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Fruit Fly Studies May Boost Understanding of Cocaine Addiction

Researchers have identified a protein in the nervous system of fruit flies that is a cousin of the molecule that cocaine targets in the human brain. Their discovery offers the possibility that researchers can genetically manipulate the protein in fruit flies to gain better understanding of how cocaine alters behavior and produces addiction.

The researchers also found that the protein they discovered, a *Drosophila* dopamine transporter (dDAT), may be a molecular ancestor of both the human dopamine transporter—which is a target of cocaine—and the antidepressant-sensitive norepinephrine transporter. Thus, say the scientists, studies of the dopamine transporter in *Drosophila* may offer insights into how drugs affect neurotransmitter transporter molecules in other organisms. The discovery of dDAT by Howard Hughes Medical Institute (HHMI) investigator Susan G. Amara and her colleagues was published in the January 2001 issue of *Molecular Pharmacology*.

Neurotransmitter transporter proteins are molecular scavengers found in the nervous systems of many organisms. After neurons release chemical signals in the form of neurotransmitters, the neurotransmitter transporter proteins attach to the neurotransmitters and recycle them back into storage compartments in neurons for reuse. Cocaine inhibits the uptake of the neurotransmitter dopamine, causing an abnormal buildup of dopamine in the region between neurons. This buildup of dopamine produces the euphoria and other affects associated with cocaine use.

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- Susan Amara

According to Amara, who is at the Oregon Health Sciences University, the search for dDAT was launched as a result of studies of the behavioral affects of cocaine on flies by one of the paper's coauthors, Jay Hirsh of the University of Virginia.

"Hirsh's work showed that flies respond to cocaine with an increase in locomotor activity that resembles the responses to cocaine seen in rodents and humans," said Amara. "His studies also suggested that there would be a transporter in the *Drosophila* nervous system that would resemble the dopamine and norepinephrine transporters in humans.

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The paper's lead author, Peter Pörzgen, used the known gene sequence of the human dopamine transporter to search for related sequences in the *Drosophila* genome database. Once Pörzgen and his colleagues identified a key genome segment, called an expressed sequence tag (EST), they used the EST to isolate and clone the full-length gene from a library of genes derived from an extract of *Drosophila* genetic material.

Hirsh and his colleagues used a probe derived from the dDAT DNA sequence to show that in fly larva the dDAT transporter gene is expressed in dopamine-containing neurons. Additional studies revealed that when dDAT was introduced into cells in culture, the dDAT transporter behaved like a hybrid of the mammalian dopamine and norepinephrine transporters.

"Based on our analysis of this transporter, we hypothesized that it could be a primordial carrier that eventually evolved into two separate carriers in

mammals—one for norepinephrine and one for dopamine," said Amara.

"Finding dDAT provides further support for the idea that flies are a potentially interesting and unique model for studying the behavioral affects of cocaine and the pathways of addiction," she said. "For example, it might be possible to generate behavioral mutants with different sensitivities to the drug, as well as knocking out the transporter to test the impact on behavior."

The discovery of dDAT could offer important new research pathways, according to dopamine transporter expert Marc Caron, an HHMI investigator at Duke University. "The proven power of *Drosophila* genetics should help us examine questions that have remained enigmatic in mammalian systems," said Caron. "We still don't know, for example, the regulatory elements that cause a given gene to be expressed solely in dopaminergic cells in the brain. In the fly, these elements should be able to be mapped easily.

"The thought of being able to study the same paradigm—such as the rewarding effects of cocaine—in flies and mammals and to compare and contrast the results is very appealing."

Also, said Pörzgen, dDAT seems to be a hybrid of two kinds of transporters, thus making it a potentially useful model for understanding basic mechanisms of drug interaction with the transporter molecules. "Presumably, the portions of the transporter that are similar to the human dopamine transporter are likely to be involved in cocaine's actions, and those that are similar to the norepinephrine transporter could be involved in binding antidepressants," he said.