Unit 5: IPS Q and A

1. Why are IPS cells so important?

Scientists are very excited about iPS cells because they offer the potential to create banks of stem cells that more closely match the genetics of patients, and they offer a way to make human stem cells without destroying a human embryo. Some people find the destruction of a human embryo immoral. There are currently government regulations that prevent government funding to be used in making an embryonic stem cell line. Government funding can also only be used on lines that were formed from excess embryos from *in vitro* fertilization procedures. IPS allows scientists to more easily make stem cell-like lines for research, and even make cells that have specific diseases in mutations.

2. Both ES cells and iPS make teratomas when they are injected into mice. If they both cause cancer why is there so much more concern about cancer and iPS cells?

Both iPS cells and ES cells form teratomas because they are pluripotent and differentiate randomly once in the body. If either of these cells is to be used in a cell transplant therapy they first must be differentiated into a more mature cell type. Once a cell is differentiated it will not form these teratomas.

The way that iPS cells are made make it more likely that they will develop into cancer even after they have differentiated. The gene c-Myc is a known oncogene and if it is continued to be expressed may cause cancer. Also, retroviruses insert their genetic information into the host cell. This could cause mutations in the DNA that could also cause cancer.

3. Why do scientists use retroviruses to transfect the cells if it might cause mutations?

Many scientists use retroviruses because they are to date the most efficient way to create iPS cells. For strictly research purposes it is far more cost effective (and easier) to use the most efficient method. It would however be unethical to use retroviruses in medicine. There are many scientists who are researching other ways to make iPS that don't use retroviruses in order to improve safety in potential therapeutic treatments. These include using other types of viruses that don't permanently insert their DNA into the host's DNA, and using chemicals to replace some or all of the Yamanka factors. Finding a safer way to make iPS cells is currently a major goal in stem cell research.

These are some of the methods scientists are currently using:

-Using a virus called the adenovirus that does not integrate its DNA into the host cell.

-Transfecting cells with circular pieces of DNA called plasmids carrying the four Yamanaka factors. Plasmids rarely integrate into the host DNA.

-Substituting some of all of the four Yamanka factors with small molecules that will turn on pluripotency genes.

4. Can we really make patient specific stem cells?

Scientifically making patient specific iPS cell is something that could be done. In reality this is unlikely. The procedure to make iPS cells is long, inefficient, and produces variable results. Making a new iPS line for every individual that needs a cell therapy would be too expensive to make it a practical option. Research on spinal cord injury treatment in rats has shown that stem cell therapy for this condition is only effective in the first two weeks of treatment, but it takes longer than two weeks to create iPS cells.

What is more likely is the creation of iPS cell banks. The most critical element in cell transplant is that the patient and the transplanted cells are an HLA match. If they don't match the body will recognize the transplanted cell as foreign and mount an immune response. The idea is to create many iPS lines from enough genetically different individuals to create matches to cover the genetic variability of the entire population. If a patient requires stem cell therapy it would only be necessary to obtain matching cells from one of the pre-existing cell lines.

5. Are skin cells the best cell type to use in iPS?

Skin cells are very good to use for creating research cell lines because they are very easy to obtain. However, skin might not be the best option for making cells for therapeutic use. The skin is constantly exposed to the elements in the external environment that might damage DNA. Any mutations could potentially cause cancer or interfere in the function or the iPS or differentiated cell. One paper summarized in this lesson showed that chimeras made from skin cells were more likely to develop tumors than those made from stomach or liver cells, but it is not known why that is the case.[1]

Some scientists theorize that adult stem cells maybe the optimal cells to use because they are more similar to embryonic stem cells than fully mature cells. In a study using neural stem cells as the starting cell types iPS cells were obtained at a higher efficiency using only Oct4 and Klf4 than skin cells transfected with all four factors.[2] However, neural stem cells are located deep in the brain and are rather difficult to obtain. More research is needed to determine what cell types can be used for iPS.

6. Are iPS cells made from different cells the same?

Gene expression analysis has shown there can be variability in iPS cells even if they originate from the same cell type. There are some concerns that iPS cells retain some features of the original cell type. For example, there may be skin specific genes that are still expressed in iPS cells but are not expressed in embryonic stem cells. If this is the case iPS cells that originate from different cell types might be slightly different. More research is needed to address this question.

7. How come so few cells actually become iPS cells?

We don't completely understand why iPS conversion is so inefficient. Only about 0.01-1% of cells become iPS cells depending on the procedure. One theory is that iPS cells are formed primarily from a small percentage of immature cell types. However, studies in the liver have shown that most iPS cells were generated from mature cells.[1] Whatever determines which cells become iPS cells and which do not is unknown at this time. It appears to be an event determined primarily by chance. More research is needed to more fully examine this question.

8. Can IPS cells replace embryonic stem cells?

Maybe, but definitely not yet. Right now iPS cells are great research tools for studying cell function *in vitro*. However at this point there are many concerns about the safety and efficacy of iPS cells. More research on the function of iPS cells is needed before it can be determined if iPS cells can be used in human cell therapies. For now embryonic stem cells are still the gold standard in pluripotent stem cells and regenerative medicine.

1. Aoi T, Yae K, Nakagawa M, Ichisaka T, Okita K, Takahashi K, Chiba T, Yamanaka S. (2008) Generation of pluripotent stem cells from adult mouse liver and stomach cells. Science Aug 1;321(5889):699-702.

2. Kim JB, Zaehres H, Araúzo-Bravo MJ, Schöler HR. (2009) Generation of induced pluripotent stem cells from neural stem cells. Nat Protoc 4(10):1464-70.