## Instructions for Requesting Small Molecule Cherry Picks at ICCB-L

All cherry-pick requests must be submitted to Dave Wrobel (david\_wrobel@hms.harvard.edu).

- You must submit the following information to Dave **before** making a request:
  - 1. Annotated primary screening data template(s). Please see <u>Step 4</u> of the Data Deposition Guidelines.
  - 2. Completed Primary Screen Report, which will include a summary of the screening protocol, controls used during the screen, a description of the analysis method, and criteria for scoring screening positives.
- You are allowed to request cherry picks for up to 0.3% of the total compounds screened. For example, if you screened 45,000 compounds in duplicate, you can request up to 135 (0.3%) cherry picks. You do not have to wait until the primary screen is complete to request cherry picks, but you must submit annotated data files for all compounds screened up to that point to make a request. Additional cherry picks at a volume of 1.5 uL/compound can be requested for a fee, requires written scientific justification and is subject to compound availability. If you would like to request cherry picks beyond the 0.3% of total compounds screened, please contact Jennifer Smith (jennifer smith@hms.harvard.edu).

Please note that for libraries with compounds plated at multiple concentrations (Prestwick 3, Selleck Bioactive, Selleck Neuronal, SYNthesis Kinase Inhibitor 2, LINCS 1 Kinase Inhibitors, LINCS 2 Kinase Inhibitors, LINCS 3 Chromatin Targeting, LINCS4 Kinase Inhibitors, Mechanism of Action), cherry picks are provided at the highest concentration, typically 10 mM. This is true even if a lower concentration well scored positive in your screen. For any of these compounds, please request the well at the highest concentration; you can find these wells by clicking on "Other Wells With Reagent" in the Well View for each compound in Screensaver. The cherry picked compounds can easily be tested at lower concentrations when using the HP D300.

- The standard format for cherry pick plates is to have the outer two columns (1, 2, 23, 24) and rows (A, B, O, P) empty. When using the HP D300, some screeners find it helpful to only fill every other column and have each plate contain the number of compounds that will be tested during one session on the HP D300 (e.g. 75 compounds). If you would like additional columns or wells to be left empty or a maximum number of compounds on each plate, please specify this to Dave at the time you request your cherry picks.
- Once your request has been submitted to Dave with all of the above conditions and in the format described below, it will be forwarded to the screening room. The screening room staff will prepare a cherry-pick microplate for you with approximately **1.5 uL** of each of the requested compounds.
- The screening room will notify the researcher via email when their cherry-pick plate is ready. This will generally be within two weeks of receiving the cherry-pick request from Dave.

**Note**: Use your cherry pick samples carefully and DO NOT use your entire cherry pick sample in a single experiment. The screening group generally does not repeat cherry pick requests and commercial vendors may not be able to supply you with compound immediately upon request. The screening room staff can assist with resealing cherry pick plates after use.

We recommend that the cherry pick plates be kept in a desiccated environment at  $-20^{\circ}$ C and handled with caution since they are DMSO-dissolved compounds. Thaw and centrifuge the plate prior to opening to ensure all of the compound is at the bottom of the well.

## **Required Format for Cherry Pick Requests**

Screeners must make their own data file in Microsoft Excel in the following format:

Example: cherry pick request file for screen # 101

Screen 101 6 cherry-picks

2,000 total compounds screened

Plate	Well	Positive	Cherry Pick	Comment, required if positive is not chosen for cherry picking.
1518	C21	М	С	
1519	D19	М		Also hit in other screens
1520	F05	W	С	
1520	M22	S	С	
1521	A11	М	С	
1522	P15	S	С	
1548	J22	М		Edge effects
1214	K10	S	С	
1226	N18	W		Very weak

Cherry pick request files should include all positives annotated as a Y, W, M, or S. The columns should be **Plate**, **Well**, **Positive**, **Cherry Pick**, and **Comment**. The file must contain the **Screen Number** for which the cherry picks are being requested. The file must also contain the **number of cherry picks** being requested and the **total number of compounds screened** to ensure compliance with the "0.3% Rule". Place a C in the Cherry Pick column for those wells that you decide to cherry pick. Enter a comment for all positives that you decide not to cherry pick.

• The Plate and Well columns are listed in ascending order, first by Plate, then by Well.

## **Important Contact Information:**

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