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CRISPR-CAS9 knockout library for CHO

Lasse Ebdrup
DTU - Technical University of Denmark, aeb@biosustain.dtu.dk

Alex Thomas
University of California, San Diego

Hooman Hefzi
University of California, San Diego

Philipp Spahn
University of California, San Diego

Kaen Karottki
DTU - Technical University of Denmark

See next page for additional authors

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Authors
Lasse Ebdrup, Alex Thomas, Hooman Hefzi, Philipp Spahn, Kaen Karottki, Helene Kildegaard, and Nathan Lewis

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CRISPR-CAS9 KNOCKOUT LIBRARY FOR CHO

Lasse Ebdrup Pedersen, NNF CFB @ Technical University of Denmark
laeb@biosustain.dtu.dk
Alex Thomas, NNF CFB @ University of California, San Diego
Hooman Hefzi, NNF CFB @ University of California, San Diego
Philipp Spahn, NNF CFB @ University of California, San Diego
Karen Julie la C. Karottki, NNF CFB @ Technical University of Denmark
Helene F. Kildegaard, NNF CFB @ Technical University of Denmark
Nathan Lewis, NNF CFB @ University of California, San Diego

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Traditionally, screening of large CHO cell population have been utilized to identify clones with desired phenotypic properties such as product quality, e.g. specific glyco forms, and population characteristics, e.g. ability to grow in high cell densities.
This has largely depended on the genomic variety naturally present in a large cell population or occasionally utilizing random mutagenesis to increase this variety.
The ability to precisely create genomic variety in mammalian cells have improved dramatically over the past decade and in the past few years the price has dropped substantially due to the CRISPR/Cas9 technology. E.g. knocking out a gene using CRISPR/Cas9 is a simple, fast and cheap process (1).
However, rational identification of which genes to target can be quite difficult and the cellular processes underlying many desired traits are simply unknown or only poorly/partially understood.
To both improve our understanding of phenotypes of interest and identify targets to modify, we have created a lentiviral guideRNA library against CHO genes.
Using this guideRNA library we subject cells to various phenotype selection assays, harvest genomic DNA from the selected cells and perform targeted next generation sequencing to identify the guideRNA sequences which led to the improved phenotype.
As an example: Using a toxic fucose binding lectin, one can potentially identify all genes required for fucosylation in one experiment, simply by identifying the guideRNA present in the surviving population.

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