# Differentiating iPSCs Which approach works best?



### Overcoming the challenges of cell differentiation and reprogramming with opti-ox<sup>™</sup>, precision reprogramming

Current methods to generate cells from human induced Pluripotent Stem Cells (iPSCs) – directed differentiation and cellular reprogramming – lack reproducibility and scalability. This raises challenges for disease research, drug discovery and cell therapy development due to unreliable data and long experimental timelines.

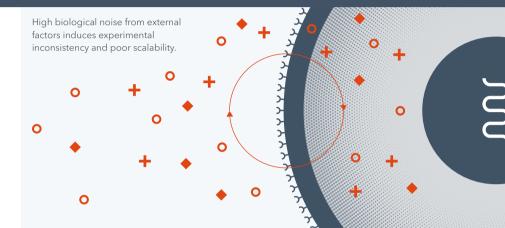
A technology that enables the reproducible, consistent and scalable production of physiologically relevant human cell models for any cell type would mean scientists can be more confident in their experiments. opti-ox, a first-of-its-kind precision reprogramming technology, meets all of these requirements by controlling the precise expression of cell-fate determining transcription factors without gene silencing.

In this infographic we outline traditional methods to generate cells from human iPSCs and compare these to the new precision reprogramming technology enabled by opti-ox.

### **Directed differentiation**

- Low purity
- Low scalability
  - Inconsistent batches
  - Long laborious complex protocols

Directed differentiation generates somatic cells from iPSCs through extracellular signalling by the addition of growth factors, cytokines or small molecules mimicking developmental signals, driving cell fate.



#### Long culture time and low scalability Long, complicated protocols combined

with the costly use of reagents (growth factors, proteins etc.) limit the ability to scale.

#### Low purity and inconsistent batches

Stem cells take a different route to differentiate each time and not all stem cells will differentiate, resulting in heterogenous populations and inconsistent differentiation. This leads to data

variability, and the collection of data that may be less relevant to the cell type of interest.

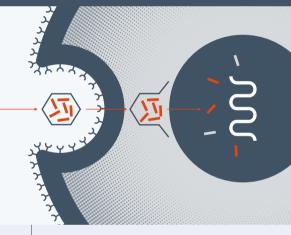


## Cellular reprogramming

- Migher purity with defined identity
- Low scalability
- Inconsistent batches
- Fast protocol

Cellular reprogramming involves the ectopic expression of transcription factors (TFs) to drive a specific cell fate. With this method cell identity is less dependent on external cues and has improved cell specificity. However the random integration of TFs can result in gene silencing meaning cell populations are inconsistent between batches.

#### Reduced biological noise from TFs drives a defined cell fate but is susceptible to gene silencing.

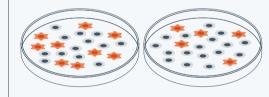


#### Shorter culture time

Cell reprogramming offers a faster protocol with differentiation occurring over weeks rather than months.



#### Higher purity, inconsistent batches and low scalability Cellular reprogramming driven by TFs results in a more homogenous population compared to directed differentiation. However the random integration of TFs leads to variable silencing resulting in inconsistent reprogramming, heterogeneity across batches and poor scalability.



### opti-ox<sup>™</sup> precision reprogramming by bit.bio

- Highly pure with defined identity
- Julia
  - Consistent batches
  - Fast protoco

opti-ox<sup>™</sup> precision reprogramming technology is a controlled gene engineering approach that enables unlimited batches of any human cell to be manufactured consistently at scale.

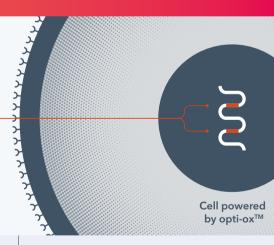
opti-ox utilises two genome safe harbour sites to protect TFs from silencing, and leverages an inducible system to control TF activation. With TFs precisely switched on in every stem cell, entire cultures consistently and rapidly convert into a defined cell type on a vast scale. Minimal biological noise as opti-ox enables inducible TF expression driving a defined cell fate while overcoming gene silencing.



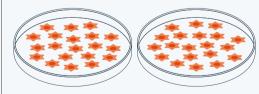
Short culture time

Using opti-ox precision reprogramming, human stem cells convert into consistent, mature and functional cells within days.





**Defined identity, consistent batches and scalability** With TFs precisely switched on in every cell without gene silencing, billions of iPSCs powered by opti-ox are reprogrammed to a new cell identity offering a consistent, scalable source of cells.



#### Who we are

bit.bio combines the concepts of cell programming and biology to provide human cells for research, drug discovery and cell therapy, enabling a new generation of medicines.

This is possible with our precision human cellular reprogramming technology opti-ox<sup>™</sup> – a gene engineering approach that enables unlimited batches of any human cell to be manufactured consistently at scale.

#### Contact us

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