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RESEARCH NEWS

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Flies Have Morning and Evening Clocks

Two groups of researchers have independently discovered the long sought dual body clocks in the brain of fruit flies that separately govern bursts of morning and evening activity.

Both research groups published their findings in the October 14, 2004, issue of the journal *Nature* . Howard Hughes Medical Institute researcher Michael Rosbash at Brandeis University led one group; François Rouyer at the Centre National de la Recherche Scientifique in France led the second group. Graduate students Dan Stoleru and Ying Peng of Brandeis were co-lead authors of the Rosbash group's article.

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>
> **— Michael Rosbash**

In an accompanying *News and Views* article in the journal *Nature* , neurobiologist William J. Schwartz of the University of Massachusetts Medical School, writes, “A truly integrative circadian biology is close at hand, as researchers learn about an adaptable, layered system that has emergent properties at many levels of organization. *Drosophila* workers, who have been so effective at taking the clock apart, are now succeeding in putting it back together.”

Biological clocks in both flies and humans operate on a 24-hour, or circadian (Latin for "about a day"), cycle. In humans, the clock's influence is far-reaching, governing such functions as sleeping and waking, fluid balance, body temperature, cardiac output, and oxygen consumption. In the fruit fly *Drosophila* , however, the circadian clock has its most overt effect on the fly's level of activity. In both flies and humans, the clocks are circuits of

neurons that naturally oscillate with a circadian periodicity. Inside these cells, the molecular components of the clock are "rewound" daily by the effects of light and other stimuli.

According to Rosbash, the central clue to the existence of dual circadian clocks in the fly was the observation that flies have two activity peaks. “It was always intriguing that flies had two peaks of activity, in the morning and evening, with a siesta during the day and not very much activity at night,” he said. “There are several ways to explain that, but one possibility was that there were two clocks running—one governing the morning peak and one governing the evening peak.”

Rosbash and his colleagues theorized that if dual circadian clocks existed, they would likely occupy different anatomical positions in the fly brain. They knew that the flies' circadian-clock neurons included one distinctive cluster called the ventral lateral neurons and another called the dorsal lateral neurons. The ventral lateral neurons were known to express an important circadian signaling molecule called pigment dispersing factor (PDF). The dorsal lateral neurons did not express a known signaling molecule, but were part of a larger group of circadian neurons that express a gene for a circadian photoreceptor protein known as cryptochrome.

Starting with those parameters, Rosbash and his colleagues used genetic techniques to selectively deliver cell death genes to ablate specific groups of neurons. They then observed how these genetic deletions affected the flies' activity. The experiments revealed that the PDF-expressing ventral lateral neurons govern the morning activity peak; while another group of neurons, including the dorsal lateral neurons, governs the evening activity peak.

In additional experiments, the researchers selectively disrupted the internal clock machinery of the circadian neurons. The studies showed that one set of circadian neurons drives the other. “It's as if there is a wiring circuit from one set to the next and, under natural light conditions, one can regulate the physiology of another,” said Rosbash. The scientists theorize that this type of coupling between two circadian oscillators coordinates the two activity peaks and aids responses to environmental factors such as seasonal changes in lightness and darkness.

Rosbash said that there is evidence that mammals, including humans, also possess such dual circadian systems and that the systems communicate with one another, but researchers have not yet been able to distinguish the systems anatomically or biochemically.

Rosbash and his colleagues will continue to explore the details of the different clocks. Such studies, he said, may offer an important opportunity for understanding general principles of neural structure and function. “The circadian neurons are one of the few circuits in neurobiology where we have a chance to understand at multiple levels how different sets of neurons communicate with each other—including understanding the wiring rules, the biochemical rules and the functional behavioral rules,” he said.