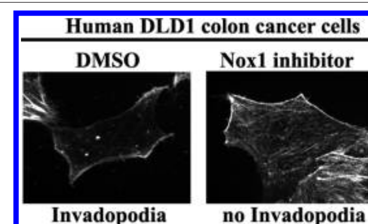
In this ISSUE

Knocking Out Nox1

Reactive oxygen species (ROS) are intriguing biomolecules that have been implicated in numerous physiological and pathophysiological processes. ROS are generated by the NADPH oxidase family, which includes enzymes Nox1−4 that are structurally similar but have different regulatory mechanisms and tissue distribution. Lack of selective inhibitors of these enzymes has hampered elucidation of their specific roles in ROS production and function. Now, Gianni *et al.* (DOI: 10.1021/cb100219n) report the identification of a novel small molecule Nox1 inhibitor and its potential anticancer activity.

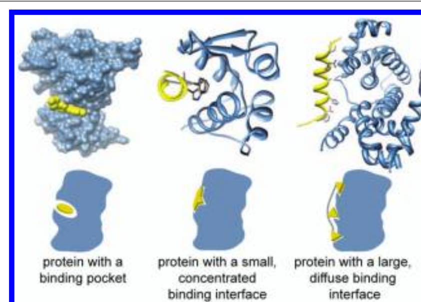
Using a cell-based high-throughput screen, 16,000 compounds were screened for their ability to inhibit ROS production in a human colon cancer cell line, which expresses only Nox-1. A phenothiazine referred to as ML171 was identified as a selective, nanomolar inhibitor of Nox-1. Moreover, ML171 prevented formation of invadopodia, cellular structures that promote the invasive character of cancer cells. ML171 and related compounds are thus valuable tools for exploration of Nox1 function and offer an exciting jumping off point for development of novel anticancer agents.



Computing the Hot Spots

Protein−protein interactions (PPIs) are tantalizing drug targets. They play key roles in numerous diseases but are notoriously difficult to disrupt with small molecules due to the large surface areas they typically span. Nevertheless, increasing numbers of small molecule PPI inhibitors have been discovered that are capable of mimicking certain "hot spot" residues (those that contribute most energetically to the interaction). Building from these examples, Jochim and Arora (DOI: 10.1021/cb1001747) develop a computational approach for determining which PPIs are likely amenable to disruption by small molecules.

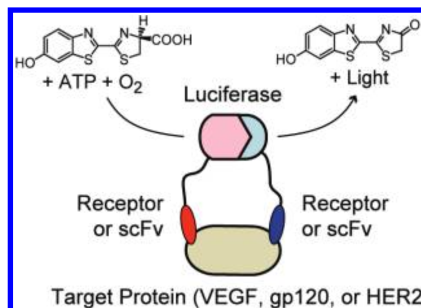
For this initial investigation, the Protein Data Bank was surveyed for all PPIs involving an α-helical interface, as α-helices are known to mediate many important PPIs. Computational alanine scanning was then used to determine the "hot spot" residues. On the basis of this analysis, the interfaces were divided into three categories according to the nature of the interactions: interfaces with binding clefts, extended interfaces, and weakly interacting interfaces. Further analysis enabled general predictions of which PPIs were the best candidates for small molecule inhibition.



Splitting Luciferase for Protein Detection

Methods to detect native proteins are important for numerous applications, including the accurate diagnosis of and development of treatment strategies for a variety of diseases. Traditional approaches, such as enzyme-linked immunosorbent assays or protein labeling strategies, require numerous steps that can increase the time, effort, and errors involved. Stains *et al.* (DOI: 10.1021/cb100143m) now report the use of a split-protein assay as a general approach for directly detecting native proteins.

The approach relies on the restoration of luciferase activity in the presence of the protein of interest. Specifically, luciferase is split in two halves, each of which is fused to a receptor or antibody that interacts with the protein. Protein binding then causes the luciferase fragments to come together, restoring activity and providing a sensitive, detectable signal. The utility of this innovative, cell-free approach was demonstrated with three clinically relevant proteins: vascular endothelial growth factor, the gp120 coat protein from HIV-1, and the human epidermal growth factor receptor 2. Detection of purified proteins as well as proteins in their cellular environments was demonstrated.



Published online October 15, 2010 • 10.1021/cb100289v
© 2010 American Chemical Society