Scientists find protein that helps synchronize fly's internal clocks

February 3 2006

Scientists have identified a receptor protein that helps the fruit fly know when to start and shut down its day, a step that should help them learn more about internal clocks in higher organisms such as humans.

Neuroscientists from Washington University School of Medicine in St. Louis identified a receptor for pigment-dispersing factor (PDF) protein, which scientists previously had recognized as a molecule that helps keep different internal "clocks" synchronized.

"Daily rhythms regulated by biological clocks shape our lives in important ways, affecting a wide range of functions including sleep, body temperature, cognitive ability, mood and sensitivity to drugs," says lead author Paul Taghert, Ph.D., professor of neurobiology. "Because these timekeeping processes have been highly conserved through evolution, what we learn from flies and other organisms often helps us better understand the same systems in higher organisms."

For example, studies of fruit flies already have helped scientists identify a human gene for advanced phase sleep syndrome, a human disorder that puts sufferers to sleep at what is normally suppertime and promotes their waking at 3 or 4 a.m.

Taghert's group was one of three to independently report identification of the PDF receptor in a recent issue of *Neuron*.

Clock cells contain a handful of proteins that interact with each other in ways that increase and decrease their own levels in the cell at various times during the course of a day. The cycle naturally repeats itself every 24 hours. Through their connections with other nerve cells and other types of tissues, clock cells regularly trigger or suppress certain physiological processes during the course of the day. Biologists call these daily patterns circadian rhythms.

Taghert's lab identifies the clock cells in fruit fly brains and traces their connections to other cells and tissues in hopes of better understanding how they affect characteristics such as the morning and evening activity peaks normally seen in fruit flies.

"We look at where the branches of these cells go, what signals they release and when they release them, and who is listening," Taghert explains. "We want to follow the chains of cells that respond to signals from the clock cells. We're hoping that path doesn't get too complicated too fast."

Working with the short-lived fruit fly, a classic model for circadian biology, allows manipulation of genes with potential circadian links and rapid assessment of the resulting effects on new generations of flies. Such manipulations helped scientists identify Period, the first gene associated with circadian rhythms. Humans have three genes analogous to Period, one of which is mutated in a critical region in patients with advanced phase sleep syndrome.

Beat the clock

PDF is a neuropeptide that originally was identified in crabs and shrimp, where it disperses pigment in light-sensing organs at the beginning of the day, adjusting the organs for the increased light levels that begin at sunrise.

In the fruit fly brain, PDF is made by 16 of the 150 brain neurons that Taghert and others have so far identified as clock cells. Taghert's group showed in an earlier study that loss of PDF altered the rhythmic behaviors of flies, changing their behavior schedule to one more appropriate for about a 22-hour day. In follow-up studies, Taghert and other scientists linked PDF to the synchronization of various kinds of clock cells.

For this study, researchers in Taghert's lab used the fruit fly genome as a guide to allow them to identify all the fruit fly peptide receptor genes, express them in cell cultures, expose them to PDF and search for receptors that are specifically activated by PDF. When they found one that interacted with PDF, they produced a line of fruit flies with a mutation in the gene for that receptor protein. The new line of flies acted like the flies in which PDF had been knocked out, demonstrating that the receptor is essential to normal PDF function.

Close relative in humans

Mammals do not have a gene directly equivalent to PDF, but the Taghert group's new findings indicate that the PDF receptor is closely related to mammalian receptors for the proteins calcitonin and CGRP (calcitonin gene-related product), a well-known molecule whose precise function has been difficult to determine, which may play a similar role in mammalian circadian systems.

"We found the fruit fly PDF receptor responded both to calcitonin, which we hadn't previously linked to circadian function, and to PACAP, a mammalian neuropeptide already recognized as a part of the circadian system," Taghert says. "This suggests that the receptor systems probably evolved from a common ancestor and that what we learn from the fruit fly may be helpful in understanding circadian biology in higher organisms."

Currently, Taghert's group is working to identify the locations and characteristics of fruit fly brain cells that make the PDF receptor and to trace the signals emitted by those cells back into the circadian system.

Source: Washington University School of Medicine