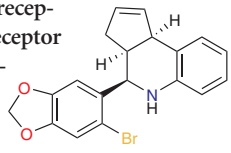
Ants go gardening

Fungus-growing ants cultivate a chosen species of fungus but must protect this fungus from a second parasitic fungus. Though this mutualistic system is known to involve actinobacteria cultured by the ants, the specific details of how the ants tend their fungal garden were unknown. Oh *et al.* have now determined the chemical basis of one of these small ecological systems with the isolation and structural characterization of dentigerumycin, a bacterially produced small molecule that is selectively toxic to the parasitic fungus. The authors used NMR to establish the overall backbone and some of the stereocenters of the molecule, which was shown to be a cyclic depsipeptide with several unusual features. As piperazic acids—represented three times in the compound—are particularly difficult to analyze, the authors further developed an improved approach to assign the stereochemistry at these sites. The identification of this molecule will allow further mechanistic investigations into this interesting interspecies system. [**Brief Communications, p. 391**] *CG*

Substituting SAM

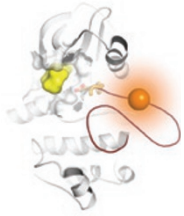
Controlling DNA methylation states represents an important pathway for epigenetic regulation of gene expression. Cytosine-5-methyltransferases such as HhaI methyltransferase (M.HhaI) catalyze the transfer of methyl groups from the cofactor *S*-adenosylmethionine (SAM) to the C5 position of cytosines in DNA in a process that involves nucleophilic activation of the target cytosine residue. Liutkevičiūtė *et al.* now demonstrate that M.HhaI can facilitate carbon-carbon bond formation reactions between cytosines and exogenous aldehydes. The authors prepared 'activated cytosine intermediate' (ACI) complexes by mixing DNA target sequences with M.HhaI in the absence of the SAM cofactor and demonstrated that aliphatic aldehydes reacted with the ACI to produce the corresponding 5-(α-hydroxyalkyl)cytosine derivatives. The authors also demonstrate that methyltransferases can drive the coupling reaction in reverse, as M.HhaI was shown to convert 5-hydroxymethylcytosine modifications back to cytosine. This observation provides compelling support for oxidative demethylation pathways that are thought to be important in epigenetic regulation and DNA repair. [**Brief Communications, p. 400**] *TLS*

Unraveling estrogen signaling



In addition to the classical nuclear estrogen receptors ERα and ERβ, the G protein–coupled receptor GPR30 can activate signaling pathways in response to estrogen. A previously identified GPR30-selective agonist has aided in understanding some of the physiological roles of GPR30. However, a pharmacological antagonist to complement the agonist studies has been missing. Using a combination of virtual and biomolecular screening, Dennis *et al.* identified a compound that is structurally related to the agonist, binds GPR30 with high affinity and selectively antagonizes GPR30-mediated signaling. With this antagonist, the authors found that in mice GPR30 contributes to regulation of depression and uterine epithelial cell proliferation, whereas uterine water content appeared to be solely determined by classical estrogen receptors. These results provide new insights into GPR30 biology and an important new chemical tool for dissecting *in vivo* responses to estrogen. [**Articles, p. 421**] *JK*

Allostery by HTS

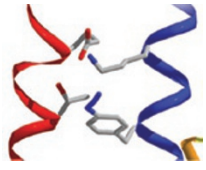


In the active, DFG-in kinase conformation, the phenylalanine of the 'DFG' motif occupies an allosteric pocket near the active site, whereas in the inactive, DFG-out conformation, this phenylalanine moves to partially block the ATP binding site. Type II kinase inhibitors, such as imatinib, bind to the newly vacated allosteric site and thus stabilize an inactive kinase form. To develop an assay for type II inhibitors, Simard *et al.* labeled the cSrc activation loop, which is sensitive to changes in the DFG motif, with an environmentally sensitive fluorophore. Movement of the DFG motif altered the environment of the fluorophore, resulting in a change in signal that could be used for screening. Using this approach, the authors identified pyrazolourea compounds that inhibited cSrc and confirmed binding to the DFG-out conformation by cocrystal structures. This method provides the first high-throughput assay for identifying inhibitors targeting the inactive kinase conformation. [**Brief Communications, p. 394**] *JK*

Tinkering with linking

In fragment-based drug discovery (FBDD), weakly binding chemical 'fragments' are covalently linked together to generate a more potent ligand. In theory, the linked compound should bind with the combined binding energy of the two fragments. However, in practice, the linker often perturbs the binding of one or both fragments. To systematically explore linker effects, Chung *et al.* synthesized a series of four linkers with varying flexibility and used them to connect uracil with aldehyde fragments previously shown to bind to human uracil DNA glycosylase. Cocrystal structures and biochemical dissection of the binding energetics were used to assess the impact of linkers on each fragment pair. This combinatorial approach revealed that the linker can have a significant impact on overall binding energy that cannot necessarily be predicted from crystal structures. Thus, experimentally optimizing the linker is as important as identifying optimal fragments for generating potent ligands. [**Articles, p. 407**] *JK*

Reading rhodopsin rearrangements



The azido group in *p*-azido-L-phenylalanine (azidoPhe) is well suited to serve as an FTIR probe, as it is both small and spectroscopically distinct from natural proteins, but it has not been used in this way. Ye *et al.* now demonstrate the incorporation of this non-natural amino acid into the membrane protein rhodopsin, a prototypical GPCR, to monitor receptor function during protein activation. The authors first tested a variety of tRNA/synthetase pairs for amber suppression technology and insertion sites to determine optimal conditions for modified protein expression. Then, by monitoring the FTIR signature in the presence and absence of light, they observed differences that varied with residue placement along the protein backbone that could be assigned as specific movements that occur during receptor activation. This methodology should be broadly applicable for studies of protein structure and function. [**Brief Communications, p. 397**] *CG*

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