CellFit

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IN VITRO 3-D TOTAL CELL GUIDANCE AND FITNESS

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Transdifferentiation of pancreatic alpha to beta cells via targeted epigenome editing by Epi-CRISPRs-s directed DNA methylation

We propose to transdifferentiate alpha to beta cells using our recently developed Epi-CRISPRs, a novel synthetic epigenetic tool. Using this methodology we are able to induce straightforward, one-step cell transdifferentiation by targeted DNA methylation and suppression of homeobox gene *Arx* that is essential for maintaining pancreatic alpha cell identity.

The Epi-CRISPR constructs with and one or four different guide RNAs for specific targeting the promoter region of *Arx*, were transiently transfected in alphaTC1-6 cells (α-cells). The success of α-cells transdifferentiation into insulin-producing cells was evaluated by measuring *Arx*, glucagon (*Glu*) and insulin (*Ins2*) mRNA level, amount of secreted insulin and by immunostaining of insulin and glucagon in the cells.

Our study will be valuable for later subsequent Epi-CRISPRs use in mouse *in vivo* model of diabetes and eventually as a potential therapy for diabetes attenuation in humans.