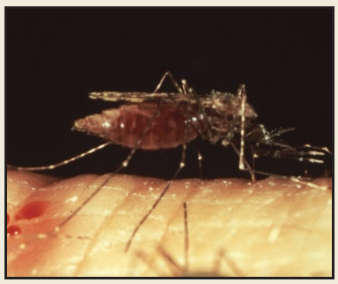
Anopheles mosquito genome sequenced



Last month, an international consortium of public and private scientists published the genome sequence of *Anopheles gambiae* (*Science* **298**, 129–149, 2002), the primary mosquito vector responsible for human infections with the malaria parasite *Plasmodium falciparum*. Using the PEST strain of *A. gambiae*, Robert Holt and his colleagues created BAC/plasmid libraries and sequenced the genome using the now well-established shotgun method. Using the assembly algorithms of Celera (Rockville, MD), the team constructed a sequence of 278 megabases representing 91% of the mosquito genome (at tenfold sequence coverage). A combination of Celera and Ensembl gene-annotation programs predicted 15,189 genes within the sequence. Analysis of expressed sequence tags from blood-fed and non-blood-fed mosquitoes identified transcripts that are upregulated or downregulated by a blood meal. The unprecedented genetic variation in the PEST genome—it contains two haplotypes of roughly equal abundance, with about 445,000 SNPs identified so far distributed along the chromosomes—posed a significant challenge for the sequencers. Holt and his collaborators believe the genome can provide a new source of targets for designing insecticides and repellants and interrupting the parasite life cycle. *AM*

NRPSs keep it simple

A US–German group has found that nonribosomal peptide synthetases (NRPSs) have a simpler-than-expected monomeric structure. The team, at Philipps University (Marburg, Germany) and Harvard Medical School (Boston, MA), constructed two variants of the NRPS gramicidin—one with a hexahistidine affinity tag, the other with a streptavidin tag. They incubated both variants with an ionic detergent to split up any dimers present. The detergent was then removed, allowing dimer molecules to re-form—some as "scrambled" dimers carrying both affinity tags. The resulting solution was then passed successively through two different affinity chromatography columns, one for streptavidin and the other for hexahistidine. The group observed that no protein molecules were captured on both columns, and concluded that no dimeric forms were present. Other NRPSs gave the same result (*Chem. Biol.* **9**, 997–1008, 2002). The discovery is unexpected because the action of related enzymes, such as fatty acid synthase and polyketide synthase (PKS), depends on their dimeric structure. It may help in understanding the synthesis of these products, some of which (like the mixed NRPS–PKS product epothilone) have potential as new drugs, according to lead author Chris Walsh. *PM*

Research News Briefs written by Aaron Bouchie, Alan Dove, Ingrid Holmes, Andrew Marshall, and Peter Mitchell.

GFP glows brighter

In order to mark and monitor proteins within cells, cell biologists currently use a system of green fluorescent protein (GFP) labeling plus photobleaching—strongly illuminating a specific cell region to extinguish the fluorescence of GFP there, which allows for the tracking of GFP-labeled proteins into or out of that area. George H. Patterson and Jennifer Lippincott-Schwartz of the National Institutes of Health (Bethesda, MD) have now developed a variant of GFP that exhibits noticeable fluorescence only when photoactivated with ~400 nm light, thus eliminating any signals from newly synthesized proteins, and may ultimately render photobleaching unnecessary (*Science* **297**, 1873–1877, 2002). The authors generated multiple GFP mutants and measured their absorbance spectra both before and after photoactivation. One variant, termed photoactivatable GFP (PA-GFP), was selected because its fluorescence was high-contrast (manifested as a ~100-fold increase in fluorescence at 488 nm after photoactivation, as opposed to a mere 3-fold increase by wild type) and maintained stability for at least one week under physiological conditions. Further experiments showed PA-GFP could label cell populations and track membrane protein exchange by lysosomes. The authors have begun to elucidate the mechanism underpinning the photoactivation process, and are continuing studies on lysosomal membrane protein dynamics. *IH*

Tissue-specific quantum dots

Quantum dots are tiny (smaller than 10 nm) inorganic particles with unique optical and electronic properties that are thought to have great potential as intravascular probes, both for diagnostic imaging and for drug delivery. Now, researchers from the Burnham Institute (La Jolla, CA) and the University of California, San Diego have combined quantum dots with peptides to improve their ability to target specific cell types and tissues, and enhance their capacity to evade the reticuloendothelial system—the body's "filter system" that removes particulates from the bloodstream (*Proc. Natl. Acad. Sci. USA* **99**, 12617–12621, 2002). By coating red luminescent ZnS-capped CdSe quantum dots with one of three "homing" peptides that target specific tissues, the researchers were able to direct the quantum dots to mouse lung tissue, tumor blood vessels, or tumor lymphatic vessels, respectively. Adding chains of the compound polyethylene glycol to the peptide-coated quantum dots also reduced the nonselective accumulation of the nanoprobes in reticuloendothelial tissues by 95%. The results of this research encourage the construction of more complex nanostructures that may eventually be able to detect specific diseased cells and deliver therapeutic molecules such as peptides, nucleic acids, lipids, or small molecules. *AB*

Gene therapy picks up the pace

Electronic heart pacemakers have come a long way in the few decades they have been available, but they still do not cure erroneous heart rhythms, only keep them in check as long as the device is working. Using a gene-therapy strategy, researchers at Johns Hopkins University School of Medicine (Baltimore, MD) now hope to turn ordinary cardiac muscle cells into specialized pacemaker cells, restoring the heart's natural pacing function. In an important proof-of-concept experiment, described in a recent issue of *Nature* (**419**, 132–133, 2002), the Hopkins team has successfully established new cellular pacemakers in the hearts of guinea pigs. The new strategy used a gene encoding a dominant-negative version of the protein Kir2.1, a potassium channel component expressed at high levels in ordinary cardiac muscle cells but not in the pacemaker cells of the sinoatrial node. Suppressing the channels caused ordinary cardiac cells to acquire pacemaker activity. In contrast to other types of gene therapy, the amount of transduction needed to create the pacemaker was very small. The researchers are now refining the approach in a pig model of heart disease. *AD*