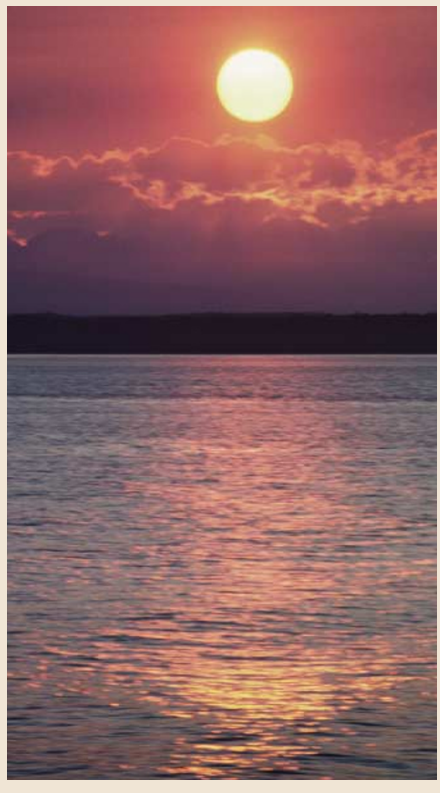
when expressed in dark-incubated seedlings, and this transcriptional activity was suppressed in the presence of light treatment. PIF1 also interacted with the active (Pfr) form of phyA and phyB, the two main phytochromes that regulate the ability of seedlings to undergo 'greening' in response to light, and the light-induced suppression of PIF1 activity required these phy proteins. As PIF1 can't interact with DNA and phyA or phyB concurrently, it seems that the phytoreceptors, when active, might function to modulate the activity of PIF1 — perhaps by sequestering or degrading it — so that chlorophyll biosynthesis can occur in the presence of light. So PIF1 seems to function as "a critical modulator by which plants optimize chlorophyll biosynthesis in response to environmental light conditions and protect against accumulation of potentially toxic levels of intermediates."

Katrin Bussell

References and links

ORIGINAL RESEARCH PAPER Huq, E. *et al.* PHYTOCHROME-INTERACTING FACTOR 1 is a critical bHLH regulator of chlorophyll biosynthesis. *Science* **305**, 1937–1941 (2004)

FURTHER READING Quail, P. H. Phytochrome photosensory signalling networks. *Nature Rev. Mol. Cell Biol.* **3**, 85–93 (2002)



GENE REGULATION

Reverse control

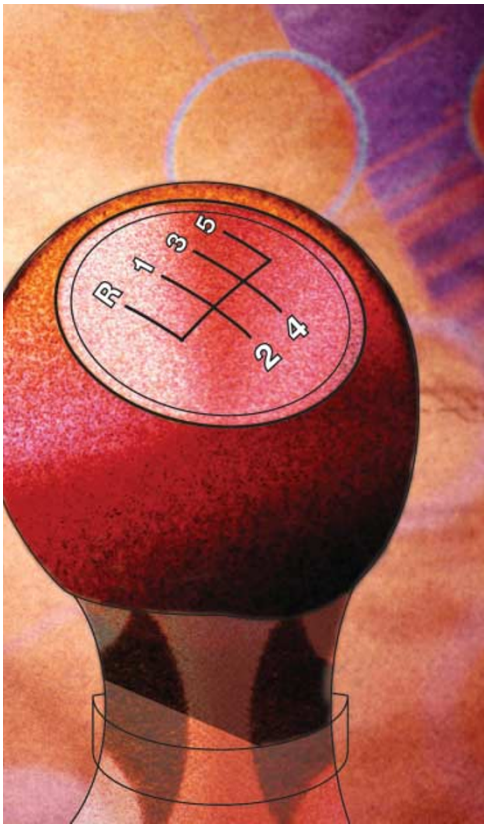
Post-translational modifications of histones, such as acetylation, phosphorylation and methylation, are thought to be dynamic and reversible. However, whereas the enzymes that help remove acetyl and phosphate groups from histone tails are known, those that counteract methylation have been elusive. But, two separate efforts have now led to the discovery that an enzyme, previously known to deiminate histone Arg residues, also regulates histone Arg methylation.

The enzyme, peptidylarginine deiminase-4 (PADI4/PAD4), was known to catalyse the removal of an imino group from peptidyl Arg to produce peptidyl citrulline. In *Science*, Allis, Coonrod and co-workers now report that, when the substrate is methylated, PADI4/PAD4 demethyliminates the methylated Arg by releasing a methylamine group and generating citrulline.

Both groups examined the substrate specificity of PADI4/PAD4. In *Cell*, Kouzarides and colleagues report that, *in vitro*, PADI4/PAD4 deiminates Arg residues, specifically Arg2, 8, 17 and 26, in histone H3. Incidentally, Arg2, 17 and 26 are known substrates for the Arg methyltransferase CARM1. The Allis and Coonrod group showed, both *in vitro* and *in vivo*, that PADI4/PAD4 targets include histone-H3 Arg8 and 17 and histone-H4 Arg3 (the latter is a known substrate for a different Arg methyltransferase known as PRMT1).

Using antibodies against specific methylated Arg residues, the Allis and Coonrod team showed that the PADI4/PAD4 activity does indeed 'undo' the methylation of Arg substrates. By tracing the radioactivity of the methyl group, they concluded that the methylimide group might be directly removed by demethylimination. Indeed, a dramatic decrease in the Arg methylation of histones was observed in response to PADI4/PAD4 activation in human HL-60 granulocytes. When treating granulocytes with *PADI4/PAD4* small interfering (si)RNA, the level of histone-H4 Arg3 methylation remained the same and little citrulline was detected. This indicated to the Allis and Coonrod group that PADI4/PAD4 is the main enzyme responsible for regulating the levels of histone Arg methylation.

When they treated synthesized peptides with PADI4/PAD4, Kouzarides and co-workers noticed that dimethylated Arg peptides could not be converted to citrulline. Unfortunately, monomethyl Arg peptides were not available, but the decrease in methylation of histone H3, as measured by an antibody that is specific for monomethyl Arg, implies that only monomethylated Arg residues could be targeted by PADI4/PAD4.



Arg methylation has been linked to transcriptional activation in response to hormone induction. Using an oestrogen-responsive promoter fused to a reporter gene, the Allis and Coonrod group showed that the presence of PADI4/PAD4 inhibited the hormone-stimulated reporter-gene activity, whereas a PADI4/PAD4 mutant protein failed to do so. As both groups confirmed, this transcriptional response coincides with the recruitment of PADI4/PAD4 to the promoter region, an increase in deiminated histone levels that is coupled to a decrease in methylated histone levels, and with the release of RNA polymerase II from the promoter.

To probe the mechanism by which PADI4/PAD4 antagonizes Arg methylation, the Kouzarides group tested a tail peptide from histone H3 in which Arg2, Arg8 and Arg17 were replaced by citrulline, and they found that the peptide remained unmethylated in the presence of CARM1. Together with the other findings, this indicates two possible mechanisms for PADI4/PAD4 action: first, it might deplete the histone-H3 substrate of CARM1, as implied by the Kouzarides group; or second, PADI4/PAD4 might reverse the methylation of monomethylated Arg. In addition, this raises the obvious question of how dimethylation of Arg residues is reversed…

Arianne Heinrichs

References and links

ORIGINAL RESEARCH PAPERS Wang, Y. *et al*. Human PAD4 regulates histone arginine methylation levels via demethylimination. *Science* 2 Sept 2004 (doi:10.1126/science.1101400) | Cuthbert, G. L. *et al*. Histone deimination antagonizes arginine methylation. *Cell* **118**, 545–553 (2004)