

# Tick-Tock Goes a Bacterial Clock

RESEARCHERS NOW BETTER UNDERSTAND THE “GEARS”  
ON THIS UNUSUAL CIRCADIAN CLOCK.

Researchers in Erin K. O’Shea’s laboratory at Harvard University lifted the face off an ancient bacterial clock and revealed how a rugged and reliable timekeeping mechanism keeps the clock ticking out daily cycles for weeks in a test tube.

The team discovered the mechanism relied on the sequential addition and subtraction of phosphate groups to one of three proteins constituting the clock.

Their study builds on dogma-smashing work by Takao Kondo and colleagues at Nagoya University in Japan. Conventional wisdom held that all circadian clocks—cellular mechanisms that regulate physiological activity on a roughly 24-hour schedule—kept time via a protein regulating its own gene expression. Kondo’s group discovered that the circadian clock governing the ancient, photosynthesizing bacteria known as cyanobacteria involves no genes and just three proteins, and can function outside the cell fueled only by a phosphate source called ATP.

While the Japanese group knew that the clock ticked by adding and removing phosphate molecules from the KaiC protein with the aid of two protein “gears”—KaiA and KaiB—they didn’t know how the addition and subtraction kept time.

O’Shea’s group revealed a cycle where, with the help of KaiA, two sites on the KaiC protein gained phosphate groups in sequence:

first one site, then the other. Once both sites on KaiC have phosphate groups, KaiB alerts KaiA, triggering phosphate removal in the same sequence. Over the course of a day, KaiC slowly cycles from no phosphate groups to two phosphate groups, back to one phosphate group, finally ending up with no phosphate groups once again. Details of the work were published October 4, 2007, in *Science Express*.

All circadian clocks make adjustments in response to changes in light, O’Shea says. For cyanobacteria—the blue-green algae responsible for 70 percent of the Earth’s photosynthesis—the clock may allow them to anticipate daylight and rev up the production of proteins needed for photosynthesis. The question is how. “Other laboratories have identified some of the proteins involved,” says O’Shea. “Our goal is to understand how the inputs from the environment feed into this clock.” ■

—LISA SEACHRIST CHIU



Precise time-keeping proteins keep the biological clock ticking steadily.

## IN BRIEF

number of genes likely to contribute to the development and progression of breast and colorectal cancers. The study suggests that each breast and colon tumor is unique and may arise through mutations in many different combinations of genes. The good news is that this large number of genes is involved in a much smaller number of biochemical pathways.

Most of the approximately 280 candidate cancer genes the researchers identified are involved in as few as 15 pathways. Studies of these pathways are already producing new ways to diagnose and treat cancer.

“We need to start thinking about cancer in terms of these pathways instead of individual genes, because that’s a more accurate view of how cancers originate,” says study leader Bert Vogelstein, an HHMI investigator at the Kimmel Cancer Center at Johns Hopkins University School of Medicine.

HHMI investigator Sanford D. Markowitz at Case Western Reserve University School of Medicine was also one of 42 authors reporting the findings, published October 11, 2007, in *Science Express*.

### IMMUNOLOGIC DEATH BLOW TO CANCER CELLS

Taking a clue from a rare disorder in which the immune system destroys a

patient’s cancer even as it attacks the nervous system, causing neurological damage, researchers have devised a strategy to fight breast and ovarian cancer. They have engineered immune cells that, in the laboratory, attack and kill cells containing a protein found in up to 60 percent of ovarian tumors and 25 percent of breast tumors.

The research team, led by HHMI investigator Robert B. Darnell at Rockefeller University, published its findings November 5, 2007, in the *Proceedings of the National Academy of Sciences*.

Darnell and his collaborators concentrated on a tumor antigen called cdr2, which, when produced by breast and ovarian tumors, can trigger paraneoplastic cerebellar disorder (PCD), in which killer T cells attack the tumors, plus cells in the nervous system. The researchers screened a large library of slightly different fragments of the cdr2 protein looking for those most strongly recognized by T cells. They identified one particularly potent version called cdr2(290). When they looked at the blood of patients with PCD, the researchers found T cells that responded to cdr2(290), confirming they had identified a molecule with diagnostic and therapeutic potential.

### IMAGES GIVE CLEARER VIEW OF ION CHANNEL

HHMI researchers have unveiled the most detailed views yet of the structure of a voltage-dependent potassium ion channel. The new images, which show the channel in a more natural environment than previous studies, reveal that the channel’s function is likely to be profoundly influenced by lipid molecules within the cell membrane where the channel is embedded.

The research team, led by HHMI investigator Roderick MacKinnon, hopes that a technique they used—lipid-detergent-mediated crystallization—will make it possible to determine the structures of other membrane proteins.

MacKinnon and his colleagues at Rockefeller University published their findings November 15, 2007, in *Nature*.

Voltage-dependent potassium ion channels are central to the function of nerves and muscles. Without them, the brain would suffer neural gridlock and the heart would seize up. In earlier studies, MacKinnon and his colleagues deduced the structure of the voltage sensor in the channels. The researchers’ latest steps entailed engineering a new form of the channel that yielded improved protein