# Polypeptides: Amyloid Formers and Molecular Motors

**Volker Knecht** 06.01.1970
**1996:** Diploma in physics (University of Kaiserslautern)
Thesis: Computer based renormalization group study of the two-dimensional XY model
**1997:** Software trainee (TECMATH GmbH, Kaiserslautern)
**1998-1999:** Software engineer (LMS Durability Technologies GmbH, Kaiserslautern)
**2003:** PhD, Physics (MPI of biophysical chemistry, Göttingen)
Thesis: Mechanical coupling via the membrane fusion SNARE protein syntaxin 1A: a molecular dynamics study
**2003-2005:** Postdoc (University of Groningen, the Netherlands)
**2005-2006:** Postdoc (MPI of Colloids and Interfaces, Potsdam)
**Since 2006:** Group Leader (MPI of Colloids and Interfaces, Potsdam)

Proteins and polypeptides in general are biopolymers composed of amino acid residues that fold into well-defined structures depending on their amino acid sequence. Folding is crucial for the function of a protein whereas misfolding can cause severe diseases. Our aim is to understand protein function and diseases on the molecular level. The information on the molecular dynamics of polypeptides accessible experimentally is very limited. Therefore we use molecular dynamics computer simulation techniques to model the process by which proteins sample conformational space. Molecular dynamics simulations which are based on iteratively solving Newton's equations of motion to propagate a system in time and semi-empirical force fields to describe interatomic interactions can provide highly detailed information about the properties of proteins in solution or at interfaces. Here, the protein and its solvent environment are described in atomic detail. Currently, we focus on the molecular basis of so-called amyloid diseases and the function of molecular motors.

## Amyloid Peptides

Amyloid diseases including Alzheimer's, Creutzfeld-Jakob disease and bovine spongiform encephalopathy (BSE) are associated with the conversion of a protein from a soluble (functional) form into higher order fibrillar aggregates rich in β-structure. The development of specific agents against amyloid diseases requires an understanding of the (mis)folding and aggregation of fibrillogenic proteins on a microscopic level. In collaboration with Gerald Brezesinski's group from the Interfaces department, we study the folding and aggregation of small amyloid peptides in solution, see **Fig. 1**, and at interfaces, see **Fig. 2 [1-4]**. The systems investigated include model peptides containing between 12 and 18 residues, see **Fig. 1(a,b)**, as well as an 11-residue fragment of the amyloid β (Aβ) peptide associated with Alzheimer's disease, Aβ(25-35), see **Fig. 1c**.

Fibrillogenic peptides, typically, are found to form β-hairpin as in **Figs. 1(a,b)** and **2(a)** or coil conformations. As shown in **Fig. 1(b)** (middle), a predominant conformation has been identified for the inner residues of the model amyloid peptide LSFD **[4]**. Knowledge of a predominant conformation may facilitate the design of possible inhibitors of LSFD aggregation as a testing ground for future computational therapeutic approaches against amyloid diseases. In ordered aggregates, β-hairpins are either placed side by side, see **Fig. 1(a)**, or dissolved often leading to extended conformations, see **Figs. 1(c)** and **2(b)**. Some of these dimer conformations might reflect the structure of fibrillar aggregates, see **Fig. 1(a,c)**.

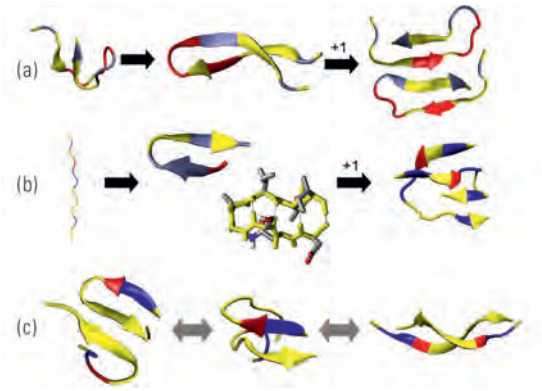


*Fig. 1: β-hairpin folding and dimerization of fibrillogenic peptides in solution in molecular dynamics simulations. The model amyloid peptides (a) B18 [1-3] and (b) LSFD [4] as well as (c) the peptide Aβ(25-35) associated with Alzheimer's disease are depicted. Initial and typical backbone configurations during the simulations are shown in ribbon representation. Colors distinguish between hydrophobic (yellow), hydrophilic but neutral (blue), and charged residues (red). In (b), the conformation of the monomer in the folded state is shown as sticks.*

For the first time, we have studied reversible β-hairpin peptide folding at an interface, see **Fig. 2(a) [4]**, the formation of side-by-side-hairpin β-sheet dimers **[3]**, see **Fig. 1(a)**, and the conformational distribution of a peptide containing more than ten amino acid residues in dimeric form at equilibrium, see **Fig. 1(c)**, using an explicit solvent model.

The studies are challenging because of the large computational expense of the simulations and the roughness of the free energy landscape underlying folding. Therefore, our initial studies have been restricted to relatively short model peptides. But, using novel methods to enhance the sampling including the coupling of simulations of various copies of the system at different temperatures (replica exchange), we are now investigating larger peptides such the 26-residue peptide Aβ(10-35) in mono- and dimeric form. A non-amyloidogenic peptide of the same size, the (antimicrobial) peptide NK-2, is investigated for comparison.

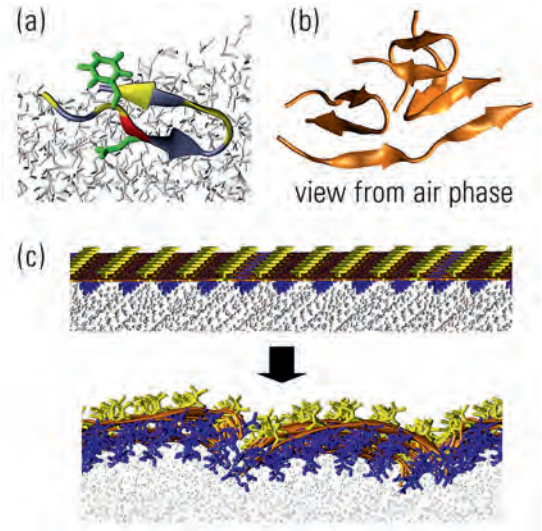


*Fig. 2: β-sheet forming peptides at water/vapor interface. An LSFD (a) monomer [4] and (b) tetramer, as well as (c) a crystalline monolayer of the peptide G(VT)$_5$ are depicted. The backbone of the peptides is shown in ribbon representation. In (a), the color code for the peptide backbone is similar to that chosen in Fig. 1, and selected side chains are shown as green sticks. In (c), the side chains of the peptides are displayed as sticks, colors distinguish between valine (yellow) and threonine (blue). In (a) and (c), water molecules are depicted as white sticks.*

The synthetic peptide with sequence G(VT)$_5$ synthesized in Hans Börner's lab in the Colloids department has been studied at a water/vapor interface where it forms β-rich crystalline monolayers. Based on data from x-ray scattering and infrared spectroscopy, we modeled an idealized monolayer in which the peptides are extended and form in-register β-sheets over their whole length, see **Fig. 2(c)**, top, and used simulations to refine the structure. We found that the β-strands are strongly bent and that β-sheets are dissolved at the termini as in **Fig. 2(c)**, bottom. When the subphase contains NaCl, both types of ions are strongly adsorbed at the termini of the peptides.

## Molecular Motors

The Kinesin molecules represent a large motor-protein family that transports cargoes within a cell by moving on microtubule filaments consisting of α- and β-tubulin dimers coupled to ATP hydrolysis, see **Fig. 3**. Conventional kinesin, henceforth denoted as kinesin, is a homodimer containing two heads by which kinesin binds ATP and walks along microtubules. The molecular details of this process are poorly understood. In order to quantify the energy levels of kinesin in all its nucleotide-binding states, we aim to determine equilibrium constants and activity scales for ATP hydrolysis from quantum-mechanical calculations using a continuum model for the solvent. Test studies to evaluate the accuracy and computational expense of different levels of theory are in progress.

In order to understand the mechanical response of kinesin to the chemical transitions, we are studying the conformational changes of a kinesin monomer in solution and at tubulin during the catalytic cycle using classical molecular dynamics simulations. Structural models based on x-ray crystallography or cryo-electron microscopy are used as input for the simulations. The timescales for each nucleotide state employed here exceed those used in previous studies of the same system by almost two orders of magnitude. We found that the region connecting the head and the adjacent neck region, the neck linker, is highly flexible and, thus, provides a hinge, for all nucleotide states. Currently, we are investigating a kinesin dimer with one of its heads in the ATP state attached to a tubulin dimer, see **Fig. 3(a)**. Within 50 ns of simulation, a large conformational change has occurred, see **Fig. 3(b)**. This study opens the perspective to investigate the mechanical step of kinesin at tubulin, the so-called "power stroke", in full atomic detail.

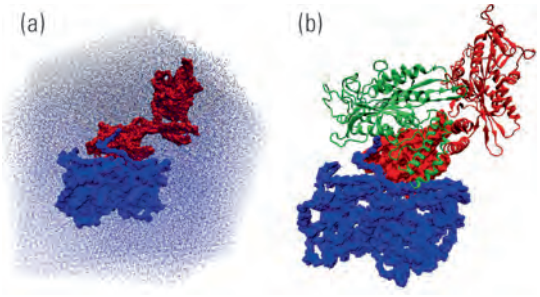


*Fig. 3: Modelling the kinesin dimer at tubulin in atomic detail. (a) The system simulated includes a kinesin dimer (red surface) as well as an α-β tubulin dimer ("tubulin", blue surface). The initial configuration of the protein complex has been modelled based on an atomic model of a kinesin monomer at tubulin and a solvated kinesin dimer using rigid body motion and solvated in explicit water (blue dots). (b) Conformation of tubulin (blue) and bound kinesin head (red surface) as well as configuration of the unbound head before (red, ribbons) and after a 50 ns simulation (green).*

V. Knecht, N. Awasthi, Y. Chai, C. v. Deuster, M. Kittner, B. Klasczyk, A. Krukau, Y. Smirnowa
*vknecht@mpikg.mpg.de*

**References:**

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**[3]** Knecht, V., Lipowsky, R.: Folding and formation of side-by-side-hairpin β-sheet dimers by a model amyloid peptide in solution, submitted.
**[4]** Knecht, V: β-hairpin folding by a model amyloid peptide in solution and at an interface, J. Phys. Chem. B **112**, 9476-9483 (2008).