Institutionen för Biovetenskaper och näringslära

Biomolecular Simulations, from RNA to Protein: Thermodynamic and Dynamic Aspects.

AKADEMISK AVHANDLING
som för avläggande av tekn. doktorsexamen vid Karolinska Institutet offentligen försvaras i sal Månen, Alfreds Nobels Allé 12.

Fredagen den 24 augusti, 2012, kl 10.00

av
Olof Allnér

Huvudhandledare:
Prof. Lennart Nilsson
Karolinska Institutet
Inst. för Biovetenskaper och Näringslära

Bihandledare:
Dr. Alessandra Villa
Karolinska Institutet
Inst. för Biovetenskaper och Näringslära

Fakultetsopponent:
Assoc. Prof. Joanna Trylska
University of Warsaw
Interdisciplinary Centre for Mathematical and Computational Modelling

Betygsnämnd:
Prof. Göran Widmalm
Stockholms Universitet, Organisk kemi

Prof. Johan Åqvist
Uppsala Universitet, Dept. Cell and Molecular Biology

Prof. Alexander Lyubartsev
Stockholms Universitet, Div. of Physical Chemistry

Stockholm 2012

ABSTRACT

The process of transforming the information stored in the DNA of genes into functional RNA molecules and proteins via transcription and translation is the most fundamental process of all known life. Even though these processes involve large macromolecules and dynamics on long time scales they all ultimately rely on atomic level interactions between nucleic acids or amino acids. Only a few experimental techniques are available that can study the large systems involved in atomic detail. Computer simulations, modeling biological macromolecules, are therefore an important tool in investigating fundamental biological processes. In this thesis, Molecular Dynamics (MD) simulations have been used to study the translation of mRNA by tRNA and the function of the regulatory riboswitches. The thesis also covers the improvement of methodology by the development of a new representation of the important Mg^{2+} ions and an improvement of the understanding of the connection between MD and experimental NMR data.

In Paper I, the effect of post transcriptional modifications of the tRNA anti codon on the decoding of mRNA in the ribosome is studied. All atom MD simulations have been performed of the ribosomal A site with and without modifications present, including extensive free energy calculations. The results show two mechanism by which the decoding is affected: The further reach provided by the modifications allows an alternative outer conformation to be formed for the non cognate base pairs, and the modifications results in increased “catalytic” contacts between tRNA, mRNA and the ribosome.

In Paper II, the folding mechanism of the *add* A riboswitch is studied under different ionic conditions and with and without the ligand bound. In addition to standard simulations, we simulated the unfolding by umbrella sampling of distance between the L2 and L3 loops. In the results, no significant effect of Mg^{2+} or Na^{+} ion environments or ligand presence can be seen. But a consistent mechanism with the P3 stem being more flexible than P2 is observed. More data might however be needed to draw general conclusions.

In Paper III, the parameters describe Mg^{2+} ions in MD simulations are improved by optimizing to kinetic data of the H_2O exchange. Data from NMR relaxation experiments was used as optimization goal. The newly developed parameters do not only display better kinetic properties, but also better agreement with experimental structural data.

In Paper IV, the dynamical data, obtained from NMR relaxation experiment of a protein is related to dynamics seen in an MD simulation. The analysis provides important information for the interpretation of experimental data and the development of simulation methods. The results show, among other things, that significant parts of the entropy are not seen by NMR due to a limited time window and inability to account for correlation of motions.

ISBN 978-91-7457-801-0