METABOLISM

A GlcNAc switch

Science **337**, 975–980 (2012)

O-GlcNAcylation, or the modification of proteins with β-*N*-acetylglucosamine, has been shown to link nutrient sensing with cell signaling. Yi *et al.* now investigate a further role for the modification in regulating cellular metabolism. The authors first observed that global increases in O-GlcNAcylation lowered rates of glucose metabolism owing to a substantial decrease in phosphofructokinase 1 (PFK1) activity. Labeling of PFK1 using a biotin or PEG tag established that PFK1 itself is O-GlcNAcylated and that the population of modified protein increases during the hypoxic conditions found in many tumors. MS further identified Ser529 as the site of modification, a residue already known to be important for PFK1 regulation by the activator fructose-2,6-bisphosphate. Analysis of PFK1 in multiple cell lines and human tissue also showed that O-GlcNAcylation was increased in tumor cells compared to nontumorigenic cells. As decreasing glycolytic flux can increase flux through the oxidative pentose phosphate pathway (the metabolites of which are necessary for DNA synthesis and protection from oxidative stress), the authors suspected this modification would be not only more prevalent in cancer cells but also necessary for their growth, as supported by impaired formation of tumors in a mouse model system harboring a S529A mutation as compared to the parent sequence. This work highlights a new regulatory mechanism in glycolysis with important implications for cancer treatment. *CG*

EPIGENETICS

Dealing with demethylases

Nature **488**, 404–408 (2012)

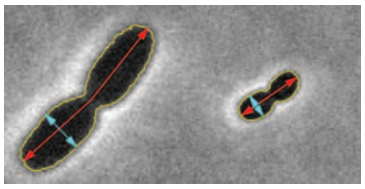
Jumonji (JMJ) family demethylases oxidatively remove methyl groups from lysine residues in histones and thereby regulate numerous gene expression pathways. To date, insights into the functions of JMJ demethylases have been hampered by the lack of potent and selective small-molecule inhibitors. A structure-based design approach now fills this gap by identifying a chemical probe that targets the KDM6 subfamily of JMJ enzymes (JMJD3 and UTX). Kruidenier *et al.* solved crystal structures of mouse and human forms of JMJD3 in complex with a peptide substrate. Building on their structural insights and subsequent mutational studies exploring substrate binding, the authors identified an inhibitor of JMJD3 and UTX activity (GSK-J1) and an inactive structural isomer (GSK-J2). Analysis of the JMJD3—GSK-J1 complex structure revealed that the compound's 2,2′-bipyridyl substructure ligates the catalytic metal ion while its propanoic acid moiety mimics the enzyme's α-ketoglutarate cofactor. Using chemical proteomics, the authors showed that immobilized GSK-J1, but not GSK-J2, specifically captured JMJD3 and UTX from whole cells. An esterified prodrug form of GSK-J1 with improved cellular efficacy maintained the amount of H3K27me3 in treated cells and was used to show that JMJD3 and UTX activity are required for the transcriptional regulation of proinflammatory genes in macrophages. In total, the studies expand the scope of chromatin-modifying enzyme inhibitors available to probe their complex cellular roles. *TLS*

MICROBIOLOGY

Size control is Fab

Proc. Natl. Acad. Sci. USA, published online 20 Aug 2012; doi:10.1073/pnas.1209742109

NATACHA RUIZ



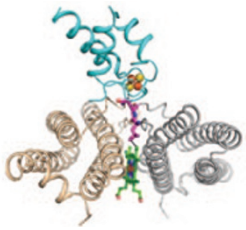
Bacteria regulate their cell size, growth rate and division depending on the availability of nutrients, growing larger and faster in nutrient-rich medium, but there is limited mechanistic information as to how *E. coli* regulate cell size in response to nutrients. Yao *et al.* shed new light on this process by examining mutations in the fatty acid (FA) biosynthetic gene *fabH*, found in a screen for suppressors of mutations in transport of lipopolysaccharide (LPS) from the inner membrane to the cell surface. Deletion of *fabH* allows survival of cells with very limited LPS at the cell surface, and FabH is essential in mutants where synthesis of the small regulatory molecule ppGpp is compromised. *fabH* mutants have a small-colony phenotype and altered FA composition that the authors detected by gas chromatography. Mutants also have a marked reduction in cell surface area (50%) and in cell volume (70%) in rich medium. Inhibition of FA biosynthesis by two additional independent strategies suggests that the slow-growth and small-size phenotypes are not related to changes in FA composition but rather are due to changes in FA biosynthesis capacity. Finally, *fabH* mutants have a 75% reduced rate of cell envelope elongation. These results suggest that FA biosynthesis regulates cell size in response to nutrients by regulating cell envelope growth and that deletion of *fabH* restores balance between LPS assembly rates and envelope biogenesis. *MB*

METALS

Get the iron out

J. Am. Chem. Soc. **134**, 13470–13481 (2012)

RITESH KUMAR



Ferritins and ferritin-like proteins such as bacterioferritin B (BfrB) in *Pseudomonas aeruginosa* store iron as the insoluble Fe^{3+} to prevent formation of reactive oxygen species by excess Fe^{2+}. Cells must also be able to retrieve this iron when needed; in *P. aeruginosa*, both ferredoxin reductase and the [2Fe-2S]-containing bacterioferritin-associated ferredoxin (Bfd) are thought to be involved in this process, but the mechanistic details are unknown. Yao *et al.* investigate this question in their report of the crystal structure of Bfd bound to the 24-mer BfrB. This first structure of Bfd shows a three-helix fold that has only previously been seen within the context of larger structures; its stability as an isolated domain is shown to depend on a phosphate ion that coordinates three cationic residues. At the largely conserved interface, Bfd inserts into a cleft between two BfrB monomers such that the [2Fe-2S] cluster is 15.1 Å from a buried heme thought to relay electrons from the surface to the core Fe^{3+}. Functional studies with purified proteins indicate that Bfd loaded with [2Fe-2S] is necessary and sufficient for Fe^{2+} release and show alternating oxidation states of the [2Fe-2S] cluster and heme, supporting a direct electron transfer mechanism. These results demonstrate that Bfd facilitates iron mobilization and sets the stage for further detailed study of this interesting system. *CG*

Written by Mirella Bucci, Amy Donner, Catherine Goodman & Terry L. Sheppard