

808.1-Pos Board # B59.1

Thermodynamic Molecular Switch in the Hydrophobic Interaction of 35 Dipeptide Pairs

Paul W. Chun¹

¹University of Florida, PO Box 100245, Gainesville, Florida 32610-0245

Applying the Planck-Benzinger methodology, the sequence-specific hydrophobic interactions of 35 dipeptide pairs were examined over a temperature range of 273-333 K. The results imply that the negative Gibbs free energy minimum at a well-defined stable temperature, $\langle T_s \rangle$, has its origin in the sequence-specific hydrophobic interactions, which are highly dependent on details of molecular structure. Each case confirms the existence of a thermodynamic molecular switch wherein a change of sign in $\Delta C_p^0(T)$ leads to true negative minimum in the Gibbs free energy change of reaction and a maximum in the related equilibrium constant. All interacting biological systems examined using the Planck-Benzinger methodology have shown such a thermodynamic switch at the molecular level, suggesting its existence may be universal.

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In Vitro and In Vivo Motilities of Nuclear Transport Cargos

Cecile Fradin¹, David Zbaida², Michael Elbaum²

¹McMaster University, 1280 Main St. W, Hamilton, Ontario L8S4M1 Canada, ²Weizmann Institute of Science, Herzl St., Rehovot, 76100 Israel

Nuclear import of proteins involves recognition of the cargo by two helper proteins, leading to the formation of a complex, which is then translocated to the nucleus. The directionality of transport is due at least in part to the small protein Ran, present in the Ran-GTP form in the nucleus and in the hydrolyzed Ran-GDP form in the cytoplasm, and able to dissociate the cargo only in the first case. Using fluorescence correlation spectroscopy, we tested the efficiency of this molecular switch by measuring the diffusion coefficient of a fluorescent cargo in presence of the two helper proteins and of increasing concentrations of either Ran-GTP or Ran-GDP. As expected we observe an increase in the cargo mobility (signature of the complex dissociation) when Ran-GTP is added, and no change when Ran-GDP is added. We then measured the mobility of a fluorescent cargo in vivo. Whereas in the nucleus the observed mobility corresponds to the expected slightly hindered diffusion of the cargo, in the cytoplasm it is too small to correspond to the diffusion of the complex. This result could be explained either by the presence of a typical mesh size within the cytoplasm, critically slowing down the complex compared to the simple cargo, or by specific interaction of the complex with cellular structures such as the microtubule network.

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The Bacterial Flagellar Hook Structure

T R Shaikh¹, D Thomas², F Samatey³, H Matsunami³, K Imada³, K Namba³, **D J DeRosier**²

¹NY Dept. Pub. Health, Empire State Plaza, Albany, New York 12201, ²Brandeis University, MS029, Waltham, Massachusetts 02454, ³ERATO, Frontier Biosciences, Osaka University, 3-4 Hikaridai, Seika, Kyoto, 619-0237 Japan

A 10-micron long complex of nine proteins makes up the sturdy, segmented, extracellular rod, hook and filament (or axial component) of the flagellum of *Salmonella typhimurium*. The sequences of the nine proteins except the cap protein (FliD) have at their N and C termini, heptad repeats characteristic of an alpha-helical bundle. Moreover, the segments characterized have a common helical symmetry. The hypothesis that these alpha-helical folds form an interlocking alpha-domain within and between the contiguous segments of the axial structure has received support from structural studies of the filament. We used electron cryomicroscopy to generate a high-resolution map of the hook. We docked atomic models for the two outer domains of the hook subunit into the corresponding features of the map. The innermost domains are interdigitated ~1 nm rods, which form a tube having a 3 nm axial lumen, a feature seen in maps of the filament. The rods are somewhat shorter than those in the filament consistent with the shorter sequences thought to generate the fold. The N and C termini of the atomic model, which lie in the middle domain, point towards the spoke of density that connects to the inner rods. Our results further support the hypothesis of a common, interlocked alpha domain for the axial proteins.

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Biological Applications of Colloidal Nanocrystals

Wolfgang J. Parak¹, Teresa Pellegrino², Rosanne Boudreau³, Mark Le Gros³, Daniele Gerion², Christine M. Micheel², Carolyn A. Larabell³, Paul Alivisatos²

¹Center for Nanoscience, Amalienstrasse 54, Muenchen, 80799 Germany, ²Department of Chemistry, Hildebrand Hall B62, Berkeley, California 94720, ³Lawrence Berkeley National Lab, MS 6-2100, Berkeley, California 94720

Colloidal nanocrystals are building blocks of the "nanoworld". Their electronic properties enable the building of single-electron transistors, their optical properties can be used to generate fluorescence labels with many different colors. Based on the principles of molecular recognition and self assembly biological molecules can be used to arrange nanoscale building blocks. Two applications will be discussed. Colloidal gold nanocrystals were conjugated with a controlled number of DNA molecules per nanocrystal. By using complementary sequences of DNA molecules that were attached to different nanocrystals, small groupings of gold nanocrystals could be formed. Biomolecule conjugated colloidal semiconductor nano-crystals also have been used to fluorescence label structural compartments of cells. These nanocrystals were found to be actively incorporated by living cells. It will be described how cells "eat" nanocrystals and an assay for cell mobility based on this fact will be introduced.