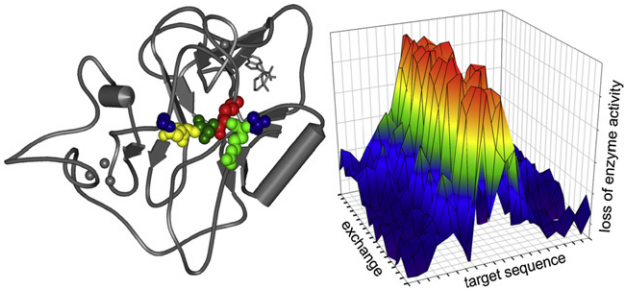
In This Issue

Determination of Substrate Specificity of Histone Methyltransferases



PAGE 5

Human cells express only a small subset of their genome. Gene expression is controlled by covalent modifications of the DNA and the histone proteins, including methylation of histones' lysine residues. Rathert et al. studied the specificity of histone methyltransferases using peptide SPOT arrays as substrates. This approach allowed simultaneous investigation of enzyme activity on 400 different substrate peptides. For the Dim-5 histone-3 lysine-9 methyltransferase from *Neurospora crassa*, authors show recognition of R8-G12 of the H3 tail, in excellent agreement with the structure of Dim-5 in complex with a target peptide. Furthermore, they demonstrate that the specificity of histone methyltransferases can be altered by protein design. (Figure credits: Rathert et al.).

A Revised Pathway for Wall Teichoic Acids Biosynthesis

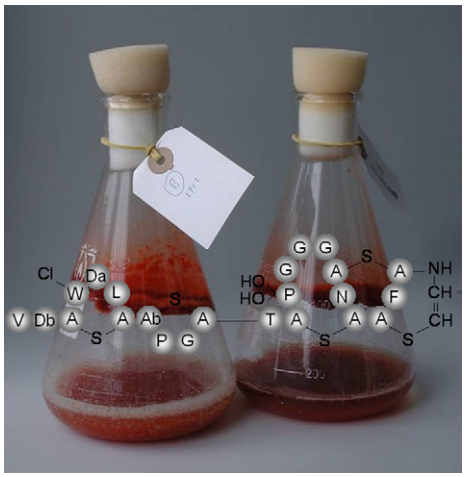
PAGE 12

Cell walls of many Gram-positive bacteria are extensively decorated by anionic polymers known as wall teichoic acids (WTAs). WTAs consist of a disaccharide-based linkage unit and a repeating polyol-phosphate polymer, synthesized in the cytoplasm and translocated to the outside of the cell. Since WTAs are indispensable for *Staphylococcus aureus* colonization and infection, their biosynthesis pathway represents a viable target for antibiotic development. To clarify the biosynthesis process for WTAs, Brown et al. performed the in vitro reconstitution of the intracellular steps of poly-ribitol phosphate WTAs biosynthesis in *S. aureus* NCTC8325. The authors assigned the function of each gene product involved in the process, which led to two significant revisions of the previously proposed biosynthetic pathway.

Microbisporicin: A Different Inhibitor of Peptidoglycan Synthesis

PAGE 22

Antibiotics blocking bacterial cell wall assembly (β-lactams and glycopeptides) are facing a challenge from the progressive spread of resistant pathogens. Lantibiotics are promising candidates to alleviate this problem. Microbisporicin, a lantibiotic produced by an uncommon actinomycete and discovered in the course of a biological activity-guided screening, is a potent antibacterial. Castiglione et al. show that it is produced as two structurally related and similarly active 24-mer peptides, containing two novel posttranslational modifications and five thioether intramolecular bridges. Its spectrum of activity covers most of the Gram-positives and some Gram-negatives of medical importance. Considering also its efficacy in vivo, microbisporicin represents a new antibiotic to treat emerging infections. (Figure credits: Castiglione et al.)



Reprogramming Genetic Code for N-methyl-peptide Synthesis

PAGE 32

N-methyl amino acids are often contained in naturally occurring peptide products and confer proteolytic stability and membrane-permeability upon them. Kawakami et al. report a new approach for the programmed synthesis of N-methyl-peptides using an *E. coli* reconstituted translation system. The key technology in this study is a ribozyme-based de novo tRNA acylation, called flexizyme, system that enables reprogramming of the genetic code. The authors demonstrate messenger RNA-directed synthesis of linear and cyclic N-methyl-peptides. This technology offers a tool for the construction of diverse N-methyl-peptide libraries, potentially leading to the discovery of in vivo compatible therapeutic peptides.

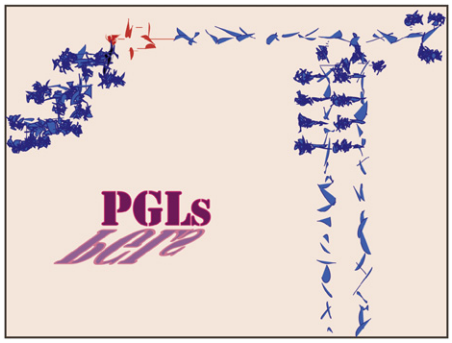
In This Issue

The Language of Host-Pathogen Interactions: Bacterially Produced NO

PAGE 43

Nitric oxide (NO) is an important signaling molecule in eukaryotic metabolism and pathogen defense. In mammals, NO is produced by nitric oxide synthases (NOSs). Genome sequencing identified truncated mammalian NOS homologs in multiple Gram-positive bacteria. The study by Johnson et al. provides the first direct evidence of in vivo NO production by bacterial NOS-like proteins in plant-pathogenic *Streptomyces* species. This NO is produced in response to host-derived signals and at the host pathogen interface, suggesting a possible role in manipulating host signaling. Since some of the human pathogens also contain NOS, the authors suggest that NOS-produced NO might be important for the host-pathogen interactions across kingdoms.

Targeting Phenolic Glycolipids Biosynthesis to Treat Mycobacterial Infections



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Phenolic glycolipids (PGLs) are polyketide-derived small-molecule virulence factors produced by mycobacterial pathogens. Ferreras et al. combined bioinformatic, genetic, biochemical, and chemical biology approaches to illuminate the mechanism of chain initiation required for assembly of the phenolphthiocerol moiety of PGLs. The insights gained allowed the authors to develop a PGL assembly inhibitor with potent activity in several mycobacterial pathogens. This work advances our understanding of the biosynthesis of an important group of mycobacterial virulence effectors and provides support for the feasibility of targeting PGL biosynthesis to develop new drugs to treat mycobacterial infections. (Figure credits: Ferreras et al.; artistic rendition of PGL structure.)

Rescue of F508del-CFTR by Solubilizing Mutations

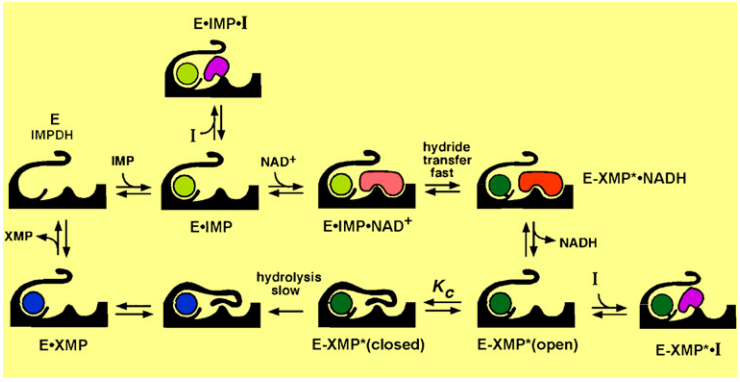
PAGE 62

The most frequent pathogenic mutation in Cystic Fibrosis (CF) is F508del, found in the first nucleotide-binding domain (NBD1) of CF transmembrane conductance regulator (CFTR) protein. F508del mutation disrupts biosynthesis, cell surface expression, and channel gating of CFTR, thus resulting in severe disease phenotype. The current high-resolution structure of F508del NBD1 domain of CFTR was determined upon introduction of a number of additional mutations to improve protein solubility. Here, Pissarra et al. investigate the effect of these additional mutations and demonstrate that these solubilizing mutations attenuate both the trafficking and functional defects of F508del-CFTR in vivo, thus suggesting that the existing structure of F508del-NBD1 corresponds to a partially corrected conformation. Accordingly, the structure of F508del-NBD1 without additional mutations is still needed to understand how F508del impacts on NBD1 conformation.

Exploiting Evolutionary Divergence of Parasite and Host Enzymes

PAGE 70

The "vicious cycle of diarrhea and malnutrition" in developing countries could be broken with effective chemotherapy against *Cryptosporidium*. This protozoan parasite also poses a credible bioterrorism threat. Eukaryotic pathogens present a particularly challenging problem for drug design because the targets resemble host proteins. Surprisingly, the purine salvage pathway of *Cryptosporiium* relies on an enzyme obtained from a prokaryote via horizontal gene transfer, which is very different from the host counterpart. Umejiego et al. designed an HTS to target the most diverged binding site. This screen identified parasite-selective inhibitors that are more effective than paromomycin, the current standard for anticryptosporidial activity. (Figure adapted from Umejiego et al.)



Engineered Biosynthesis of Oxo-Amphotericins

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Amphotericin B is an effective but toxic antifungal antibiotic that is produced by *Streptomyces nodosus*. In addition, the compound is also active against enveloped viruses, protozoan parasites, and pathogenic prion proteins. Therefore, there is considerable interest in production of nontoxic analogs by both chemical modification and by genetic manipulation of *S. nodosus*. Here, Power et al. report engineering of amphotericin biosynthetic genes to introduce ketone groups at different positions of the macrolactone core. This approach provides starting material for a range of interesting semisynthetic derivatives, with optimized therapeutic properties.