

# **Generation of human induced Pluripotent Stem cells in microfluidics**

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Induced pluripotent stem cells (iPSCs) are generated via the expression of the transcription factors OCT4 (also known as POU5F1), SOX2, KLF4 and cMYC (OSKM) in somatic cells. In contrast to murine naive iPSCs, conventional human iPSCs are in a more developmentally advanced state called primed pluripotency. Here, we report that human naive iPSCs (niPSCs) can be generated directly from fewer than 1,000 primary human somatic cells, without requiring stable genetic manipulation, via the delivery of modified messenger RNAs using microfluidics. Expression of the OSKM factors in combination with NANOG for 12 days generates niPSCs that are free of transgenes, karyotypically normal and display transcriptional, epigenetic and metabolic features indicative of the naive state. Importantly, niPSCs efficiently differentiate into all three germ layers. While niPSCs can be generated at low frequency under conventional conditions, our microfluidics approach enables the robust and cost-effective production of patient-specific niPSCs for regenerative medicine applications, including disease modelling and drug screening.