DRUG DISCOVERY

Mutants muffled

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Somatic mutations in the active sites of isocitrate dehydrogenases (IDH), enzymes that interconvert isocitrate and α-ketoglutarate (αKG), confer new activity that results in the reduction of αKG to *R*-2-hydroxyglutarate (*R*-2HG). *R*-2HG inhibits αKG-dependent dioxygenases, including histone and DNA demethylases, altering the epigenetic state of cells and thereby promoting cancer cell proliferation. It would be desirable to identify compounds that block the activity of mutant IDH enzymes. Wang *et al.* and Rohle *et al.* now report AGI-6780 and AGI-5198, potent and selective inhibitors of R140Q IDH2 and R132H IDH1, respectively. Structural and biochemical studies revealed that AGI-6780 binds the IDH2 dimer interface and is an allosteric inhibitor. Both AGI-6780 and AGI-5198 reduced the amounts of *R*-2HG. AGI-6780 induced the differentiation of leukemic blasts derived from patients, and AGI-5198 blocked the growth of *IDH1*-mutant glioma cells in soft agar and xenograft mouse models. High doses of AGI-5198 promoted gene expression changes that were consistent with differentiation and reduced repressive histone marks on promoters of these genes. These new inhibitors can be applied in additional preclinical experiments to determine whether the inhibition of mutant IDH enzymes is a viable approach to treating various cancer types and whether these inhibitors are leads that can be used to generate anticancer drugs. *AD*

IMMUNOLOGY

Taking on the pHagosome

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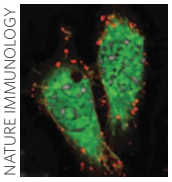

Clearance of microbes by the immune system requires their delivery into acidic degradative organelles, called phagolysosomes, within macrophages. Acidification of microbe-containing compartments occurs rapidly after phagocytosis and requires vacuolar H^+ ATPase (V-ATPase), though the mechanism by which pH is regulated is poorly defined. To determine the nature of the acidification event, Sokolovska *et al.* focused on the activity of the inflammasome, a complex that is assembled after the immunological receptor NLRP3 senses pathogens or danger signals. When *Staphylococcus aureus* was introduced to macrophages, the authors found active caspase-1, the effector of the inflammasome, near bacteria-containing phagosomes. Caspase-1 activation required the inflammasome and occurred immediately after internalization independently of V-ATPase activity. From a bioinformatics analysis of phagosome-associated proteins, the authors identified several candidate caspase-1 substrates, including ROS-generating NADPH oxidase, NOX2, which neutralizes phagosomal pH. They verified that at least one of the predicted caspase-1 cleavage events within the NOX2 complex occurs *in vivo* during infection. Cleavage by caspase-1 decreases NOX2 activity and the production of ROS that is associated with phagocytosis, thereby directly controlling the accumulation of protons in the phagosome. A role in regulating phagosome pH by counteracting the buffering activity of the NOX2 complex adds a crucial function to the known roles of caspase-1 in the release of proinflammatory cytokines and immune cell death. *MB*

BIODEGRADATION

Making the cut

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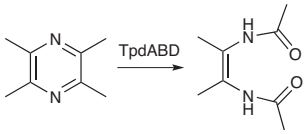

The catabolism of pyrazines—odorous compounds produced by bacteria and plants—is poorly understood. Pathways for the degradation of di- and trisubstituted pyrazines have been proposed, but these related routes consistently include a hydroxylation step that cannot be performed on a tetrasubstituted pyrazine such as tetramethylpyrazine (TTMP). To establish a pathway for these pyrazines, Kutanovas *et al.* first looked for genes that were induced by TTMP in the TTMP-degrading bacterium *Rhodococcus jostii* TMP1. MS/MS analysis of the primary upregulated protein led to a locus encoding this protein (named TpdA), along with seven other open reading frames. Sequence analysis suggested that the putative proteins included seven enzymes and one transcriptional regulator with similarity to the LuxR transcription factor family. Expression of TpdABC or TpdAB in a non–TTMP-degrading strain led to the production of oxidatively cleaved intermediates, supporting the authors' predictions that TpdA is a flavin monoxygenase and TpdC is a hydrolase, whereas TpdD, a putative flavin reductase, could apparently be replaced by an endogenous enzyme in the nondegrading host. TpdE was confirmed to be a short-chain reductase, yielding *N*-(3-hydroxybutan-2-yl) acetamide as a potential substrate for the final uncharacterized enzymes. Although further study will be needed to clarify the specific mechanisms of each enzyme, including a potential role for TpdB in destabilizing the aromatic ring, this enzymatic sequence provides the first validated pathway for pyrazine degradation. *CG*

PROTEIN ANNOTATION

Not the usual suspect

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Candida albicans stochastically alternates between two phenotypic states in a process called 'white-opaque switching'. Nearly 20% of the genome is differentially regulated in the two states, with five transcriptional regulators known to be involved in the switch. To search for additional regulators, Lohse *et al.* combined RNA-seq transcriptional profiling data of cells in the two different states and ChIP-chip binding data for the master regulator, white-opaque regulator 1 (Wor1), to find genes that were both upregulated in opaque cells and regulated by Wor1. Overexpression of the only gene identified caused switching to the opaque phenotype; given similar behavior to known regulators, the authors named the protein encoded by this gene Wor3. Surprisingly, genetic deletion of Wor3 also promoted the opaque state, altering the expression levels of 125 genes. Analysis of two libraries of 8-mer DNA strands identified a Wor3-specific 5′-ATAACC-3′ DNA-binding sequence. Comparison of these libraries with Wor3 ChIP-chip results highlighted a correlation with the location of Wor1's preferred sequences, suggesting that the two regulators may cooperate. Bioinformatics analysis and structure prediction algorithms identified Wor3 family members but did not indicate homology to any other protein family except through similarity to Wor3's eight CXXC motifs. Phylogenetic analysis suggested that Wor3 evolved and was then lost, given its presence in a subset of fungal genomes only. The discovery of a new participant in fungal state regulation serves as a reminder that surprises lurk in unannotated DNA. *CG*