

ANALYTICAL CURRENTS

A colorful way to detect SNPs

Lewis Rothberg and Huixiang Li at the University of Rochester have discovered a new phenomenon that allows them to detect specific DNA sequences and single nucleotide polymorphisms (SNPs) within 10 min simply by looking at the color of a DNA-gold-nanoparticle colloid.

Rothberg and Li observed a previously unknown attraction between single-stranded DNA (ssDNA) and colloidal gold nanoparticles. Short DNA strands and high temperatures were found to speed up this adsorption process.

Gold nanoparticles aggregate when they are exposed to salt, which causes them to change color from pink to blue-gray. But when Rothberg and Li added enough ssDNA and then the salt, the nanoparticles did not aggregate and the color change did not occur.

The researchers used the color change as an indication that PCR-amplified test DNA contained specific sequences. A pink colloid indicated that hybridization between short ssDNA probes and test DNA did not occur and that aggregation was prevented. A blue-gray colloid indicated that the sequences were complementary; hybridization occurred, and the probes were no longer free to prevent aggregation.

To detect the single base-pair changes characteristic of SNPs, the researchers used four probes that would dehybridize from test DNA at the same temperature if the sequences were perfectly complementary. Two control probes bound to sequences outside the SNP region, but the other two probes overlapped the SNP region. Because of a mismatch, probes that



(a) A blue-gray color indicates that a control probe is bound to DNA. (b) A pink color indicates that a mismatched probe has melted off the SNP-containing DNA.

bound to a SNP region melted away from the test DNA at a lower temperature than did control probes. The color change was tested at various temperatures.

The assay was successfully used to identify SNPs associated with a hereditary cardiac arrhythmia, and it could be used to detect SNPs implicated in other diseases and conditions without expensive instruments or labor-intensive DNA modifications. (*J. Am. Chem. Soc.* **2004**, *126*, 10,958–10,961)

Probing the yeast cell wall

James Gimzewski and colleagues at the University of California, Los Angeles, have measured the motions of the cell wall in baker's yeast, *Saccharomyces cerevisiae*. The investigators showed, for the first time, that cooperative action between motor proteins inside the yeast cell propagates through the cell wall. The work demonstrates that local motion observed in the yeast cell wall can be related to processes occurring inside the cell.

Gimzewski and colleagues trapped single, live yeast cells in $\sim 5 \mu\text{m}$ pores of a polycarbonate membrane under physiological conditions. Using a cantilever in an atomic force microscope, they probed the cells and measured the spring constant of the yeast

cell wall as $\sim 0.06 \text{ N/m}$. The spring constant of the cantilever was 0.05 N/m , which allowed them to study the coupling of the cantilever and cell wall movements as two springs in a series.

The motion of the cell wall was temperature-dependent between 22 and 30 °C, which suggests that the movements were due to either active metabolic processes occurring inside the cell or Brownian motion. Gimzewski and colleagues treated the yeast cells with sodium azide to inhibit ATP production and confirmed that the cell wall motions were due to ATP-dependent, active metabolic processes.

The investigators calculated an activation energy of 58.15 kJ/mol, which is con-

sistent with the activation energy known for motor proteins such as myosin, kinesin, and dynein. Because a force of 10 nN was measured at the cell wall, which is too large for a single motor protein to exert, they suggested that the motor proteins acted together in a cooperative fashion. The cooperative action of the motor proteins inside the cell propagated through the cell wall.

The researchers hope to next probe the mechanical properties of mammalian cell membranes. Because the membranes have a very low spring constant ($\sim 0.002 \text{ N/m}$), special cantilevers will be needed to carry out the experiments. (*Science* **2004**, *305*, 1147–1150)