GLYCOMICS

## Highlights for carbohydrates

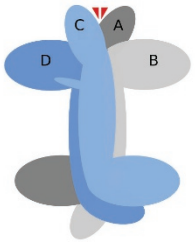
*Nat. Methods* **13**, 528–534 (2016)

Glycans are a diverse class of carbohydrate–biomolecule conjugates that have many different biological functions. Yet, because of their complex and heterogeneous structures, glycoconjugates have been less tractable to chemical synthesis and analytical characterization than other biomolecules, a fact that has hindered mechanistic glycobiology. Song *et al.* now report an approach called ORNG (oxidative release of natural glycans), which enables scalable glycomic profiling of biological samples. ORNG uses sodium hypochlorite (NaClO), the reactive component of household bleach, to release glycans from protein and lipid glycoconjugates into products that can then be tagged with visualization reagents, analyzed by HPLC and MS, or used to construct glycan microarrays. In the case of N-glycans from sources including eggs and human saliva, NaClO treatment oxidatively cleaves the asparagine glycosidic bond to produce glycans with free reducing ends. ORNG can also identify sites of protein O-glycosylation, by releasing glycans terminated with O-linked glycolic or lactic acid moieties, and was used to profile O-glycans from mouse gastrointestinal tissues. ORNG was also effective in fragmenting glycosphingolipids from porcine brain tissue to produce cyanomethyl glycosides, which could be further derivatized by selective chemistries. Taken together, ORNG offers a straightforward and preparative method for future analyses of the biological roles of glycoconjugates. *TLS*

ANTIBIOTICS

## I want a new drug

*Proc. Natl. Acad. Sci. USA* http://dx.doi.org/10.1073/pnas.1600630113 (2016)



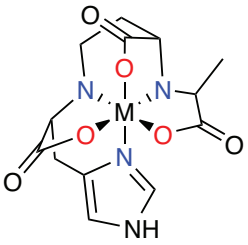

PNAS

Although tuberculosis is one of the leading infectious causes of mortality and morbidity in the world, reports of drugs that kill its causative bacterium, *Mycobacterium tuberculosis*, through a new mechanism of action are very rare. Kasbekar *et al.* used a fluorescence-based assay to screen a library of 479,984 small molecules for inhibitors of fumarate hydratase, an essential enzyme in the tricarboxylic acid cycle. *M. tuberculosis* and human fumarate hydratase are highly homologous, with an overall sequence identity of 53% and identical amino acid residues in their active sites, so it has been very difficult to find selective inhibitors of *M. tuberculosis* fumarate hydratase. The authors identified two structurally similar molecules that were potent inhibitors of the bacterial enzyme *in vitro*, and comparison of the X-ray crystal structures of the enzyme in the absence and presence of one of the inhibitors revealed that two molecules of the compound bind in the same allosteric pocket. The small molecule did not inhibit the human enzyme *in vitro*, which the authors believe is because the amino acids in the allosteric sites of the two homologs are quite different. Although the molecule inhibited the growth of *M. tuberculosis* under aerobic conditions, the authors believe that significant optimization is still required before they will have an advanced probe for future studies. *JMF*

BIOSYNTHESIS

## Metal-pilfering pathogens

*Science* **352**, 1105–1109 (2016)




Pathogenic bacteria scavenge essential metal nutrients from their host organisms, often through the biosynthesis and excretion of metal-chelating siderophores followed by re-uptake of the metal–siderophore complexes. Ghssein *et al.* have elucidated the biosynthesis of staphylopine, a metallophore produced by *Staphylococcus aureus*, which is structurally similar to the metal chelator nicotianamine found in plants. Comparison of the wild-type strain to strains carrying mutations within the staphylopine biosynthetic gene cluster by liquid chromatography and mass spectrometry led to characterization of metal–staphylopine complexes and identification of three key enzymes that define its biosynthetic pathway. A histidine-specific racemase first converts L-His to D-His, and a nicotianamine synthase homolog catalyzes the addition to D-His of α-aminobutyric acid derived from SAM. An opine synthase homolog then catalyzes the NADPH-dependent condensation of this intermediate with pyruvate to yield staphylopine. Once secreted out of the cell through a dedicated exporter, staphylopine can bind copper, nickel, cobalt, zinc or iron; a transporter also encoded in the gene cluster is responsible for importing the resulting metal–staphylopine complex. As homologous gene clusters have also been identified in other pathogens, this enhanced understanding of bacterial metal acquisition could aid efforts to treat infections by these strains. *CD*

PROTEOSTASIS

## TSA1 to the rescue

*Cell* http://dx.doi.org/10.1016/j.cell.2016.05.006 (2016)

The failure to remove and degrade stress-induced protein aggregates by the Hsp70 chaperone and Hsp104 disaggregase has been associated with premature aging in yeast. However, it is not clear how Hsp70 and Hsp104 are recruited to these aggregates in times of oxidative or heat stress. Hanzén *et al.* performed a yeast synthetic lethal genetic screen and observed interactions of the yeast cytosolic peroxiredoxin Tsa1 with the protein quality-control system. Elevated dosage of Tsa1 promoted increased lifespan independent of caloric restriction and hydrogen peroxide ($H_2O_2$) scavenging and was associated with a low level of protein aggregates. The authors found that $H_2O_2$ promoted the direct recruitment of Hsp70 and Hsp104 to age-induced protein aggregates. This recruitment was facilitated by the hyperoxidation of Tsa1 at a specific cysteine residue by $H_2O_2$. *TSA1* mutants showed rapid accumulation of protein aggregates and ubiquitinated proteins, resulting in premature aging. Hanzén *et al.* furthermore noted that the timing of aggregate clearance coincided with sulfiredoxin (Srx1)-mediated reduction of Tsa1. Consistent with this observation, *srx1* mutants exhibited slower aggregate clearance, suggesting that Srx1-mediated reduction of Tsa1 is required to facilitate the removal of these aggregates. Overall, these findings propose a redox switch defense mechanism for maintaining proteostasis in aging cells and in response to $H_2O_2$ stress. *GM*

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