003b). Figure 3 shows the most interesting result obtained from the QM calculations on the SH2 domain.

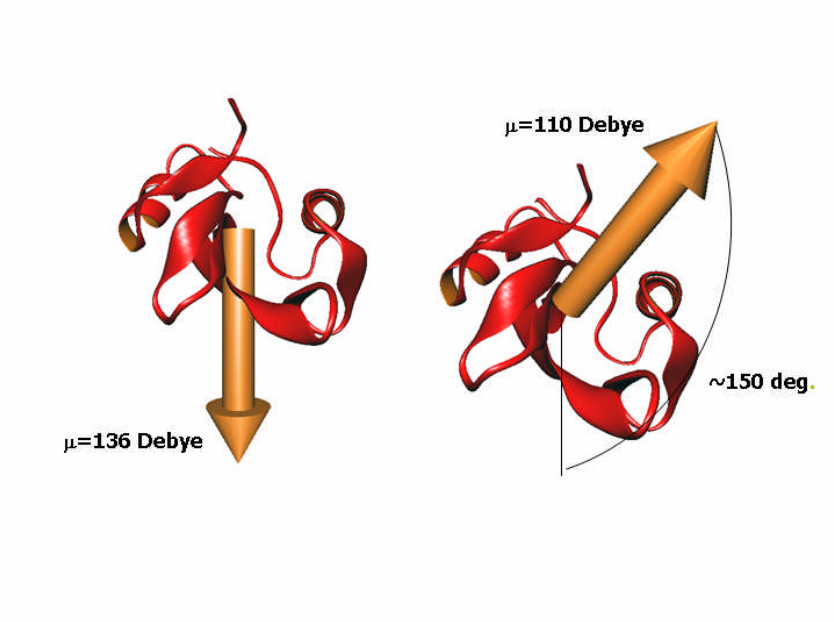

Figure 3. The macrodipole of the SH2 domain (left) rotates by about 150 degrees upon ligand binding with the phospho-peptide (right).

The calculations indicate that, upon binding, the dipole moment vector of the protein changes its orientation by rotating about 150 degrees around the axis parallel to that of one alpha-helix. The magnitude of the dipole moment decreases only slightly, namely from 136 to 110 Debye. This result represents the signature of a sizeable quantum mechanical effect that takes place in a specific ST event (Pichierri, 2004a).

4. ST-based nanoelectronics

With the above background and results we are now in a position of proposing possible implementations of ST-based processes for 21st century nanoelectronics. The most ambitious goal would be that of implementing a full ST pathway so as to achieve either signal integration (Figure 1) or signal amplification (Figure 2). This implies the possibility of employing whole cells so as to perform cellular computing (Amos, 2004). Working with large ensembles of cells, however, might be quite difficult since little or no external control can be exercised on the ongoing intracellular ST processes. Furthermore, since ST pathways are directed from the outside to the inside of cells, the problem of how to extract the processed signal may be quite hard to solve. A simpler and more practical approach is that of isolating some elements of an ST pathway whereby sizeable QM effects are known to operate. Above we have seen how the change in the direction of the dipole moment upon ligand binding may act as a switch or a two-state (ON/OFF) system. It is worth noticing that the term switch is commonly employed by biologists to indicate those protein-ligand complexes that are found in the bound-unbound state. Not all the know protein-ligand complexes, however, are likely to produce the sizeable QM effect that is observed for the SH2 domain (see Figure 3).

A further extension of the single biomolecular switch is that of building an array of dipoles so as to achieve signal amplification by means of vector addition. This could be done by attaching the protein on a metallic surface such as that of gold, which can form stable covalent bonds with the thiol groups of cysteine residues. The resulting supramolecular device might be sensible enough to interact with an external probe or circuit, as shown in Figure 4.

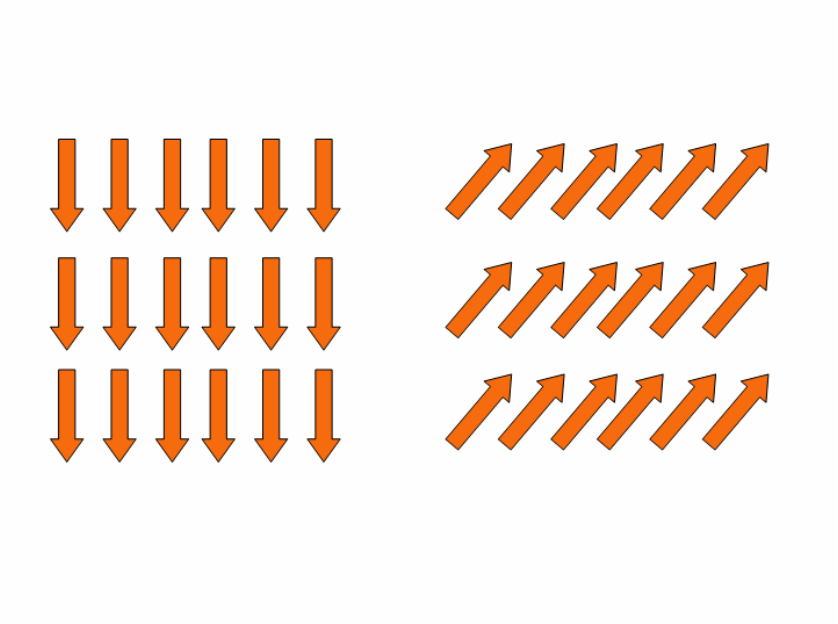

Figure 4. An array of protein dipoles with which it could be possible to achieve a significant amplification of the signal via a vector summation effect.

The protein array displayed in Figure 4 might be useful for the development of protein-based chips that could be employed in the nanoelectronic devices of the 21st century. Protein-based chips have already been constructed and employed to perform immunoassays for medical diagnostics (Kojima et al., 2003).

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