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Pilot Project Proposal
(Not to exceed two pages)

Name of Project: Precision human genome engineering of disease-associated noncoding variants

Proposer and Contact Information:

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Background:

New CRISPR/Cas-based genome engineering technologies have enabled complete gene knock-out in a rapid and efficient manner (Shalem*, Sanjana* et al., *Science*, 2014; Sanjana*, Shalem* et al., *Nature Methods*, 2014). In this case, gene knock-out takes advantage of the efficient non-homologous end-joining repair pathway to create frameshift mutations that abolish protein production. However, it remains a challenge to make precise single nucleotide variants using homology-directed repair, which tends to occur at ~100-fold lower efficiencies.

Technical Idea:

We propose to develop novel selection techniques to enrich for homologous recombination repair in disease-relevant cell types, such as human pluripotent stem cells (hPSCs). Given the growing set of relevant CRISPR systems (e.g. SpCas9, SaCas9, AsCpf1, LbCpf1, etc.), we will also develop bioinformatic tools to identify the best CRISPR system for introduction of a particular mutation/edit.

Utility and “Fit” For GP-write:

Our goal is to create a complete pipeline for rapid engineering of disease-specific variants into human cells with significantly higher efficiency than previously possible.