Kuhlman Lab Protocol for Storing Plasmids

Introduction | Kuhlman Lab members have plasmids that fall into three categories:

1. Plasmids significant enough to be mentioned in a paper. These must be made available via AddGene (see instructions below), the company that efficiently curates and distributes plasmids.

2. Plasmids significant enough to be requested by other lab members or friends of the lab. These, as well as all plasmids sent to Addgene, must be entered in Quartzy and saved in our plasmid inventory racks in our -20 °C freezer.

3. Plasmids which are so specific or unremarkable as to be only useful to the lab member who created them. These need not be saved in the lab-wide inventory.

Addgene Instructions | Addgene has its own set of instructions for submitting plasmids on their website here.

Again, Kuhlman Lab members must add plasmids to Addgene that are cited in our publications so that interested readers can request them from Addgene. In addition, we can add pre-publication or unpublished plasmids, however this costs time and money.

Addgene will ask for a name, type, and description of the plasmids, as well as other plasmid information including:

- Sequences, maps, and files
- Gene and insert
- Cloning information
- Growth and distribution information

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Lab-Wide Plasmid Inventory: Creating Quartzy Entries | For those plasmids in the first two categories above, Kuhlman Lab members must save an aliquot in our Kuhlman Lab inventory on a yearly basis. Experience has shown that it is important not to wait until preparing to leave the lab. Lab members can, of course, handle all of this work themselves, including initiating Quartzy entries and making tube labels. However, the lab manager and work-study students are willing to help with these tasks.

Lab members who would like help must fill out an Excel sheet for work-study students to use to begin Quartzy entries for their plasmids. On this Excel sheet, Kuhlman Lab members must provide information for the following fields in Quartzy:

- Plasmid name
- Backbone
- Insert
- Date made
- Concentration in ng/ul
- Tags
- Restriction sites used
- Source (Made by someone in the lab? Gift of collaborator? Ordered from a vendor?)

All of this information will be entered into Quartzy by a work-study student, at which point a Quartzy number will be assigned to each plasmid for unique identification. It then becomes the responsibility of Kuhlman Lab members to proofread and enhance Quartzy entries with additional information, including but not limited to:

- Gene Product
- Primer Sequences
- Size of Insert
- Solvent
- Use
- A vector map

For more information about Quartzy, please see “How lab members and work-study students navigate and edit the online Quartzy database”
Lab-Wide Plasmid Inventory: Making Tube Labels | In addition, a work-study student will prepare labeled tubes with our label maker. It is crucial that label-maker made tubes are used for the following reasons:

1) Unlike labeling with Sharpies, the labels last a long time at low temperatures, and
2) More legible information can be included on the tube, making the physical inventory much more accessible.

Lab-Wide Plasmid Inventory: Aliquoting into Tubes | Finally, the Kuhlman Lab member, with a work-study student if desired, can add the plasmids to the inventory in the -20 °C freezer. A work-study student can transfer aliquots of the plasmids into labeled tubes if plasmids are organized according to the Quartzy number. However, since there is a great risk in this process of tubes getting mixed up, it is generally recommended that lab members aliquot their own plasmids. The tubes are then placed in the appropriate box in the appropriate rack in our -20 °C freezer.