Research in the Department of Theory & Bio-Systems

Denken ohne Erfahrung ist leer, Erfahrung ohne Denken ist blind.
Immanuel Kant

The main objective of our research activities is to understand the hidden dimensions of self-organization in biomimetic and biological systems. The molecular building blocks of these systems join „by themselves" and form a variety of supra-molecular assemblies, which then interact to produce even larger structures and networks.

Reinhard Lipowsky 11.11.1953
1978: Diploma, Physics, (University of Heidelberg)
1982: PhD (Dr. rer. nat.), Physics (University of Munich)
1979-1984: Teaching Associate (University of Munich)
1984-1986: Research Associate (Cornell University)
1986-1988: Group leader (FZ Jülich)
1987: Habilitation, Theoretical Physics (University of Munich)
Thesis: Critical behavior of interfaces: Wetting, surface melting and related phenomena
1989-1990: Associate Professorship (University of Munich)
1990-1993: Full Professorship (University of Cologne), Director of the Division "Theory II" (FZ Jülich)
Since Nov 1993: Director (Max Planck Institute of Colloids and Interfaces, Potsdam)

The associates of the department form several research groups. At present, the research group leaders and topics are:

· *Rumiana Dimova:* Biophysics Lab;
· *Thomas Weikl:* Proteins and Membranes;
· *Mark Santer:* Carbohydrates and Polysaccharides;
· *Christian Seidel:* Polymers and Polyelectrolytes;
· *Andrea Grafmüller:* Multiscale Simulations;
· *Angelo Valleriani:* Stochastic Processes;
· *Stefan Klumpp:* Regulation of Bioprocesses.

The main results of these research groups are described in separate reports on the following pages. These reports are ordered in a bottom-up manner, i.e., from small to large length scales, and related to five research areas: Biopolymers, Motors and Filaments, Membranes and Vesicles, Soft Interfaces, and Complex Systems. Here, the results of the research groups will be briefly summarized and some additional results will be highlighted.

Biopolymers

Carbohydrates and polysaccharides have been studied by the research group of *M. Santer* using molecular dynamics simulations. The focus was on two types of polysaccharides: GPI-anchors, which can link a variety of proteins to cell membranes, and lipopolysaccharides, which protect bacteria against the infection by bacteriophages. The conformational freedom of the different glycosidic bonds between the subunits of the polysaccharides was determined by calculating free energy landscapes as a function of glycosidic torsion angles: even short oligosaccharides were shown to be relatively flexible.

Proteins that act as enzymes must first bind the reaction partners as ligands. The group of *T. Weikl* considered the temporal ordering of these binding processes with respect to conformational changes of the enzymes: the enzyme may undergo conformational changes before ligand binding or the ligands may first bind and then induce conformational changes of the enzyme. The temporal ordering has no effect on the binding equilibrium, but affects the binding kinetics and, thus, may be revealed by mutations of the protein.

Molecular Motors and Filaments

Intracellular cargo is transported by teams of molecular motors that pull on the cargo via elastic stalks. The simplest case corresponds to cooperative transport by two identical motors as shown in **Fig. 1**. The influence of this elastic coupling between the motors on the transport properties has been addressed in the framework of chemomechanical networks and semistochastic models (see the report by *F. Berger* and *C. Keller*). The chemomechanical networks are relatively complex but involve only two additional parameters that can be deduced from the cargo trajectories. The semi-stochastic models reveal different interference regimes, in which the motors stall each other or pull each other from the filament.

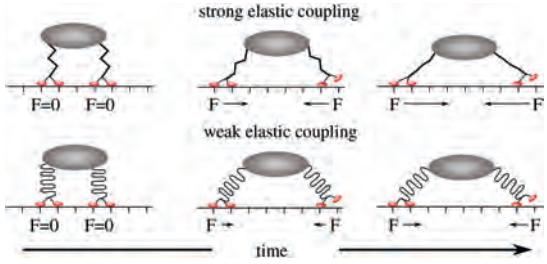

Fig.1: Two molecular motors coupled to a cargo particle via their elastic stalks or linkers. The motors step forward stochastically and stretch their linkers, thereby inducing an elastic interaction that generates the mutual strain force F. Strong elastic coupling leads to a fast buildup of large forces, whereas weakly coupled motors experience only relatively small forces. [F. Berger et al, Phys. Rev. Lett. (2012)]

Another molecular motor, for which a chemo-mechanical network has been constructed, is myosin V that steps along actin filaments, see **Fig 2**. Furthermore, stochastic tug-of-wars between two teams of molecular motors were experimentally confirmed for the transport of early endosomes in fungi. In this case, dyneins that bind to a cargo or unbind from it can change the cargo's direction of motion, see **Fig 3**.

In the context of actin filaments, we have addressed a recent controversy about the depolymerization of actin filaments (see the report by *T. Niedermayer*). Using single filament experiments, it was shown that the depolymerization of actin filaments typically proceeds in a bi-phasic way, with an initial fast phase interrupted by a slow phase, see **Fig. 4**. In contrast to previous proposals, the interruptions were shown to be caused by the local and random dimerization of actin subunits. The theoretical analysis of the stochastic interruption times and pause durations provides a general method to determine rather small changes in the molecular interactions between the subunits of actin filaments.

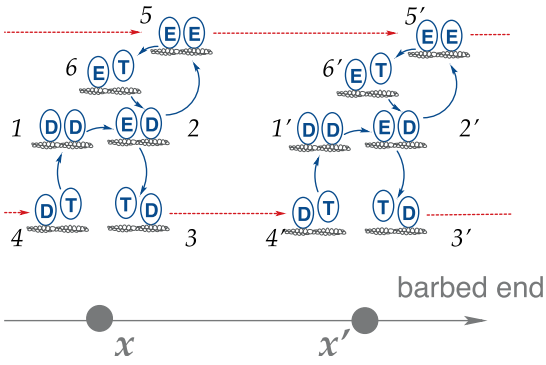

Fig. 2: Chemomechanical network for myosin V that steps along actin filaments. At each filament position x, x',..., the chemical network of the motor consists of six states. The motor can perform two types of mechanical forward steps, l34') and l55'), towards the barbed end of the filament. [V. Bierbaum et al, Biophys. J. (2011); PLoS ONE (2013)]

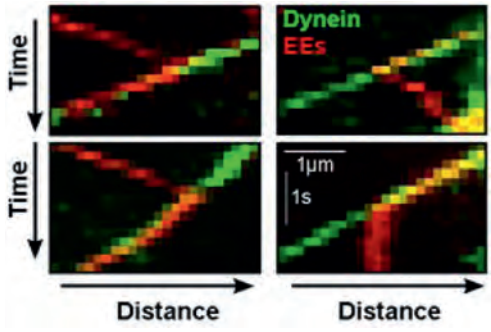

Fig 3: Changes in the direction of motion for red-labeled cargo particles by green-labeled dynein motors that bind to (left) or unbind from (right) the cargo. [M. Schuster et al, PNAS (2011)]

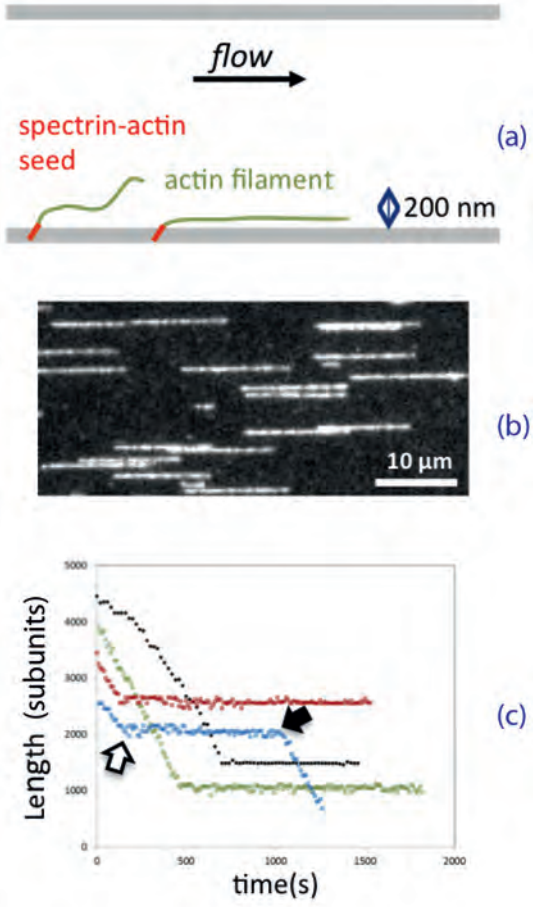

Fig.4: (a) Actin filaments are anchored to the chamber wall and aligned by a continuous microfluidic flow. Actin depolymerization is induced by fast switching to a flow channel without actin; (b) The filaments are imaged using TIRF or epifluorescence microscopy; and (c) The length of the filaments as measured during depolymerization; black data points correspond to a filament grown from MgATP-actin whereas red, green, and blue data points were obtained for three filaments grown from MgADP-actin. One pause in depolymerization occurs between the white and black arrow in (c). [T. Niedermayer et al, PNAS (2012)]

Ribosomes and Protein Synthesis

Ribosomes are rather complex molecular machines that synthesize proteins by translating the codon sequences of mRNA molecules into peptide chains. In order to do so, the ribosomes move along the mRNAs and translate one codon after another by binding and processing cognate tRNA molecules that are charged with the correct amino acids.

In order to understand this process of translational elongation, one has to take two important molecular features into account. First, the ribosome has three binding pockets for tRNA molecules, the A-, P-, and E-sites, see **Fig. 5**(a). As indicated in this figure, these three sites are aligned along the mRNA that is translated by the ribosome. Second, a tRNA can only bind to the ribosome after it has formed a ternary complex with an EF-Tu protein and a GTP nucleotide, see **Fig. 5**(b).

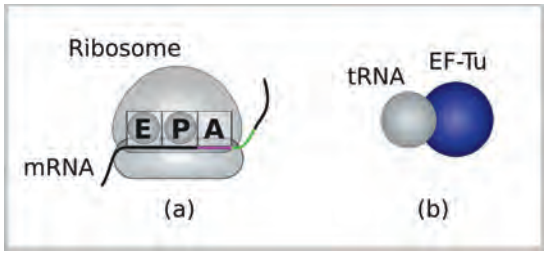

Fig.5: (a) Schematic view of a ribosome (grey dumbbell) that translates a mRNA molecule (black strand). The ribosome has three binding pockets for tRNA molecules, the A-, P-, and E-sites. These three sites are aligned along the mRNA and their separations are equal to the length of a single codon; (b) Ternary complex formed by a tRNA molecule (small grey ball), an EF-Tu protein (large blue ball), and a GTP nucleotide (not shown).

Each tRNA molecule that is processed by the ribosome first binds as a ternary complex to the A-site and is then translocated from the A- to the P-site. During the next elongation cycle, this tRNA is moved from the P- to the E-site, from which it is finally released.

A single, codon-specific cycle of translational elongation involves several ribosomal states as depicted schematically in **Fig 6**. At the beginning of the elongation cycle, the ribosome contains two tRNA molecules in its P- and its E-site but has an empty A-site, which is located at the codon to be translated. In **Fig. 6**, this codon is colored in red. When a cognate tRNA molecule arrives at the ribosome by diffusion, it first binds loosely to the A-site but becomes fully accommodated into this site after its correct anticodon has been recognized. During the latter substeps, the E-site tRNA and the EF-Tu molecule are released from the ribosome and a new peptide bond is formed. However, before the translating ribosome is able to bind and process a cognate tRNA, it typically samples through a large number of noncognate tRNAs that also bind to the ribosomal A-site and, thus, impede binding of the cognate tRNA, see upper left cartoon in **Fig. 6**.

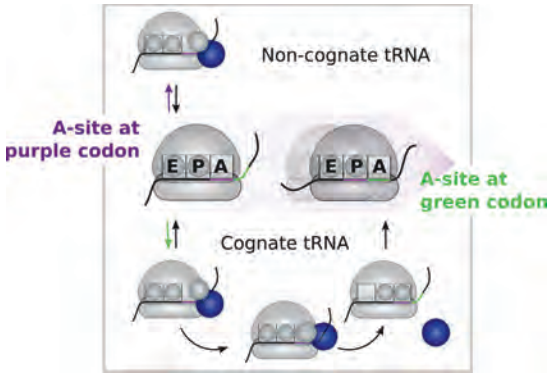

Fig.6: Elongation cycle of a ribosome corresponding to the translation of a single codon (red mRNA segment): Initially, the ribosome dwells at this codon with an empty A-site until a ternary complex arrives and occupies this site. This ternary complex is typically non-cognate (upper left cartoon) and is then released from the A-site without further processing. If a cognate ternary complex is bound (lower left cartoon), it is recognized by its anticodon and is then fully accommodated into the A-site (lower right cartoon). During this accommodation step, the EF-Tu molecule is released. After the formation of a new peptide bond (not shown), the ribosome undergoes translocation and moves to the subsequent codon (green mRNA segment) in order to start the next elongation cycle. [S. Rudorf et al, to be published]

We have recently developed a quantitative theory that takes both the formation of ternary complexes and the competitive binding between cognate and non-cognate tRNAs into account and allows to calculate the codon-specific elongation times of the ribosome. Another interesting aspect of translation that we studied theoretically is the robustness of protein synthesis with respect to variations of individual tRNA concentrations.

Membranes and Vesicles

Lipid molecules in aqueous solution self-assemble into bilayer membranes that have a thickness of about 4nm. In order to desorb from the membrane again, a single lipid has to overcome a large free energy barrier that has been determined in the group of *A. Grafmüller* using atomistic molecular dynamics simulations. Unexpectedly, the desorption free energy was found to increase with membrane tension because of the conformational entropy of the lipid tails.

Lipid vesicles exposed to different aqueous phases exhibit unusual morphologies and morphological transitions as discovered in the group of *R. Dimova*: wetting transitions, droplet-induced budding processes, and spontaneous tubulation, i.e., the formation of membrane nanotubes that are stable even in the absence of external forces. The latter process provides direct evidence that the polymer/lipid interactions lead to a spontaneous membrane curvature that generates a large membrane tension. In fact, one unique feature of aqueous phase separation in vesicles is the possibility to directly determine the membrane tensions from the (effective) contact angles as visible in the optical microscope, see **Fig. 7**, and from the interfacial tension between the two liquid phases.

The interactions of nanoparticles with membranes and vesicles have been studied by the group of *T. Weikl* using Monte Carlo methods to minimize the free energy of the membrane/particle systems. These studies revealed strongly attractive interactions between nanoparticles adsorbed onto vesicles. As a result of these interactions, the adhering nanoparticles aggregate on the vesicle membranes and often form linear chains enwrapped by membrane nanotubes.

Another membrane system that has been addressed is provided by double-membrane structures that play an important role in cellular processes such as autophagy, reproduction, and viral infection. In these processes, one typically starts from double-membrane sheets that become unstable and close up into double-membrane vesicles. The stability of a double-membrane sheet depends primarily on its lateral size and the spontaneous membrane curvature along its rim [R. Knorr et al, PLoS ONE (2012)].

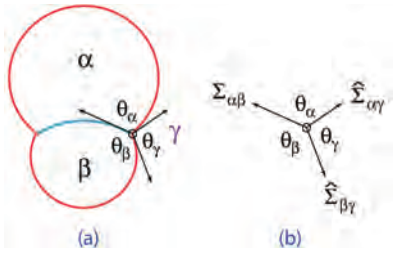

Fig.7: (a) Morphology of vesicle membrane (red) enclosing two liquid droplets. The upper droplet contains the PEG-rich α phase, the lower one contains the dextran-rich β-phase. The interface (blue) between the droplets meets the membrane along the three-phase contact line, with the exterior phase denoted by γ. The two membrane segments and the interface define three (effective) contact angles, θ_α, θ_β, and θ_γ that can be directly measured by optical microscopy; (b) These contact angles and the interfacial tension $\Sigma_{\alpha\beta}$ of the ($\alpha\beta$) interface determine the two membrane tensions $\hat{\Sigma}_{\alpha\gamma}$ and $\hat{\Sigma}_{\beta\gamma}$.

Interfacial Phenomena

Polymer Brushes consisting of diblock copolymers undergo micro-phase separation and provide surfaces with stable nanoscale patterns, which can be used to control the organization of nanoparticles into larger aggregates as studied by the group of *C. Seidel*. Using dissipative particle simulations, a variety of different morphologies for these aggregates has been identified as well as morphological transitions, which resemble wetting transitions of liquid droplets at chemically patterned surfaces.

Morphological wetting transitions can be induced, in a rather simple way, by increasing the volume of the liquid droplets. As a consequence, these transitions also have a strong effect on surface nucleation and lead to non-isomorphic nucleation pathways. One example for such a pathway has been studied in the context of edge melting of alkane monolayers (*H. Kusumaatmaja* et al, Phys. Rev. Lett. (2012).

Complex Systems

Most macromolecules within the living cell are continuously synthesized and degraded. Experimental data on mRNA degradation and translation have been analyzed in the group of *A. Valleriani* using stochastic modeling. In the context of mRNA degradation, it was shown that the experimentally determined decay patterns for the mRNA degradation can be used to determine the age-dependent decay rates and the life time distributions of the mRNA molecules. In the context of translation, data on ribosomal profiling have been analyzed for different growth and stress conditions.

The independent research group of *S. Klumpp* addressed the interplay of physical constraints and functional requirements in living systems, with a focus on molecular machines involved in gene expression, genetic circuits, and cellular dynamics. Genetic circuits in bacteria are intimately coupled to cellular growth because many parameters of gene expression depend on the growth rate. These dependencies have been studied for the replication control system of plasmids. Some bacteria can respond to magnetic fields via organelles called magnetosomes that contain magnetic nano-particles. Robust chain formation was found to require both magnetic interactions and active transport.

Another, more abstract class of complex systems that has been studied is provided by binary or 'spin' variables on scale-free networks with correlations between their vertex degrees. In assortative and dissortative networks, the high-degree vertices are primarily connected to other high-degree and low-degree vertices, respectively. In both cases, the networks can be decomposed into vertex layers, which are ordered at low temperatures and undergo successive phase transitions as the temperature is increased, see **Fig. 8**.

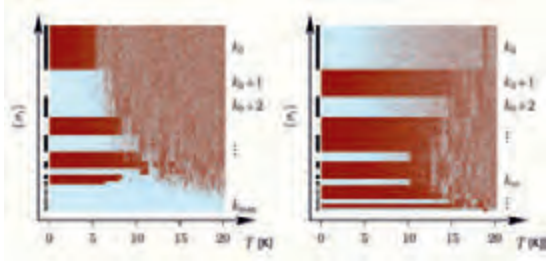

Fig 8: Typical configurations of binary or 'spin' variables on assortative (left) and dissortative (right) scale-free networks as a function of temperature T. 'Spin-up' and 'spin-down' states are shown in red and blue, respectively. Each column parallel to the y-axis shows the 'spin' states { i} of all vertices in the network. As the temperature increases, the ordered vertex layers become disordered one after another.
[J. Menche et al, Phys. Rev. E (2011)]

International Max Planck Research Schools

The department of Theory & Bio-Systems was in charge of the International Max Planck Research School (IMPRS) on "Biomimetic Systems", which was in operation from 2000 until 2012, and is also in charge of the new IMPRS on "Multiscale Biosystems", which will start in July 2013.

For additional information about research at the Department of Theory & Bio-Systems, see the following reports and www.mpikg.mpg.de/th/.

Reinhard Lipowsky
Director, Theory & Bio-Systems Department