VIBRATION IN MICROTUBULES

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INTRODUCTION

Living cells are structurally and dynamically organized by networks of protein polymers called the cytoskeleton. The cytoskeleton is a highly dynamic structure which reorganizes continuously as the cell changes its shape, divides, and responds to the environment[1,2]The cytoskeleton is composed of intermediate filaments, actin filaments (which are also called microfilaments), and of microtubules. The filaments and the microtubules are mutually connected and form a three dimensional network in the cell. The cytoskeleton enables transport of molecules, structures, and organelles (by means of motor proteins), receives information signals from cellular environment through the membrane proteins, and has a basic role in the mitotic spindle.

The microtubules are the main organizers of the cytoskeleton. They are formed by cylindrical structures (with a diameter of about 30nm - Fig. 1a) composed of 13 (or 14) protofilaments which are one dimensional chains of heterodimer tubulin molecules. A tubulin molecule is formed by two globular proteins - α- and β-tubulins; each of them has the relative molecular mass of about 55000 ("molecular weight" in daltons) but the masses are not equal. Conformation of the heterodimer molecule can be changed from the α state (non tilted) to the β state (tilted by 29 degrees from the microtubule axis) as is shown in Fig. 1b. The heterodimer molecule is an electric dipole[3-5]. Therefore, the microtubules are polar structures with the plus end (fast growing) and the minus end (slow growing) embedded in the centrosome - a center of the cytoskeleton. The half-lifetime of a microtubule is of about 10min.

After polymerization the guanosine triphospate molecules (GTP) bound to the β-tubulins in a heterodimer are hydrolyzed to guanosine diphosphate (GDP). The majority of the hydrolysis energy is stored in the microtubule[2,6]. The hydrolysis energy is not used for polymerization.

The cytoskeleton exerts forces and generates movements without any major chemical changes[2]. The mechanisms of the cytoskeleton activity are still not well understood. Physical mechanisms are probably a background for the cytoskeleton function. We may assume that the cytoskeleton can use the energy from hydrolysis of GTP stored in microtubules. We will analyse vibrations in microtubules as they may be one of possible mechanisms underlying the cytoskeleton function.

ANALYSIS OF VIBRATION

The tubulins in a protofilament form a one dimensional chain with translation symmetry. The tubulins have internal vibrations (exerted by atoms and by parts of the tubulins) and external vibrations in the chain (i.e. vibrations exerted by each tubulin as a whole). We will analyse the external vibrations in the protofilaments using a method similar to that employed in the solid state physics[7]. (We will not discuss here the differences between symmetrical crystalline solids and microtubules as it is beyond the scope of this short contribution; it may be found in[8].) The external vibrations will be analyzed without including dynamic deformations of tubulin globules.

Fig. 2 shows a scheme of the one dimensional chain with translation symmetry (e.g. of a protofilament). The circles denote the centers of mass of tubulins in equilibrium positions, the black spots show the theoretical equidistant control

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points (the distance between them is a), Δ_1, Δ_2 are distances of tubulins from the corresponding control points, m_1, m_2 are masses of tubulins, and f_1, f_2 are the elastic force constants which need not be equal. If only the interactions between the closest neighbours are assumed then the one-dimensional equations of motion can be written in the form

$$m_1 \ddot{q}_j = f_2(q_{j-1} - q_j) + f_1(q_{j+1} - q_j)$$

$$m_2 \ddot{q}_{j+1} = f_1(q_j - q_{j+1}) + f_2(q_{j+2} - q_{j+1}) \qquad (1)$$

where q_{j-1}, q_j, q_{j+1}, q_{j+2} and $\ddot{q}_{j-1}, \ddot{q}_j, \ddot{q}_{j+1}, \ddot{q}_{j+2}$ are the displacements from equilibrium positions and their second time derivatives, respectively. (The equations (1) have linear form but for $f_1 \neq f_2$ the vibrations are anharmonic as will be discussed further.) Assuming the solution of Eq. (1) in the form of propagating waves we get the dispersion relation

$$\omega_{\pm} = \frac{(f_1 + f_2)(m_1 + m_2)}{2m_1 m_2} \pm \sqrt{\left[\frac{(f_1 + f_2)(m_1 + m_2)}{2m_1 m_2}\right]^2 - 4\frac{f_1 f_2}{m_1 m_2}\sin^2 ka} \qquad (2)$$

where k is the absolute value of the wave vector, and ω_+ and ω_- denote the optical branch and the acoustical branch of vibrations, respectively. In the acoustical branch the neighbouring particles (tubulins) move in the same direction and in the optical branch the particles move in opposite directions under condition that the center of gravity remains at rest (similar results are proved in [7]). The term vibration mode will be used for the waves propagating in both directions along the chain with the same absolute value k of the wave vector. The wave vector can acquire certain number of discrete values on account of discreteness of the chain.

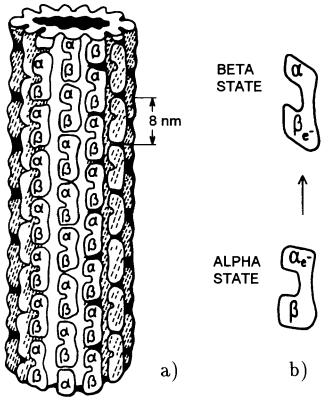


Figure 1. A schematic picture of a microtubule (a) and of the conformation change of a tubulin heterodimer from the α state to the β state. After[4].

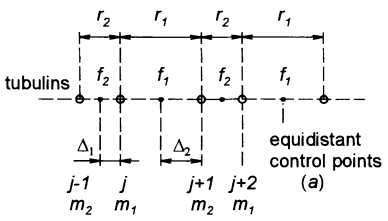


Figure 2. Translation symmetry in a protofilament.

Protofilaments in a microtubule are bound and build up a wall of a cylinder. Vibrations in particular protofilaments are not independent and form a common vibration system in a microtubule. As a microtubule is composed of 13 protofilaments we assume, that a microtubule "particle" has 13 times greater mass than a protofilament "particle". Nevertheless, the elastic force constants in isolated parts of protofilaments (oligomers) and in microtubules may be different. Frequency versus absolute value of the wave vector evaluated from Eq. (2) is shown in Fig.3a for $f_1 = f_2$ and in Fig. 3b for $f_1 = 10f_2$. The elastic force constants are parameters of the curves. The elastic force constants might be assessed from the shear strain-stress experimental data [9-11]. The dynamic elastic shear moduli for microtubules are of about $5 Nm^{-2}$. [10] Using relation between the elastic shear modulus and Young's modulus and assuming the distance between neighbouring particles about 4nm the elastic force constants might be about 10^{-5}-$10^{-6} Nm^{-1}$. Nevertheless, filaments in the cytoskeleton have different stiffness. The large elastic modulus appears to depend on the fact that filaments are arranged orthogonally and cross-linked to each other at junction points. Changes of viscoelasticity very likely come about by changing the nature of the cross-links or the filament length. The elastic force constants can vary in a wide range of values. To show their effect on frequency we used several values of the elastic force constants in Fig. 3. For a microtubule the lowest frequency for $\nu \neq 0$ of the acoustical branch is in the band of 5 - 50MHz if the length of a microtubule is 10μm (=λ /2) and velocity of the acoustical waves is in the range from 10^2 to $10^3 ms^{-1}$. If the elastic force constants along the microtubule chain are not equal (i.e. $f_1 \neq f_2$) the

potential of "the tubulin particle" as a function of distance is asymmetric with respect to the equilibrium position. Therefore, the system is non linear and higher harmonic components (i.e. $\omega = n\omega$) and components with combination frequencies (i.e. $\omega = n\omega + m\omega_i + \ldots$, where n, m,... are integers) are generated. The dispersion relation (2) does not contain these harmonic and combination terms.

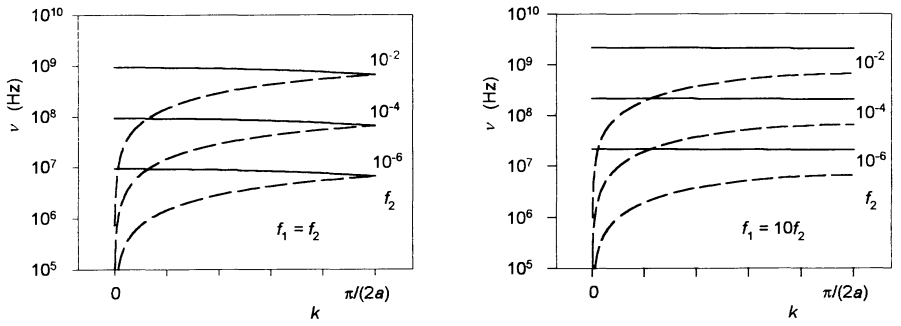


Figure 3. Dispersion relation for a microtubule (frequency versus wave vector). The full lines and the dashed lines denote the optical and the acoustical branches, respectively. Elastic force constants are parameters of the curves: $f_1 = f_2$ (a), $f_1 = 10f_2$ (b). After[8].

The biological reality can certainly yield more complicated patterns of vibrations in microtubules than those derived in this contribution as we used a number of simplifications. For example, internal vibrations were neglected and simple structure of microtubules was assumed. The internal vibrations are combined with the external vibrations giving rise to complicated spectral patterns. On account of small mass and great values of the elastic force constants frequencies of internal vibrations are much higher than those of external vibrations. The frequency spectrum can contain frequency lines in the millimeter and in the far infrared region too.

EXITATION OF VIBRATION

When a microtubule grows tubulin heterodimers add to the free plus end and GTP's attached to the β-tubulins are hydrolyzed to GDP's. The rate of polymerization may be greater than the rate of hydrolysis and an energy rich cap (a cap of heterodimers where GTP is not hydrolyzed) can be formed at the end of the microtubule. The energy rich cap at the end of the microtubule is hydrolyzed later. The energy released by hydrolysis of GTP in solution, in the tubulin heterodimers, and in the tubulin subunits in the microtubules is 5.18, 3.79, and 0.7kcal/mol (i.e. 0.22, 0.16, and 0.03eV for one entity), respectively[6,12]. Therefore, the energy of about 4.48kcal/mol (i.e. 18.8kJ/mol) is potentially available. A part of the energy may be used for transport of the phosphate group but the majority of it is stored in the microtubule lattice[2,6].

Utilization of hydrolysis energy is not well understood. The energy can change bond strengths[3] or excite soliton or soliton like waves[3,13]. It can be used for conformation changes of tubulin heterodimers (transition from untilted to tilted conformation) and for kink wave reorientation of heterodimer dipoles[3]. It can excite coherent latice vibrations by the Fröhlich mechanism[13] (the Frohlich mechanism is described e.g. in[14-16]) and/or by the intrinsic non linear mechanism. Regardless of the primary mechanism of energy utilization, in the final stage a part of it is always supplied to the vibrations. Besides energy stored in the cytoskeleton during polymerization a part of energy from ATP hydrolysis supplied to motor proteins moving along a microtubule can be transferred to the microtubules as random excitations of vibrations.

We assess the amount of energy which might be supplied to the microtubule vibrations from hydrolysis of GTP. The energy supplied by hydrolysis of GTP per one entity is about 7 times greater than the energy in the thermodynamic equilibrium in the frequency region from 10^7 to 10^{11}Hz (for hydrolysis energy 4kcal/mol). On account of non linear mechanism the energy can be accumulated in certain vibration modes which can shift the system far from the thermodynamic equilibrium.

CONCLUSIONS

The microtubules are polymer structures with translation symmetry. The symmetry conditions enable derivation of dispersion relation determining frequency of vibrations as a function of the wave vector. The lowest frequency can be below

10MHz. The tubulin heterodimers are polar and vibrations in the microtubules are accompanied by polarization waves and generate an oscillating electric field. A similar vibration mechanism can be found in the actin filaments.

The generated field mediates interaction with the surroundings and enables the energy transfer. Therefore, the microtubules might be capable of the spectral energy transformation and of the energy condensation in certain vibration modes e.g. on account of the Fröhlich mechanism. The intrinsic non linear mechanism in the microtubules can also cause the spectral energy transformation. Energy from hydrolysis of GTP to GDP can be transferred to the vibrations. The role of excited vibrations in the cytoskeleton dynamic activity has to be determined by future experimental and theoretical work.

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