In This Issue

Ligands In Situ via HiSBE Synthesis

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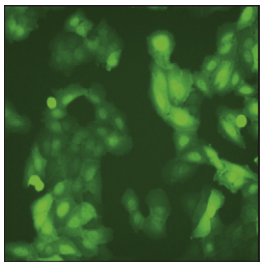
Many molecules that could manipulate cellular function are not practical due to their large size and concomitant undesirable pharmocokinetic properties. Here Shin et al. describe a bioorthogonal reaction, highly stable boronate ester (HiSBE) synthesis, and use this reaction to assemble a biologically active molecule from smaller precursors in a physiological context. The rapid rate of HiSBE synthesis suggests that it may be useful for assembling a wide variety of biologically active molecules in physiological solutions.

Riluzole Enhances Wnt/β-catenin Signaling in Melanoma



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Wnt/β-catenin signaling is a major regulator of melanocyte differentiation. In melanoma, elevated β-catenin signaling has been correlated with favorable patient outcome and shown to promote a transcriptional profile consistent with differentiation toward a less proliferative, melanocyte-like state. Now, Biechele et al. show that riluzole, an FDA-approved drug recently shown to be efficacious in metastatic melanoma, enhances Wnt/β-catenin signaling and examine effects of Wnt/β-catenin signaling on melanoma cells. The results of this study, combined with prior data that β-catenin signaling has complex effects in different cancers, should stimulate further investigation into the use of riluzole for melanoma as well as other cancer therapies.

Inhibition of Huntingtin by a miRNA-like RNAi Mechanism

PAGE 1183

Inhibiting expression of huntingtin (HTT) protein is a promising strategy for treating Huntington's Disease (HD), but indiscriminant inhibition of both wild-type and mutant alleles may lead to toxicity. An ideal silencing agent would block expression of mutant HTT while leaving expression of wild-type HTT intact. Hu et al. now explore switching the RNAi mechanism towards that used by miRNAs by introducing mismatched bases, which leads to potent and highly selective inhibition of mutant HTT expression in patient-derived cells. Potent allele-selective inhibition of HTT by mismatched RNAs provides a new option for developing HD therapeutics.

Pan-Caspase Inhibitors in Huntington's Disease Models

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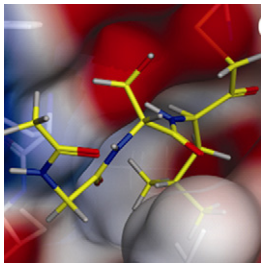
Huntington's Disease (HD) is characterized by a mutation in the huntingtin gene encoding an expansion of glutamine repeats on the N terminus of the huntingtin (Htt) protein. Numerous studies have established cleavage of Htt as a critical pathological event, and caspases have been identified as enzymes that carry out this cleavage. Leyva et al. report the development of three pan-caspase inhibitors that block cleavage of Htt. In HD models, these inhibitors rescued striatal and cortical neurons from cell death. These results confirm the role of caspase-mediated Htt cleavage in HD and further implicate caspases as promising targets for HD therapeutic development.

Blocking *C. difficile* Toxin Production

PAGE 1201



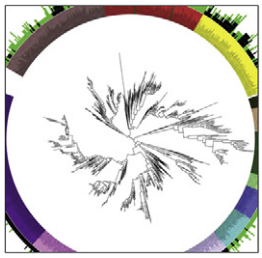
The large glucosylating toxin TcdB is a primary virulence factor of the antibiotic-resistant pathogenic bacterium *Clostridium difficile*. Here, Puri et al. screen a focused library of covalent small molecules to identify potent inhibitors of the toxin cysteine protease domain (CPD), which autoproteolytically activates the toxin. The authors discover inhibitors potent enough to block toxin function in cell culture and convert these inhibitors into probes that can monitor toxin allosteric activation. By solving the crystal structure of inhibitor-bound TcdB CPD, this study provides a framework for developing antivirulence therapeutics that target *C. difficile* infection.

Bioorthogonal Chemical Proteomics Playing Tags

PAGE 1212

Development of bioorthogonal ligation methods has afforded new opportunities to explore small molecule protein interactions using specific probes of enzymes or reporters of protein modifications. The identification of the proteins that are targeted by small molecule probes/reporters is essential for these functional studies. Yang et al. present here the synthesis and characterization of selectively cleavable affinity tags for versatile and robust bioorthogonal chemical proteomic studies. The azide/alkyne-functionalized, ortho-hydroxylated-diazobenzene-cleavable affinity tags enabled selective enrichment and efficient elution of alkyne- and azide-labeled proteins/peptides from complex mixtures and allowed protein identification and modification-site mapping using diverse proteomic platforms.

Broad Spectrum Antibiotic Activity Unmasked



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While natural products have proven to be the most robust source of antibiotics, those with broad spectrum have become increasingly difficult to identify, and the narrow spectrum of others is typically assumed to be an intrinsic limitation of their scaffold or target. In contrast, Smith et al. demonstrate that the apparently narrow spectrum of the arylomycin natural products results not from intrinsic limitations, but from target mutations. The arylomycins have a much broader spectrum of activity than previously thought, with an even broader spectrum if optimized to bind their target with higher affinity.

Engineering Complete Erythromycin A Biosynthesis

PAGE 1232

The biosynthetic pathway for the complex antibiotic erythromycin A has been reconstituted through *E. coli* by Zhang et al. To do so, the authors designed 23 foreign genes for coordinated expression within an *E. coli* strain engineered to support biosynthesis. As a result, the most clinically relevant form of erythromycin is now available through a production platform that offers multiple next-generation engineering opportunities. As an early step toward this potential, the current system was reconfigured to produce two new erythromycin analogs. Future research will continue to mine this biosynthetic pathway for native and altered forms of erythromycin to be applied toward established and emerging pathogenic microorganisms.

Affinity Patterns of Small Molecule Kinase Inhibitors

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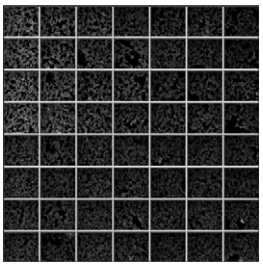
Interactions between kinases and small molecule inhibitors can be activation state dependent. Wodicka et al. have systematically explored the effects of ABL1 activation loop phosphorylation and PDGFR family autoinhibitory juxtamembrane domain docking on inhibitor binding affinity. For a diverse compound set, the binding affinity patterns correctly classify inhibitors as having type I and type II binding modes, and the authors show that juxtamembrane domain docking can have dramatic negative effects on inhibitor affinity. The results have allowed authors to associate ligand-induced conformational changes observed in cocrystal structures with specific energetic costs. The approach described should facilitate indication-specific kinase inhibitor design.

O-GlcNAcase Inhibitors with Selective Tastes

PAGE 1250



O-GlcNAc is a posttranslational modification that regulates several cellular processes in metazoa. The work by Dorfmueller et al. describes the design of chemical tools that inhibit *O*-GlcNAcase in live cells and can be used to study the role of *O*-GlcNAc in signal transduction pathways. The inhibitors, GlcNAc-statin derivatives, are nanomolar inhibitors of the enzyme and 9×10^5-fold selective over the structurally related lysozomal hexosaminidases. These small molecules are shown to be cell-penetrant and raise cellular *O*-GlcNAc levels when applied at low nanomolar concentrations and are thus valuable tools for studying the role of *O*-GlcNAc in a range of cellular processes.

Normalizing Cardiovascular Function in Hypertension through FAAH Inhibition

PAGE 1256

Godlewski et al. developed a compound, AM3506, which normalizes the elevated blood pressure and cardiac function in hypertensive rats. These effects are due to blockade of fatty-acid amide hydrolase (FAAH): an enzyme that catalyzes the degradation of endocannabinoid anandamide and controls its action at cannabinoid receptors in brain and liver. AM3506 is able to block FAAH activity in the brain without affecting the liver enzyme and, therefore, is devoid of unwanted metabolic effects observed in obesity, e.g., increased blood glucose level (hyperglycemia). This unique activity profile of AM356 makes it attractive for potential therapeutic use as an antihypertensive in obese subjects.

