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Control of catalytic activity of proteins in vivo by nanotube ropes excited with infrared light

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Abstract

We discuss the possibility of controlling biological systems, by exciting in the near infrared region *hybrid* metallic nanotube ropes, dressed with proteins and embedded in the biosystems. If one nanotube, in a double-tube rope, is filled with metallofullerenes and the other is empty, the two tubes change their opposite equilibrium charging during the irradiation. The resulting change of the local electric field can deform proteins attached to the tubes, and change their catalytic properties.
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1. Introduction

Over billions of years, a fascinating internal and external complexity evolved in myriads of competing biological species [1]. For example, many bacteria and multicellular organisms use structure-sensitive proteins to biomineralize nanocrystals [2], that form unique nanodevices. These *cold*-growth techniques [3] could greatly complement growth methods developed by humans. In general, biosystems and artificial nanosystems can coexist and supplement each other in a number of other directions, in particular, during the release of drugs [4]. Their coevolution can lead to the formation of new hybrid *bio-nano* (BIONA) systems, with unprecedented organizational and functional level.

In this Letter, we discuss possible forms of communication between BIONA components. The (direct) 'talk' from the nanosystem to the biosystem could be realized by controlling catalytic activity of its proteins. The (backward) 'talk' could be done by the cells, if they change their local microenvironment or emit (electromagnetic) signals. The direct talk, that we explore here in more details, could be based on electrochemical methods, used in bioelectronics [5]. Unfortunately, these techniques require the presence of electrodes [6], that cannot be easily applied inside cells. More promising is thus their combination with *contactless* methods. Optical techniques [7] are convenient for their selectivity, but the sensitive interior of cells prohibits the use of large optical frequencies that can easily manipulate chemical bonds. We

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could thus use the fact that cells are transparent (up to 5 mm thick samples) for the *near-infrared* (NIR) radiation (0.74–1.2 μm) [8].

The activity of biosystems could be manipulated via artificial nanosystems, embedded in them, that absorb in the NIR region. This approach is followed in photodynamic therapies, where tumor cells are destructed chemically, via NIR excitation of porphyrin-based molecules with many extended electronic states [8]. Similar results can be obtained by heating the system locally with ultrasound or microwave radiation or via NIR-radiation heated metallic nanoparticles [9]. Silver and gold nanoparticles [10], that can be produced biologically [11], are naturally excellent candidates for use in biocontrol. In order to control tiny cellular sections, one could also think of using nanotubes. Metallic C nanotubes form sensitive detectors in liquid environments [12], and when dressed with biomolecules, via structure-selective [13,14] or less specific hydrophobic coupling [15], they can work as sensitive biosensors [16].

2. NIR-radiation control of protein activity

The biocontrol could be elegantly realized by a *hybrid* nanotube rope, heated by NIR-radiation, that is formed of two adjacent metallic C nanotubes, where one is (peapod) filled with *metal-*
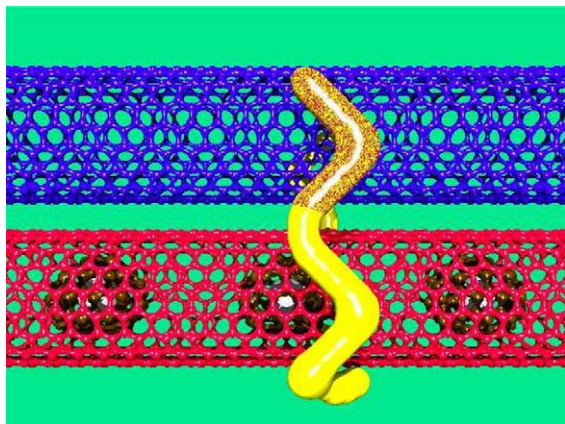

Fig. 1. Scheme of a hybrid nanotube system that controls protein activity in vivo by near-infrared radiation, as described in the text.

lofullerenes [17] and the other is empty. In peapods, electrons can be transferred to/from the C_{60} fullerenes under electric bias [18]. In isolated metallofullerenes like $Dy@C_{82}$, several electrons are passed from Dy to C_{82}. When these are used in a peapod, the transferred electrons can be passed further to the nanotube [19]. In a double-rope formed by this peapod and a 'twin' (empty) nanotube, the last would absorb the excessive charge too, so the two would become oppositely charged. This process can be partly *inverted* at elevated temperatures [19], since the metallofullerenes have levels close to the Fermi level [20,21].

In Fig. 1, we schematically show the NIR-control of protein (enzyme) activity, based on this process. The NIR excitation heats the two nanotubes, so that electrons, released in equilibrium from the fullerenes to the peapod [19] and the twin tube, become transferred *back*. This transfer is accompanied by recharging of the tubes, and the resulting change of the local electric field causes deformation of proteins [22,23], that are selectively attached to the nanotubes. Their new conformation can have a very different catalytic activity [24]. The system thus works in an opposite way than some biosensors [25], where antibodies bind to proteins attached to material surfaces, bend them, and thus change the surface electric parameters. We can tune the system by using different nanotubes, fullerenes and their filling, and especially proteins, to be controlled. The attached proteins, that in general could be much bigger than the tubular system, might help to dissolve the hydrophobic nanotubes in water.

3. Modeling of the control

We consider that the system is formed by two metallic (10,10) carbon nanotubes of the radius $r_t \approx 0.68$ nm, where *one* of them is the peapod. In a double-rope [26,27], their centers are separated by $D_T \approx 1.7$ nm, which determines the tunneling time, $\tau_t \approx 1$ ps, of electrons between the tubes. The fullerenes are separated one from another by $d_F \approx 2$ nm [21], and their charging strongly but locally deforms electronic bands of the peapod [20].

We can excite the metallic nanotubes at deliberate NIR frequencies. Their absorbtion fits the Drude formula for the dielectric function [28]

$$\varepsilon(\omega) = \varepsilon_\infty \left(1 - \frac{\omega_p^2}{\omega^2 + i\omega/\tau} \right), \tag{1}$$

where $\hbar\omega_p = 0.86$ eV is the plasma frequency, $\varepsilon_\infty = 4.6$ and $\tau = 5 \times 10^{-15}$ s. The irradiation, needed to induce reabsorption of electrons by the fullerenes, would have to heat the nanotubes by several tens of degrees [19]. This should not be harmful in vivo, if we use, for example, short (few nanometers long) nanotube *capsules* [29], and isolate them in liposomes [30], that can be delivered to the cells by special techniques.

In the lack of available data, we assume here that one electron *per* fullerene is reabsorbed during the irradiation, and $\approx 20\%$ of those come from the empty tube. This gives the NIR-radiation induced *recharging* density $\sigma = 0.2\ e/d_F \approx 0.1e$ nm^{-1}. From this σ, we can calculate the change of electric field between the two tubes. If we assume that these two are *ideal* metallic cylinders of length L, their electric capacity is [31] ($\varepsilon = \varepsilon_0 \varepsilon_r$)

$$C = \frac{\pi\varepsilon L}{\cosh^{-1}(D_T/2r_T)}. \tag{2}$$

Thus, the potential difference between the nanotubes due to the induced charge transfer is

$$\Delta\varphi = \sigma L/C \approx 0.1 \text{ V}, \tag{3}$$

where we use the permitivity of water $\varepsilon_r \approx 4.6$. Similar voltage was used, for example, in manipulation of proteins attached to metals [24].

Activation of the attached protein by this NIR-radiation induced potential can be realized by moving a *charged tip* of one of its domains [25] (see Fig. 2). We model this process, by evaluating
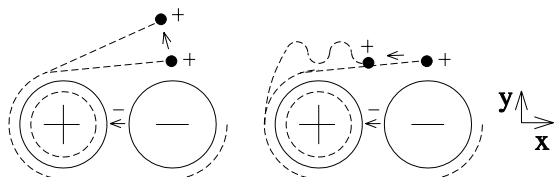

Fig. 2. Scheme of two (hinge and shear) configurations for the protein control, as described in the text.

first the potential energy of a charge q at the position $\mathbf{r} = (x, y)$. The two cylinders with charge densities σ and $-\sigma$ have their centers are at $\mathbf{r}_1$ and $\mathbf{r}_2$, respectively. The potential energy of the charge q is formed by the *direct* Coulombic component [31]

$$V_C(\mathbf{r}) = -\frac{\sigma q}{2\pi\varepsilon} \ln\left(\frac{|\mathbf{r} - \mathbf{r}_1|}{|\mathbf{r} - \mathbf{r}_2|} \right), \tag{4}$$

that can be either positive or negative, depending on which of the oppositely charged tubes is closer. It also has a negative *screening* component [32], originating in the reflection of the external charge in the metallic tube, that close to the surface of both tubes has the form

$$V_S(\mathbf{r}) \approx -\frac{q^2}{16\pi\varepsilon} \left(\frac{1}{|\mathbf{r} - \mathbf{r}_1| - r_T} + \frac{1}{|\mathbf{r} - \mathbf{r}_2| - r_T} \right). \tag{5}$$

Here, we simply add the screening potentials of the two tubes, neglecting thus multiple reflections.

Typically, structural domains in proteins perform *hinge* or *shear* motion [22,23]. These domains are often formed by (rigid) α-helices, connected by (flexible) β-sheets. The structures (conformations) of deformed proteins in nature are usually close in energies, so that they can be flipped over by room temperature energies $k_B T$ [22]. The conformations of the externally controlled proteins should have different catalytic properties and be more energetically distant, so that they are not changed at room temperatures.

In the present system, where the control of protein's motion is realized via dynamical charging of nanotubes, we can also consider these two generic (hinge and shear) configurations, shown schematically in Fig. 2. Since, the tubes are different and become charged in equilibrium, the proteins should be able to *distinguish* them and deposit on them *asymmetrically* (see Figs. 1 and 2). In the bend configuration (left), the trajectory of the controlled protein domain is practically vertical, toward one of the tube's centers. In the shear configuration (right), the trajectory of the domain goes approximately in parallel with the vector connecting the tube's centers of masses.

We assume that the balance of internal forces in the protein, in the presence of equilibrium

charging of the tubes, adjusts the charged tip to a position $\mathbf{r}_0 = (x_0, y_0)$. The dependence of the protein energy around (close to) this position can be considered to be parabolic

$$V_R(\mathbf{r}) \approx C_x(x - x_0)^2 + C_y(y - y_0)^2, \qquad (6)$$

where the constants C_x, C_y describe rigidity of the deformed protein [23]. Their values should be such, that the difference in energies ΔE between the used conformations is $k_B T < \Delta E < 100$ kJ/mol (1 eV), where the last value is the lower energy limit required to deform *individual* protein domains [23].

4. Discussion of the protein motion

In order to estimate which of the configurations, in Fig. 2, can be more easily controlled, we calculate the *distance* over which the protein domains move during the NIR-radiation induced charge transfer. The tip moves from the equilibrium position $\mathbf{r}_0$ to a new position $\mathbf{r}_0'$, given by the local minimum of the total potential

$$V_T(\mathbf{r}) = V_C(\mathbf{r}) + V_S(\mathbf{r}) + V_R(\mathbf{r}). \qquad (7)$$

In Fig. 3 (up), we search this minimum for the 'hinge' configuration, shown in Fig. 2 (left). The tube centers are located at $\mathbf{r}_{1,2} = (x_{1,2}, y_{1,2})$, $x_{1,2} = \mp 1.1 r_T$, $y_{1,2} = 0$. We present the dependence of the potentials V_C, V_S, V_R and V_T on the y distance from the center of the right nanotube, and assume that a unit charge $q = e$ is at the tip of the domain [25]. The results are calculated for the (effective) charge density $\sigma = 0.1$ e/nm (equilibrium) and $\sigma = 0$ (irradiation). We position the domain tip at $x_0 = x_2$, $y_0 = 2.5 r_T$ and use the rigidity constant $C_y = 2$ eV/nm^{-2}. We can see that the *change* of the V_C potential energy, due to the induced charge transfer, is small, while V_S is rather large. With the above parameters, the V_R potential can *locally* compensate the steep V_S, so that close to the tubes their sum is almost flat. Then, the weak V_C potential can control the position of the local minimum in V_T, but the overall motion is quite small (thin and thick solid line correspond to equilibrium and irradiation, respectively). The magnitude of motion could be enlarged, if we let the V_T potential to loose the local minimum in the *absence* of irradiation. Then
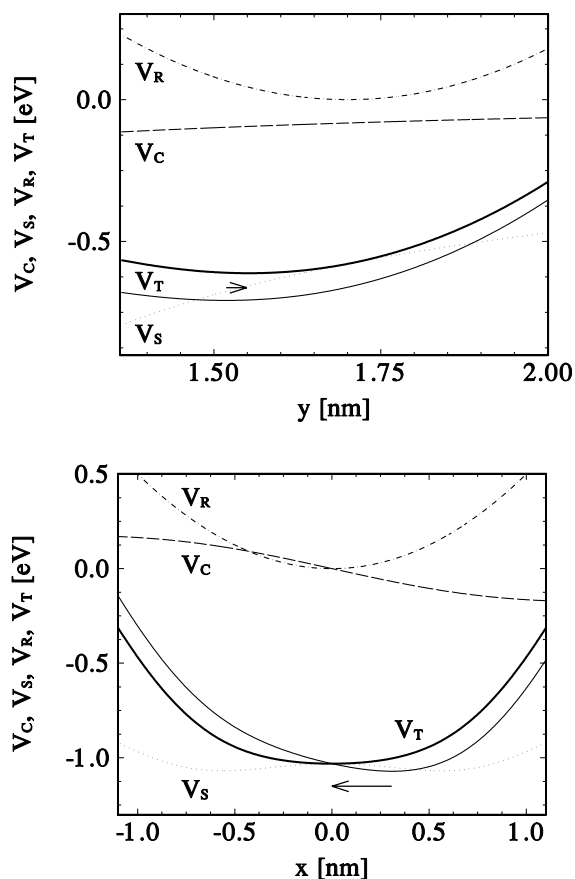

Fig. 3. The calculated potentials of the proteins in the hinge (up) and shear (down) configurations for the parameters in the text. From the local minima of V_T, corresponding to the change $\Delta \sigma = -0.1$ e/nm of the charge density on the tubes, we obtain the protein displacement ≈ 0.04 and ≈ 0.35 nm, respectively, shown by the arrows.

the domain position would *fluctuate* from being adjacent to the tube to being almost at $\mathbf{r}_0$.

Such a large motion can be obtained directly in the 'shear' configuration, shown in Fig. 2 (right). Here, the in-plane (of the tubes) components of the screening forces largely cancel each other, and the out-of-plane components are not effective. Thus the system responds more sensitively to the charging given by V_C, as we show in Fig. 3 (down). We use parameters, $x_0 = 0$, $y_0 = 1.7 r_T$ and $C_x = 0.5$ eV/nm^{-2}, so that V_R can *flatten* the two-well minima of V_S. In equilibrium, the tube charging causes that V_T develops a minimum, close to one of the V_S minima. During the irradiation, the charging decreases and

the minimum shifts to $x = 0$. Since the two conformations are shifted in energy by $\Delta E \approx 50$ meV, they would not flip one to another at room temperatures $k_\mathrm{B}T < 30$ meV. This domain motion is large enough to control the protein (enzyme) activity. It could open or block pockets on the 'back side' of the protein, that is not exposed to the nanotube, and change the catalytic strength of the proteins.

We have demonstrated that NIR-radiation excited hybrid nanotube ropes could control the activity of proteins in vivo. Nanotube systems might also *directly* activate chemical reactions used in phototherapy [8], in particular, if special 'porphyrin-like' defects are formed in the nanotube walls. In vitro and possibly in vivo, one could also collect electronic signals [33] or bias nanotubes externally in order to control biochemical reactions or use them in other applications on the nanoscale [34].

Acknowledgements

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