

W0167

**Substrate Recognition and Catalysis in Thimet Oligopeptidase.** Nicholas Noinaj, David Rodgers, Univ. of Kentucky Dept. of Molecular and Cellular Biochemistry and Center for Structural Biology, Lexington, KY.

GPCR signaling is crucial for the regulation of neuronal communication. Here, neuropeptides serve as signaling molecules which transduce a signal from one neuron to the next and are then terminated through several mechanisms, one of which involves neuropeptide degradation via a class of enzymes known as neuropeptidases. Thimet oligopeptidase (TOP) is a neuropeptidase that hydrolyzes many diverse substrates with cleavage sites that have no apparent consensus. The TOP crystal structure has previously been reported and despite significant efforts, specificity remains a mystery. Recently though, we have determined crystal structures of TOP bound with fragments from seven different peptides. Our results indicate that TOP is able to recognize diverse substrates via a novel multipurpose binding surface that is relatively flat and composed mainly of aromatic and hydrophobic residues that interact primarily with C-terminal backbone atoms. This surface is located on a wall opposite the active site, providing further indication that a hinge-like conformational change is required for catalysis. Our work has also shown that a previously reported potent inhibitor of TOP, dynorphin A<sub>1-13</sub>, is in fact hydrolyzed and that a product fragment acts as the true inhibitor. *NIH/NIDA/NINDS/ACS-PRF.*

