BE.342/442 Thursday, December, 2005
Topic: Prof. Zhang's Research and Guest Lecture by Andreas Mershin: "Applying Biomimicry to Nanotechnology."

Administrative
Take-home midterms will be returned next Tuesday.

Stabilization of membrane proteins with detergents
(Continued from last time.)

Structure of bovine rhodopsin: transformation in optical properties in photo cycle.
Two distinctive absorptions are observed in bovine rhodopsin: in the dark, near 500 nm, and under light, at 380 nm. Heating, as well as illumination, changes the relative ratios of the absorbance peaks at these two frequencies.

Rhodopsin tends to degrade over time. Detergents such as A6D and DM can stabilize the protein to some extent, but many require cold temperatures to impart stability. A6D alone can stabilize the protein at 55°C for more than 11 hours. Discovery of new detergents could be critical to further study of membrane proteins. Without stabilization by detergents, membrane proteins tend to aggregate out of solution due to the driving force for hydrophobic bonding in their nonpolar regions, which are normally embedded in membranes. Detergents can coat these regions with their hydrophobic tails, presenting hydrophilic head groups to the solvent and stabilizing the protein in its native conformation.

Joanne Yeh and co-workers used A6DA6K with 200 mM NaCl and OG with 200 mM NaCl to stabilize a matrix protein and allow it to crystallize.

> Source: Joanne I. Yeh et al, "Peptergents: Peptide Detergents that Improve Stability and Functionality of a Membrane Protein, Glycerol-3-Phosphate Dehydrogenase." *Biochemistry*, 2005 (in press).

Prof. Zhang looked into 3-D membrane lattices with membranes linked by bridging proteins into a stack. The proteins set the distance between the membranes, and can be made to bend the proteins. These structures could prove useful for membrane protein stabilization.

Alpha-helical Materials

Andreas Lomander and colleagues attempted to create an alpha helical molecular switch, in which gold nanoparticles are coupled to the ends to two alpha helices (a coiled coil) by Cys residues. Although this strategy did not work out directly, the work did lead to formation of fractal patters of gold on mica. With an enormous surface area, the branching gold structure resembles the leaves of a fern.

Using simulations, they discovered that the coiled coils coupled to each other by disulfide bridges, forming a cross-shaped structure with no more than six pairs. The structures of six coiled-coils likely formed clusters that deposited into the mica substrate at low pH (at least, this is the theory; this has not been proven). Experiments using hydrophobic denaturant (DTT and beta-mercaptoethanol) to separate the alpha-helices of the coiled coils showed that the structure disappeared, supporting the theory of self-assembly of the coiled coils. Copper chloride nucleated by the peptide also forms fractal structures.

Source: Lomander, et al. *Nano Lettrers,* July 2005.

DNA and "electronic ink"

Two or three years ago, Prof. Zhang's students coupled gold clusters onto DNA and regulated the DNA denaturation by application of radio-frequency waves (RFMF). The gold clusters allowed them to "tag" the molecules to follow their denaturation.

Source: *Nature*

Nanocrystal Necklace

Chains of nanocrystals (e.g., gold or silver nanoparticles, ~2nm in size) were coupled onto DNA, and later onto protein strands using a single amino acid: lysine. Gold-coupled lysine was synthesized, and microscopy confirmed a ratio of one lysine per gold in up to 15% of the residues (but, alas, the yield got no higher); the lysine could then be incorporated into proteins.

The discoveries of the next generation are up to you.

Guest lecture: Andreas Mershin, Ph.D.
"Applying Biomimicry to Nanotechnology."

Prof. Zhang's designer peptides have been useful for some recent project, including:

Nano-Bio-Photovoltaic cell, now in the Boston museum of science.
Dog on a chip: smelling odorants using a chip.

The strategies for these designs come from **biomimicry**: imitating living things. Evolution used billions of years to build spectacular organisms, but we can take a shortcut.

For example…

Birds use the lift of air above their curved wings to fly: this concept inspired the Wright Brothers to build a first plane, and later technology surpassed evolution to fly at twice the speed of sound.

Velcro is thought of as a first deliberately biomimetic design. In 1948, Swiss engineer Georges Mestral, a mountaineer, noticed that burrs stick to him and his dog with tiny hooks. He decided to go into industry for Velcro to computer with the huge YKK zipper industry. (Check your zipper – it probably says YKK on it, the initials of the 3 Japanese inventors of the zipper.) And sure enough, his invention was a hit.

Color television can be encoded by RGB: just three colors yield the whole spectrum. The cones in your eyes, too, only detect three possible wavelengths. BUT the coding scheme of vision had not yet been solved at that time: humans had re-invented the scheme of vision!

Earth's energy budget

Earth could only provide 200 more years of fossil fuels, but the sun produces 10,000 times more energy that we consume. To harvest this much energy, we'd need to cover the area covered by the US's highway system. So if we could make a solar cell as cheap as asphalt, we could survive on solar energy alone!

However, solar cells are only up to 40% efficient, and far too expensive. Could biology provide a cheaper alternative? Here are some examples of electricity in biology:

Eels can generate up to 600V and 1A (or 600 Watts) in a single jolt. That's not enough to kill you, but it can stun you and make you drown, and the many jolts that the eel can produce can power a light bulb or a laptop. The exact mechanism is unknown, but seems to use ATP and the movement of the eel's own muscles.

In 2001, the bacterium Geobacter (Desulfuromonas acerixidas) has been shown to generate up to 1 V at the mud-seawater interface, and has been used to power a light bulb! The exact mechanism is still unknown, but seems to be protein-based.

Plants, of course, act as photovoltaic cells, generating an exciton in the photosynthetic complex to eventually pump a proton across a membrane. After a complex reaction of photosynthesis, the plant generates ATP.

But can we harness just the initial exciton in the photosynthetic complex?

Susan Linquist (MIT Biology Professor) at Prof. Zhang's Crete Meeting, 2003: "About 10,000 years ago, man began to domesticate plants and animals. Now it's time to domesticate molecules." Indeed, man has force the evolution of animals and crops to best fit the application of food production. Can a similar selection be applied to molecules?

Can we "domesticate" the photosynthetic complex to yield electricity?

Prof. Zhang's self-assembled peptides might be able to eliminate the need for a membrane protein: the detergents stabilize the membrane proteins, "tricking" them into thinking they are in embedded into a membrane, and stabilizing them as a result.

Source: Das, et al. “Integration of Photosynthetic Protein Molecular Complexes in Solid-State Electronic Devices” *Nanoletters* **4** (6), 1079-1083, 2004.

Electricity out of spinach?

In the Boston Museum of Science, you’ll find a 1cm x 1cm chip containing spinach extract capped with a gold electrode. On the bottom a silver electrode gathers electrons generated by the organic semiconductor.

How hard is it to make electricity out of spinach? Is it time to start painting highways with spinach extract? Not quite. The molecules have to orient in a specific orientation. However, adding His tags with DSSP/NTA peptide sheaths can orient the molecules to the gold surface. The engineered surfactants created in Prof. Zhang’s were the key to lab stabilizing PS1 “dry” on an “unnatural” surface.

The current in the tiny cell follows the absorption spectrum of the protein, indicating that the protein is responsible for 0.12 mA/cm^2 at $10W/cm^2$, with $\lambda = 808$ nm. With a maximum power conversion of 20%, the lightweight photovoltaic cell beats Si on weight and, eventually, on cost.

Making it efficient: into the electric nanoforest

The thicker the photosynthetic layer, the better the light absorption, but the worse the charge extraction (due to electron/hole recombination). How can a thin, ordered layer contain more protein per area?

In nature, diatoms (photosynthetic organisms that live in the sea) trap light with a silica shell. A series of silica ridges give photons many chances to get absorbed.

Micromachining ridges into the glass containers for the photosynthetic protein (3-5 microns in height, with 40-micron “roofs” surrounded by 10-micron “streets”) led to a noticeable enhancement, but also increased cost excessively. Carbon nanotubes, with a huge aspect ratio and functionalizable ends, had desirable chemical, electrical, and mechanical properties, but absorbed light too strongly to be useful.

Is there anything they can use that’s both transparent and conducting, to mimic diatom ridges? Yes: zinc oxide nanowires. Essentially, they wanted to build an electric nanosforest: trunks of zinc oxide with leaves of photosynthetic proteins to absorb light. In fact, this even outdoes nature, because with *transparent* tree trunks, more light can be absorbed, with leaves even near the base receiving light below the “canopy.”

An additional bonus: why is zinc oxide used in sunscreen? It also absorbs well in the UV, generating current on its own to augment the current produced by the photosynthetic protein!

To grow the nanoforest, gold nanoparticles are sputtered onto an aluminum oxide substrate and used to grow zinc oxide nanowires coated with (detergent-ensheathed) PS1 monomers, which eject electrons in the direction of the nanowire.

Ways to increase power:

Choose more efficient photosynthetic complexes, such as those of seaweed.

Use a ZnO-binding peptide sequence that Angela Belcher uses to bind single crystals of ZnO, instead of the Au and DTSSP/NTA/His "glue".

Still, efficiency is the greatest obstacle for this technology.

The Nose Chip

We still don't know how smell works: how we identify different odors, etc. However, we have recently learned the mechanisms of the receptors. Molecules with identical bonds but different vibrational behavior can trigger different receptors (e.g. regular water vs. D_2O or "heavy water"), so detection of vibrations may be key to smell.

One team of scientists at Amersham Hospital, Buckinghamshire, trained a dog to smell bladder cancer, correctly identifying all 52 of the patients with bladder cancer. (In fact, the 52^{nd} patient was undiagnosed at the time, and ended up developing cancer after what they thought was a mistake of the dog!) Can a chip be built for this purpose? We'll see.