RNAi genetic screens provide an unbiased approach to better understand disease progression and gene-disease association, identify functionally important transcriptionally activated genes driving responses, characterize drug mechanisms of action, look for genetic sensitizers or blockers for compounds, and find novel drug targets. However, the complexity of high-throughput (HT) genome-wide screening mandates properly constructed libraries and well-designed screening assays to generate good results. Cellecta has developed effective, scalable, HT RNAi screening services using pooled lentiviral-based shRNA libraries. Cellecta will perform all steps required for a comprehensive RNAi screen with your cell models.

**All-Inclusive RNAi Screening Process**

1. Library transduction in your cell model (*in vitro* or *in vivo*)
   - You provide frozen cells to Cellecta
   - Choose to use a pre-made library or have us design a custom library for the target genes that interest you
2. Cell culture and selection, or screens with compounds, depending on the type of screen
3. Genomic DNA isolation
4. Sequencing of samples from various assay points
5. Data deconvolution

**Dropdown Viability Screens: Identify Novel Drug Targets or Determine Mechanism of Resistance**

- Transduce cells with lentiviral shRNA expression library for which the distribution of shRNA sequences has been characterized
- Grow cells over a period of 6-12 divisions
- Assay shRNA sequences again by HT sequencing of the bar codes in the remaining infected population
- Compare the number of each shRNA sequences remaining in the cultured cells with the original shRNA representation in the library
- Underrepresented shRNAs inhibit cell growth

**HT Sequencing and Analysis Services**

Cellecta shRNA and barcode libraries contain barcodes compatible with the Illumina HT Sequencing platform. For laboratories needing support for this analysis, without the need for a full RNAi screen, Cellecta can provide HT Sequencing and Barcode Enumeration of samples. You provide a frozen cell pellet after performing the RNAi screen, and Cellecta does the rest.
**Rescue Screens:**
To identify genes functionally required for cell sensitivity to a treatment

- Infect cells with the shRNA expression library
- Treat the cells with a factor that under normal conditions kills virtually 100% of the cells
- Then determine which shRNA are present in the surviving cells
- The genes targeted by the shRNA in the surviving cell population are essential for the killing response caused by the treatment

**Fluorescent Detection Screens:**
To identify elements regulating activation of a particular pathway or cell response

- Use for cellular responses not involving cell growth or cell death
- Detect responses with a fluorescent reporter system or fluorescent immunoassay and select using FACS
- HT sequencing on sorted cells to determine the genes targeted by the shRNA in the responding cells
- Compare to the original library, unselected cells, or cells undergoing different treatments

**In Vivo Dropout Screens:**
To identify genes essential for xenograft tumor proliferation

- Screen mice growing xenografted tumors
- Inquire for details

**Cellecta Can Validate Hits as an Extra Service**

- Custom cloning of individual shRNAs to targets in lentiviral vectors
- Use our novel Tet-ON shRNA vector for inducible studies
- Lentiviral packaging services available
- Functional assays such as proliferation assays available