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Advancing human cell reprogramming synthetic mRNA technology to study and treat human disease.

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Abstract:

Dr. Michael Edel: (*mRNA iPSC to NSC for Spinal Cord Injury)* The development of a clinical grade method to efficiently generate genetically stable induced pluripotent stem cells (iPSC) and derived cells that engraft with no evidence of pathology remain key challenges to accurately model and reliably treat human disease with iPSC 1-5. To develop a clinical grade protocol, we adopted a synthetic mRNA transfection method using clinical grade cell culture materials that is robust, efficient and maintains parental cell CNV. Human iPSC made with our new method and derived neural stem cells (iNSC) have significantly reduced genetic instability: (i) lower multi-telomeric signal, (ii) reduced double strand DNA breaks (DSB) measured by H2AX expression, (iii) correct nuclear localization of Rad51 protein expression and (iv) whole genome sequencing (WGS) reveals reduced DNA structural variations implicating less coamplifications of cancer genes. In vivo analysis demonstrates reduced teratoma growth and proliferative rate of our iPSC and iNSC. The iNSC derived from our iPSC engraft in a spinal cord injury microenvironment, have a high percentage of survival, differentiate properly to neurons, oligodendrocytes and astrocytes over time with no evidence of pathology. In conclusion, we have developed a method to generate iPSC to generate improved genetically stable iPSC and iNSC that we offer as a potential standard operating procedure (SOP) for future clinical applications to accurately model and treat human disease.

Dra Ana Belén Alvarez Palomo: (*mRNA iPSC and direct cell reprogramming for lung and limbal disease)* Regenerative medicine opens a whole new world of possibilities in the treatment of degenerative diseases and conditions in which the endogenous capacity of regeneration of the organ has been exhausted. For many of these conditions and diseases don´t have a cure nowadays and the treatment is symptomatic or palliative. Given the high number of patients with ocular surface or lung disease poses a heavy burden in society and the national health systems, especially as the world population ages. The discovery of cell reprogramming that includes two

different approaches: (i) induced pluripotent stem cells (iPSC discovered by S. Yamanaka 10 years ago) 6 and (ii) direct cell reprogramming 7 (by-passing the pluripotent cell step), we are able to generate any cell type of the body to treat many diseases of many tissues of the human body. The iPSC approach takes 2-3 months to generate cells and is slower than the direct cell reprogramming approach that takes 1 month. This has opened the possibility of restorative cell therapy to regenerate the patient's tissues with his own cells, avoiding cell immune rejection (no need for immune suppression drugs). We have advanced the methodology and designed a clinical grade cell reprogramming protocol to produce multipotent stem cells with the capacity to produce lung or ocular cells by reprogramming from adult patient skin cells. We aim to use both cell reprogramming approaches, (i) iPSC and (ii) direct cell reprogramming methods to lung stem cells or ocular cells using synthetic mRNA transfection of key tissue specific factors 6,7.

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