**Equipment**

**Arkin Lab/Small Molecule Discovery Center**

Dr. Arkin’s laboratory contains modern robotic instrumentation for screening and up-to-date equipment for biochemistry and cell biology research. Her laboratory shares routine biochemical and cell culture equipment with the adjacent laboratory of Dr. Jim Wells. Equipment includes 2 biosafety cabinets and 4 incubators for mammalian tissue culture; 4 bacterial culture incubators, 2 insect cell culture incubators, and a Beckman Coulter J-26 XP centrifuge for protein expression; 3 Akta FPLC instruments, gel electrophoresis systems, AlphaInnotek imager, a Nanodrop spectrophotometer; a Flexstation III injection-capable, multimodal plate reader (Molecular Devices); an EL406 washer/dispenser (Biotek).

The laboratory is fully capable of running and evaluating protein/compound interactions by mass spectrometry (Waters UPLC/XevoGS) and high-throughput SPR (Biacore 4000). The lab has access to crystallography equipment in Genentech Hall, including a Mosquito for setting up crystal screens and x-ray generators. We also regularly use the Stanford Synchrotron Radiation Lightsource and the Lawrence Berkeley National Laboratory Advanced Light Source (ALS).

This project will make use of the automation laboratory at the SMDC. The HTS lab has a fully automated system that utilizes an F5 six-axis arm and Momentum scheduling software (Thermo). Liquid handling is accomplished with a Biomek FXp (Beckman Coulter) equipped with dual (96/384-well and Span8) pipetting arms and pin tool (VP Scientific) for nanoliter volume transfers (10, 50, 200 nL). Additional liquid handling and plate washing is provided by an EL406 and a Multiflow bulk dispenser (Biotek). Plate hotels include a Cytomat MPH, Cytomat 2C incubated hotel and Liconix 104-plate incubated hotel. Plates are read in an Envision plate reader (Perkin Elmer) or in an InCell Analyzer 2000 high-content imager (GE Healthcare), which includes objective lenses from 4x – 40x, 8 filter sets for fluorescence, bright-field modes, and automated data analysis software. Data are stored and analyzed in the SMDC’s MYSQL database and visualized with a custom-built web interface called HiTS.

The SMDC houses a ~200,000-compound small-molecule screening library. This compound collection contains a 177,700-compound diversity library compiled from three commercial sources (Chembridge, ChemDiv, Specs), 24,000-compounds targeted towards kinases, proteases, and RNA (proprietary sources), and 4000 drugs and “bioactive” compounds (SelleckChem, Microsource, and proprietary sources) with known mechanisms of action. Libraries conform to the current standards for lead-likeness. For the diversity collection, molecules were selected to have at least four lead-like properties (MW < 350 Da; cLogP < 5; H-bond donors < 5; H-bond acceptors < 8; rotatable bonds < 8; polar surface area < 100A2). Compounds were also filtered to remove reactive functionalities (e.g. Michael acceptors and thiols) and known toxicophores (e.g. nitroxides), and “promiscuous” scaffolds have been flagged [e.g. 6305 compounds contain chemotypes flagged as Pan Assay Interference (PAIN) compounds (Baell, 2010)]. More than 100 screens have been run with most of the compounds in this collection, providing a rich data set for selecting against toxic compounds, frequent hitters, and reporter-based artifacts; HiTS also provides a link for compounds found in ZINC and PubChem/Chembl, thereby allowing us access data for hundreds of additional screens. The fragment-screening libraries include 1600 disulfide fragments synthesized at the SMDC and a 3000-member fragment library optimized for surface plasmon resonance (SPR). The tethering and SPR libraries were selected for diversity, rule-of-three compliance, and three-dimensionality. The screening libraries are stored in an ample number of -40oC Revco freezers located in the SMDC. All of SMDC’s libraries and equipment will be available for this project, as needed.

**Renslo Laboratory/Small Molecule Discovery Center**

For synthetic chemistry, analytical and chromatographic instrumentation located in the SMDC includes a Waters/Micromass Alliance LC/MS system, a Biotage SP1 high performance flash chromatography (HPFC) system, and a Waters AutoPurification semi-preparative HPLC system. Parallel and rapid-serial synthesis is supported by a CEM Explorer microwave reactor with 24-sample autoloader and a ThalesNano H-cube flow-hydrogentation apparatus. For multiplexed evaporation of reaction mixtures or HPLC fractions we utilize a Genevac EZ-2 evaporator and a Virtis 4K series lyophilizer. For NMR spectra determination, SMDC chemists have access to a Varian 400 MHz NMR spectrometer located in neighboring Genentech Hall in addition to a Bruker 300 MHz instrument resident in the SMDC chemistry laboratory. Other relevant equipment resident in the SMDC includes the pION parallel artificial membrane permeability assay (PAMPA) system, which includes pION software and plate stirrer ("gut-box"). This equipment can be used along with and Analyst multi-modal plate reader (Molecular Devices) to make determination of passive permeability.

**UCSF Data Management**

For storage and manipulation drug-discovery data, the SMDC employs a custom-designed MySQL database running on a high-performance Linux server (8 CPU, 32 Gb RAM, 4-146 Gb storage). Accelrys Pipeline Pilot and/or Dotmatics Vortex software is used to manage and analyze these data. A tailor-made web application, HiTS, integrates these tools and will provide all team members regardless of their research site a web-based user-interface to view and query chemical structures and biological results. In addition, the SMDC has a data management program that has been operational for the past several years using the Collaborative Drug Discovery (CDD) database (Collaborative Drug Discovery Inc., South San Francisco, CA) which is web-based, password protected, and can be used by all team members to mine and share data.

**Core facilities**

We have access to well-equipped core facilities throughout the UCSF Mission Bay, Parnassus, and Mt Zion campuses. Facilities that may be relevant to this project include DNA sequencing, microarray, confocal microscopy, mouse blastocyst injection service, flow cytometry, mass spectrometry, and the preclinical therapeutics core, located in the Helen Diller Comprehensive Cancer Center.