In This Issue

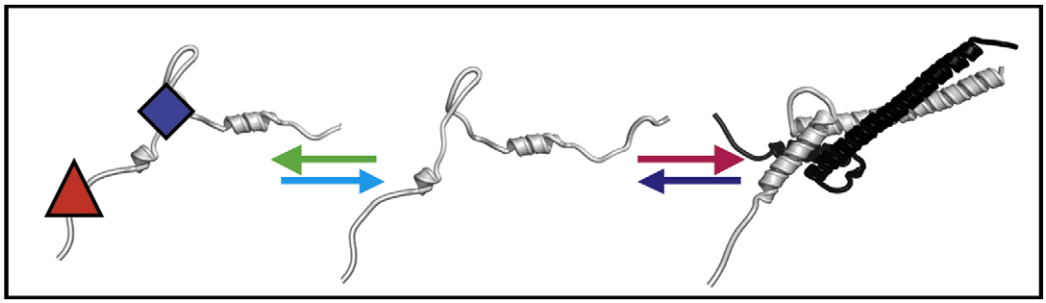
Pssst, Bacteria are Talking

PAGE 1141

Bacterial populations use quorum sensing (QS) to coordinate their behavior and execute a number of functions according to the density of their population. This phenomenon of intercellular communication is mediated by different classes of small molecules, including *N*-acylhomoserine lactones. This minireview by Cooley et al. discusses the most recent findings that some soild bacteria have the ability to employ host-derived compounds and produce alternative *N*-acylhomoserine lactones. Additionally, this minireview discusses the recently described mechanism of how *N*-acylhomoserine lactones affect host inflammatory signaling pathways to promote bacterial survival.

Modulating Intrinsic Disorder

PAGE 1149



Several small molecules that disrupt c-Myc-Max heterodimerization act by specifically binding c-Myc and thus stabilizing the intrinsically disordered (ID) protein monomers over the highly ordered heterodimer. The characteristics of sites within an ID region capable of specific binding by small molecules were unknown. Here, Follis et al. describe the interactions of two small molecules that form soluble, reversible complexes with c-Myc. These findings suggest that potential binding sites may be prevalent in ID proteins and that the discovery of small molecules capable of modulating the functions of these proteins may be practicable. (Figure adapted from Follis et al.)

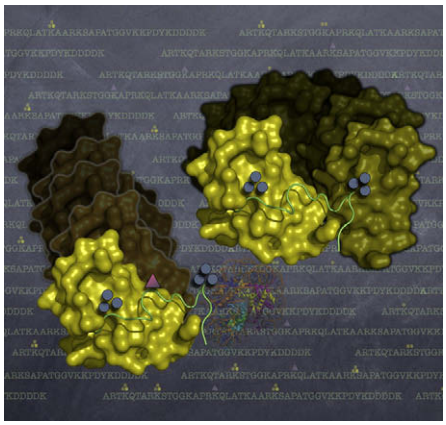
Protein-Protein Interactions within the Type II PKS Complex

PAGE 1156

Myriads of biological processes depend on the dynamic protein/protein and protein/ligand interactions. In metabolic pathways, enzymes catalysing sequential reactions often transiently associate to increase reaction rates and protect labile intermediates by channelling them directly from one active site to another. Castaldo et al. now investigate complex protein interactions involved in the biosynthesis of the aromatic polyketide components of daunorubicin, an anticancer drug, using a yeast two-hybrid system, tandem affinity purification, and computer simulation. The approach resulted in gaining valuable insight into the protein-protein interactions within the type II PKS complex, which might lead to improved set of design rules for the synthesis of aromatic polyketides through combinatorial biosynthesis.

Wagging Histone H3 Tails

PAGE 1166



By redefining the genetic code, one can make the ribosome incorporate nonproteinogenic amino acids into peptides according to information encoded in mRNAs. Using this genetic code reprogramming, Kang et al. have prepared a series of N-terminal tails of histone H3 containing combinatorial lysine modifications, including mono-, di-, and tri-methylation and acetylation. The full-length 38-mer H3 tails with distal combinatorial modification enabled Kang et al. to reveal the possible crosstalk among epigenetic markers upon HP1 chromodomain binding. Thus, the approach described here could be a general tool to investigate lysine modification-protein interaction relationships. (Figure provided by Kang et al.)

Microbial Symbiont Investigated

PAGE 1175

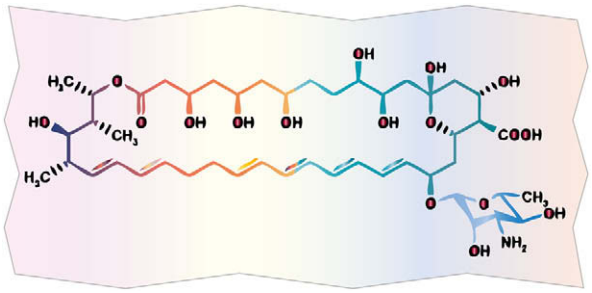
Many pharmacologically active natural products are generated by microbial symbionts of marine invertebrates. In most cases, these microorganisms remain refractory to cultivation, and thus gene disruption methods cannot be employed to establish that a sequenced biosynthetic cluster specifies production of a particular secondary metabolite. In this study, Lopanik et al. demonstrate that several of the genes from the putative *bry* biosynthetic system responsible for assembly of the bryostatins, polyketide metabolites with anticancer and neurological activity, operate according to their hypothesized role based on in vitro biochemical analysis and genetic complementation in a heterologous host. This work illustrates the power of combined approaches to provide important insights into enzyme function of complex microbial symbiont-derived biosynthetic systems.

Genetically Encoding N^{ε}-(*o*-Azidobenzyloxycarbonyl)lysine

PAGE 1187

Yanagisawa et al. report here a method to incorporate a nonnatural, chemically reactive amino acid, N^{ε}-(*o*-azidobenzyloxycarbonyl)-L-lysine (AzZLys), site specifically into proteins in *Escherichia coli* cells. *Methanosarcina mazei* pyrrolysyl-tRNA synthetase (PylRS) esterified an amber suppressor tRNA ($tRNA^{Pyl}$) with several nonnatural lysine derivatives, in addition to the natural substrate pyrrolysine. Some of the lysine derivatives were incorporated into proteins by a mutant PylRS●$tRNA^{Pyl}$. Then, structure-based and random mutagenesis of PylRS enabled 10 mg-scale production of proteins that contain AzZLys site specifically. The AzZLys residue in the produced protein can be modified with a fluorescent label, more efficiently than the *p*-azido-L-phenylalanine residue with a shorter side chain.

Improved Anti-Fungal Polyene Macrolides



PAGE 1198

Efficient and safe antifungal agents are urgently needed due to the growing number of life-threatening systemic fungal infections. Brautaset et al. generated new analogs of the polyene macrolide antibiotic nystatin using biosynthetic engineering and tested the obtained compounds for antifungal activity and toxicity both in vitro and in vivo. Two nystatin analogs were shown to be effective in treatment of disseminated candidosis in mice, while being considerably less toxic compared to amphotericin B, the only polyene macrolide currently used for treatment of systemic fungal infections. The two analogs might therefore represent promising lead compounds for further development of antifungal drugs for human therapy.

Structural Requirements for Cannabinoid Receptor Binding

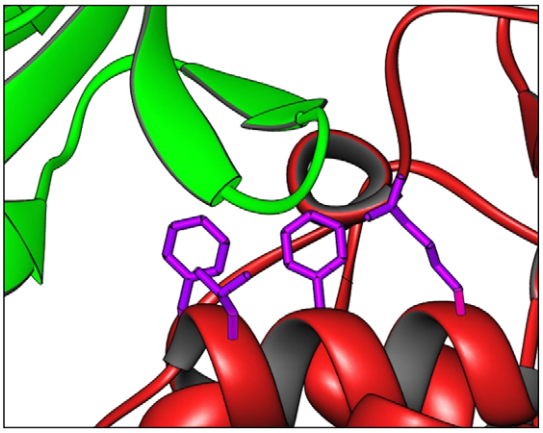
PAGE 1207

Activity of the CB2 cannabinoid receptor has been implicated to be of benefit in neuropathic pain, inflammation, and immune disorders. Thus, small-molecule ligands that bind to and activate the CB2 receptor have promise as potential therapeutics for these and other disorders. In order to design ligands that are potent and selective for the CB2 receptor without inducing off-target effects, it is important to understand the ligand-binding architecture of the CB2 receptor. By using a "ligand-assisted protein structure" experimental approach, this work by Pei et al. helps characterize directly the structural requirements for ligand binding to the human CB2 receptor critical to pharmacotherapeutic CB2 receptor modulation.

Isoform Specific Activators of AMPK

PAGE 1220

The AMP-activated protein kinase (AMPK) is a key player in the regulation of cellular and whole-body energy balance and is now recognized as a promising target for new drugs to fight the growing epidemic of obesity and Type 2 diabetes. The thienopyridone drug A769662 has been shown to activate AMPK by an AMP-independent mechanism, revealing the existence of an alternate regulatory site. In this study, Scott et al. demonstrate that A769662 selectively activates specific isoforms of AMPK, highlighting the feasability of developing isoform-specific activators of AMPK that can target AMPK in particular tissues. (Figure adapted from Scott et al.)



Stereochemistry of Enoylreduction in Modular PKS

PAGE 1231

When an enoylreductase (ER) enzyme of a modular polyketide synthase (PKS) reduces a propionate extender unit that has been newly added to the growing polyketide chain, the resulting methyl branch may have either *S*- or *R*-configuration. Kwan et al. have now uncovered a correlation between the presence or absence of a unique tyrosine (Y) residue in the ER active site and the chirality of the methyl branch that is introduced. When this position in the active site is occupied by Y, the methyl branch has *S*-configuration, otherwise it has *R*- configuration. Thus, a mutation (Y to valine [V]) in an erythromycin PKS-derived ER caused a switch from *S*- to *R*-. However, V to Y mutation in a rapamycin-derived PKS ER was insufficient to achieve a switch from *R*- to *S*-, suggesting that additional residues also participate in stereocontrol of enoylreduction.