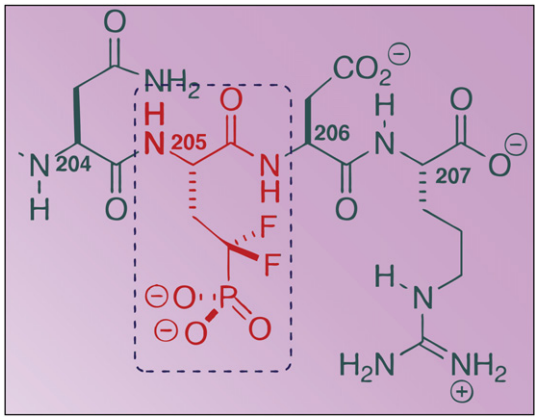
# In This Issue

## Tick-Tock Goes the Clock

PAGE 921

Most of us get up every morning and go to bed each night, thus succumbing to the beckoning of our internal clock mechanism. Such a clock, regulating daily physiology of the majority of organisms on the planet, is called a circadian clock, and the molecular core of its mechanism is made of a network of transcription factors that generate circadian rhythms of gene expression. Posttranslational modifications, such as phosphorylation of clock proteins, provide another layer of clock regulation. Hirota and Kay now provide discussion of a circadian clock and elaborate on the recent chemical biology-driven efforts to extend our understanding of its mechanism.

## In Review: Mimicking Phosphoserine



PAGE 928

Protein kinases are enzymes that catalyze transfer of a phosphate group from ATP to a side-chain hydroxyl group of a specific serine, threonine, or tyrosine residue in a target protein. The review by Panigrahi et al. now focuses on the ($\alpha$,$\alpha$-difluoromethylene)phosphonate mimic of phosphoserine ($pCF_2Ser$) and its application to the study of kinase-mediated signal transduction. The review describes both the synthetic chemistry schemes that lead to production of $pCF_2Ser$ phosphoserine mimic and the means to incorporate this mimic into the protein of interest. Using tumor suppressor p53, DNA damage response signaling cascade, and AANAT protein stabilization as examples, the authors illustrate the type of information that can be accessed by $pCF_2Ser$-based strategy.

## Poisoned by Mercury, Recovery on "Budgeted" Release

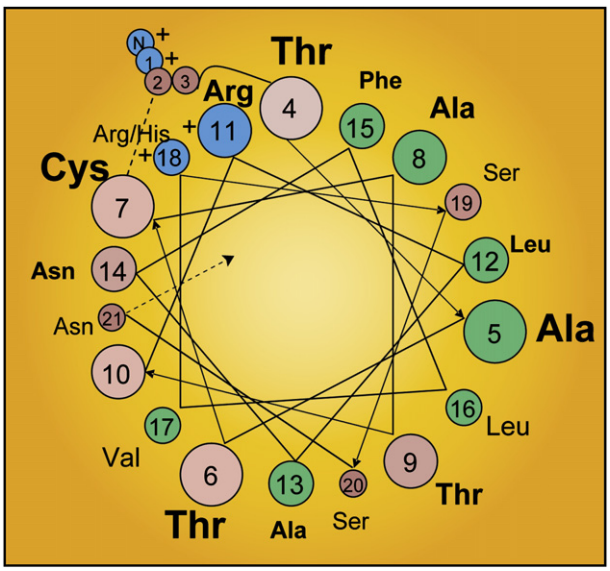
PAGE 937

Chelation therapy is a strategy used to treat mercury poisoning, and a number of different chelation agents have been previously developed. However, for the majority of chelators, inappropriate dosages can increase toxicity and, to date, no single platform can be used to detect and adjust the dosage of chelators simultaneously. Yigit et al. now describe a proof-of-concept study of a multiple-use, liposome-based system that can detect inorganic mercury and release mercury chelators only if the mercury concentration is above a certain limit. This "budgeted" release profile will be particularly useful in situations where the local levels of Hg contamination vary in a location- and time-dependent manner.

## $\alpha$-Helical Mimetic Prevents Amyloid Fibril Formation

PAGE 943

Amyloid formation is a process by which a normally soluble protein misfolds and polymerizes into a $\beta$-sheet-rich fiber. This process contributes to the pathology in a number of diseases such as Alzheimer's, Parkinson's, and diabetes. Here, Hebda et al. use a newly developed small molecule $\alpha$-mimetic compound to target the precursor protein structures in type II diabetes. Remarkably, this $\alpha$-helical mimetic interferes with the $\beta$-sheet fiber formation and ameliorates cytotoxicity in insulin-secreting cells. The work establishes that $\alpha$-helical, membrane-bound states are central to diabetic pathology and identifies a structure-specific target for the development of small molecule therapies. (Figure adapted from Hebda et al.)




## Reporting on Butyrolactones in *Streptomyces*

PAGE 951

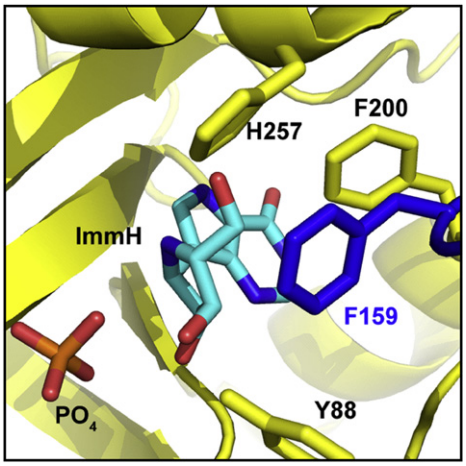
In this report, Hsiao et al. characterize two $\gamma$-butyrolactones, bacterial hormones that stimulate antibiotic production in *Streptomyces coelicolor*. In addition, the authors identify the specificity determinants of these compounds for the $\gamma$-butyrolactone receptor using a small signaling molecule reporter system they developed. Understanding the ligand specificity determinants will allow improved targeted design of new compounds to inhibit or promote antibiotic production. The described reporter system displays superior sensitivity and can be applied to detect $\gamma$-butyrolactones, which are only produced in small quantities and thus difficult to detect. The improved sensitivity was critical for identification of two $\gamma$-butyrolactone producers among the commercial antibiotic-producing *Streptomyces* reported here.

## LasR Quorum Sensing at Atomic Level

PAGE 961

*Pseudomonas aeruginosa* is an opportunistic pathogen that has proven recalcitrant to traditional antibiotics. *P. aeruginosa* utilizes quorum sensing as a strategy for intercellular communication, and disruption of this signaling pathway is shown to attenuate virulence. Zou and Nair now describe crystal structures of the LasR quorum-sensing receptor bound to its cognate autoinducer and to a class of structurally distinct compounds that can interact specifically and potently with the receptor. These structural data provide the molecular bases for understanding how chemically distinct compounds can be accommodated by a highly selective receptor and establish a framework for further development of quorum-sensing modulators.

## Ribocation Transition State: Leaky Active Site View




PAGE 971

The reactivity of enzymatic transition states is one of the most difficult and unexplored subjects in the chemical biology of catalysis. Ghanem et al. now test the reactivity of the ribocation transition state for purine nucleoside phosphorylase with water and other nucleophiles. Additionally, the authors prepared a set of glycine mutants to introduce catalytic-site solvent leaks. Even with the chemical phosphorolysis reaction occurring millions of times, no hydrolysis was observed. The results support a transition state lifetime shorter than the timescale required for water diffusion. Surprisingly, an unprecedented N9-to-N3 isomerization was observed with two of the mutants. Relaxed catalytic-site geometry permits misalignment after transition state formation. (Figure adapted from Ghanem et al.)

## HDAC3 in Friedreich's Ataxia Gene Silencing

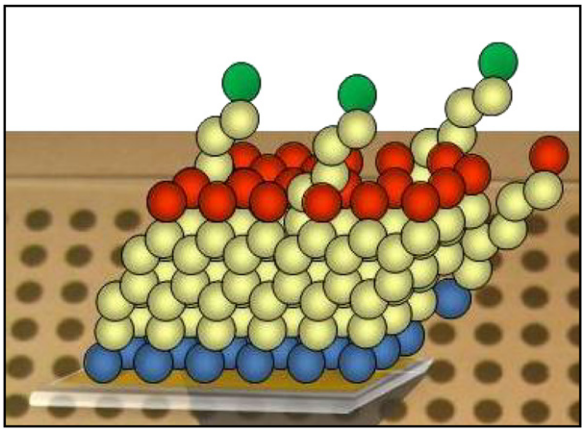
PAGE 980

Numerous studies have pointed to histone deacetylase (HDAC) inhibitors as potential therapeutics for various neurodegenerative diseases. In the course of studies on Friedreich's ataxia (FRDA), the authors identified a novel class of HDAC inhibitors (pimelic diphenylamides) that reverse heterochromatin-mediated silencing of the *frataxin* gene in this disease. In the present study, Xu et al. identify HDAC3 as the likely cellular target of the pimelic diphenylamides and provide evidence that other potent HDAC inhibitors that target different HDAC enzymes are without effect on *frataxin* gene expression in cells derived from FRDA patients. The results suggest that HDAC3 is a target for therapeutic intervention in FRDA.

## Relaxed Specificity of αIIbβ3 Integrin

PAGE 990

Fibrinogen mediates platelet aggregation and adhesion by binding to the platelet integrin αIIbβ3. Despite significant study, the sites on Fibrinogen that bind the receptor are still unresolved. Here, Sánchez-Cortés and Mrksich report the use of self-assembled monolayers to identify and characterize peptide motifs that mediate cell adhesion. This work reveals that αIIbβ3 has a relaxed specificity for its ligands and recognizes peptides of the form XGD where X is either a basic or hydrophobic residue. (Figure credit: Sánchez-Cortés and Mrksich)



## Probing Activity of Proteases in Cytosol of Apoptotic Cells

PAGE 1001

Lysosomal proteases were not normally thought of as participating in a programmed cell death termed apoptosis; however, cathepsin proteases can be released from the lysosome and then participate in cell death. Pratt et al. report here the development of an activity-based probe that reports on cathepsin B activity only in apoptotic cells by reading out its release from the lysosomes. This provides the first direct method for visualizing this release in intact cells. The probe and other biochemical data further support roles for cathepsin B in the cytosol during cell death and provide a chemical tool for future investigations.

## Today Sequential Steps, Tomorrow Enzyme-Based Device

PAGE 1013

Retaining the activities of surface-immobilized enzymes is a major challenge in the development of hybrid organic-inorganic devices. To address this issue in relation to enzymatic energy production, Mukai et al. used a biomimetic strategy, copying a design found in the flagellum of mammalian sperm, in which the enzymes of glycolysis are organized on a cytoskeletal element. The authors produced recombinant forms of hexokinase and glucose-6-phosphate isomerase capable of oriented immobilization on a nickel-nitrilotriacetic acid surface and demonstrated their activities in series when immobilized on the same surface. This is the initial report of sequential steps in a biological pathway being tethered to a single solid support and should serve as a launching pad for future studies.