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Review

Quantitative approach of Min protein researches and applications: Experiments, mathematical modeling and computer simulations

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Cell division in prokaryotes is a process (known as binary fission) where the parent cell divides into daughter cells. In this process, the dynamics of Min proteins is an important factor in the accurate positioning at the midcell in *Escherichia coli.* This site specificity is regulated by the oscillatory behavior of Min proteins. Numerous studies of Min protein dynamics have focused on dynamic spatial-temporal pattern formation, the movement mechanism and the biochemical basis function mostly using wet lab experiments, but the quantitative data remains limited. Thus in this research review, focus is on quantitative methodologies. Up-to-date information and findings regarding Min proteins, particularly MinD proteins obtained by using quantitative approaches such as experiments, modeling and simulations were provided. This review of quantitative techniques is expected to benefit not only those who want to conduct research in this area using more quantitative approaches, but also those who are interested in using qualitative data to support their findings.

Key words: Min protein, quantitative characterization, protein oscillation, ***Escherichia coli***, spot tracking technique, modeling, simulation.

INTRODUCTION

Cell division in prokaryotes is the process where the parent cell divides into daughter cells after its DNA has been duplicated and distributed into the two regions that will later become the future daughter cells. This process is also known as binary fission. For successful cell division to take place, the cell has to determine the optimal location for cell separation and the optimal time to start cell cleavage. In ***Escherichia coli***, the oscillatory dynamics of Min proteins have played an important role in determining the default site of septal placement in cell division, usually at midcell. This septum formation is initiated through the polymerization of the FtsZ protein into the Z-ring, a process crucially facilitated by the Min proteins (Justice et al., 2000; Lutkenhaus and Addinall, 1997).

The Min protein system consists of MinC, MinD and MinE expressed from the MinB operon (de Boer et al., 1989). They are responsible for facilitating the accurate

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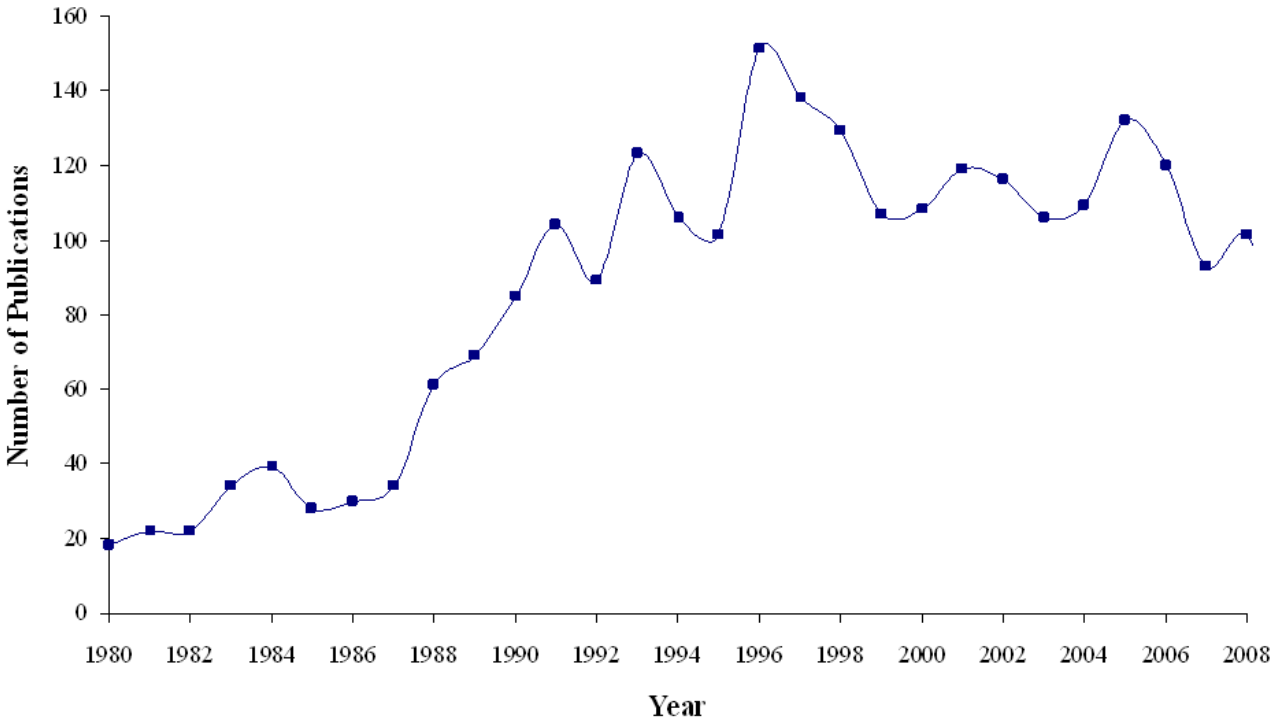

Figure 1. The graph of number of papers vs. year of the search keyword "min proteins in ***E. coli***" getting from SCOPUS database during year 1980 to 2008.

location of the division site at midcell through the oscillatory cycle from pole to pole (Rothfield et al., 2005). MinC proteins prevent septum formation by inhibiting FtsZ polymerization ***in vitro*** (Hu and Lutkenhaus, 1999). In other words, MinC is an antagonist of FtsZ polymerization and a specific inhibitor of Z-ring formation. While MinD plays a role in inhibiting MinC-mediated division, it is sensitive to suppression by MinE. ***In vivo***, MinC co-localizes and co-oscillates with MinD (Hu and Lutkenhaus, 1999; Raskin et al., 1999a) which acts together as a negative regulator of Z-ring assembly (Hu and Lutkenhaus, 1999; Raskin et al., 1999a, 1999b). Since MinC binds to MinD, the movement of MinC from pole to pole with relatively long polar dwell times and a short transit time results in blocking the formation of polar Z-rings. The ATPase activity of MinD is presumed to provide the driving force for the pole-to-pole oscillation of the MinC division inhibitor. MinE acts as a topological specificity factor to prevent the division inhibitor from acting at the midcell site, while permitting it to block septation at polar division sites. If MinE is absent, MinD will be distributed evenly over the cell membrane (Raskin et al., 1999a, Rowland et al., 2000).

The past two decades have seen tremendous effort put into Min protein research, with thousands of research articles and several dedicated journals. In fact as of October 26, 2009, a keyword search of "Min protein in ***E. coli***" between year 1980 - 2008 results in records as shown in Figure 1 (also see references such as Rothfield et al., 2005; Shih and Rothfield, 2006; Fange and Elf, 2006; Kanthang, 2009). Research conducted on Min proteins is highly multidisciplinary, covering the biological and physical sciences, as well as the fields of mathematics and engineering. Hence in this review of Min protein, references from archival scientific literature which could be valuable to those conducting future experiments and/or developing new theories and applications were included. Elaboration was made specifically on MinD, which has been studied using quantitative techniques, including the spot tracking technique, modeling and simulations. Image processing and data analysis were discussed to provide qualitative and quantitative interpretations on the dynamics of MinD. In addition, examination of quantitative methodologies (both experimental and computational) that have emerged over the past year for studying Min protein dynamics as well as highlighting of some recent results was carried out.

EXPERIMENTS USING THE QUANTITATIVE SPOT TRACKING TECHNIQUE

Two important innovative works that have contributed to advancing molecular biology and related research in both pure and applied sciences are the development of fluorescent proteins that allow researchers to selectively label single proteins and the development of high-resolution fluorescent imaging, which was made possible by the

new generation of bright-field and confocal microscopes (Pierce et al., 1997; Endow, 2001; Kain and Kitts, 1997). Thanks to these new technologies as scientists are now able to study molecular dynamics within the living cell at sub-micron resolution or even at the molecular scale. Researchers can now record a time-lapse series to study the dynamics of molecular transport or the conformation changes within the cell. While these methods offer enormous potential for increasing our understanding of biological systems, they also constitute a challenge for researchers in the field who have not yet devised efficient ways to exploit and quantitatively interpret the unprecedented flow of technical data.

Regarding Min protein studies, the large majority of data analysis and feature extraction is currently still done manually. This is very time consuming and the data are prone to human and systematic errors. In the present view, although a number of previous studies of Min protein oscillations focus on spatial-temporal pattern formation and the biochemical-based function (Rothfield et al., 2005; Shih et al., 2002), the experimental data have been inadequately interpreted for quantitative study. Therefore optimal and friendly-user quantitative techniques, together with data acquisition and image processing, are being developed to crack this short-coming. It should be noted that some previous quantitative results on Min protein dynamics that deserve to be mentioned were mostly obtained by using in-depth experimental techniques or by modeling and simulation (Rothfield et al., 2005; Shih et al., 2002; Meinhardt and de Boer, 2001; Howard et al., 2001; Kruse, 2002; Howard and Rutenberg, 2003; Huang, 2003).

To deal with this shortcoming, application for the first time of the spot tracking technique (STT) by applying the more well-known technique called single particle tracking (Saxton and Jacobson, 1997; Qian et al., 1991) to explore the dynamics of GFP-Min proteins as the indicator of MinD and MinE dynamics was done in this study. The analysis concentrated on the ensemble positions of GFP: MinD and GFP: MinE, as well as dynamics and pattern formation, by tracking the ensemble positions of Min proteins. In addition, data analysis was carefully performed to provide qualitative and quantitative interpretations of the behavior of Min oscillation. It is believed that the obtained quantitative results should benefit biological, biomedical and biotechnological researches.

Spot tracking technique (STT)

For the sake of brevity, STT will be briefly described as more details can be gotten from the references. The STT (spot tracking technique) is an image processing technique used to follow the spot-like particle in the fluorescent image under intense fluorescence signals (Unai et al., 2009). In other words, STT tracks the maximum distribution of a particle ensemble. Data from STT measurements generally provide key characteristics of a tracked micro-particle's (or organism's) temporal and spatial dynamics. The STT can be exceptionally useful for biophysical research and biotechnological applications to measure the trajectory of individual proteins or organelles inside the cell, or in cell membranes such as plasma and nuclear membranes-in a manner similar to that of the more powerful single particle tracking technique (Saxton and Jacobson, 1997), which has been used in the nuclear trafficking of viral genes (Babcock et al., 2004), in chromosome dynamics (Sage et al., 2005) and in the motion of bacterial actins (Kim et al., 2006). Materials and experimental procedures for demonstrating how STT can be applied to study Min protein dynamics in ***E. coli*** is discussed below.

Here, focus will be on MinD only. In the experiments conducted, ***E. coli*** with MinD labeled with green fluorescent proteins (GFP) were used. A starter of RC1/ pFx9 cells was grown in optimal condition media until the OD_{600nm} was approximately 0.4 (log phase). Centrifugation was performed to collect the cells. The sample was treated with isopropyl-β-D-thiogalactopyranoside (IPTG) for protein induction and diluted with media before use. Each ***E. coli*** sample (5 - 7 µl) was dropped on a glass slide coated with Poly-L-lysine and covered with a cover slip at room temperature before examination. Fluorescence microscopy was used with ***in vivo*** software to obtain fluorescent image sequences. In this process, a charge-coupled device (CCD) camera was attached to the microscope video port to acquire images and movies. After images were obtained, the STT technique was used to follow the region of interest (ROI) which consists of the highest GFP-MinD concentration signal. The data obtained in STT measurement were supported by the Spot Tracker Java plugin provided by public domain ImageJ software (SpotTracker). Typically, the acquired images were in the configuration of a fluorescent signal that can fade after about 4 - 5 min. Subsequently, the final image sequence was noisy. Hence, to improve the quality of the acquired images, the software's Gaussian filter to reduce noise was used. The improved images were further enhanced by using the rescaling option of the Spot Tracker plugin. Tracking of ROI with the SpotTracker plugin was performed to collect the positions at given times in the text file (Sage et al., 2005). Ensemble positions were then analyzed by MATLAB software. The procedure is summarized in Figure 2.

With the STT, the main quantitative results can include the position and velocity profile, oscillator period, dynamic local and global pattern formations, mean-square displacement and sub-diffusion measurements, power spectrum and/or correlation function, energy landscape, waiting time distribution and so on. For example, obtained data for ensemble GFP-MinD oscillations from pole to pole with ~ 45 s at time 0 - 50 s are shown in Figure 3. Figure 3(Top) is the 2D image sequence of pole-to-pole MinD oscillations at each successive time for

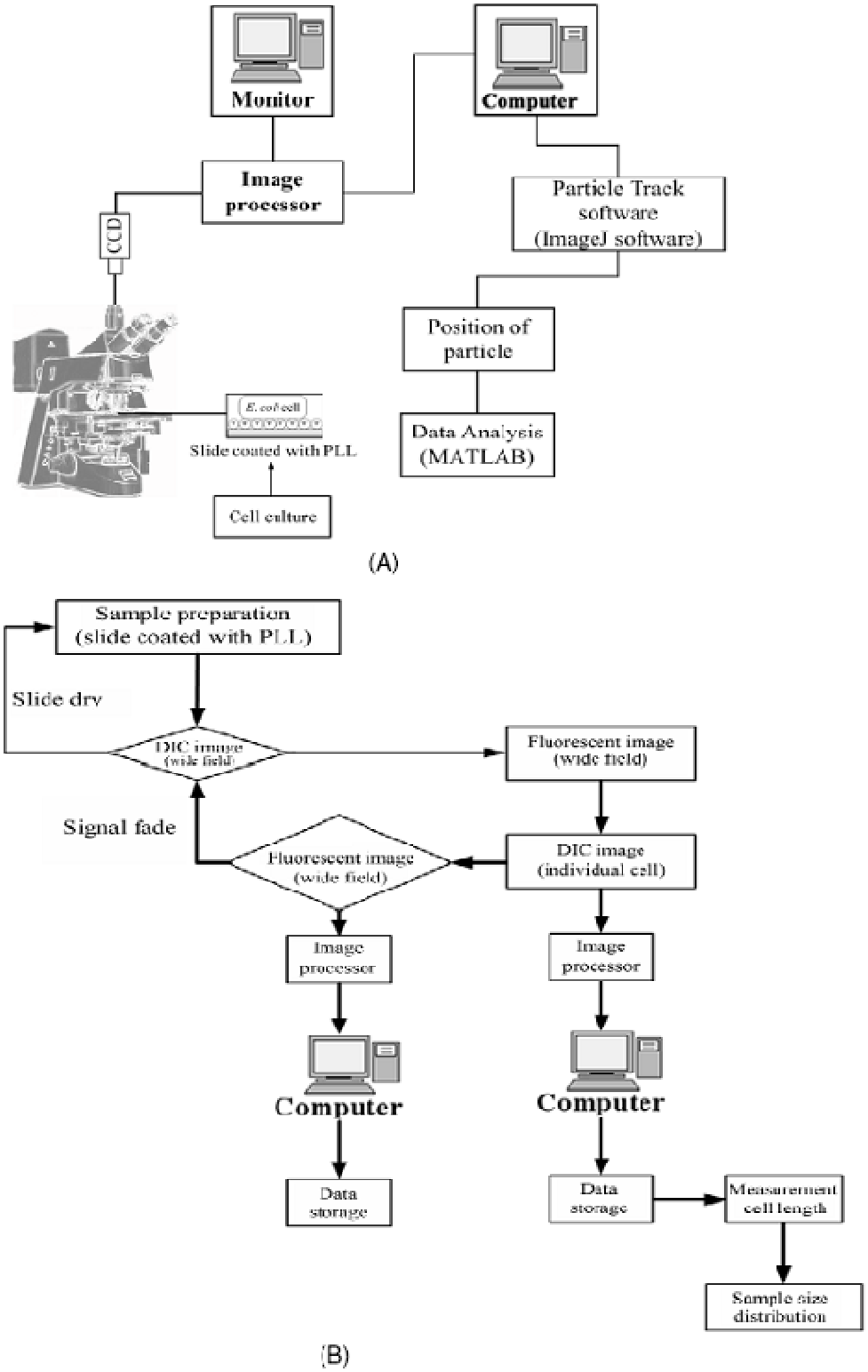

Figure 2. (A) Diagram of the procedure for STT of MinD protein oscillation. (B) More details of the process for image acquisition are depicted in.

the rescaled and enhanced signal. Each fluorescent image represents the ensemble of the GFP-MinD signal located at the polar zones. The time(s) label on the left side of the column is the first time at which GFP-MinD

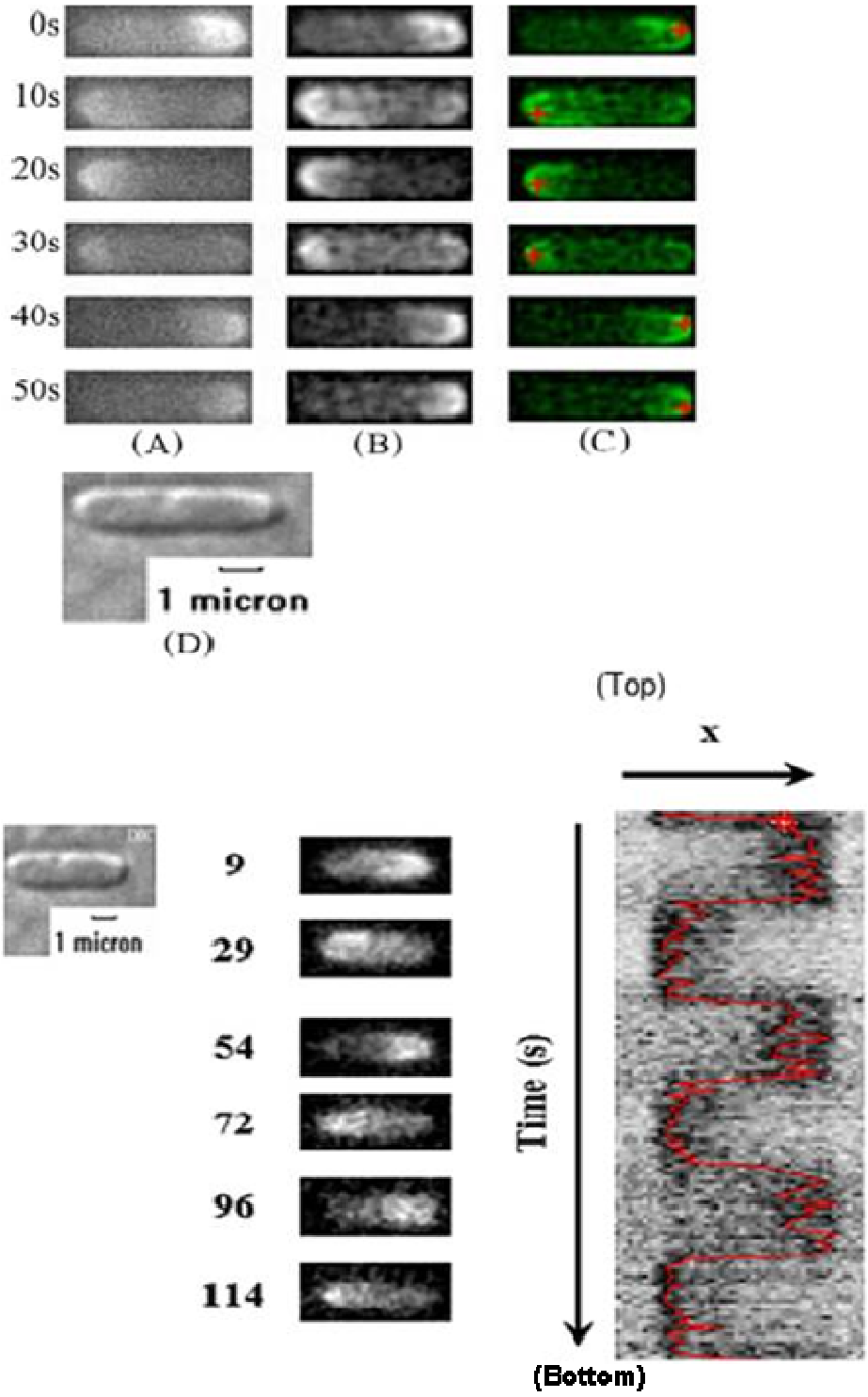

Figure 3. The ensemble GFP-MinD oscillations from pole to pole. (Top) From left to right during 0s-50s in ***E. coli*** c: (A) 2D image sequence at each successive time for the resize, (B) Enhanced signal, each fluorescence image represents the cluster of GFP: MinD signal locating at polar zones, (C) result after tracked, position of cluster of GFP: MinD denote as red cross. (Bottom) The time evolution trajectories of MinD oscillations along the x(t) and y(t) direction.

assembles after switching to the new pole. The sequence of positions at successive times can be used to determine the trajectory of GFP-MinD in x and y components as shown in Figure 3 (Bottom). It is believed that the STT

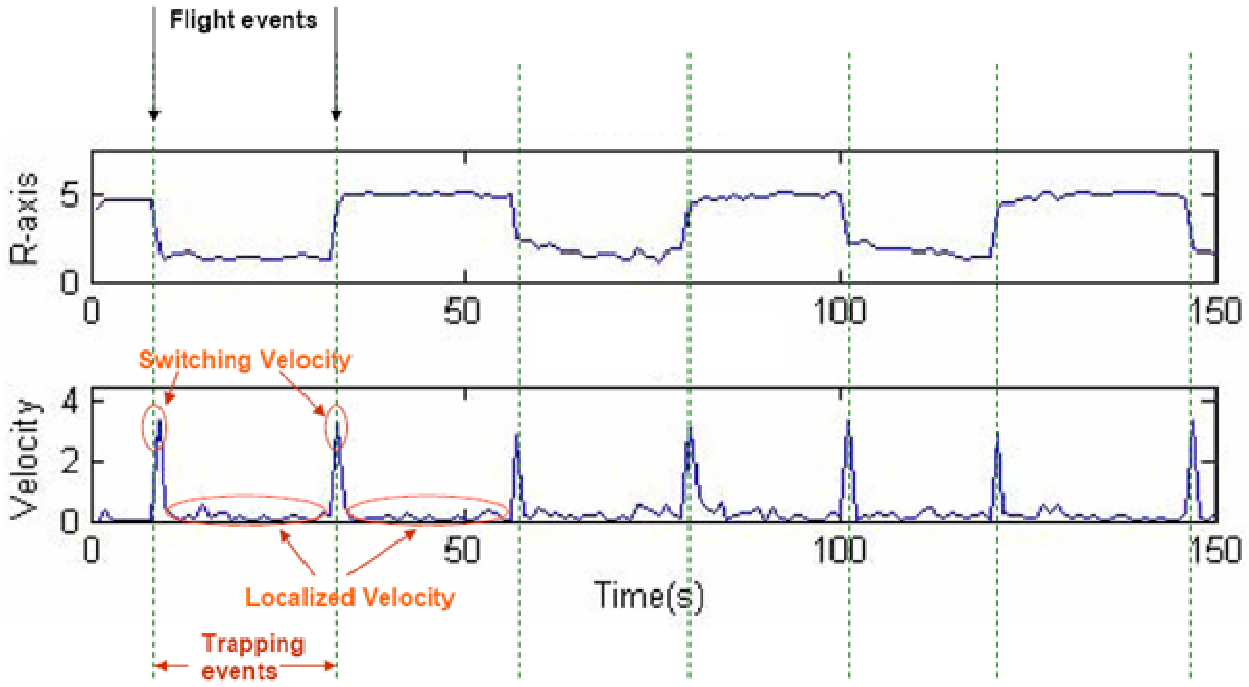

Figure 4. The trajectory and velocity time evolution of GFP-MinD for acquisition time over 150s. (Top) Cluster Position Trajectory of GFP-MinD along x or cell length. (Bottom) Absolute Velocity time evolution plot of cluster GFP-MinD oscillation pole-to-pole; velocity peak called the ***switching velocity*** (Flight events), the other is called the ***localized velocity*** (Flight events).

can provide accurate enough predictions to suggest useful biological features in predicting protein motion. With improvements to the STT (such as improved data acquisition and data analysis), the technique could become a very well-accepted one. It is reasonable to say that the quantitative information the STT yields could contribute to improvements in the dynamic model of protein oscillation, as well as improved experimental procedures.

Quantitative results of the movement dynamics and pattern formation of MinD proteins via STT

If the time series of MinD positions denoting $\vec{r}(t) = (x(t), y(t))$ were recorded, the mean squared displacements (MSD) can be calculated, $\left\langle \left|\Delta\vec{r}\right|^2 \right\rangle$, with the average being taken over time within a single trajectory (one also could do ensemble average). It is theoretically well known that the MSD of a diffusing particle varies with time as $\left\langle \left|\Delta\vec{r}(t)\right|^2 \right\rangle \propto t^{\alpha}$, where the dynamic exponent α distinguishes the type of diffusion encountered; $\alpha = 1$ indicates normal Brownian diffusion; $0 < \alpha < 1$ subdiffusion and $\alpha > 1$ superdiffusion (Saxton and Jacobson, 1997; Qian et al., 1991; Metzler and Klafter, 2000).

Recently, Unai and co-workers (Unai et al., 2009) used the STT to quantitatively investigate MinD dynamics at the level of particle cluster. Their studies provided information on position alterations and time sequences which can be used to analyze GFP-MinD dynamics. They quantitatively found that the ensemble GFP-MinD protein dynamics can be separated into 2 types: trapping event and flight event (Figure 4). The trapping event mostly occurs at the polar zones, while the flight event takes place between the trapping events in the space between the polar zones. Therefore, high concentrations of GFP-MinD are mostly found in the Polar Regions. In previous studies, this phenomenon was explained by stating that the dynamics of trapping events at polar zones involves the formation of MinD polymerization at the cytoplasmic membrane (Hu and Lutkenhaus, 2001; Hu et al., 2002). Flight event behavior, on the other hand can be identified by the peaks of each time interval mentioned earlier. Although MinD proteins globally perform oscillatory pole-to-pole motion, the local membrane-bound motion, which exhibits a horseshoe structure in the polar zone (Raskin et al., 1999a; Rowland et al., 2000), is another trademark. That's why it is believed that this dynamic relates to the formation of MinD polymerization at the cytoplasmic membrane (Suefuji et al., 2002; Hu et al., 2002).

Recently, Kanthang, (2009) applied the STT to reveal the underlying spatial-temporal pattern formation dynamics and energy landscape of MinD proteins. They specifically focused on the physical quantities of MinD cluster dynamics, including mean square displacements (MSDs), time memories, spatial distributions and effective

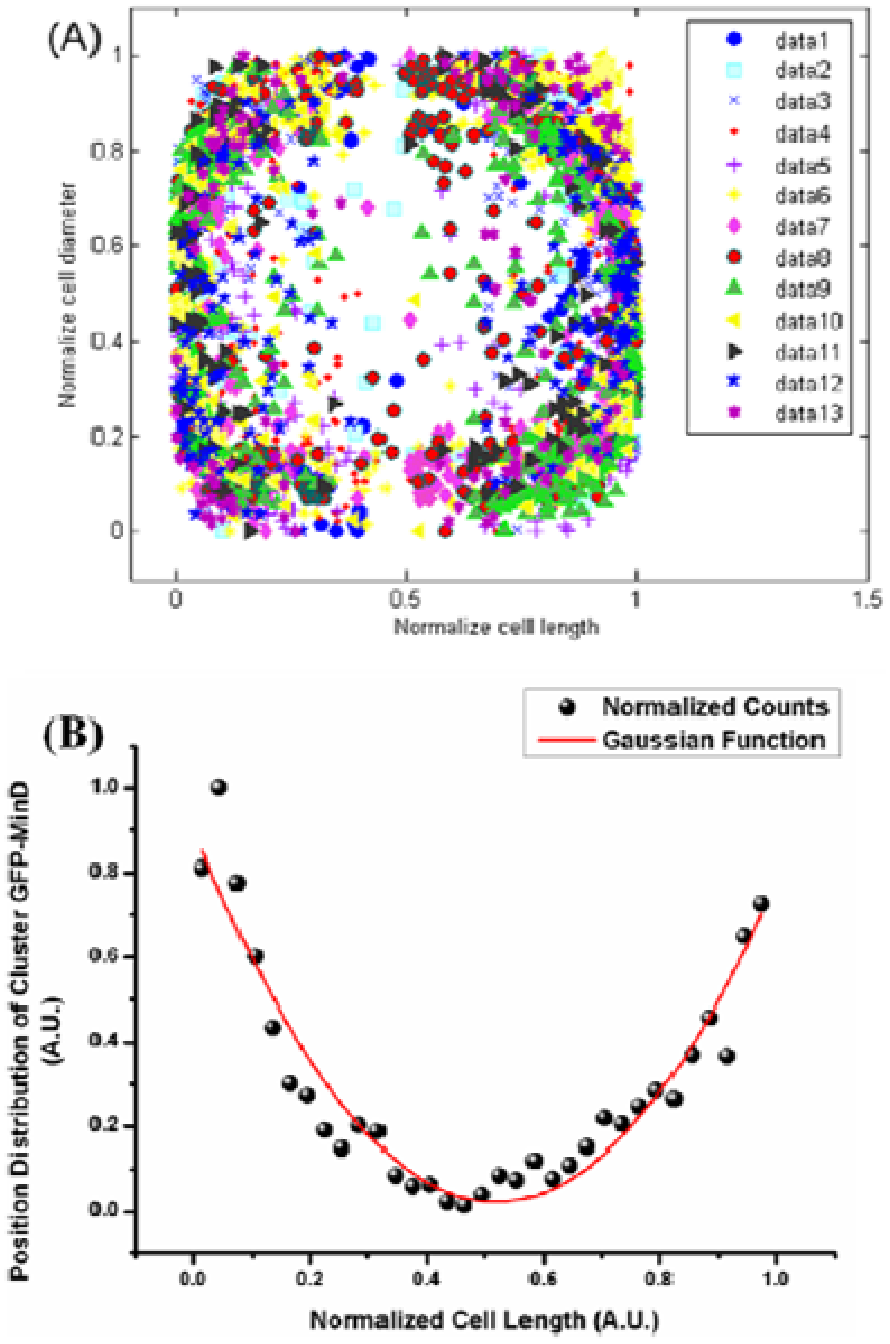

Figure 5. The position scattering and histogram of GFP: MinD localization. (A) Shows the position scattering normalization plot of x-y (13 individual cells). (B) The solid line is a fit to the Gaussian Distribution Function of the cluster GFP: MinD position along x axis for 13 individual cells, $x_c = 0.52$ and $R^2 = 0.9$.

potential profiles. Moreover, they revealed the relation between the potential profiles and diffusion modes which typically follow sub-diffusion. Analyzed ensemble GFP-MinD localization through a normalization position scattering plot is shown in Figure 5(A). The probability distribution plot shown in Figure 5(B) revealed that the region near the midcell had the lower concentration. The midpoint of cell length with GDF fitting is $x_c = 0.52, R^2 = 0.9$. This fiting value provides the lower concentration point of GFP- MinD located at 51.685% of cell length**.** These results correspond to previous reports (Justice

Table 1. Some physical quantities of MinD proteins.

Physical quantities	MinD finding	References
Oscillation Periods	~55 s	Hale et al. (39)
	~40 s	Raskin et al. (45)
	~60 s	Shih et al. (15)
	~55 s	Unai et al. (9)
Velocities	Polar Zones ~ 0.3 μm/s	Unai et al. (9)
	Pole to Pole ~ 3 μm/s	Unai et al. (9)
Diffusion Coefficients	Cytoplasmic Diffusion ~ 17 $\mu m^2/s$	Meacci et al. (47) ^{FCS}
	Cytoplasmic Membrane ~ 0.2 $\mu m^2/s$	Meacci et al. (47) ^{FCS}
	Polar Zones ~ 0.3 $\mu m^2/s^{\alpha}$	Unai et al. (9) ^{STT}

^{FCS} = Fluorescence correlation spectroscopy; ^{STT} = Spot tracking technique. $\alpha = 0.34 \pm 0.18$.

et al., 2000; Hu and Lutkenhaus, 1999; Raskin et al., 1999a, 1999b) which suggested that the time-averaged concentration of MinD (or the division inhibitor) is lowest at midcell. A summary of previous MinD dynamic findings both from fluorescence correlation spectroscopy and STT is shown in Table 1.

To recap, the spot tracking technique (STT) was used here to track the maximum distribution of a particle ensemble. The STT can provide accurate enough predictions to suggest useful biological features in predicting protein localization. With improvements to the STT (in data acquisition and data analysis), the technique could become a very well-accepted one. It is reasonable to say that this quantitative information could contribute to improvements in the dynamic model of protein oscillation.

COMPUTATIONAL METHODS

To gain greater insight into the nature of the mechanism of MinD dynamics, numerous computational researchers have proposed models to fill in the missing links. In this section, a review the on computational methods used to study MinD dynamics is made. They include deterministic mean-field mathematical modeling, the mesoscopic lattice Boltzmann method and stochastic modeling and simulation.

Deterministic mathematical reaction-diffusion models for Min protein oscillation

A number of mathematical models of Min protein oscillation have been proposed and studied (Howard et al., 2001, 2003; Meinhardt and de Boer, 2001; Kruse, 2002; Huang et al., 2003, Huang and Wingreen, 2004; Drew et al., 2005). Due to the fact that these Min proteins can move diffusively and interact with each other, these models were based on macroscopic nonlinear reaction-diffusion equations (RDE) and were solved using conventional grid-based finite difference method (Strikwerda, 1989). Howard et al. (2001) proposed an RDE model in which the reaction consisted of a protein's association to the membrane and its dissociation from the membrane (Figure 6). This model was primarily based on the experimental results of Raskin et al. (1999a). The model incorporates a series of events where MinE was recruited to the membrane by membrane-associated MinD using a set of four non-linear coupled reaction-diffusion equations. Though the model is straightforward and relatively simple, it gives the correct placement of the division septum in ***E. coli***. The mechanism is governed by the time rates of change of protein densities due to the diffusions of MinD and MinE and the mass transfer between the cell membrane and the cytoplasm, as schematically shown in Figure 6. In dimensionless form, the dynamics may be given by the following equations:

$$\frac{\partial \rho_D}{\partial t} - D_D \nabla^2 \rho_D = R_D = -\frac{\sigma_1 \rho_D}{1+\sigma_1' \rho_e} + \sigma_2 \rho_e \rho_d \quad (1)$$

$$\frac{\partial \rho_d}{\partial t} - D_d \nabla^2 \rho_d = R_d = \frac{\sigma_1 \rho_D}{1+\sigma_1' \rho_e} - \sigma_2 \rho_e \rho_d \quad (2)$$

$$\frac{\partial \rho_E}{\partial t} - D_E \nabla^2 \rho_E = R_E = \frac{\sigma_4 \rho_e}{1+\sigma_4' \rho_D} - \sigma_3 \rho_D \rho_E \quad (3)$$

$$\frac{\partial \rho_e}{\partial t} - D_e \nabla^2 \rho_e = R_e = -\frac{\sigma_4 \rho_e}{1+\sigma_4' \rho_D} + \sigma_3 \rho_D \rho_E \quad (4)$$

Where, ∇^2 = Lapacian operator and $s = \{D, d, E, e\}$ symbolizes the cytoplasmic MinD, the membrane bound MinD, the cytoplasmic MinE and the membrane bound MinE, respectively; ρ_s = mass density of particles of species s at time t and position $\vec{r}$; R_s = reaction term which depends on the density of the species (ρ_s) and

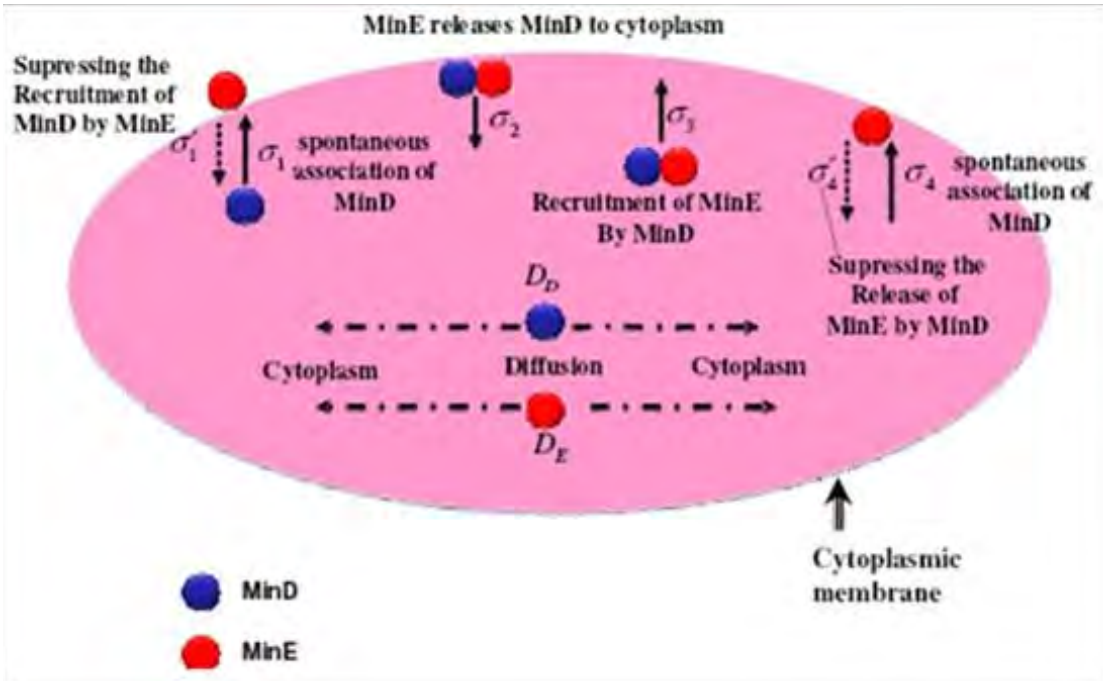

Figure 6. A schematic diagram of the MinDE dynamics according to the model proposed by Howard et al**.** (2001)**.** It depicts reactions of Min proteins in the cytoplasm and cytoplasmic membrane.

on the density of the other species that react with species s; D_s = diffusion coefficient; σ_1 = parameter connected to the spontaneous association of MinD to the cytoplasmic membrane; σ_1' = connected to the suppression of MinD recruitment from the cytoplasm by the membrane-bound MinE; σ_2 = the rate that MinE on the membrane drives the MinD on the membrane into the cytoplasm; σ_3 = the rate that cytoplasmic MinD recruits cytoplasmic MinE to the membrane; σ_4 = the rate of dissociation of MinE from the membrane to the cytoplasm and σ_4' = cytoplasmic MinD suppression of the release of the membrane-bound MinE.

The diffusion on the membrane occurs at a much smaller time scale than that in the cytoplasm. It seems, therefore, reasonable to set D_d and D_e at zero. The Min protein is expected to bind/unbind from the membrane, but not be degraded in the process. Thus, the total amount of each Min protein type is conserved. The zero-flux boundary condition is imposed, which requires a closed system with reflecting or hard-wall boundary conditions. The total concentration of Min proteins is conserved.

In addition, Meinhardt and de Boer, (2001) proposed the model and showed later that the pattern formation of the Min system requires the interaction of a self-enhancing component with its long-ranging antagonists. They included the dynamics of FtsZ proteins in their model. More recently, Kruse (2002) found that pole-to-pole oscillation depends on the membrane-bound MinD's tendency to cluster and attach to (and detach from) the cell wall. However, the Kruse model requires unrealistically rapid membrane diffusion of MinD. Since most of the models mentioned above were applied only to uniformly rod-shaped wild-type cells, Huang and Wingreen, (2004) proposed a model to reproduce experimental oscillations in not only rod-shaped cells, but also in round and ellipsoidal cells. In all these models, oscillation patterns were successfully generated and in qualitative agreement with experimental observations. Lastly, Huang et al. (2003) formulated a 3 dimensional model based on their experiments to describe Min protein oscillation, while Drew et al. (2005) proposed a mathematical model to describe polymerization and depolymerization behavior of MinD. Drew's team found that MinD bind to the membrane, followed by subsequent binding of MinE (Drew et al., 2005).

Mesoscopic lattice Boltzmann method (LBM)

The lattice Boltzmann method (LBM) is a recently developed efficient numerical tool for simulating fluid flows and transport phenomena based on kinetic equations and statistical physics (Zhaoli et al., 2005). The LBM is based on particle dynamics that focus on the averaged macroscopic behavior, leaving out the fluctuation. With

the LBM, it is relatively easy to implement more complex boundary conditions, such as the curved boundary (Mei et al., 1999), when compared to conventional grid-based numerical integration. In addition, for models where the dynamics is very complex, use of parallel computing (Chen and Doolen, 1998) in combination with the LBM algorithm is greatly beneficial in terms of delivering simulation times in a straight forward manner. The LBM solves a problem at the microscopic level in order to recover particle density and velocity from macroscopic properties (Zhou, 2004), while traditional computational methods in fluid dynamics (such as the finite element method, finite difference method and finite volume method) solve macroscopic fluid dynamics equations.

The LBM consists of simple arithmetic calculations, so it is easy to implement the algorithm. In this method, the space is divided into a regular Cartesian lattice grid as a consequence of the symmetry of the discrete velocity set. Each lattice point has an assigned set of velocity vectors with specified magnitudes and directions connecting the lattice point to its neighboring lattice points. The total velocity and particle density are defined by specifying the number of particles associated with each of the velocity vectors. The microscopic particle distribution function, which is the only unknown, evolves at each time step through a two-step procedure: convection and collision. The first step, convection (or streaming), simply advances the particles from one lattice site to another lattice site along the directions of motion according to their velocities. This feature is borrowed from kinetic theory. The second step, or collision, is to imitate various interactions among particles by allowing for the relaxation of a distribution towards an equilibrium distribution through a linear relaxation parameter (Baoming and Daniel, 2003). The averaging process uses information based on the whole velocity phase space. The lattice Boltzmann equation can be viewed as a discretized version of the Boltzmann equation. LBM can be derived directly from the simplified Boltzmann Bhatnagher-Gross-Krook (BGK) equation (He et al., 1997a, 1997b).

Ngamsaad et al. (2009) used the lattice Boltzmann method (LBM) to study the dynamics of the oscillations of the Min proteins in ***E. coli***. Their results indicated that the LBM can be an alternative computational tool for simulating problems dealing with complex biological systems that are described by reaction-diffusion equations. They also used the LBM on a two dimensional system to investigate the possible evolutionary connection between the shape and cell division of ***E. coli***. The results showed that for the dimension 1 x 2 the oscillatory pattern is most consistent with experimental results. They also suggested that as the dimension of the system approaches a square shape, the oscillatory pattern no longer places the cell division of ***E. coli*** at the proper position. LBM simulation results were found to agree well with experimental results as shown in Figure 7. Both experiment and simulation implied that the position division site of ***E. coli*** locates at the midcell via the pattern formation of MinD. This suggestion is supported by previous reports (Hu and Lutkenhaus, 1999; Raskin et al., 1999a, 1999b; de Boer et al., 1990). As mentioned before, the first two computational methods neglect the fluctuation effects generally involved in protein dynamics.

Stochastic modeling and simulations

The methods mentioned in the previous sections of this review only deal with macroscopic features and provide average or trend behavior. To take into account more microscopic details, stochastic models are more relevant and suitable. Typically, the stochastic modeling approach is used in order to take fluctuation or noise into account. This approach allows us to understand how the intrinsic chemical fluctuations in spatially extended systems can cause different properties than what would be described by a mean-field model or deterministic counterpart.

Howard et al. (2003) studied the fluctuation effect due to the low copy number of Min proteins and focused on a stochastic model of oscillating in a Min protein system. They found that for some parameter regions, the copy number of Min proteins is low enough that fluctuations are essential for the generation of the oscillation pattern. Later, Modchang et al. (2005, 2008) investigated the response of oscillatory dynamics to various electric field strengths and the total number of Min proteins. Figure 8 shows a space-time plot of number of MinD proteins at lattice sites along the cell under the influence of an electric field. It can be seen that in the case of no external field (J = 0.0) concentrations, MinD were symmetric around the midcell. MinD has a minimum at the midcell which is in good agreement with that which was reported in previous studies. When the external electric field is turned on, a shift in the minimum of MinD was observed to be dependent. The positions of MinD concentration minimum are more pronouncedly shifted toward the left pole as J increases. These results are consistent, at least qualitatively, with those obtained with a deterministic partial differential model (Modchang et al., 2005). In Figure 8, two different numbers of total MinD protein copies were used. It can be clearly seen from the figure that using lower number of Min proteins (N = 200) can still produce the oscillation pattern but with a higher level of noise. They also reported that using fewer number of Min proteins degrades the cell division accuracy but using too much number of Min proteins costs ***E. coli*** to use more energy in order to maintain the oscillation pattern, so there should exist an optimal number of Min protein in ***E. coli*** (Howard et al., 2003; Modchang et al., 2008). These studies may be of significant importance in the development of new technological processes in the fields of agriculture, food processing and medicine. The electric field may stop ***E. coli*** cell division or may even kill ***E. coli***. Lastly, Kerr et al. (2006) studied a large number of protein oscillations in a stochastic model by using the

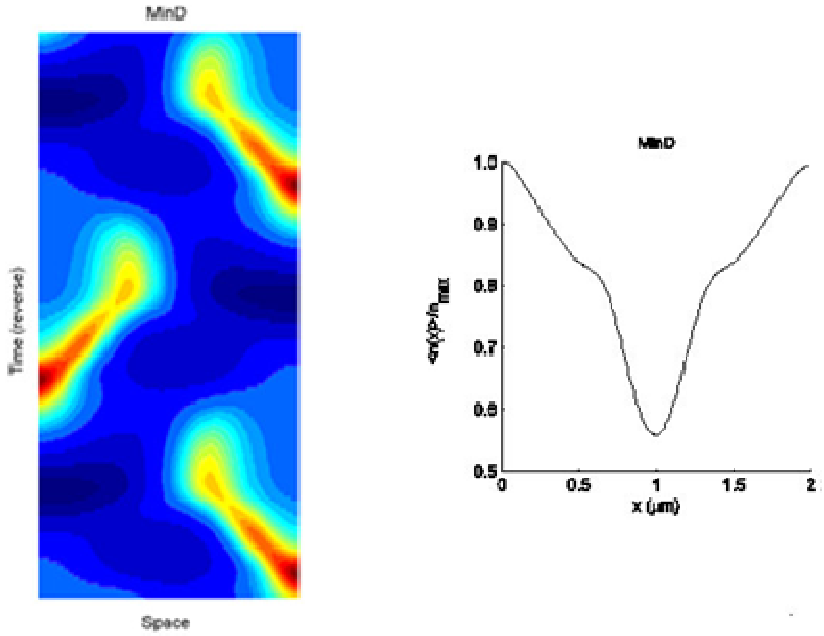

Figure 7. The LBM simulation results for MinD protein oscillation. (Left) Space-time plots of the MinD densities. The color scale runs from the lowest (blue) to the highest (red). The MinD depletion from midcell is immediately evident. Times increase from top to bottom, and the pattern repeats indefinitely as time increases. The vertical scale spans a time 1000 seconds. (Right) The time average of MinD densities $\langle n(x) \rangle / n_{\max}$, related to their respective time-average maxima, as a function of position x (in μm) along the bacterium.

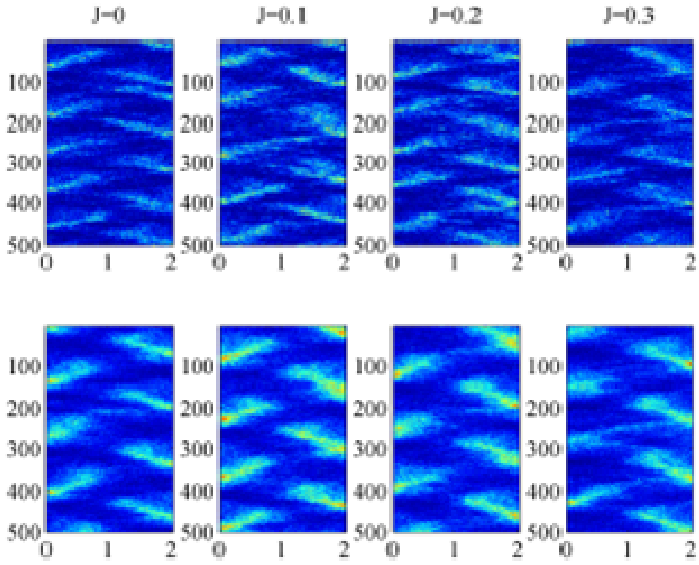

Figure 8. Space-time plots of the number of MinD proteins at lattice sites along the bacterium cell for J = 0.0 μm/s to J = 0.3 μm/s with total number of MinD protein molecules N = 200 in upper panel and N = 800 in the lower panel. The color scale, running from blue to red, denotes an increase in the numbers, and hence concentrations, of Min proteins from the lowest to the highest. The vertical scale spans a time of 500 s. The time increases from top to bottom. The horizontal scale spans the bacterial length 2 μm. The oscillation pattern is robust. Even at the low number of Min protein copies N = 200 the oscillation pattern still can be generated but with a larger amount of noise as compare to N = 800.

Monte Carlo simulation and showed that their results agree well with the results from a deterministic treatment of the equations. Ultimately, more realistic (and complicated) models still need to be developed.

CONCLUDING REMARKS

Understanding bacteria cell division is central to understanding microorganisms, as well as the origin of life. However, the functions and mechanisms underlying the transport process and movement still remain unclear. ***E. coli*** is a prokaryotic microorganism well appropriate for research and biotechnological applications. In these bacteria, chromosome replication and cell division appear to be coordinated such that the frequency of initiation of replication determines the rate of cell division. To control the number of bacteria via the division process through interrupting the protein expression or protein dynamics is one of the practical means.

Using quantitative techniques to conduct research would provide more precise and useful information toward understanding and applying this prokaryote. Achieving this goal requires more experimental and theoretical work, especially more realistic (and complicated) model development using deterministic and stochastic approaches. Therefore the quantitative approach may be of significance in the development of new technological processes in the fields of biology, agriculture, food and medicine. Given the significance of protein oscillation in correlation with cell division, another interesting question is: how is abnormal or unsuccessful cell division affected by the abnormal protein oscillation? More specifically, under external stresses such as pH, heat, electric fields, or magnetic fields, how does each perturbation or combined perturbation affect protein oscillation in correlation with cell division? Answering such questions will be significant in the development of these new technological processes. Researchers can expect that the quantitative approach will help them define the roles of these factors in affecting ***E. coli*** division more accurately. It may also help minimize the number of experiments necessary when conducting future ***in vitro*** studies of cell division. Moreover, establishing a correct link to other compare-tments like signal transduction or system biology, will contribute greatly to the health and well-being of our communities. More quantitative tools could contribute to great improvements in the dynamic models of protein oscillation, cell division and related applications. Thus these quantitative approaches will benefit biotechnological research, which includes multidiscipline research tools, including biological, physical and mathematical approaches.

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