

# Protein Disposal: Gumming Up the Works

*Researchers discover a class of molecules that can prevent proteins from being degraded.*

**FOR A CELL, DESTROYING PROTEINS IS AS ESSENTIAL AS** building them, says Rati Verma, a researcher in the Caltech laboratory of HHMI investigator Raymond J. Deshaies.

The job of mincing proteins is performed by enzymatic machines called proteasomes. “Proteasomes affect almost all biological processes in the cell,” Verma says. By their deliberate destruction of regulatory proteins, they orchestrate activities from cell division to cell death.

Ubiquitin is the protein that hands down the death sentences. “Ubiquitin is the most highly conserved protein in eukaryotes [organisms whose cells contain a distinct nucleus],” Verma says, “with only three amino acid differences between yeast and mammals.” A chain of ubiquitins gets attached to doomed proteins, marking them for destruction and ushering them to the gates of proteasomes.

Last year, the Caltech researchers and colleagues discovered a class of small molecules that can block proteasomes from degrading proteins. The finding could open new avenues for treating diseases.

Few researchers considered proteasomes likely candidates as drug targets. Because proteasomes control so many processes, most researchers thought that perturbing them with drugs could only wreck havoc with the body, Deshaies says. “Even I initially felt that by inhibiting the proteasome you would just kill everything.”

Then, 2 years ago a proteasome inhibitor called bortezomib received quick approval for treating multiple myeloma. “Everyone in the proteasome field started paying attention once the drug was fast-tracked by the FDA,” Verma says.

Randall W. King, Deshaies’ colleague at Harvard University, initiated the hunt for inhibitors by screening close to 110,000 different small molecules for their ability to prevent proteasomes from destroying proteins. Three of the compounds showed promise as proteasome inhibitors.

“What was different about what Randy did,” says Deshaies, “is that the target was not defined at the outset of the screen. The ‘target’ was an entire system—a large group of proteins.” That left open the possibility that compounds King identified blocked protein degradation at any of a number of steps. It was up to Verma and Deshaies to determine which one.

They analyzed King’s compounds further using purified proteasomes and ubiquitin-tagged proteins. Verma and Deshaies systematically narrowed down the possible steps of the protein death march that the inhibitors were blocking. Their first experiments eliminated the two most obvious enzymatic possibilities: removal of the ubiquitin chain and degradation in the core of the proteasome.

The final answer was a surprise. Verma discovered that the inhibitors coated the ubiquitin chain, rendering it unrecognizable to the proteasome’s ubiquitin receptors, thereby halting protein destruction before it could even begin.

Small molecule inhibitors rarely work that way, she notes. Most, like bortezomib, clog the tight spaces at the reactive cores of enzymes. But a small molecule disrupting the broad surface contacts between proteins is like the fairy-tale pea poking the princess through a stack of mattresses.

Deshaies cautions that the compounds (which they named ubistatins) are still unsuitable as drugs. In fact, some of their chemical properties suggest that they may never enter the pharmaceutical pipeline. Instead, he says, “our research offers a proof in principle that the ubiquitin chain-receptor interaction is an Achilles’ heel of the ubiquitin-proteasome system that is potentially inhibitable by small molecules.” With that established, he says, pharmaceutical researchers might be able to accurately measure the binding of purified ubiquitin chains and ubiquitin chain receptor and then “screen a library of half a million compounds to find one that inhibits that binding.”

Such a compound could treat diseases besides cancer, he says. Proteasomes play a key role in regulating inflammation, and proteasome inhibitors might have potential in treating inflammatory conditions, such as rheumatoid arthritis, or as anti-tissue rejection drugs for transplant patients. ■

—Paul Muhlrad-

BELOW \_ RATI VERMA (LEFT) AND RAYMOND DESHAIES FOUND A NEW WAY TO THINK ABOUT HOW A CELL DESTROYS PROTEINS.



# DEGRADING PROTEINS

In this artist's conception, by medical illustrator Graham Johnson, a proteasome (lower left) degrades a protein.

**1.** A chain of ubiquitin molecules (green) ushers the protein (yellow) to the entryway of the proteasome (lavender).

**2.** The proteasome then unfolds the protein and feeds the amino acid chain into its digestive core (blue) where it degrades the protein into amino acids.

**3.** Ubistatin molecules (red) can bind ubiquitin chains (at right), blocking them and their attached proteins from entering the proteasome.

