

## E0056

**Drug Discovery at Adaptive Signaling Interfaces: Moving Targets.** James A. Wells Sunesis Pharmaceuticals, 341 Oyster Point Blvd., S. San Francisco, CA 94080.

Small molecule drug discovery at signaling interfaces including protein:protein interactions and enzymes is often challenged to identify novel and selective chemotypes that will interact with them. In many cases the binding surfaces are highly adaptive which complicates structure-based methods for discovery. Selectivity can be an issue for many enzymes, such as kinases, which share a promiscuous substrate like ATP. In other cases the highly charged nature of the substrate make it challenging for finding drug-like chemotypes such as for some proteases and phosphatases. We have developed a fragment-based approach to drug discovery, called Tethering®. This approach allows us to find weak drug-like fragments (MW~200 Da) that can nucleate the drug discovery process to targets for which has been traditionally difficult to get hits and advance them by medicinal chemistry. A native or engineered thiol in a protein is allowed to react reversibly under thiol exchange conditions with a small library of disulfide-containing small molecules at concentrations that are typical for drug screening. The thiol-captured ligands, which are identified by mass spectroscopy, represent the most stable complexes even though in the absence of the covalent tether the most stable ligand may bind very weakly ( $K_d \sim 0.1$  to 2 mM). The method provides binding stoichiometry and site location for the tethered compounds, data that are not immediately available by HTS. Moreover, the site-directed character of the approach can focus the discovery process on unactivated enzymes, such as kinases, and allosteric sites which would otherwise be difficult to selectively target. The application of this technology toward discovery of small molecules interfaces will be discussed.