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Biophysical aspects of cancer – Electromagnetic mechanism

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Hypothesis of coherent vibration states in biological systems based on nonlinear interaction between longitudinal elastic and electric polarization fields with metabolic energy supply was formulated by Fröhlich. Conditions for excitation of coherent states and generation of electromagnetic fields are satisfied in microtubules which form electrical polar structures. Numerical models are used for analysis of Fröhlich's vibration states in cells.

Reduction of activity and of energy production in mitochondria, and disintegration of cytoskeleton structures by phosphorylation on the pathway of cancer trasformation can diminish excitation of the Fröhlich's vibration states and of the generated electromagnetic field, which results in disturbances of the interaction forces between cells. Interaction forces between cancer cells may be smaller than interaction forces between healthy cells and cancer cells as follows from numerical models. Mechanism of malignity, i.e. local invasion, detachment of cancer cells, and metastasis, is assumed to depend on the electromagnetic field.

Keywords: Cancer cells, Coherent states, Fröhlich's theory, Microbules

Introduction

Cancer reflects disturbances of properties and behavior of cells in a multicellular system. Cancer changes inner mechanisms in cells as well as their interactions in the tissues. The vast majority of cancers are connected with mutagenesis, i.e. with changes of DNA (deoxyribonuclec acid). Epigenetic origin of cancer is an extremely rare event. Cancer transformation is a multistep microevolutionary process that triggers a vast spectrum of biological, biochemical, and biophysical changes[1]. Long term transition from healthy to malignant behavior of cells lasting even several decades is indicated by medical observation. Proliferation is a general feature of cancer. The initial noninvasive local growth of cancer very often forms a structure lacking the typical organized pattern of corresponding normal tissue[2]. The most dangerous developmental steps – malignant steps – start with local invasion. Cancer cells burrow into their immediate surroundings. Process of local invasion is assumed to be a result of active locomotion rather than of passive transport, for instance not a result of pressure. Cancer cell displays changed properties of the cytoskeleton, of the cytosol, of the plasma membrane, loss of cohesion and of contact inhibition, and tendency to separation. Loss of adhesiveness plays a major part in the detachment of cancer cells and their arrest in other parts of the body. Establishment of metastasis involves the whole concept of specific adherence between cells.

Alteration of proto-oncogenes to oncogenes results in production of cancer protein kinases. Phosphorylation of proteins in normal cells is a ubiquitous mechanism of regulation, but phosphorylation in cancer cells is different from that in healthy cells. Phosphorylation replaces a hydrogen atom in the terminal hydroxyl group OH in the side chain residues of serine, of threonine, and of tyrosine by a phosphate group (by inorganic phosphate). The phosphate group has about 80 times greater mass than hydrogen atom and represents an electric multipole with negative charge of three oxygen atoms at the outer surface. The electrostatic interaction with positively charged side chain amino acid residues can change protein conformation and can also cause increased coupling with the surroundings (with the heat bath).

Regardless of a great amount of the biological and biochemical research results mechanisms underlying processes of malignity are not well understood. Fröhlich assumed that disturbances of electromagnetic coherent states lead to evasion of cancer cells from control and regulation in the tissue[3]. The Fröhlich's electromagnetic mechanism is based on nonlinear interaction between longitudinal elastic and

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polarization fields and energy supply to the system from metabolic sources. Fundamentals of the biophysical theory were presented by Fröhlich[4-6]. He described a vibration model too[6-10]. The mechanism has a parallel in solid state ionic crystals. A mechanical stroke on the side of the NaCl crystal excites synchronized motion of atoms–of the natrium and chloride ions[11]. The mechanical shock wave interacts with polarization wave generated by motion of charges. Polarization current displays coherent signals (greater than noise) whose frequency depends on direction of wave propagation in the crystal (the frequency is of the order of magnitude of 10 THz). Coherent vibrations based on nonlinear interaction between elastic and polarization fields and on energy supply seems to be a general property of electric polar structures.

The Fröhlich theory contains general considerations on coherence and long range control[12-17]. Energy storage and action of specific forces are important consequences[5, 9, 18-19].

Endogenous electromagnetic field generated by vibrations in electrical polar structures may have fundamental function in organization of biological systems including morphological features, transport of macromolecules and particles, biochemical reactions, and polymerization of large structures[20-25]. Changes of the generated endogenous electromagnetic field may belong to the pathway of cancer transformation. Cytoskeleton disintegration and blocking of mitochondria activity in cancer cells are a signature of cancer.

The Fröhlich's hypothesis of a fundamental role of coherent polar vibrations in living cells processes is now supported by direct and indirect measurements of the generated electromagnetic field and of mechanical vibrations of cellular membrane. Attraction of small dielectric particles with high permitivity to cells was observed[26-28]. Pohl[29] explained the observed phenomenon by dielectrophoretic. The greater the permittivity of the particles and the smaller the conductivity of the suspension with cells, the greater the number of attracted particles. In the M phase yeast cells attract the greatest number of particles. Collection of attracted barium titanate particles to the tips of elongated alga *Monoraphidium griffithii* was observed by Hölzel and Lamprecht[30]. At the tips there are regions of the highest intensity and of the greatest inhomogeneity of the electric field. Albrecht-Buehler investigated the ability of cells to detect electromagnetic signals of other cells in the red or in the near-infrared range[31]. Baby hamster kidney (BHK) elongated cells were plated on both sides of a glass film. If the film was thin (1 – 2 μm) than the cells plated later (when the cell layer on the opposite side was fully developed) oriented themselves in transverse direction with respect to the cells on the opposite side. The thick glass layer (150 μm) prevented communication between cells. Using also some other methods to suppress transfer of red or near infrared fields between the cells on opposite sides of the glass film Albrecht-Buehler concludes, that interaction between cells is mediated by the electromagnetic field. The Swiss 3T3 cells are able to sense near infrared radiation (with specific wavelengths) and to determine direction of individual sources[[32]. Scattering latex particles (3.22 μm) illuminated by the near infrared radiation served as a source of the electromagnetic field.)ne forth of the cells extended lamellipodia towards a single infrared scatterer and 47 % of the cells towards two infrared scatterers next to each other. The strongest response was elicited by the electromagnetic field at the wavelengths 800 nm, intermittently modulated (60 periods/min with rectangular or sinusoidal variations of the amplitude). Illumination by a light with wavelengths in the range 510 – 560 nm did not had a noticeable effect. Electromagnetic attraction between 3T3×cells (a variant derived from Swiss 3T3 cells) by near infrared field results in formation of aggregates[33]. Diameters of the cells are 22±5 μm. After ingestion of scattering particles (latex particles, polycrystalline diamond particles of 1 – 3 μm in diameter) the cell were hyperscattering. Intensity of scattering (I_{sc}) from hyperscattering cells was measured by fiber optics cable during irradiation by gallium aluminium-arsenide laser at the wavelength 830 nm. Cells were randomly plated on an adhesive strip of a substrate. The attached cells migrate along the strip and eventually form aggregates. The range of aggregation was five fold greater than the size of the cell at the wavelength of 800 nm and for the particle size 3 μm.

Interaction forces between red blood cells up to a distance of about 1 μm were also observed[34-39]. Interaction between erythrocytes is weakened or disappears if the cells are deprived of metabolic energy stores, if their membrane is disorganized, and if the quasi-static membrane potential is considerably lowered. Cell membrane fluctuations of human erythrocytes with frequency up to 30 Hz were

measured by point dark spectroscopy[40-44]. The dominant component of membrane fluctuations is metabolically excited and depends on a dynamic mechano-chemical coupling of the membrane-skeleton network. Nanoscale oscillations of membranes of erythrocytes are correlated in a certain time period and conditioned by energy supply, e.g. from intracellular MgATP. The actin's ATPase, located at the end of short actin filaments in spectrin submembrane skeleton, is responsible for the MgATP stimulation of red blood cell fluctuations. Mechanism of transformation of chemical to vibrational energy is not yet revealed. Levin and Korenstein conclude that the low frequency fluctuations of the cell membrane observed in erythrocytes may be a general property of living cells[40].

Coherent motion in biological systems was experimentally investigated using femtosecond laser spectroscopy. Nuclear motion in sub-millimeter wave bands was probed by stimulated emission in the near infrared band. Coherent vibrational motion performed in bacterial photosynthetic reaction centers was measured[45-51]. Membranes of bacteria (e.g. of *Rhodobacter capsulatus*) with genetically modified photosynthetic systems were used. Coherent oscillation at 15 and 77 cm^{-1} (0.45 and 2.3 THz) were observed on the time scale of about 2 ps[46]. Nonlinear effects and generation of signals with combined frequencies were observed. Pelling *et al.*[52-53] measured membrane mechanical oscillation of yeast cells (of *Saccharomyces cerevisiae*) using atomic force microscope (AFM). Frequency of oscillations depends on temperature (1.63 and 0.87 kHz at the temperature 30° and 22°C, respectively) and on the properties of the membrane (1.61 kHz of the undisturbed membrane and 0.73 and 0.86 kHz of the bud scar). Amplitudes of oscillation of an undisturbed membrane are up to 3-4 nm. After application of sodium azide (NaN_3), which switches off ATP production in the mitochondria but does not change the mechanical properties of the cell membrane, the cells do not display oscillatory motion. Pelling *et al.* assumed that large-scale forces are generated in yeast cells through the action of many proteins working in a concerted and cooperative manner and suggested the concerted action of motor protein[52].

Vibrations in cells may display special distribution corresponding to the condition of the minimum energy of the generated electromagnetic field[54]. Therefore, vibration of the plasma membrane may have a zonal character and only detection of vibrations at a very small area (very likely smaller than 0.5 × 0.5 μm) can yield a result such as the measurement using small dielectric particles[26], measurement by a light beam [40], or by a cantilever tip of AFM[52-53].

Regardless of the fact that measurement of the electromagnetic field generated by living cells represents only a limited partial picture of a variety of possibilities in nature, it proves existence of the biological electromagnetic fields and interactions mediated by them.

Fröhlich's theory of polar modes and coherent states

Fröhlich considered an electric polarization field $\boldsymbol{P}(\boldsymbol{r},t)$ coupled nonlinearly to an elastic field $\boldsymbol{A}(\boldsymbol{r},t)$ where $\boldsymbol{r}$, t are space and time coordinates[4-6]. The densities of kinetic (K) and potential (W) energy are given by the relations:

$$K = \frac{1}{2}\left(\dot{\mathbf{P}}^2 + \dot{\mathbf{A}}^2\right) \quad \ldots (1)$$

$$W = \frac{1}{2}\omega_0^2 P^2 + \left[\frac{1}{2}s^2\left(\nabla.\mathbf{A}\right)^2 + cP^2\nabla.\mathbf{A}\right] \quad \ldots (2)$$

where ω_0 is the circular frequency, s is the velocity of sound, and c is a coupling constant (Fröhlich also introduced a correction factor into the relation for W to provide reasonable behavior of the solution).

From the Lagrangean density $L = K - W$ equations of motion can be derived. For justified simplifications and for appropriate value of the coupling constant two branches of polarization waves exist – one with vanishing and a second with non-vanishing mean polarization. A metastable ferroelectric state may be formed. Due to electric field of the polarization waves long range nonlinear interactions can exist – interactions between vibration modes in the system and in the heat bath. Energy can be transferred between modes with different frequencies with participation of the heat bath.

The longitudinal polarization waves have discrete spectrum of normal modes whose frequencies ν_i are given by

$$\nu_1 \leq \Lambda \leq \nu_i \leq \Lambda \leq \nu_z \quad (3)$$

where $i = 1, 2, 3, \ldots z$.

Energy transfer between vibration modes of the polarization waves may be described by the rate equations[6-8]:

$$\dot{n}_i = s_i - \phi_i\left[n_i \exp(\beta\nu_i) - (n_i + 1)\right] - \sum_j \chi_{ij}\left[n_i(n_j + 1)\exp(\beta\nu_i) - (n_i + 1)n_j \exp(\beta\nu_j)\right] \quad \ldots (4)$$

where n_i is the occupation number of the i-th normal mode, s_i is the number of energy quanta supplied in unit time from the energy source (rate of energy supply), ϕ_i and χ_{ij} are the linear and the nonlinear transition probabilities, respectively, and $\beta = h/kT$, h is the Planck constant, k is the Boltzmann constant, and T is the temperature. A stationary solution of Eq. (4) may be derived for $\phi_i = \phi$, $\chi_{ij} = \chi$, and $s_i = s$ in the form:

$$n_i = A\frac{1}{\exp[\beta(\nu_i - \mu)] - 1} \quad \ldots (5)$$

where μ is the excitation potential ($0 \le \mu \le \nu_1$). For $\nu_1 \sim \mu$ energy condensed in the lowest frequency mode may be high. The stationary solution (5) displays energy condensation in the lowest frequency mode, which is a fundamental property of the Fröhlich's vibration system.

Two Fröhlich's systems mutually interact. The interaction energy I per one system may be expressed by an approximate relation[20]:

$$I = \frac{h}{2}\left[\frac{1}{2}\left(\sum_k n_k\nu_k\right)_{\mathrm{I}} + \frac{1}{2}\left(\sum_\lambda n_\lambda\nu_\lambda\right)_{\mathrm{I}} - \left(\sum_i n_i\nu_i\right)_0 - \left(\sum_j n_j\nu_j\right)_0\right] \quad \ldots (6)$$

where ν_i, ν_j (index 0) and ν_k, ν_λ (index I) are the frequencies of the systems before interaction and in interaction, respectively. The frequencies of the interacting systems are given by a simplified relation:

$$\nu^2 = \frac{\nu_i^2 + \nu_j^2}{2} \pm \sqrt{\left(\frac{\nu_i^2 - \nu_j^2}{2}\right)^2 + \left(Q^2\nu_i\nu_j\right)^2} \quad \ldots (7)$$

where Q is the modified interaction coefficient and ν represents ν_k or ν_λ.

The force acting between the vibration systems is given by the relation:

$$F = -2\frac{\mathrm{d}I}{\mathrm{d}R} = -2\frac{\mathrm{d}I}{\mathrm{d}Q}\frac{\mathrm{d}Q}{\mathrm{d}R} \quad \ldots (8)$$

where R is the distance.

Relations (4), (6) – (8) will be used for numerical modeling of the Fröhlich's vibration systems and interaction between them.

Models of the coherent systems

Numerical analysis enables us to assess effects of various parameters and conditions on the vibration system. Coupling of the system to the heat bath may depend on the condensed energy in the system. Let us assume that ϕ_i (χ_{ij}) have the same values for all the modes($\phi_i = \phi$ and $\chi_{ij} = \chi$) and that the ϕ transition probability depends on the occupation number of the lowest frequency mode in the form

$$\phi = \phi_0 \exp\left[\left(\frac{n_1 - n_{01}}{n_{\max}}\right)^p\right] \quad \ldots (9)$$

where n_{01} is occupation number in the thermodynamic equilibrium, $n_{\max}$ is the limit value of the occupation number in the lowest frequency mode (600 is used), and ϕ_0 is the coefficient of the linear transition probability. The parameter p determines the degree of the nonlinear dependence of the transition probability ϕ on excitation. The nonlinear transition probability is assumed to be given by the relation $\chi = 0.1\,\phi_0$.

Figure 1 shows excitation of vibration modes of a model Fröhlich's system (in mm and submm wave range). Energy condensation in the lowest frequency mode depends on energy supply from metabolic sources. The χ term in Eq. (4) describes energy transfer to the lower frequency modes as well as to the higher frequency modes. But due to different transition probabilities the resulting energy flow is directed from the higher frequency modes downwards

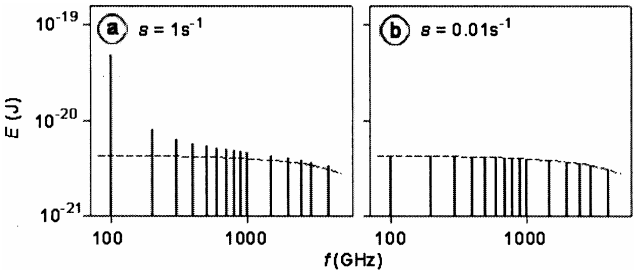

Fig. 1—Effect of energy supply on excitation of a Fröhlich's system in the mm and sub mm wave bands (energy versus frequency). The dashed lines denote thermal equilibrium values. Resulting energy flow is downwards along the frequency scale. Parameters: Coefficient of the linear transition probability $\phi_0 = 0.2$ s^{-1}, ϕ is given by Eq. (9) for $p = 4$ and $n_{\max} = 600$. Energy supply $s = 1$ (a) and $0.01 s^{-1}$ (b)

along the frequency scale. Some higher frequency modes are excited above the thermodynamic equilibrium values too. Condensation decreases with diminished energy supply. The greater the ϕ_0 value (i.e. the coupling to the heat bath), the smaller the excitation of the vibration system (Fig. 2).

The interaction forces between two vibration systems for p = 4 are shown in Fig. 3. The p value used represents strong nonlinear dependence of damping on excitation. (a similar model of interaction forces was analyzed by Pokorny[55]). For small values of ϕ_0 and sufficient excitation the interaction forces are negative, i.e. attractive. If one of the interacting systems is strongly excited the attractive forces are greater than in the case of weak excitation (excitation depends on the coupling to the heat bath and on the energy supply). The absolute values of the interaction forces decrease with increasing transition probabilities and with decreasing energy supply. Position of the rising part of the interaction forces curves is shifted to

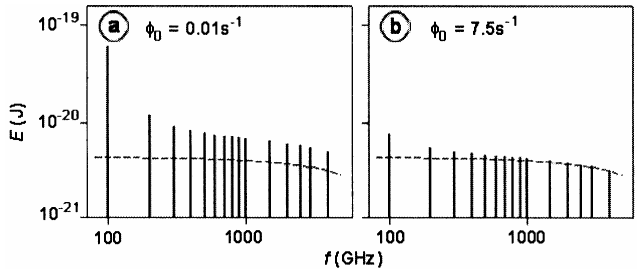

Fig. 2—Effect of ϕ_0 on excitation of the Fröhlich's system in the mm and sub mm wave bands (energy versus frequency). The dashed lines denote thermal equilibrium values. Parameters: s = 1 s^{-1}, p = 4, and n_{max} = 600. ϕ_0 is 0.01 (a) and 7.5 s^{-1} (b)

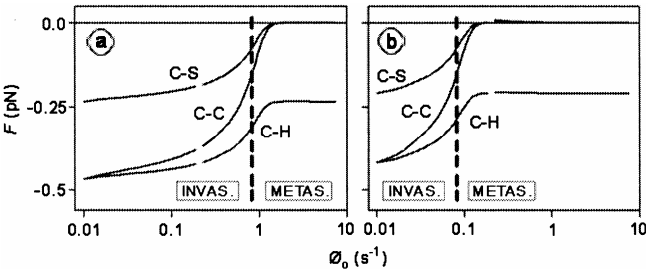

Fig. 3—Electromagnetic interaction forces F between two Fröhlich's systems versus coefficient of the linear transition probability ϕ_0 (coefficient of coupling to the heat bath). [C-H – interaction between a system with weak coupling to the heat bath (ϕ_0 = 0.01 s^{-1}) and a second system with ϕ_0 given on the abscissa; C-C - interaction between two systems with ϕ_0 given on the abscissa; C-S - interaction between a system with ϕ_0 given on the abscissa and a second system with strong coupling to the heat bath (ϕ_0 = 7.5 s^{-1}); Parameters: p = 4, n_{max} = 600. Energy supply s = 1 s^{-1} (a) and 0.1 s^{-1} (b)]

smaller values of ϕ_0 by decreased energy supply. For large values of ϕ_0 and small energy supply (s = 0.01 s^{-1}) the C-C and C-S curves have values near zero.The interaction forces are very small - attractive or even repulsive.

Measurement of mechanical and electrical vibrations of yeast cells – Preliminary results

Cold sensitive β-tubulin mutant *tub2-401* of *Saccharomyces cerevisiae* was used (strain CUY67 Mata *tub2-401 ura3-52 ade2-101*). The temperature dependence of microtubule polymerization is used to synchronize the cell culture. At the restrictive temperature (< 14°C) the cells cannot polymerize microtubules. The cells cultivated at the restrictive temperature continue in their develepment up to the beginning of the M phase where the cells are arrested. Increase of the temperature above the permissive one (above 25°C) triggers the synchronized entry of the arrested cells into the M phase. Non-synchronized cells are cultivated at the permissive temperature. At the permissive temperature microtubules can be polymerized and the cell evolution along the cell cycle is not influenced.

Mechanical vibrations of the membranes of the synchronized yeast cells were investigated by atomic force microscope (AFM) – MultiMode IV, Veeco. A cell in the sedimented layer was measured at the temperature of 28°-30°C. Figure. 4 shows oscillations at about 800 Hz.

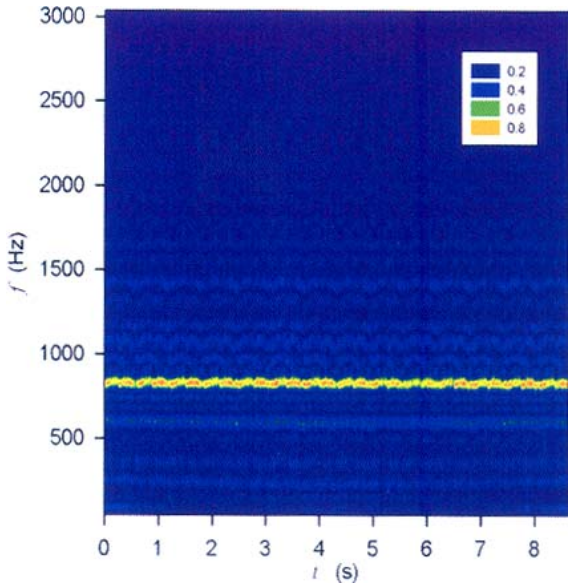

Fig. 4—Mechanical vibrations of the yeast cell membrane measured by AFM. (f – frequency, t – time).

Electrical oscillations in the frequency range 400-1600 Hz were measured by a spectrum analyzer Agilent E4448A controled by PC. Reference level was -80 dBm, and the resolution (RBW) and the video (VBW) bandwidths were 1 Hz. The investigated frequency range was examined by two scans, each of 600 Hz bandwidth and with the sweep time 1.85 s. The sum of both sweep times together with the time for transfer of the trace data into the PC memory was 6 s. A schematic picture of the measurement sytem is given in Fig. 5. The power gain of the preamplifiers is of about 110 dB. The electric oscillations were detected by a short segment of Pt wire (with diameter of 200 μm) obliquely cut to form a tip of about 50 nm. The tip of the wire was positioned 8 μm above a Pt covered bottom (grount contact) of a cuvette in the suspension with yeast cells. A cell in the layer of the sedimented cells was measured at the temperature of 28°-29°C.

Mean electric power of synchronized yeast cells, evaluated from 10 scans (i.e. within one min) at about 20 min after beginning of measurement, is shown in Fig. 6. The frequency of the spectral line at 800 Hz coincides with that of mechanical vibrations (Fig. 4).

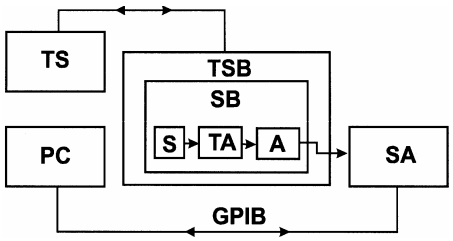

Fig. 5—Schematic diagramme of measurement system of electric potential at acoustic frequencies [SA=spectrum analyzer, TSB=temperature stabilized box, SB=threefold screened box, S=sensor, TA=transformation amplifier, A=amplifier, PC=computer, TS=temperature stabilizer]

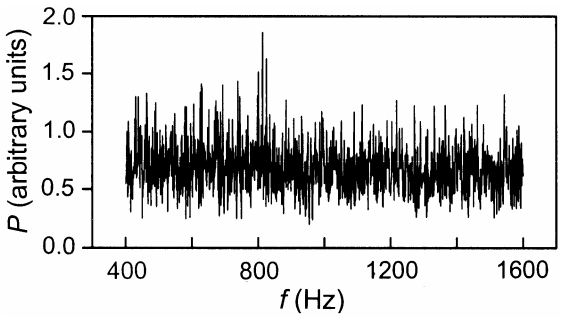

Fig. 6—Mean electric power of synchronized yeast cells versus frequency evaluated from 10 scans (in one min) at about 20 min after beginning of measurement.

Data from 400 double scans in the frequency range 400-1600 Hz were recorded for each individual measurement and the mean power evaluated. Figure 7 shows mean power of individual measurements of synchronized and non-synchronized yeast cells. Figure 8 shows the average value of the power of synchronized and of non-synchronized yeast cells evaluated from individual measurements in the frequency range 400 – 1600 Hz. The average value of the power of synchronized cells is of 9 % greater than the power of non-synchronized cells with statistical significance of 0.001.

Mitochondrial energy production

From thermodynamic point of view biological systems are far from equilibrium. Organization of living cells is conditioned by continuous energy

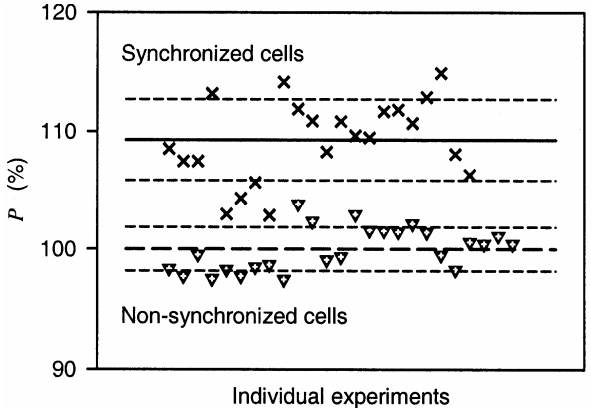

Fig. 7—Mean power of yeast cells measured 40 min in the frequency range 400-1600 Hz with 1 Hz RBW and VBW of the spectrum analyzer (400 double scans – one from 400 to 1000 and the other from1000 to 1600 Hz). Synchronized cells – crosses, non-synchronized cells – triangles.

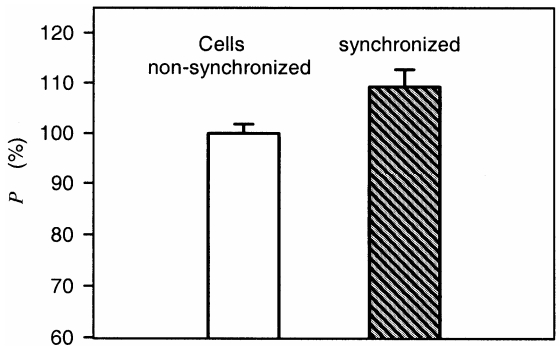

Fig. 8—The average value of the mean power of synchronized and of non-synchronized yeast cells evaluated from individual measurements in the frequency range 400 – 1600 Hz. The average value of power of synchronized cells is of 9 % greater than the power of non-synchronized cells with statistical significance 0.001.

supply from energy sources. Energy production in living cells is closely connected with basic processes of life. In the absence of sufficient energy producing activity of mitochondria the anaerobic glycolysis is stimulated.

Study of energy production system and its changes are included in investigation of cancer cells. In normal cells mitochondrial ATP production exceeds production by glycolysis. Pyruvate, the final output of fermentation after entering a mitochondrion is transformed to acetyl coenzyme A (acetyl CoA). Nearly 60 % of acetyl CoA energy is not utilized for ATP production in a mitochondrion. Wasted energy flows out from mitochondria (which are assembled along microtubules) in the form of heat and may have effects on microtubules and their ambient medium. At least a part of the heat energy may be absorbed by microtubules.

Energy production in mitochondria is supressed by cancer transformation and the vast amount of energy production is switched to glycolysis in cytoplasm of the cell – the Warburg effect[56-57]. Structure and functioning of mitochondria are changed and the pathway for energy production is blocked. The energy deficiency, under which the cells operate, is a driving force to the increase of fermentation. It is proved that alterations in respiratory activity and mtDNA (mitochondrial deoxyribonucleic acid) abnormalities appear to be a general feature of malignant cells[58]. For instance, the function of mitochondria is repressed in human liver, kidney, and colon carcinogenesis[59]. Energy substrate availability could play an important role in mitochondrial disfunction. Defective mitochondrial system in cancer cells can be improved by changing substrate availability[60]. Iron-sulfur clusters processes in mitochondria are important biosynthetic pathway of life[61], but their role in cancerogenesis is not yet fully understood.

Frataxin can activate mitochondrial energy production[62]. Formation of frataxin increases the activity of the electron transport chain, the mitochondrial membrane potential, and ATP production[63], and diminishes malignant transformation *in vitro*[64]. Impaired mitochondrial function and tumor growth is connected with disruption of frataxin expression[65]. In cancer cells blockade of ATP production in mitochondria is caused by phosphorylation of pyruvate dehydrogenase (PDH) enzyme family converting pyruvate to acetyl CoA. The ability of mitochondria to generate energy is booted by DCA (dichloracetate) which activates PDH[66] Functioning mitochondria help to restore the normal cellular functions and to switch on the apoptosis of aberrant cells.

In cancer cells ATP production by mitochondria is replaced by production in glycolytic processes. Warburg predicted, that the cells need their respiratory energy production to preserve their structure[57]. If the activity of mitochondria is important, then the main influence may be connected with the wasted energy efflux and/or with the electric field of the mitochondrial membrane.

Energy absorbed from mitochondrial heat efflux, together with energy supplied by motor proteins, and from hydrolysis of GTP to GDP in β tubulins after polymerization can excite vibration states in microtubules. It seems reasonable to assume that microtubules form the main structure generating electromagnetic field in eukaryotic cells that contributes to interactions and organization inside the cell and between cells. Therefore, reduction or cut off the wasted energy efflux from mitochondria may be one of the main agents causing disturbances of the coherent vibrations and reduction of coherence of the endogenous electromagnetic field.

Disintegration of the cytoskeleton

Actin filaments, intermediate filaments, and microtubules are the cytoskeleton components which together with accessory proteins participate in cell motility, ribosomal and vesical transport, mitosis, and transduction of pressure and tension. Elastic modulus of living cell is determined by the state of the cell and can be changed in dependence on internal cellular organization, on biochemical conditions, and on pathological state. The cytoskeleton functions are well organized, regulated, and synchronized. Morphological changes caused by the cytoskeleton on the pathway of malignant transformation are used for cancer diagnosis. Cancer evolution induces cytoskeleton defects before malignant properties of the cell are created.

Mechanical stiffness of nontumorigenic human epithelial breast cells MCF-10, nonmetastatic cancer cells MCF-7, and modified MCF-7 cells (denoted as modMCF-7 cells) with increased metastatic potential by treatment with TPA (12-*Q*-tetradecanoylphorbol-13-acetate) was measured using a stretcher with optically induced forces by two counterpropagating divergent laser beams published by Guck *et al*.[67]

(Fig. 9). Chemically modified modMCF-7 cells display increased metastatic potential. Deformability of the nonmetastatic cancer MCF-7 cell is twice greater compared with healthy MCF-10 cell, and deformability of modMCF-7 cell with increased metastatic potential is three times greater. The stiffness of the epithelial breast cells displays alterations of the cytoskeleton properties and organization.

The relationships among biomechanical properties, cellular structure, and cancer evolution are analyzed by Suresh and coworkers[68,69]. Figure 10 contains description of changes of mechanical properties of a cell on its cancer evolution pathway[69]. We assume that the cytoskeleton changes are followed by disturbances of the Fröhlich's coherent states. Changes of the cytoskeleton structure followed by deformability and cellular adherence defects are characteristic precursors of cell malignant properties.

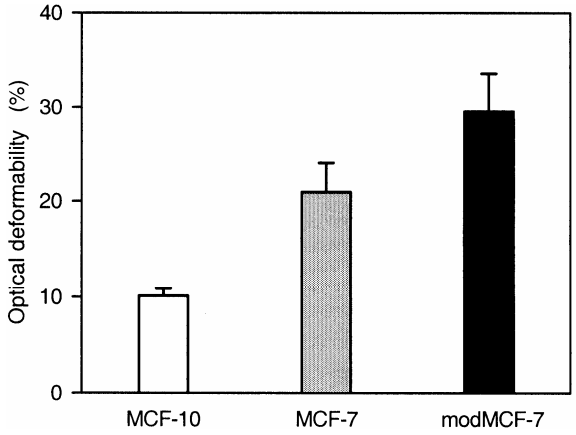

Fig. 9—Effect of TPA on cancer cells. Relative optical deformability of healthy (MCF-10), cancer (MCF-7), and TPA modified metastatic cancer (modMCF-7) human breast epithelial cells. Adapted from Guck *et al.*[67]

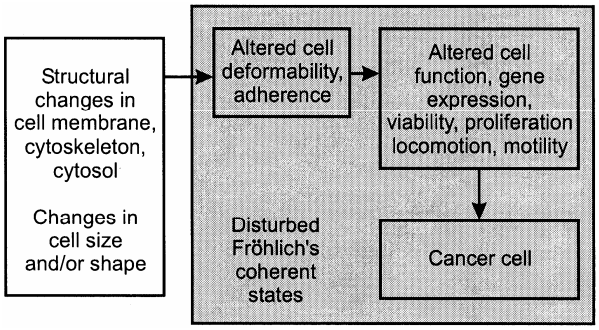

Fig. 10—A schematic diagramme of the changes of the mechanical properties in cells on the cancer evolution pathway as were described in [69]. Cytoskeleton structural changes might result in the changes of the Fröhlich's coherent states.

Cytoskeleton defects and mechanical properties of human cancer pancreatic cells Panc-1 were investtigated[68,69]. Keratin network in the cell is reorganized and disturbed after treatment with sphingosylphosphorylcholine (SPC) which plays critical role in invasion and metastasis of gastrointestinal tumors[70]. Treatment of Panc-1 cells with SPC leads to phosphorylation and reorganization of keratin into a ring-like structure around the nucleus within 45 min. After application of SPC the normalized distance between the outer edge of the keratin structure and the nucleus membrane is reduced to about 50 % of the unperturbed value (Fig. 11). Deformability of the Panc-1 cells before and after treatment with SPC was measured using the microplate mechanical stretcher method. The relative elastic spring constant decreases to about 40 % (Fig. 11) and the energy dissipated per one cycle of displacement increases to 400 % after SPC treatment. The Fröhlich's coherent vibrations may be disturbed as a consequence of cytoskeleton disintegration.

Organization of the mitotic spindle can be disturbed by external electric field with intensity 1-2 V/cm and frequency 100 – 300 kHz in the cytokinetic phase of cell division as was published by Kirson *et al.*[71,72] and Cucullo *et al.*[73]. In the narrow midbody region where actin filament contracting ring creates clevage furrow the external electric field has

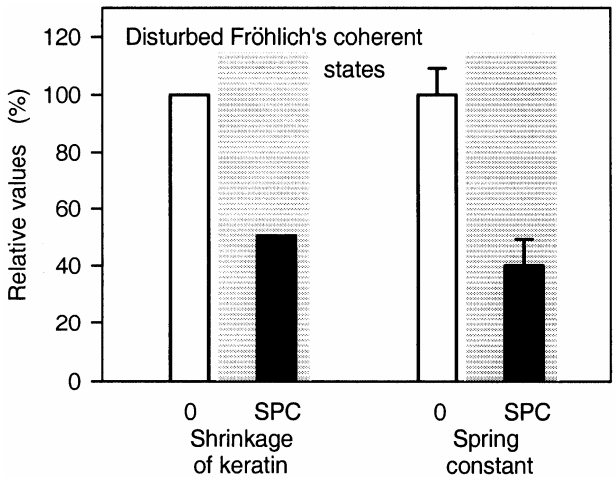

Fig. 11—Collapse of keratin network – biomechanical properties of Human Panc-1 cancer cells before (the white columns) and 60 min after (the black columns) SPC treatment. The normalize distance between outer edge of the keratin cytoskeleton distribution and the membrane of the nucleus and the effective elastic spring constant of the cells are shown. Data on keratin collapse and on spring constant are taken from Suresh and coworkers[68,69]. Assumed disturbance of the Fröhlich's coherent states is included.

greater intensity than the electric field generated by the polar microtubular tip. The forces exerted by the external field on tubulin heterodimers prevent their correct orientation and attraction to the close vicinity of the tip and, therefore, disturb polymerization of microtubules. The cells (*e.g.* melanoma cells) are arrested in mitosis by the external field and/or the cells are disintegrated. This effect, *i.e.* disruption of normal polymerization-depolymerization process, shows biological significance of endogenous electric field generated by microtubules.

Malignity of cancer cells

At certain stage of cancer development cancer cells begin to burrow their way into the surrounding tissue. A sprout insinuates itself amongst the surrounding normal cells moving outwads from the locus of its origin. The cell surface and the surrounding structures with which the cell surface comes into contacts are of profound importance for the cell invasive movements. Invasion seems to be a result of force effects. We will not disscussed here the molecular and biochemical interactions. We present biophysical hypothesis of electromagnetic field forces between cells. Fig. 12 shows possible force effects. Cell as a whole represents the Fröhlich's vibration system generating electromagnetic field which mediates interaction between cells[55]. If the electromagnetic forces between cancer cells are smaller than forces between healthy cells and cancer cells, then the cancer cells are pulled by healthy cells inside the healthy tissue (Fig. 12).

We assume that along the pathway of cancer evolution the Fröhlich's vibrations are progressively deteriorated by the increase of the linear transition probability ϕ_0 (indicating energy losses from the Fröhlich's system)[20], by the decrease of enegy supply, and by disitegration of the cytoskeleton. Values of ϕ_0 on the horizontal axes of Fig. 3 may also be understood as representing a transformed level of phosphorrylation. The C-H curves determine the interaction forces between a „current" cancer cell (C) with a value of ϕ_0 given on the abscissa and a healthy cell (H), the C-C curves the interaction forces between two current cancer cells, and C-S curves the interaction forces between a „current" cancer cell and a cancer cell with strong coupling to the heat bath (S). If the attractive forces between a healthy cell and a cancer cell (C-H curves) are considerably greater than attractive forces between two cancer cells (C-C curves), cancer cells are pulled by healthy cells out of the tumor (Fig. 3). For higher ϕ_0 values (higher phosphorylation level) the forces acting between cancer cells are very small (attractive or repulsive) and cancer cells can detach. The vertical dashed lines may delineate approximate regions of local invasion and of metastatic growth.

Reduced energy supply to the Fröhlich's coherent vibrations can shift the regions of local invasion and of metastasis to smaller values of ϕ_0 (as is shown in Fig. 3b).

We may conclude that adherence forces between cells determine their behavior – whether cells adhere to or separate from one another.

Figure 13 shows a schematic picture of a pathway of cancer transformation of a cell. Cytoskeleton, the locus of the Fröhlich's coherent vibrations, is phosphorylated and disorganized and wasted energy efflux from mitochondria is diminished. Coherent states are disturbed. Therefore, cancer is a disease including biophysical processes. Malignity of a cancer cell can be explained by defects of the Fröhlich's coherent states and of the generated endogenous cellular electromagnetic field.

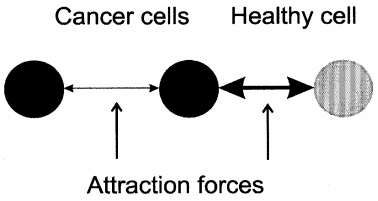

Fig. 12—A model of force interactions between cancer and healthy cells. A cancer cell is pulled into a healthy tissue by a force exerted by a healthy cell overcoming cancer cell attraction. In a biological system the forces are acting between cancer and healthy tissues.

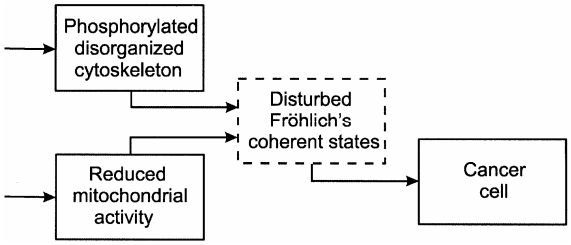

Fig. 13—A schematic hypothetical diagramme of the role of disturbances of the Fröhlich's coherent states in cancer transformation of a cell. Phosphorylation of proteins resulting in increased coupling of protein and protein structures to the ambient medium, disorganization of the cytoskeleton, and diminished mitochondrial activity with reduced wasted energy efflux are assumed to cause low excitation and disturbances of coherent vibrations and of generated bioelectromagnetic field.

Conclusion

The Fröhlich's hypothesis of coherent polar vibrations in living matter is based on nonlinear interaction of electric polarization and longitudinal elastic fields. Energy supplied from metabolic sources is not thermalized and coherent state is formed. Microtubules in the cytoskeleton satisfy requirements for excitation of coherent vibrations and generation of the electromagnetic field, which has organization role in the cell, mediates directed transport of dielectric particles and of electrons and interactions between cells.

On the pathway of cancer evolution activity of mitochondria is reduced, wasted energy effux to the vicinity of microtubules diminished, and cytoskeleton in the cell disintegrated. As a result endogenous electromagnetic field and its coherence are disturbed. Organization inside cell and interactions between cells are damaged. Cancer is a disease of the energy producing system with reduced mitochondrial activity (the Warburg effect) and of cytoskeleton disintergration. We assume that cancer is a disease of coherent electrical polar states (of the Fröhlich's vibrations), and of the endogenous electromagnetic field too. Biological electromagnetic field is already measured and its existence proved. Further progress in measurement based on nanotechnological detection and amplification systems may disclose electromagnetic differences between healthy and cancer cells.

References

1 Alberts B, Bray D, Lewis J, Raff M, Roberts K & Watson J D, *Molecular biology of the cell*. third ed. (Garland Pub., New York & London) 1994.

2 Abercrombie M & Ambrose E J, The surface properties of cancer cells: A review, *Cancer Res.* 22 (1962) 525.

3 Fröhlich H, Coherent electric vibrations in biological systems and cancer problem, *IEEE Trans*, MTT-26 (1978) 613.

4 Fröhlich H, Collective behaviour of non-linearly coupled oscillating fields (with applications to biological systems), *J. Collective Phenom,* 1 (1973) 101.

5 Fröhlich H, Long range coherence in biological systems, *Riv. del Nuovo Cimento,* 7 (1977) 399.

6 Fröhlich H, The biological effects of microwaves and related questions, *Adv Electronics Electron Phys,* 53 (1980) 85.

7 Fröhlich H, Bose condensation of strongly excited longitudinal electric modes, *Phys. Lett*, 26A (1968) 402.

8 Fröhlich H, Long-range coherence and energy storage in biological systems, *Int. J. Quant. Chem,* II (1968) 641.

9 Fröhlich H, Organisation and long range selective interaction in biological and other pumped systems, in *Synergetics– Cooperative phenomena in multi-component systems*, edited by H Haken (B G Teubner, Stuttgart) 1973, 241.

10 Fröhlich H, Coherent excitations in biological systems (in Russian: Kogerentnyje vozbuzhdenija v biologicheskikh systemach.) *Biofizika XXII*, (1977) 743.

11 Reed E J, Soljačić M, Gee R, Joannopoulos J D, Coherent optical photons from shock waves in crystals, *Phys. Rev. Lett*, 96 (2006) 013904-1.

12 Fröhlich H, Quantum mechanical concepts in biology, in *Theoretical physics and biology,* edited by M Marois (North Holland, Amsterdam) 1969, 13; (Proc. 1st Int.Conf. on Theor. Phys. Biol., Versailles, 1967).

13 Fröhlich H, The connection between macro- and microphysics, *Riv. del Nuovo Cimento,* 3, (1973), 490.

14 Fröhlich H, Biological Control through long range coherence, in *Synergetics*, ediyted by H Haken, (Springer, Berlin-Heidelberg-New York) 1977, 241.

15 Fröhlich H, General introduction, in *From theoretical physics to biology*, edited by M Marois, (S. Karger, Basel) 1973, 2.

16 Fröhlich H, Long range coherence and the action of enzymes, *Nature,* 228 (1970) 1093.

17 Fröhlich H, The extraordinary dielectric properties of biological materials and the action of enzymes, *Proc. Natl. Acad. Sci. USA 72*, 1975, 4211-4215.

18 Fröhlich H, Selective long range dispersion forces between large systems, *Phys. Lett,* 39A, (1972) 153.

19 Fröhlich H, Possibilities of long- and short-range electric interactions of biological systems, *Neurosci. Res. Program Bull,* 15 (1977) -72.

20 Pokorný J& Wu.-M. *Biophysical aspects of coherence and biological order* (Academia, Praha, Czech Republic; Springer, Berlin - Heidelberg - New York) 1998.

21 Pokorný J, Endogenous electromagnetic forces in living cells: Implications for transfer of reaction components, *Electro-Magnetobiol,* 20 (2001) 59.

22 Pokorný J, Viscous effects on polar vibrations in microtubules, *Electromag. Biol. Med,* 22, 2003, 15.

23 Pokorný J, Excitation of vibration in microtubules in living cells, *Bioelectrochem,* 63 (2004) 321.

24 Pokorný J, Hašek J & Jelínek F, Electromagnetic field in microtubules: Effects on transfer of mass particles and electrons, *J. Biol. Phys,* 31 (2005) 501.

25 Pokorný J, Hašek J & Jelínek F, Endogenous electric field and organization of living matter, *Electromag. Biol. Med,* 24 (2005) 185.

26 Pohl H A, Oscillating fields about growing cells, *Int. J. Quant. Chem. Quant. Biol. Symp,* 7 (1980) 411.

27 Pohl H A, Braden T, Robinson S, Piclardi J & Pohl D G, Life cycle alterations of the micro-dielectrophoretic effects of cells, *J. Biol. Phys.,* 9 (1981)133.

28 Roy S C, Braden T & Pohl H A, Possibility of existence of pseudoferroelectric state in cells: Some experimental evidence, *Phys. Lett,* 83A (1981) 142.

29 Pohl H, A, *Dielectrophoresis* (Cambridge Univ. Press, London) 1978.

30 Hölzel R & Lamprecht I, Electromagnetic fields around biological cells, *Neural Net. World,* 4 (1994) 327.

31 Albrecht-Buehler G. Rudimentary Form of Cellular 'Vision'. *Proc. Natl. Acad. Sci. USA 89*, 1992, 8288-8293.

32 Albrecht-Buehler G, Surface extensions of 3T3 cells towards distant infrared light sources, *J. Cell Biol,* 114 (1991) 493.

33 Albrecht-Buehler G, A long-range attraction between aggregating 3T3 cells mediated by near-infrared light scattering, *PNAS* 102 (2005) 5050.

34 Rowlands S, Sewchand L S, Lovlin R E, Beck J S & Enns E G, A Fröhlich interaction of human erythrocytes, *Phys. Lett,* 82A (1981) 436.

35 Rowlands S & Sewchand L S, Quantum mechanical interaction of human erythrocytes, *Can. J. Physiol. Pharmacol*, 60 (1982) 52.

36 Rowlands S, Sewchand L S & Enns E G, Further evidence for a Fröhlich interaction of erythrocytes, *Phys. Lett,* 87A (1982) 256.

37 Rowlands S, Coherent excitation in blood, in *Coherent excitation in biological systems*, edited by H Fröhlich and F Kremer (Springer, Berlin-Heidelberg-New York) 145.

38 Sewchand L S & Rowlands S, Specificity of the Fröhlich interaction of erythrocytes, *Phys. Lett,* 93A (1983) 363.

39 Rowlands S, The interaction of living red blood cells, in *Biological coherence and response to external stimuli*, edited by H Fröhlich (Springer, Berlin-Heidelberg-New York) 1988, 171.

40 Levin A & Korenstein R, Membrane fluctuations in erythrocytes are linked to MgATP-dependent dynamic assembly of the membrane skeleton, *Biophys. J,* 60 (1991) 733.

41 Tuvia S, Almagor A, Bitler A, Levin S, Korenstein R & Yedgar S, Cell membrane fluctuations are regulated by medium macroviscosity: Evidence for a metabolic driving force, *Proc. Natl. Acad. Sci. USA,* 94 (1997), 5045.

42 Tuvia S, Bitler A & Korenstein R, Mechanical fluctuations of the membrane-skeleton are dependent on F-actin ATPase in human erythrocytes, *J. Cell Biol,* 141 (1998) 1551.

43 Tuvia S, Moses A, Gulayev N, Levin S & Korenstein R, β-Adrenergic agonists regulate cell membrane fluctuation of human erythrocytes, *J. Physiol,* 516 (1999) 781.

44 Bitler A & Korenstein R, Nano-scale fluctuations of red blood cell membrane reveal nonlinear dynamics, *Biophys. J*, 86 (2004) 582.

45 Vos M H, Lambry J C, Robles S J, Youvan D C, Breton J & Martin J-L, Direct observation of vibrational coherence in bacterial reaction centers using femtosecond absorption spectroscopy, *Proc. Natl. Acad. Sci. USA,* 88 (1991) 8885.

46 Vos M H, Rappaport F, Lambry J C, Breton J & Martin J-L Visualization of coherent nuclear motion in a membrane protein by femtosecond spectroscopy, *Nature,* 363 (1993) 320.

47 Vos M H, Jones M R, Hunter C N, Breton J, Lambry J C & Martin J-L, Coherent dynamics during the primary electron-transfer reaction in membrane-bound reaction centers of *Rhodobacter sphaeroides, Biochem,* 33 (1994) 6750.

48 Vos M H, Jones M R, Hunter C N, Breton J & Martin J-L, Coherent nuclear-dynamics at room temperature in bacterial reaction centers, *Proc. Natl. Acad. Sci. USA,* 91 (1994) 12701.

49 Vos M H, Jones M R & Martin J-L, Vibrational coherence in bacterial reaction centers: spectroscopic characterisation of motions active during primary electron transfer, *Chem. Phys,* 233 (1998) 179.

50 Vos M H, Rischel C, Jones M R & Martin J-L, Electrochromic detection of a coherent component in the formation of the charge pair $p^+h_l^-$ in bacterial reaction centers, *Biochem,* 39 (2000) 8353.

51 Vos M H, Jones M R, McGlynn P, Hunter C N, Breton J & Martin J-L, Influence of the membrane environment on vibrational motions in reaction centers of *Rhodobacter sphaeroides, Biochem. Biophys. Acta,* 1186 (1994) 117.

52 Pelling A E, Sehati S, Gralla E B, Valentine J S & Gimzewski J K, Local nano-mechanical motion of the cell wall of *Saccharomyces cerevisiae, Science,* 305 (2004) 1147.

53 Pelling A E, Sehati S, Gralla E B & Gimzewski J K, Time dependence of the frequency and amplitude of the local nanomechanical motion of yeast, *Nanomedicine: Nanotechnol. Biol. Med,* 1 (2005) 178.

54 Pokorný J, Fiala J & Vacek K, Fröhlich coherent vibrations and raman scattering, *Czechoslovak J. Phys,* 41 (1991) 484.

55 Pokorný J, The role of Fröhlich's coherent excitations in cancer transformation of cells, in *Herbert Fröhlich, FRS: A physicist ahead of his time*, edited by G J Hyland and P Rowlands (The University of Liverpool) 2006, 177.

56 Warburg O, Posener K & Negelein E, Über den Stoffwechsel der Carcinomzelle, *Biochem Z,* 152 (1924) 309.

57 Warburg O, On the origin of cancer cells, *Science,* 123 (156) 309.

58 Carew J S & Huang P, Mitochondrial defects in cancer, *Mol. Cancer,* 1 (2002) 9.

59 Cuezva J M, Krajewska M, López de Heredia M, Krajewski S, Santamaria G, Kim H, Zapata J M, Marusawa H, Chamorro M & Reed J C, The bioenergetic signature of cancer: A marker of tumor progression, *Cancer Res,* 62 (2002) 6674.

60 Rossignol R, Gilkerson R, Aggeler R, Yamagata K, Remington S J & Capaldi R A, Energy substrate modulates mitochondrial structure and oxidative capacity in cancer cells, *Cancer Res,*64 (2004) 985.

61 Lill R & Mühlenhoff U, Iron-sulfur-protein biogenesis in eukaryote, *Trends Biochem. Sci,* 30 (2005) 133.

62 Ristow M, Pfister M F, Yee A J, Schubert M, Michael L, Zhang Ch-Yu, Ueki K, Michael M D II, Lowell B B & Kahn C R, Frataxin activates mitochondrial energy conversion and oxidative phosphorylation, *Proc. Natl. Acad. Sci. USA*, 97 (2000) 12239.

63 González-Cabo P, Vázquez-Manrique R P, Garcia-Gimeno M A, Sanz P & Palau F, Frataxin interacts functionally with mitochondrial electron transport chain proteins, *Human Molec. Genetics,* 14 (2005), 2091.

64 Schoichet S A, Bäumer A T, Stamenkovic D, Sauer H, Pfeiffer A F H, Kahn C R, Müller-Wieland D, Richter Ch & Ristow M, Frataxin promotes antioxidant defense in a thiol-dependnt manner resulting in diminished malignant transformation *in vitro*, *Human Molec. Genetics,* 11 (2002) 815.

65 Thierbach R, Schulz T J, Isken F, Voigt A, Mietzner B, Drewes G, von Kleist-Retzow J-Ch, Wiesner R J, Magnuson M A, Puccio H, Pfeiffer A F H, Steinberg P & Ristow M, Targeted disruption of hepatic frataxin expression causes impaired mitochondrial function, decreased life span and tumor growth in mice, *Human Mol. Genetics,* 14 (2005) 3857.

66 Bonnet S, Archer S L, Allalunis-Turner J, Haromy A, Beaulieu Ch, Thompson R, Lee Ch T, Lopaschuk G D, Puttagunta L, Bonnet S, Harry G, Hashimoto K, Porter Ch J, Andrade M A, Thebaud B & Michelakis E D, A mitochondria-K^+ channel axis is suppressed in cancer and its

normalization promotes apoptosis and inhibits cancer growth, *Cancer Cell,* 11 (2007) 37.

67 Guck J, Schinkinger S, Lincoln B, Wottawah F, Ebert S, Romeyke M, Lenz D, Erickson H M, Ananthakrishnan R, Mitchell D, Käs J, Ulvick S & Bilby C, Optical deformability as an inherent cell marker for testing malignant transformation and metastatic competence, *Biophys. J,* 88 (2005) 3689.

68 Suresh S, Spatz J, Mills J P, Micoulet A, Dao M, Lim C T, Beil M & Seufferlein T, Connections between single-cell biomechanics and human disease states: gastrointestinal cancer and malaria, *Acta Biomaterialia,* 1 (2005) 15.

69 Suresh S, Biomechanics and biophysics of cancer cells, *Acta Materialia,* 55 (2007) 3989.

70 Beil M, Micoulet A, von Wichert G, Paschke S, Walther P, Omary M B, Van Veldhoven P P, Gern U, Wolff-Hieber E, Eggermann J, Waltenberger J, Adler G, Spatz J & Seufferlein T, Sphingosylphosphorylcholine regulates keratin network architecture and visco-elastic properties of human cancer cells, *Nature Cell Biol*, 5 (2003) 803.

71 Kirson E D, Gurvich Z, Schneiderman R, Dekel E, Itzhaki A, Wasserman Y, Schatzberger R & Palti Y, Disruption of cancer cell replication by alternating electric fields, *Cancer Res*, 64 (2004) 3288.

72 Kirson E D, Dbalý V, Tovaryš F, Vymazal J, Soustiel J F, Itzhaki A, Mordechovich D, Steinberg-Shapira S, Gurvich Z, Schneiderman R, Wasserman Y, Salzberg M, Ryffel B, Goldsher D, Dekel E & Palti Y, Alternating electric fields arrest cell proliferation in animal tumor models and human brain tumors, *PNAS,* 2104 (2007) 10152.

73 Cucullo L, Dini G, Hallene K L, Fazio V, Ilkanich E V, Igboechi Ch, Kight K M, Agarwal M K, Garrity-Moses M & Janigro D, Very low intensity alternating current decreases cell proliferation, *Glia,* 51 (2005) 65.