FRM 2018 Abstract **466/500 Words**

Making Sperm and Eggs from Skin Cells

IPS cells (induced pluripotent stem cells) can be derived from skin cells and then can be differentiated into primordial germ cells (PGC-like cells) or PGCLCs. From there you can make spermatogenic stem cells (SSCs) when they are transferred to the fetal or neonatal testis, or viable oocytes after culture with fetal granulosa cells. This is not science fiction. This is being done daily in the lab in mice with many normal babies (pups). SSC's derived from IPS cells (that are derived from skin cells) can be transplanted into the sterile testis, and there they colonize the seminiferous tubules and form spermatogonia. PGC's are now being made successfully in humans as well as mice.

However, there are a few provisos to be worked out before this technology in mice can be applied clinically to humans. Firstly, if ES cells (embryonic stem cells) or IPS cells (induced pluripotent stem cells) are injected directly into fetal testis (or ovary) without being at least 95% converted to PGC-like cells, they will just form a tumor. So the IPS cells must firstly be differentiated in culture into PGC-like cells (PGCLCs), i.e., primordial germ cells. This is fairly easy to do, in both mice and humans.

IPS cells can be made readily by incubating somatic cells (like the skin) with just four known genes (which can be readily purchased commercially). One can use just three genes, but all four originally described genes are best: KLF4, SOX2, OCT4, and C-MYC. You can then maintain those IPS cells in just three genes: LIF, FGF-i, and GSK2B-i.

Next, you incubate those IPS cells in three genes (again all commercially available) to make epiblast-like cells. The three genes required to make epi-L cells are activin A, b FGFi, and KSR. Then the epi-L (epiblast) cells are incubated with five genes, and behold, you have in vitro PGCs. The five commercially available genes needed to do this are BMP4, SCF, LIF, EGF, and BMP8a. So with this simple in vitro culture, you now have primordial germ cells that can be made into functionally normal spermatogonia, or oocytes.

These PGCL cells need fetal or neonatal gonadal somatic cells to make sperm or eggs, just like in real life. To make oocytes in vitro from PGCL cells, you must incubate the PGC's in fetal granulosa cells for 3 weeks (in the mouse). Fetal granulosa-like cells in humans will have to be made (via a different pathway) from IPS cells. Alternatively, fetal gonadal cells can be harvested from abortuses. Sperm is much more difficult than making oocytes because the PGCs need to be surrounded by fetal or neonatal somatic cells in a specific structural array. Nonetheless, the plasticity of IPS cells and PGC's in vitro should eventually lead clinically to making sperm and eggs from the skin in vitro.