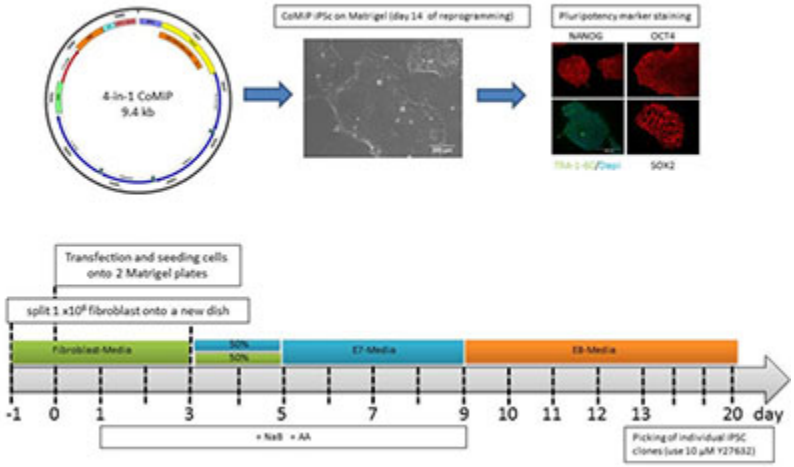
Docket #: S13-138

4-in-1 Plasmid for Efficiently Generating iPS Cells

Stanford researchers developed a single plasmid reprogramming system called CoMiP carrying codon optimized sequences of the canonical reprogramming factors (OKSM) and short hairpin RNA against p53. They have successfully derived >10 different mouse and human iPSC lines from fibroblast of young and old subjects by performing a single transfection using either electroporation or Lipofectamine LTX. Together with an additional vector encoding for *MYC, LIN28* and *NANOG* they were also able to reprogram blood derived PBMCs into iPSCs. The novelty of this system is that it is highly efficient, integration free, easy to use, and inexpensive to produce. Furthermore, the construct is color labeled and is free of any antibiotic selection cassette which might be beneficial for future application in regenerative medicine.



The figure is showing the novel reprogramming plasmid and the expected reprogramming result. Furthermore, this construct is so efficient that the first colonies appear after 5 days and can be picked after 16 days when cultured in chemical defined media.

Stage of Research
The utility of this vector has been demonstrated by transforming different human cell lines including blood derived PBMCs into iPS cells. Recently the technique was used in a cancer vaccination study.

Applications

- **iPS Cells** - create human and mouse iPSCs from young or old cells

Advantages

- One reprogramming plasmid (fibroblasts, keratinocytes, renal tubular cells)
- Antibiotic free
- All bona fide iPSCs (no intermediates)
- Color labeled (Tomato)
- Inexpensive
- Efficient (200-300 iPS colonies/ 1 x106 fibroblasts)
- Lipofectamine LTX
- Blood reprogramming (2 plasmid system)
- Integration free

Publications

- "Novel codon-optimized mini-intronic plasmid for efficient, inexpensive, and xeno-free induction of pluripotency" Sebastian Diecke, Jiamiao Lu, Jaecheol Lee, Vittavat Termglinchan, Nigel G. Kooreman, Paul W. Burridge, Antje D. Ebert, Jared M. Churko, Arun Sharma, Mark A. Kay and Joseph C. Wu. Jan 2015. *Scientific Reports*. DOI: 10.1038/srep08081

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