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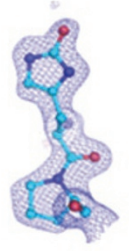
Proteasome target practice

For proteolysis of cellular proteins via the ubiquitin proteasome system (UPS) to occur, two structural features must be present in the substrate: a covalent polyubiquitin modification and an unstructured region. By fusing ubiquitin moieties and unstructured regions in different configurations to the RNase barnase and its protein inhibitor barstar, Prakash *et al.* show that these two structural features can act in *trans* within multisubunit protein complexes to induce targeted proteolysis by the proteasome *in vitro*. When the two features are separated onto different subunits, UPS subunit specificity is determined by the location of the unstructured region, not the ubiquitin modification. Although the biological relevance of in *trans* targeting is yet to be established, this study provides new insights into UPS subunit specificity and suggests a potential mechanism by which adaptor proteins could target other proteins for proteolysis. [**Articles, p. 29; News & Views, p. 3**] *KS*

Observing histidine oxidation



Cellular hydrogen peroxide sensing typically involves oxidation of a sensor cysteine residue. Unusually, the peroxide sensor for *Bacillus subtilis*, PerR, senses peroxide by an Fe^{2+}- or Mn^{2+}-dependent oxidation of a histidine residue. Detailed investigations of PerR have been complicated because two distinct histidines can be oxidized, and purified protein is often a mix of unoxidized, singly oxidized and doubly oxidized forms. Following optimization of conditions for obtaining singly oxidized protein, Traoré *et al.* report the first crystal structure of PerR with a 2-oxo-histidine modification, which, along with mass spectrometry, demonstrates that His37 is the primary site of oxidation. Spectroscopic and biochemical studies revealed that oxidation of His37 in PerR results in reduced affinity for Mn^{2+} and no detectable DNA binding activity. This study provides a foundation for investigating the formation and function of this post-translational modification. [**Articles, p. 53**] *JK*

Fluorination two-step

Many drugs have fluorine substituents that have been shown to confer useful pharmacological properties such as enhanced potency, improved metabolic stability and increased membrane transport. Though synthetic approaches exist for introducing fluorine into organic compounds, fluorination of unactivated sites remains a challenge. Rentmeister, Arnold and Fasan now report a versatile two-step chemo-enzymatic method for organofluorine synthesis. The authors screened a library of mutated bacterial P450 enzymes for their ability to hydroxylate selected carbon scaffolds. The most active variants were applied to the preparative scale synthesis of alcohols that could readily be converted to the target alkyl fluorides by treatment with diethylaminosulfur trifluoride. The combination of directed evolution strategies with synthetic transformations offers promise for regio- and stereochemical introduction of fluorine substituents into a diverse array of organic scaffolds. [**Brief Communications, p. 26; News & Views, p. 6**] *TLS*

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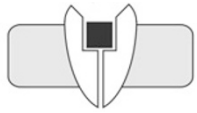
Profiling peptides

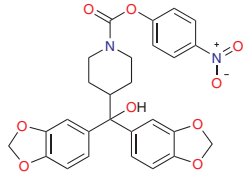
Dipeptidyl peptidase 4 (DPP4) cleaves peptides containing an N-terminal penultimate proline residue. Some plasma substrates of the enzyme are known, but substrates of the membrane-bound form have not been identified. Tagore *et al.* used LC-MS to profile global peptide levels in wild-type and DPP4-null mice kidneys and were able to find ten cleavage products whose signal intensities differed between the two genotypes. By looking in the wild-type tissues for cleaved versions of longer peptides found in the DPP4-null tissues, the authors were able to identify two new physiological substrate-product pairs for DPP4. They used a similar LC-MS–based approach to demonstrate that aminopeptidase action generates DPP4 substrates in the kidney, thus revealing a previously unknown peptide salvage pathway of the kidney epithelium. [Brief Communications, p. 23; News & Views, p. 5] *KS*

Revising nutrient sensing



Transceptors are integral membrane proteins that sense nutrients and either initiate signaling cascades or transport these nutrients into the cell, or both. Though several transceptors have been identified, their behavior is not well understood on a mechanistic level. Van Zeebroeck *et al.* assayed a panel of 319 amino acid analogs against Gap1, a general amino acid permease, and identified specific compounds that inhibit transport via competitive and noncompetitive pathways. A different but overlapping group of the original compounds agonized intracellular signaling by Gap1. Though these results demonstrate that there are many functional categories for these amino acids, labeling experiments surprisingly suggested that all of the compounds bind to the same site on the protein. These combined results demonstrate that binding, signaling and transport with Gap1 are not necessarily coupled, thus leading the authors to propose a new model for transceptor function. [**Articles, p. 45**] *CG*

Enhancing endocannabinoids



2-Arachidonoylglycerol (2-AG) and anandamide are endogenous ligands of the Δ^9-tetrahydrocannabinol (THC)-activated cannabinoid receptors CB1 and CB2, but their respective roles in nervous system function remain unclear. Enzymatic hydrolysis regulates this endocannabinoid signaling, and inhibitors have been identified for the hydrolase FAAH, the primary regulator of anandamide. However, so far there are no potent, selective inhibitors of MAGL, the primary regulator of 2-AG. Long *et al.* used activity-based protein profiling to identify an inhibitor of brain serine hydrolases and then chemically modified the compound to obtain the potent, selective MAGL inhibitor JZL184. They further showed that inhibition of MAGL by JZL184 raises 2-AG levels and induces a broad array of cannabinoid behavioral effects in mice. With inhibitors for both FAAH and MAGL in hand, it should now be possible to tease apart the roles of 2-AG and anandamide. [Articles, p. 37; News & Views, p. 8] *KS*