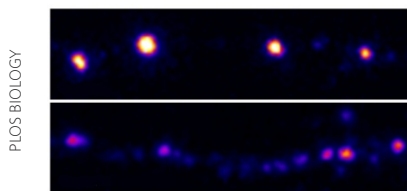


NEURONAL RECEPTORS

A lipid closes a loop

PLoS Biol. **12**, e1001908 (2014)



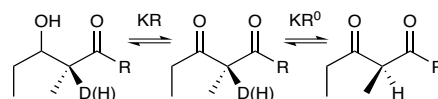
GABAergic synapses, characterized by the presence of GABA_A receptors (GABA_ARs), are involved in inhibition of neuronal signals. Gephyrin is a scaffold protein that regulates the clustering of, and therefore the number of, GABA_ARs at synapses, which is important for functional plasticity. A recent proteomic study showed that gephyrin may be palmitoylated at inhibitory synapses. To learn more about how gephyrin is regulated, Dejanovic *et al.* used fractionation experiments, metabolic labeling in conjunction with biochemistry as well as small-molecule inhibition of palmitoyl transferases (so-called DHHC enzymes) to confirm that gephyrin is palmitoylated *in vivo*. Mutation of various gephyrin residues combined with MS identified the palmitoylated residues as Cys212 and Cys284. Mutation of both amino acids abolished gephyrin palmitoylation and the postsynaptic clustering mediated by gephyrin. Complementary screens in HEK cells and in primary neurons followed by the use of dominant-negative DHHC-12, its overexpression and its knockdown identified this palmitoyltransferase as the main gephyrin-palmitoylating enzyme. DHHC-12 expression

in primary hippocampal neurons increased the size of postsynaptic gephyrin clusters and increased the amplitude of miniature postsynaptic currents, indicative of an increase in the GABA_AR pool, whereas a GABA_AR antagonist inhibited gephyrin palmitoylation. These results suggest that GABA_AR-mediated increases in gephyrin palmitoylation promote postsynaptic clustering of the receptor itself. *MB*

BIOSYNTHESIS

Redox not required

J. Am. Chem. Soc. **136**, 10190–10193 (2014)



Ketoreductase domains are responsible for reducing carbonyl groups during polyketide biosynthesis, thus determining the stereochemistry of these centers; some ketoreductases also function as epimerases to invert adjacent methyl-bearing stereocenters. Ketoreductases require NADPH, yet several multimodular polyketide synthases also contain enigmatic ‘redox-inactive’ homologs of ketoreductases that have lost the capacity to bind NADPH. Garg *et al.* suspected that these proteins might still perform epimerizations. To test this idea, the authors adapted a previously reported equilibrium isotope exchange assay used to determine whether individual ketoreductases are also epimerases to work as a tandem assay in which an NADPH-dependent, nonepimerizing ketoreductase (‘KR’ in image) is used to oxidize a representative substrate,

and then epimerization of the transiently generated ketoester by the redox-inactive KR⁰ domain was detected by monitoring washout of deuterium from the reduced substrate. The authors observed that both redox-inactive KR⁰ domains tested were able to deplete deuterium from the substrate, and tests with a different NADPH-utilizing KR confirmed the generality of the assay. Finally, to explore the epimerization mechanism used by redox-active and redox-inactive ketoreductases, the authors introduced a series of mutations into an epimerase-active KR domain that disrupted NADPH binding, thereby abolishing the native redox activity. These mutants were still able to epimerize the deuterated substrate. These results define a function for this little-studied group of enzymes and rule out an existing NADPH-dependent mechanistic proposal for epimerization. *CG*

PROTEIN DEGRADATION

Savior or executioner?

Nature doi:10.1038/nature13527

Thalidomide and its derivatives lenalidomide and pomalidomide interact with cereblon (CRBN), a substrate recognition component of the Cul4 ubiquitin E3 ligase complex, to promote the ubiquitin-dependent degradation of two members of the Ikaros family of transcription factors, IKZF1 and IKZF3. However, the location of the thalidomide-interacting site on CRBN was not known. Fischer *et al.* obtained crystal structures of a Cul4 complex component made up of the human DDB1–chicken CRBN co-complex with thalidomide and its derivatives. The S enantiomer of all three compounds bound a conserved pocket on the C-terminal domain of CRBN and required their glutarimide moiety for proper engagement. Thalidomide occupation on CRBN was known to be required for IKZF1 and IKZF3 degradation, but its effect on endogenous substrates was unknown. To identify these, the authors performed a biochemical screen and identified the homeobox transcription factor MEIS2, which is involved in brain and eye development, as a target for ubiquitination. In this case, ubiquitin-mediated degradation of MEIS2 was blocked upon treatment with lenalidomide. Multiple lines of evidence on the biochemical and cellular levels support the finding that lenalidomide and MEIS2 compete for the same binding site. The binding of thalidomide-like derivatives to CRBN prevents access to endogenous CRBN substrates such as MEIS2, resulting in their stabilization while promoting the degradation of IKZF1 and IKZF2. Overall, thalidomide-like derivatives can modulate both the stability and degradation of particular proteins. *GM*

HISTONE ACETYLATION

On-demand production

Cell **158**, 84–97 (2014)

Histone acetylation, a post-translational modification and epigenetic regulator of gene expression in eukaryotes, is maintained by acetyltransferases that use acetyl-coenzyme A (Ac-CoA) as a substrate. The primary source of Ac-CoA in cells is the pyruvate dehydrogenase complex (PDC), a multiprotein enzyme that is typically localized in mitochondria. Because Ac-CoA contains a reactive thioester, it has remained unclear how the nucleus is able to obtain enough Ac-CoA to support histone modification. Sutendra *et al.* now show that PDC undergoes transport to the nucleus in a cell cycle-dependent manner where it generates Ac-CoA and facilitates histone acetylation. Using antibodies against all components of PDC, the authors visualized the complex in the nuclei of human sperm, normal fibroblasts and cancer cell lines. Immunoblotting identified assembled PDCs in highly purified nuclei preparations, and isotopic labeling and MS analysis revealed that the nuclear PDCs generate Ac-CoA in the presence of pyruvate. Nuclear synthesis of Ac-CoA is correlated with elevated histone acetylation, as validated by siRNA knockdown of a PDC component and selective inhibition of mitochondrial versus nuclear PDC. Cellular analysis showed that PDC from mitochondria is translocated to the nucleus at the S phase of the cell cycle in a process that is facilitated by the Hsp70 chaperone. The nuclear translocation of PDC is induced by signals including epidermal growth factor or mitochondrial stress, leading the authors to suggest that PDC-mediated nuclear production of Ac-CoA and histone acetylation is a key regulator of S phase cell-cycle entry. *TLS*