

LISA IMB-internal services:

... what we need from you and what you get from us.

Last Update 15 March 2011

1. Inform client about GIPZ-shRNAmir that are available in the facility's collection.

Clients: Specify your genes of interest with their NCBI Entrez Gene ID, official symbol and biological species in an Excel sheet and submit it to LISA.

LISA: Extract and compile from our 125,000 shRNAmir random library all shRNAmir constructs that target your genes of interest. Provide a brief summary with average shRNAmir per gene, any genes that might be underrepresented, and a quote for obtaining this set as glycerol, DNA, and/or lentivirus.

2. Individual GIPZ-shRNAmir as 200 µl or 1 ml E. coli glycerol cultures; any number of constructs

Clients: Specify your genes of interest with their NCBI Entrez Gene ID, official symbol and biological species in an Excel sheet and submit it to LISA.

LISA: Cherry-pick GIPZ-shRNAmir *E. coli* glycerols from random 125,000 shRNAmir library. Provide inoculated small LB/carbenicillin culture to client.

3. Individual GIPZ-shRNAmir as 1 ml lentiviral bulk preparations; any number of constructs.

Clients: Specify your genes of interest with their NCBI Entrez Gene ID, official symbol and biological species in an Excel sheet and submit it to LISA.

LISA: Cherry-pick GIPZ-shRNAmir *E. coli* glycerols as above, then culture and isolate DNA. Transfect HEK293T producer cells with GIPZ-shRNAmir and helper plasmids in a single T-75 to produce GIPZ lentivirus. Harvest, filter, concentrate, and determine viral titre by its ability to re-infect HEK293T cells. Provide lentivirus to client.

4. Boutique shRNAmir libraries, microplates only

Clients: Specify your genes of interest with their NCBI Entrez Gene ID, official symbol and biological species in an Excel sheet and submit it to LISA.

LISA: Extract and compile from our 125,000 shRNAmir random library all shRNAmir constructs that target your genes of interest. Provide a brief summary with average shRNAmir per gene, any genes that might be underrepresented, and a quote for obtaining this set as glycerol, DNA, and/or lentivirus.

5. Glycerol copy of boutique shRNAmir library, microplates only

Clients: Confirm your order after reviewing the LISA GIPZ-shRNAmir datasheet.

LISA: Cherry-pick GIPZ-shRNAmir *E. coli* glycerols from random 125,000 shRNAmir library. Replicate and grow in fresh *E. coli* glycerol 96 well microplates. Provide one set of 96 well microplates (some 200 µl per well) to client and maintain a second copy within LISA as a backup.

6. DNA of boutique shRNAmir library, microplates only

Clients: Confirm your order after reviewing the LISA GIPZ-shRNAmir datasheet.

LISA: Cherry-pick GIPZ-shRNAmir *E. coli* glycerols as above and use to inoculate *E. coli* cultures in 1.8 ml deep well plates. Isolate DNA mini in a 96 well format. Determine and normalize DNA concentration. Provide DNA to client.

7. Lentivirus of boutique shRNAmir library, microplates only

Clients: Confirm your order after reviewing the LISA GIPZ-shRNAmir datasheet.

LISA: Cherry-pick GIPZ-shRNAmir *E. coli* glycerols and prepare DNA as above. Transfect HEK293T producer cells with GIPZ-shRNAmir and helper plasmids in a 96 well format to produce GIPZ lentivirus. Determine the average viral titre of two randomly selected columns (16 wells) by their ability to re-infect HEK293T cells. Provide lentivirus to client.