In This Issue

Natural Product-Based Antifungal Discovery

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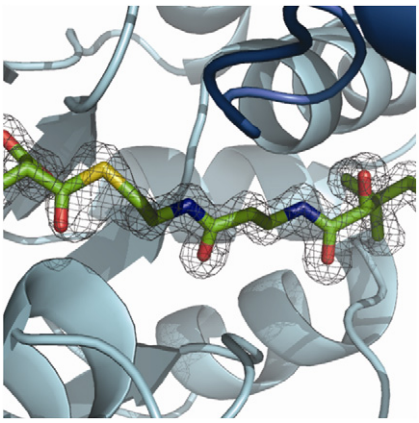
Natural products (NPs) have been a rich source of antibiotics and other pharmaceuticals. Unfortunately, discovery of structurally novel and chemically tractable NPs has slowed down, resulting in decrease in industrial efforts NP discovery and development. In this perspective, Roemer et al. discuss currently employed strategies for antifungal NP discovery and present potential solutions for barriers in target identification and validation. The authors focus the discussion on a *Candida albicans* chemical genetics-based NP screening approach and describe a dereplication strategy to predict the presence of known or new bioactive molecules within NP extracts that might lead to resolution of issues that continue to disrupt NP-based antimicrobial discovery.



An Enzymatic Extender Unit Generation Machine

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In vitro experiments using polyketide synthases (PKSs) are limited by the cost and availability of extender units such as methylmalonyl-CoA. Hughes and Keatinge-Clay show that the malonyl-CoA ligase *Streptomyces coelicolor* MatB can be employed to help overcome this obstacle, synthesizing all combinations of five diacids and three thiol acceptors. Methylmalonyl groups linked to CoA, D-pantetheine, or N-acetylcysteamine were incorporated by a PKS module into a triketide pyrone. Two MatB ternary complex structures provide structural insight into the broad substrate specificity of this invaluable ligase. The research presented marks significant progress towards the economical and preparative in vitro synthesis of polyketides.

c-Abl Activation through Myristoyl Binding Site

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Here, Yang et al. describe a small-molecule activator of the c-Abl kinase DPH, which was identified in an HTS campaign. DPH binds to the myristoyl binding site of c-Abl and prevents the formation of its autoinhibited conformation. DPH displays potent enzymatic and cellular activity in stimulating c-Abl activation, therefore representing the first cell-permeable, small-molecule tool compound for c-Abl activation. It has been suggested that c-Abl activation may play a role in processes such as tumorigenesis, metastasis, and normal myelopoiesis. Therefore, identification of this small-molecule c-Abl activator allows further investigation of the physiological as well as pathophysiological functions of endogenous c-Abl.

Flavonoid Metabolism Linked to Vacuolar Trafficking Defects

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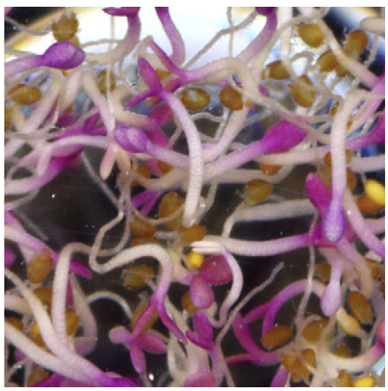
In this report, Rosado et al. assign two important functions to flavonoids: as metabolites required for the maintenance of the vacuolar integrity under oxidative stress conditions and as regulators of the delivery of vacuolar targeted cargoes. These characteristics might be used to enhance or inhibit the accumulation of products of interest in the large central vacuole either by genetic or chemical manipulation of the flavonoid biosynthetic pathway. Further analysis of the Sortin1 chemical structure may provide useful information regarding the Sortin1 binding partners and may also allow the uncoupling of vacuolar cargo and flavonoids transport pathways. More broadly, the study demonstrates that hypersensitive screens, forward and reverse genetics, and chemical genomics are powerful tools to identify and dissect crosstalks between metabolic pathways.



Primary and Secondary Metabolism Crosstalk

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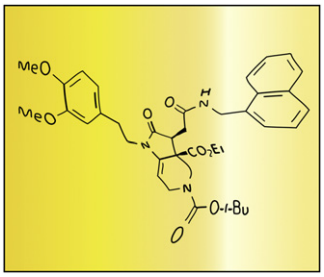
The regulation of gene expression and product formation of the only PKS-NRPS present in the genome of the filamentous fungus *Aspergillus terreus* has been elucidated. Although a transcriptional activator for PKS-NRPS expression was identified, induced overexpression remained negatively dominated by glucose repression. Using β-galactosidase reporter strains, the natural conditions required for gene activation and product formation were identified by Gressler et al., revealing a crosstalk of primary metabolism through regulators such as CreA, PacC, and AreA with secondary metabolite production. The products are isoflavipucine and dihydroisoflavipucine, and isotope labeling experiments unveiled an unusual heterocycle rearrangement during metabolite biosynthesis.

Myricetin Blocks DnaJ Regulation of DnaK

PAGE 210

DnaK is a molecular chaperone important for maintaining cellular proteostasis. The intrinsically slow ATPase activity of DnaK is stimulated by its cochaperones, DnaJ and GrpE. To identify inhibitors of the DnaK-DnaJ combination, Chang et al. screened plant-derived extracts against the reconstituted complex. These efforts led to the identification of flavonoids, including myricetin. Interestingly, Chang et al. found that myricetin bound DnaK and prevented stimulation of ATP turnover by DnaJ, but that it had no effect on DnaK alone or GrpE-DnaK. Based on results from NMR, computational simulations, and mutagenesis, myricetin appears to allosterically interrupt the interaction between DnaJ and DnaK. Together, these results highlight the potential utility of a "gray box" screening approach for targeting protein-protein interactions in complexes.



Halting the Glucose Transport

PAGE 222

The Warburg effect, in which glycolysis remains active even under aerobic conditions, is considered a key driver for cancer cell proliferation, malignancy, metastasis, and therapeutic resistance. Currently, there is a limited arsenal of small molecules that can target aerobic glycolysis. Ulanovskaya et al. developed a new approach for rapid discovery of glycolytic inhibitors from structurally diverse chemical libraries and successfully identified two chemical inhibitors of the GLUT family of facilitative transmembrane transporters. Such compounds would be useful not only to study the system-level organization of energy metabolism, but could also facilitate development of drugs targeting upregulation of aerobic glycolysis in cancer.

Correcting Trafficking Defect of F508del-CFTR by a Pharmacological Chaperone

PAGE 231

The most common mutation causing cystic fibrosis (CF), F508del-CFTR, is retained in the endoplasmic reticulum and degraded rather than being trafficked to the cell surface, where it functions as a chloride channel. One therapeutic approach is to identify small molecule pharmacological chaperones that bind directly to the F508del-CFTR protein and correct the trafficking defect. Sampson et al. use differential scanning fluorimetry to identify a novel pharmacological chaperone, RDR1, which binds to the first nucleotide binding domain (NBD1) of F508del-CFTR. RDR1 partially corrects F508del-CFTR trafficking in multiple cell models and in a mouse model of CF.

Sulfonylureas Are Isoform Selective Activators of Epac2

PAGE 243

Sulfonylureas (SUs), a common antidiabetes treatment, were recently identified as activators of exchange protein directly activated by cAMP 2 (Epac2). Here, Herbst et al. investigate the underlying mechanisms of this molecular action and identify a key residue, R447, to be critically involved in SU binding. SUs are further identified as isoform-selective activators of Epac2, as they do not activate the closely related Epac1. As Epac proteins emerge as key regulators of numerous cAMP-mediated signaling processes and as new therapeutic targets, these findings should provide a basis for the development of more effective therapeutic agents and enhance our fundamental understanding of Epac signaling.

TGF-β Signaling Inhibitors via Profiling in Whole Organisms

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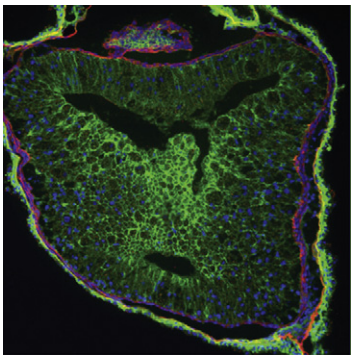

Identifying small molecules with predictable in vivo efficacy remains a major challenge in drug discovery. Dush et al. developed a novel phenotypic screen in frog embryos that revealed pyridine derivatives with highly desirable in vivo antitumor properties. Using a combination of independent developmental phenotypes, immunohistochemical analyses, embryonic gene expression patterns, and biochemical assays, the pyridine compounds were identified as a new class of TGF-β signaling inhibitors. This work illustrates that multiphenotype profiling in whole organisms is a powerful strategy for identifying bioactive small molecules and identifying the pathway-level mechanism of action of novel compounds.

Inhibitors of the Uncoupling Proteins

PAGE 264

The uncoupling proteins (UCPs) are mitochondrial transporters that provide the cell with an energy dissipating mechanism used, for example, to maintain body temperature. The UCPs also participate in the defense against oxidative stress and, thus, UCP2 plays a protective role in tumor cells. Rial et al. have identified a series of chromane derivatives that inhibit the UCPs and demonstrated that inhibition of UCP2 in HT-29 human carcinoma cells causes oxidative stress. The chromanes can act synergistically with chemotherapeutic agents increasing the efficacy of drugs such as arsenic trioxide. These findings open a promising line in the development of novel anticancer agents.

