

by the different dependences of these two time scales on the load. The model further reveals the dependence of the motor dynamics on the number of stators. In particular, we show that the maximum speed of the motor is independent of the number of stators, which agrees with recent resurrection experiments at near zero loads (Yuan & Berg, PNAS 105, 1182-1185, 2008). We have also used the model to study stepping statistics in single flagellar motor and different noise sources for rotational speed fluctuation. In general, we believe the model may be useful to study other molecular motor systems with multiple asynchronous power generating units. [Part of the work was published in (Meacci & Tu, PNAS 106, 3746-3751, 2009)].

824-Pos

Understanding Kink Propagation in Spiroplasma

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Spiroplasma are helical bacteria of the class Mollicutes, that lack cell walls and swim by propagating a kink of handedness-change along their helical shaped body. Recent microscopy studies indicate that the major structural component of the cell is a multistranded protein ribbon, bound to the inner cell membrane. The ribbon runs along the whole cell body, following the shortest helical path on the membrane inner surface. Kink propagation is believed to be driven by conformational changes in the ribbon subunits (itself possibly driven by unidentified motor proteins), but the microscopic mechanism is largely unknown. We use simple mechanical models to understand kink propagation in Spiroplasma. Our conclusions differ from earlier work based on purely geometrical considerations in several important ways. This leads us to propose new microscopic mechanisms for the handedness change, and a new interpretation of the observed bend angle in kinked cells.

We further model the mechanochemistry of kink propagation, and find that the kink speed might be limited either by protein friction or a chemical event in the mechanochemical cycle of a ribbon subunit. The two mechanisms might be distinguished based on the randomness of the kink propagation.

Our results offer a qualitatively new understanding of existing observations, and several useful suggestions for future experiments.

825-Pos

Swimming Hydrodynamics of a Run-And-Tumble Bacterium with Helical Flagella

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To study the swimming of a peritrichous bacterium such as Escherichia coli, which is able to change its swimming direction actively, we simulate the "run-and-tumble" motion using a bead-spring model to account for the hydrodynamic and the mechanical interactions between the cell body and multiple flagella, the reversal of the rotation of a flagellum in a tumble and the associated polymorphic transformations of the flagellum. The cell body and each flagellum are connected by a flexible hook, so that the flagella can take independent orientations with respect to the cell body. This simulation reproduces the experimentally observed behaviors of E. coli, namely, a three-dimensional random-walk trajectory in run-and-tumble motion and steady clockwise swimming near a wall. We show that the polymorphic transformation of a flagellum in a tumble facilitates the reorientation of the cell, and that the time-averaged flow field near a cell in a run has double-layered helical streamlines, with a time-dependent flow magnitude large enough to affect the transport of surrounding chemoattractants. This new model, which can be refined by using more beads if more quantitative predictions are desired, strikes a balance between accuracy and simplicity that will permit it to be used to determine the migration behavior of particles near a swimming cell, cell-cell hydrodynamic interactions, the effect of the number and geometric distribution of flagella on migration, the mechanism of circular swimming near a wall, details of the tumbling motion, and the effect of the Brownian motion on swimming. We also develop minimal models, inspired by the simple model of Najafi and Golestanian, that contain only 3-5 beads, and can simulate simple "pusher" and "puller" micro-swimmers, and are also able to include helical flow typically produced by rotary flagellar motion.

826-Pos

Swimming Microorganisms in Gels

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Many swimming microorganisms must move through viscoelastic fluids and gels. I present work on swimming in gels. First, unlike incompressible fluids, a gel can have compressional modes with relative motion between polymer and solvent fractions. In a continuum model for a gel, we show that compress-

ibility can increase the swimming speed of Taylor's swimming sheet. The zero-frequency shear modulus of a gel requires altered boundary conditions on the swimmer. Second, many biological gels are heterogeneous on the lengthscale of swimming microorganisms, necessitating non-continuum models that treat the gel network and swimmer on equal footing. We show that a random network modeled as dilute, immobile spherical obstacles increases the average swimming speed of a Golestanian three-sphere swimmer.

827-Pos

A Tug-of-War Mechanism for Bacterial Surface Movement

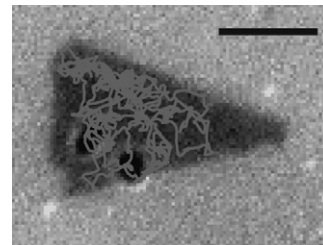
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In various bacterial species surface motility is mediated by cycles of type IV pilus motor adhesion and force generation, but it is unclear whether multiple motors cooperate mediate movement. Here we show that 7 ± 1 pili/cell are required for persistent movement of *Neisseria gonorrhoeae* with $MSD \sim t^{1.5 \pm 0.1}$. The unbinding force of individual pili from the surface $F < 30$ pN was considerably lower than the stalling force $F > 100$ pN, suggesting that density, force, and adhesive properties of the pilus motor have evolved to enable a tug-of-war mechanism for bacterial movement. Consistently, we found that bacteria were unable to move on fluid lipid membranes, most likely because force generation was not translated into bacterial movement due to slippage. Using microcontact printing, we confined the surface motility and microcolony formation to non-fluid islands within a fluid lipid membrane. Our patterning technique used physico-chemical surface properties that did not interfere with bacterial tug-of-war mediated motility and we anticipate that it will be useful for studying differentiation and gene expression within dynamical bacterial clusters and biofilms.

Figure: BSA-triangles on coverslips surrounded by with a DOPC membrane. The trace shows a path over 2min. Scale bar: 5µm.



828-Pos

Examples of X-Ray Scattering Studies of Biological Systems under Extreme Conditions

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Biological systems under extreme conditions often resort to unusual structures to achieve special functions for survival. The "bioengineering" principles of such "extreme" structures may inspire biomimetic designs of functional materials. Here we give several examples of such naturally occurring unusual structures probed with x-ray scattering. i) In bacterial spores, the spore coat appears to comprise of laminar layers of quasi-2D crystals with periodicity of ~1 nm. Such ordered assembly may be responsible for the spore resistance to heat, toxic chemicals, and mechanical disruption. ii) In starved bacterial cells, the DNA packaging protein (DPS) is over-produced to compact the chromosomal DNA close to crystalline density to protect the genomic integrity and facilitate homologous recombination. iii) In bacterial cells with over-produced DNA, mild treatment with antibiotics can lead to liquid-crystalline DNA that responds to external osmotic stress. Future work includes 1) uncovering the molecular basis of characterized structures and 2) extending into systems such as cells under radiation or heat and cancer cells.

829-Pos

Redundant Mechanisms for Stable Cell Locomotion Revealed by Minimal Models

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Crawling of eukaryotic cells on flat surfaces is underlain by the dynamics of the actin network and graded adhesion to the substrate and is regulated via a complex biochemical network. Some crawling cells maintain roughly constant shape and velocity. The paradigm of this stable crawling is the fish keratocyte, a rapidly moving cell that maintains a half-moon shape while translocating. Here we use moving boundary simulations to explore 4 different, minimal mechanisms for cell locomotion and show that all of these are sufficient to produce steady shapes and movements with resulting features that resemble the keratocyte morphology. We begin by considering a diffusion-limited actin model where G-actin transport to the leading edge controls the rate of protrusion of the leading edge.

The second model treats actin contraction by myosin as the primary determinant of shape and speed. Simulations show that this model is able to produce very realistic keratocyte-like shape and also reproduces the bistability that is observed in keratocyte fragments. We next consider a simple biochemical regulation model that has been proposed for the polarization of the Rac/Rho system. Here a high concentration of Rac at the front of the cell stimulates actin polymerization, and Rho at the rear of the cell induces contraction of myosin. Interestingly, volume conservation is not required for this model to work, yet the model behaves very differently if the cell has fixed volume than when the volume is self-regulating. The final model assumes that microtubule-based transport of vesicles to the leading edge limits the rate of protrusion. As all of these models are able to produce steady crawling locomotion, it suggests that these mechanisms may serve redundant roles in driving cell motility.

830-Pos

Crowding Effects on Association Reactions at Membranes

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The effect of macromolecular crowding on the binding of ligands to a receptor near membranes is studied using Brownian dynamics simulations. The receptor is modeled as a reactive patch on a hard surface and the ligands and crowding agents are modeled as spheres that interact via a steep repulsive interaction potential. When a ligand collides with the patch it reacts with probability. The association rate constant can be decomposed into contributions from diffusion-limited and reaction-limited rates. The simulations show that the diffusion-limited rate is a non-monotonic function of the volume fraction of crowding agents for receptors of small sizes. This is because crowding decreases the rate of diffusion to the surface but inhibits the escape of the ligand from the vicinity of the surface. The association rate constant has a qualitatively different dependence on the macromolecular crowding, for different values of the reaction probability. The simulation results are used to predict the velocity of the membrane protrusion driven by actin filament elongation. Based on the simple model where the protrusive force on the membrane is generated by the intercalation of actin monomers between the membrane and actin filament ends, we predict that crowding increases the local concentration of actin monomers near the filament ends and hence accelerates the membrane protrusion.

831-Pos

The Equilibrium and Nonequilibrium Mechanics of Cytoskeletal Networks

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The cytoskeleton is a complex, chemical heterogeneous network of semiflexible protein filaments, cross-linking proteins, and molecular motors that control the mechanics of some eukaryotic cells. Investigations of the mechanics of simplified biopolymer networks have shown that these materials differ substantially from better understood polymer gels in at least two respects: (i) they show large deviations from the predictions of continuum elasticity in sparsely cross-linked networks but undergo a non-affine to affine cross-over in denser networks and (ii) when endogenous molecular motors are present to drive the network out of equilibrium, the elastic moduli of the nonequilibrium network increase by more than one hundred fold.

In this talk we report analytic calculations and numerical simulations of equilibrium and non-equilibrium networks. Previous theoretical studies of the non-affine to affine transition in cytoskeletal networks have been confined to statistically isotropic random networks of monodisperse filaments. Biologically relevant and experimentally realizable networks are highly polydisperse and are frequently comprised of filaments with a preferred orientation. We examine, via numerical simulation, the individual effects of uniaxial order, filament polydispersity, and motor activity on the non-affine to affine transition. Finally, we demonstrate analytically how one can use the correlated motion of pairs of tracer particles embedded in the network (i.e. two-particle microrheology) to experimentally determine the density of active motors in *in vitro* networks and in living cells.

832-Pos

Fiber Network Elasticity as Function of Crosslinker Density

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Crosslinkers determine the architecture of polymer networks and thus are of great importance for the resulting mechanical properties. A simple morphological model is proposed for the investigation of the linear elastic response of 3D

fiber networks to randomly disconnecting network nodes. Isotropic ordered and disordered, 4-coordinated networks are modeled as homogeneous bodies in the shape of a network with a given volume fraction with locally isotropic elastic moduli. The 4-coordinated nodes are randomly split into two locally unconnected fibers representing a morphology change of the network at constant volume fraction. The effective shear modulus is studied using a voxel-based finite element method. Our results show a strong, continuous decrease of the shear modulus with decreasing number of nodes in the network without a percolation transition. The morphology of the networks is characterized by the Euler number that linearly depends on the fraction of split nodes, and that is easily extracted from 3D confocal microscopy data. Associating all network nodes with fiber junctions connected by a crosslinking molecule, this approach is a first order model for elasticity of biological networks with varying crosslinker density.

833-Pos

Heterogeneity and Flow in Biological Networks and Implications for Cargo Transport

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Single molecule experiments have measured stall forces and procession rates of molecular motors on isolated cytoskeletal fibers in Newtonian fluids. But in the cell, these motors are transporting cargo through a highly complex cytoskeletal network. To compare these single molecule results to the forces exerted by motors within the cell, an evaluation of the response of the cytoskeletal network is needed. Using magnetic tweezers, we quantify force-velocity curves for magnetic beads moving through entangled F-actin networks [12 μ M]. Below a certain critical force, we see an elastic response with a plateau indicating a shear modulus of 0.1 Pa, comparable to bulk rheological measurements. Above this critical force we find a viscous response with a viscosity of approximately 0.3 Pa.s. The exact value of the critical force ranges from roughly 6-14 pN, reflecting the spatial heterogeneity of the network. This non-Newtonian force-velocity relationship, as well as the considerable heterogeneity in the network response, suggests the local cytoskeletal environment is an important factor when considering cargo transport inside the cell.

834-Pos

Regulation of Nonmuscle Myosin IIA Assembly

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In eukaryotic cells, nonmuscle myosin II filament assembly is critical for essential motile processes such as cytokinesis, cell motility, and the maintenance of cell morphology. Although myosin II filament dynamics are believed to be under strict spatial and temporal control, the mechanisms modulating filament assembly and disassembly are poorly understood. In this work, we examined the molecular mechanisms regulating myosin IIA filament assembly that rely on the intrinsic dynamics of the C-terminal coiled-coil of the myosin IIA heavy chain and on non-covalent interactions with S100A4 protein, a major metastasis factor. Sedimentation equilibrium, hydrogen-deuterium exchange, and thermal melt CD spectroscopy showed that the C-terminal coiled-coil of the nonmuscle myosin IIA heavy chain exhibits significant conformational plasticity. Based on these observations, we propose that the plasticity of the C-terminal coiled-coil is an inherent regulatory mechanism for modulating myosin IIA filament assembly. We are testing this hypothesis by introducing stabilizing mutations into the C-terminal coiled-coil and examining how the loss of plasticity modulates salt-dependent oligomerization of myosin IIA and filament assembly as assayed by turbidity and critical concentration measurements.

835-Pos

Myosin II is an Active Stress Sensor at the Core of a Cell Division Control System

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Mechanosensing is important in many cellular processes such as cell motility and cell division. Cells experience mechanical stress from the surrounding environment and also its internal cytoskeleton structure. Previous studies from our lab showed that cellular mechanosensing is crucial for regulating cytokinesis shape change. Using micropipette aspiration (MPA) to generate stress on the cell cortex, we discovered that mechanical stress stimulates the accumulation of myosin-II (a contractile force generating protein) and cortexillin-I (an actin bundling protein) at the deformation site. These proteins then contract the cortex, correcting the shape of the cell. Both myosin-II and cortexillin-I are required for this mechanosensory system during cytokinesis. Recently, we