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MASTER

NONLINEAR ELECTRODYNAMICS IN CYTOSKELETAL PROTEIN LATTICES

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INTRODUCTION

Nonlinear electrodynamic theories predict dynamic organization of biomolecular activities at all cellular levels: DNA, membranes, extracellular glycoproteins and the interconnecting cytoskeletal lattice.¹⁻⁵ Cytoskeletal lattice proteins including microtubules are particularly involved in dynamic regulation of intracellular movements and activities.^{6,7} This paper considers possibilities and implications of biological information processing due to coupling of Davydov solitons, Frohlich coherent oscillations and other nonlinear electrodynamic phenomena to conformational states of the grid-like polymer subunits of cytoskeletal microtubules.

CYTOSKELETAL PROTEIN LATTICES

Cytoskeletal lattices (Figure 1) including protein polymers microtubules (MT), actin, intermediate filaments and the microtubular lattice interconnect and organize cell components and dynamic

Fig. 1. Schematic representation of cytoskeletal lattice elements in vicinity of cell membrane. MT: microtubule, M: membrane, MP: membrane protein, MF: microfilaments, MTL: microtrabecular lattice, GP: extracellular glycoprotein.

activities.⁵⁻⁷ Intracellular movement and coordination, growth, mitosis, transport and turnover of membrane proteins are accomplished by activities of cytoskeletal lattices and attachments. The major cellular architectural elements, MT are cylindrical polymers which also comprise cilia, mitotic spindles, and other organelles.⁶⁻¹¹ MT are intimately involved in dynamic biological activities, but mechanisms of "real time" regulation and control by MT or other cytoskeletal filaments are unknown. The particular structural geometry of MT will be considered as a potential site for functionally significant nonlinear electrodynamic phenomena.

Most MT are assemblies of 13 longitudinal protofilaments which are each a series of polar, tubulin dimers (Figure 2). Each dimer consists of two slightly different classes of 55,000 molecular weight monomers (α and β tubulin) which are 4 nm in diameter.¹²⁻¹⁴ Tubulin binds eighteen calcium ions (Ca^{++}) per dimer, and MT polymerization depends on Ca^{++} and other factors. Hexagonal packing of the dimers

Fig. 2. Microtubule (MT) structure from x-ray crystallographic data.¹⁷ MT subunits are 8 nm dimers of α and β tubulin in a hexagonal, cylindrical lattice.

results in 3, 5 and 8 start helical patterns.^{15,16} MT outer diameters are 25-30 nm and inner diameters are 14-15 nm.^{17,18} MT outer surfaces are surrounded by a "clear zone" of several nm which apparently represents electronegative charges on tubulin¹⁹ and may also organize cytoplasmic water and enzymes.¹⁹ Functional MT lengths may range from hundreds of nm to micrometers and meters in some mammalian neurons. Contractile (ATPase dynein) or enzymatic proteins (microtubule associated proteins: "MAPs") may be attached at specific dimer sites resulting in various helical patterns.²⁰ Numerous proteins and other substances are transported by MT and attached contractile proteins at rates from one to 400 millimeters/day.^{21,22}

Fig. 3. Coordinated transport by MT based ATPase contractile sidearm proteins. Signaling for contraction may occur by solitons propagating through the MT lattice. In this representation, inclusion of sidearm base dimers in a "diamond" (4 contiguous aligned dipoles) connotes signal to contract. Diamonds or other structures may derive from coherent oscillations (see automaton model) resulting in solitons.

Phosphate nucleotides including GTP and cyclic GMP bind reversibly at specific dimer sites.²⁴⁻²⁵

Fig. 4. Dimer subunits in automaton neighborhood. Assuming α and β monomers share electron, dimer is in α state (represented by a) if dipole orientation is toward α monomer, and dimer is in β state (represented by $\cdot$) if dipole orientation is toward the β monomer. Relative distances among neighbor loci may be represented by r , x , and y .

Extensive reviews on MT and cytoskeletal structure and function are available.^{8,11} Intimate involvement of MT with dynamic cellular activities and their grid-like polymer structure suggest capabilities for information processing which may be explained by nonlinear electrodynamic phenomena.

SOLITONS, COHERENT OSCILLATIONS, AND DYNAMIC ORGANIZATION OF PROTEIN CONFORMATIONAL STATES

Solitons

Biological solitons have been theorized as couplings of phonons and excitons along molecular chains producing nonlinear vibrations which are extremely long-lived in comparison to linear oscillations.¹ Davydov proposed that biological solitons could be energetically initiated by hydrolysis of ATP phosphate bonds and propagate along linear molecules such as the alpha helix portion of proteins.^{2,4} Nonlinear coupling of electronic bond energy (possibly anharmonic stretching of amide I bonds,^{4,26} orbital changes in hydrophobic aromatic regions, and/or disturbance of the electronegative charge field on the MT outer surfaces) to mechanical oscillations of protein peptide chains can account for wave-like soliton propagation and a control mechanism for protein conformational states. Calculations and simulations have suggested that solitons may propagate through an alpha helix protein sequence at subsonic velocity, occupying nine peptide bond lengths (~4 nm) per soliton.^{4,26-28}

Cytoskeletal lattice arrays may provide significant pathways for soliton propagation. Phosphate nucleotides such as GTP bind rapidly and reversibly at specific tubulin monomer sites. GDP, incapable of energy donation, binds tightly at other tubulin monomer sites.^{29,30} Thus solitons may be introduced or supported at specific "preprogrammed" GTP binding subunits. Tubulin monomers are 4 nm in diameter and are comprised of about 22 percent alpha helix chains.^{14,31} Thus solitons initiated by GTP and other factors could propagate between α and β tubulin dimer regions, altering conformational states and binding of Ca^{++} and other factors. Soliton propagation among neighboring MT grid dimers might depend on numerous additional factors including dimer-dimer neighbor interactions, dimer conformational and Ca^{++} binding states, attachment of GTP, MAPs and contractile proteins, and perhaps the directional orientation of alpha helix chains within the globular tubulin subunits. Solitons propagating through an MT lattice would encounter gates and switches at every dimer-dimer interface. Dynamic patterns of solitons propagating and switching through MT lattices have been compared to computer and information processing technologies (Boolean switching matrices, charge coupled devices, bubble memories, etc.) and theoretically linked to dynamic organization of cellular activities.⁶ Programming modes governing soliton information patterns within cytoskeletal lattices may include genetic (e.g., primary protein structure) and "environmental" (e.g., Ca^{++} flux) influences.

Biological activity which may be explained by soliton propagation and switching in MT lattices is exemplified by the possible orchestration of contractile proteins attached at specifically arrayed MT lattice sites and which are active in transport of mole-

Table 1. Automaton rules derived from $\sum y/r^3$ which may be calculated from MT neighbor geometry distances.

Neighbor	Center State	Neighbor State	Distance (x)	Distance (y)	y/r^3
N	alpha	alpha	0.	8.	.0156
N	alpha	beta	0.	4.	.0625
N	beta	alpha	0.	12.	.0069
N	beta	beta	0.	8.	.0156
NW	alpha	alpha	-5.	4.9231	.0314
NW	alpha	beta	-5.	0.9231	.0064
NW	beta	alpha	-5.	8.9231	.0083
NW	beta	beta	-5.	4.9231	.0314
SW	alpha	alpha	5.	-3.1769	-.0152
SW	alpha	beta	5.	-7.1769	-.0108
SW	beta	alpha	5.	0.9231	.0070
SW	beta	beta	5.	-3.1769	-.0152
S	alpha	alpha	0.	-8.	-.0156
S	alpha	beta	0.	-12.	-.0069
S	beta	alpha	0.	-4.	-.0625
S	beta	beta	0.	-8.	-.0156
SE	alpha	alpha	5.	-4.9231	-.0314
SE	alpha	beta	5.	-0.9231	-.0083
SE	beta	alpha	5.	-8.9231	-.0064
SE	beta	beta	5.	-4.9231	-.0314
NE	alpha	alpha	5.	3.1769	.0152
NE	alpha	beta	5.	-0.9231	-.0070
NE	beta	alpha	5.	7.1769	.0108
NE	beta	beta	5.	3.1769	.0152

cules and organelles. Figure 3 depicts such orchestration by the signaling of propagating solitons. Dynamic organization of intracellular transport, ciliary movement, Ca^{++} fluxes, patterns of cell growth, differentiation and other intricate biomolecular actions requiring temporospatial organization could thus be explained.

Coherent Dipole Oscillations

Frohlich has theorized that nonlinear dipole excitations among molecular subunits of membranes and proteins of high dielectric

Fig. 5. Computer simulation of MT automaton based on rules derived from electrostatic force contributions from neighbors. Protofilaments 0-12 are on vertical (x) axis, and rows 0-64 are on horizontal (y) axis. An all beta (') pattern is invaded by an initial pattern of alpha states which grows. Diamond patterns of alpha states are highlighted by a larger central dot. Generations 1-4 are depicted.

strength will result in coherent oscillations (10^{-10} to 10^{-11} sec fluctuations), lower frequency metastable states, coherent polarization waves ("solitons"), and long-range order.^{2,32-34} Coherent oscillations of MT and other cytoskeletal lattice subunits may derive from intrinsic oscillations dependent on a high dipole moment and dielectric strength across the MT wall^{19,35} or coupling to oscillations in membranes or other structures. Del Giudice et al.^{35,36} have suggested that MT and other cytoskeletal filaments entrain and focus depolarization waves as solitons by nonlinear waveguide behavior.

Frohlich also theorized that protein conformational states are coupled to electron dipole oscillations.³³ Protein conformational

Fig. 6. Generations 33-36 of the patterns from Fig. 1. The alpha states have grown and reached the boundaries causing alpha diamonds and beta diamonds, highlighted by line connections. The pattern is otherwise orderly and alternating alpha and beta.

coupling to coherent dipole excitation among individual subunits in biomolecular arrays conceptually resembles soliton propagation and would apparently depend on genetic and environmental biochemical factors as well as neighbor interactions. Protein polymer neighbor interactions can be mediated by factors which include electrostatic dipole interactions, conformational states, binding of water, ions, MAPs, phosphate nucleotides and associated proteins, intrinsic subunit factors, or solitons. Net effects of these influences could alter oscillatory phases of particular subunits in coherent biomolecular arrays, possibly resulting in dynamic patterns, polarization waves and long-range order. Dynamic and stable patterns of dipole excitation and conformational states in MT could have direct, profound effects on regulation and organization of cellular

Fig. 7. Generations 40, 45, 55, and 70 subsequent to Fig's 5 and 6. Alpha and beta diamond patterns develop further from boundary interactions and move towards each other at velocities of $8 \text{ nm}/10^{-10} \text{ sec}$ ($\sim 80 \text{ m/sec}$).

processes. As proposed for soliton traffic, patterns of resonant excitation and conformational states of cytoskeletal lattice subunits could mediate protein binding sites, ion fluxes, and organization of cellular activities. Self-organizing patterns and information processing based on neighbor interactions at discrete time intervals among lattice subunit states have been described and are defined as cellular automata.³⁷⁻³⁹ Coherent oscillations in tubulin dimer subunits whose phases at discrete time intervals depend on lattice neighbor interactions may be considered as an automaton model of dynamic biological organization.

AUTOMATON MODEL OF MICROTUBULE INFORMATION PROCESSING

Dynamic patterns and information processing in automata depend on neighbor interaction rules among lattice subunits.³⁷⁻³⁹

Fig. 8. Generations 75, 80, 85, and 90. Collision and interference of diamond pattern with triangle, parallelogram, and trapezoid structures.

Frohlich coherent excitations (10^{-11} sec) could effect discrete time intervals in MT automaton neighborhoods. Tubulin dimers as basic units would have a 7-member neighborhood on a hexagonal cylindrical lattice (Figure 4). The cylinder has a circumference of 13 units and the start pattern of the leftward helix would be one and one-half dimers (three tubulin monomers).

Each dimer may be viewed as a dipole with an electron shared by the two monomers. At each time-step (e.g., 10^{-10} sec), the electron's average position is in the center of one or the other monomer. The electrostatic force exerted on the electron by its nearest neighbors would depend on the neighbor states at the previous time interval, and may serve as "rules" for an automaton. The magnitude of the force exerted by any one neighbor is:

$$\frac{-k(e^2)}{r^2}$$

where k is $1/4\pi\epsilon_0$, e is the charge on an electron and r is the distance between the electrons.

The location of the electron at any time can determine the conformational state of the dimer. We will assume each dimer is in either an alpha state or beta state at each 10^{-10} interval, depending on dipole orientation. The dimer will change state if the sum of the forces on its electron are sufficient to push it to the other side. The dimer neighbors are labeled with "north" (n) in the longitudinal direction ("y" axis, now horizontal) of the MT cylinder.

sw	nw	-x	↑
s	c	n	y →
se	ne		

Only the relative magnitudes of the forces along the axis of the MT are important. Taking the MT axis to be the y direction, the force $F(y)$ in the y direction on the electron in the center dimer is proportional to:

$$\sum \sin \theta / r^2$$

taken over all neighbors.

$$\sin \theta = y/r$$

therefore $F(y)$ is proportional to y/r^3 . Along the MT axis, tubulin subunits are 5nm apart, and around the circumference they are 4 nm apart. The helical twist yields a y offset of

$$4\text{nm}(3/13) = .9231\text{nm}$$

for nw and sw neighbors and $-.9231\text{nm}$ for ne and se neighbors. MT automaton rules are shown in Table 1.

There are two parameters which can be varied in this model relating to the transition region where the force on the electron is near zero. The "center" provides a bias which prefers one state or the other due to, perhaps, protein structure, ion binding, or other factors. The "width" determines a neighborhood around the center where the force is not great enough to influence the next state. It is in some sense a measure of the "viscosity" of the system, the tendency for a state to remain stable (or oscillatory) until a certain threshold is reached. "Width" and "center" are both in units of y/r^3 or nm^{-2} . Forces less than the value of "center" minus "width" yield alpha states. The fate of forces within "width" of "center" is determined by the sign of "width." A negative width implies that a dimer will switch states regardless of the force on it (as long as the force is less than "width" from "center"). This implies that some other mechanism is causing

Fig. 9. Generations 91-93 with further interference and annihilation of structures and altered background patterns.

Fig. 10. Generations 100, 110, and 120. Minimal structure persists with altered background patterns as residue (? memory).

oscillation of the dipole. A positive width implies that for very small forces the state will remain the same. In the case of "center" at zero and a very small positive "width," several static, oscillatory and dynamic structures appear. All other variations of the rules will be treated as perturbations of the behavior of these rules.

In a field of dimers in the same state, occurrences of dimers in the opposite state "invade" the others until an approximately equal mixture exists. Figures 5-10 illustrate alpha states invading beta states. Except for the abnormal hexagonal geometry, the progress of the invasion is like the ripples from a stone dropped in a pond. The front wave advances steadily while standing waves oscillate behind. Since the rules are by nature quantized, any "width" (viscosity) less than the magnitude of the smallest possible unbalanced force will yield undamped oscillation. This means that wave fronts will travel forever and the standing waves left behind will continue to oscillate indefinitely. There appears to be a small set of structures which exist independent of initial conditions. Alpha and beta sites tend to alternate along the MT axis while they tend to align (a-a,b-b) along the (nw-se, sw-ne) axes. This yields rows of alternating bars which oscillate from alpha to beta and back. Disturbing this regularity can exist kinks and change of axis (nw to ne) which are also oscillatory with period two. The one and one-half start helix geometry prevents continuous bands longer than one revolution of the cylinder to occur in the sw-ne direction without a kink or direction reversal. In addition to kinks and change of direction, opposed pairs of triangles and other structures consisting of alpha and beta sites appear and oscillate also. These pairs also align themselves along the two off axes (ne-sw, s-nw).

The dynamic structures which exist in this environment appear to all be stable and/or interacting combinations of two basic structures. The simplest is just a pair of alpha or beta sites along the main axis. Alpha pairs travel south along the axis while beta pairs travel north. The other structure is a diamond arrangement, four alpha or beta sites surrounded by their opposite. These diamonds travel in opposite directions (alpha north, beta south). Both types of structures interact with their environment and can be destroyed by each other as well as absorbed by the stable triangle pairs. They also disturb the background pattern as they travel, reorganizing the structure as they go. These structures have also been observed to "bounce" off of each other or exchange phase. Figures 5-10 show boundary conditions causing reflection and initiation of patterns of diamonds and other structures which then interfere, resulting in altered patterns. These traveling patterns have velocities ($8 \text{ nm}/10^{-10} \text{ sec} \approx 80 \text{ m/sec}$) comparable to solitons and/or nerve conduction velocities, and may serve to transport binding sites or intracellular information.

CONCLUSION

Nonlinear electrodynamic phenomena including Davydov solitons and Fröhlich coherent oscillations provide theoretical mechanisms for dynamic organization of biological activities. Cytoskeletal lattice proteins including microtubules are intimately involved in dynamic biological organization, and have structural geometries conducive to information processing by nonlinear electrodynamic phenomena. This paper has presented possible modes of microtubule information processing and dynamic organization. Fröhlich coherent oscillations can result in automaton-like information patterns, propagating solitons, and regulation of a wide variety of dynamic cellular activities.

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