Stem Cell Therapy: The Ultimate Cure

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Introduction

Death is a human destiny that every single person in our population has to go through eventually. There is no one that can avoid death, but, medicine can extend the life expectancy to effectively change the human survival state. There are a lot of causes that lead to death, including aging, accidents, and diseases, which account for the top causes of death within the human population. When considering only natural causes of death, scientists have reached ways to prevent diseases and slow down aging. Why do humans age? Why do we have shorter or longer life expectancies compared to other species?

A human is a collection of cells. The cells have the finite replicative capacity of human fibroblasts. What that means is that cells have a limited number of duplications. The limited duplication potential of a cell is caused by the decrease in the amount and the length of telomeric DNA in the human chromosome resulting from many duplications. After a cell division, the daughter cells have exactly same DNA with shorter telomerase. Telomerase is the part of DNA that protects each end of the DNA sequence. After around 50 cycles of replication, the telomeric DNA is too short to protect the genes, causing cell death (Harley, Futcher, Greider 1990). Because the cell has an expiry date, humans, the collections of cells, eventually are going to die as soon as the cells die, unless we can repair and replace our old and dead cells.

Because people think aging and accidents happen naturally, most people tend to focus on the prevention of diseases in order to extend their lifespan. A primary

reason for disease is a change in genetic information, caused by a mutation in certain genes. A mutation in genetic information has the ability to cause a loss of gene expression or the production of a non-functional or incorrectly functioning protein in the body, which has the potential to make people ill. Fortunately, all healthy humans have their own repair mechanism in order to deal with certain mutations (Jane B. Reece, Lisa A. Urry, Michael L. Cain, Steven A. Wasserman, Peter V. Minorsky, Robert B. Jackson 2010).

If a mutation happens in the body cells, the cells become damaged and cannot work as they are supposed to. As a result, adult stem cells must replace the damaged cells. Adult stem cells, also known as somatic stem cells, are cells that have not yet differentiated into specific cell types. Thus, differentiation signals from damaged cells cause the pluripotent adult stem cells to replace the damaged cells (Jane B. Reece, Lisa A. Urry, Michael L. Cain, Steven A. Wasserman, Peter V. Minorsky, Robert B. Jackson 2010).

Sometimes, adult stem cells are unable to replace the damaged cells. If these damaged cells remain in the body, they become cancerous cells, which replicate repeatedly in order to fit into their surroundings. The collection of these cancerous cells can possibly lead to cancer if left to replicate unimpeded (Jane B. Reece, Lisa A. Urry, Michael L. Cain, Steven A. Wasserman, Peter V. Minorsky, Robert B. Jackson 2010).

In order to treat cancer, an investigative procedure known as a biopsy has been used to determine the stage of cancer and how severe it is. Biopsy also has

been used to remove certain damaged parts of the body. Advances in biopsy techniques have allowed modern surgery to become safer. However, there are still risks and side effects involved with the biopsy procedure. Bleeding and damage to nearby tissues and other organs can occur during the biopsy. Pain and infection are commonly reported after biopsy as well. Similarly, surgery also can be used to remove non-malignant solid tumors (National Cancer Institute, December 12, 2012).

Doctors also use chemotherapy to treat cancer. Chemo drugs are strong drugs that are cancer-specific. They may be used to prevent the cancer from spreading, slow the rate of tumor growth, and kill cancerous cells all in an attempt to eventually cure the cancer. Common chemo side effects include nausea, vomiting, hair loss, a drop in blood cell counts following bone marrow changes, a loss in sexual desire, and emotional changes (National Cancer Institute, December 12, 2012).

Other procedures used to treat cancer include radiation therapy, targeted therapy, immunotherapy, hyperthermia, and photodynamic therapy. With these therapies, however, not all diseases can be treated (National Cancer Institute, December 12, 2012). Diseases that could not be treated with the treatment types mentioned previously were considered incurable diseases until researchers discovered a new advanced technique known as stem cell therapy. For the first time, stem cell therapy provided a method to treat the full range of incurable diseases and possibly slow down the aging process, resulting in longer and healthier life expectancy.

Using the idea of how nature (the adult stem cell) works, researchers came up with the idea of adult stem cell therapy. Adult stem cell therapy is a therapy using transplanting as a method to replace the damaged cells. However, cell types that the adult stem cells can differentiate into are restricted to their organ sources and the number of adult stem cells is very few for each tissue. For example, adult somatic stem cells cannot possibly regenerate lung cells that are damaged from undergoing cancerous stages. Additionally, in many cases, the transplants using adult stem cell therapy are rejected. These results lead researchers to search for other methods to replace damaged cells successfully (Henningson Jr., Stanislaus, Gewirtz 2003).

What the adult stem cells are missing is pluripotentiallity, which is the ability of a cell to differentiate into any kind of cell. By using the cells with pluripotentiallity, scientists believe that stem cell therapy can be accomplished more successfully. In mammals, cells that exhibit the characteristic of pluripotentiallity are restricted to the oocyte, the zygote, early embryonic stem cells, and primordial germ cells (Reubinoff and others 2000). Because of the pluripotency of embryonic stem cells, embryonic stem cell therapy gives a promising donor source for transplantation of cells or organs for a lot of diseases. Using human embryos, however, brings up some controversial issues. The main issue surrounds what the criteria is to consider something as 'life'; more specifically, it involves the question of whether or not human embryos are considered 'life'. Because the embryo will die during the embryonic stem cell therapy procedure, there are ethical issues involved

(Takahashi and others 2007). Because of the issues involved and low availability of embryos, the scientists have trouble further executing their research.

Induced pluripotent stem cells are a breakthrough in stem cell research that allow researchers to circumvent these ethical issues. By reprogramming already differentiated cells into an embryonic-like state, the ethical issues surrounding the use of human embryos can be avoided. The problem is generation of such cells. The induction of pluripotent stem cells is the area that stem cell researchers are still working on. There are currently available methods to generate induced pluripotent stem cells, but these methods confer both advantages and disadvantages. For the case of induced pluripotent stem cells from human somatic cell lines, there is the potential to generate tumor cells. Even with the difficulties that surround finding the perfect stem cell therapy, researchers believe it will be the best transplantation therapy for the treatment of a full range of diseases and slowing down the rate of aging in the future (Takahashi and others 2007).

Embryonic stem cells

Unlike adult stem cells, embryonic stem cells have the ability to differentiate into any cells in our body. Figure 1 draws a comparison between embryonic stem cells and somatic stem cells. Embryonic stem cells are derived from the blastocysts of the mammalian embryo, which has the capacity to differentiate into all three germ layers, the endoderm, the mesoderm, and the ectoderm. In contrast, somatic stem cells only have the ability to differentiate into certain types of cells. Both stem cells are pluripotent and self-renewable (Evans and Kaufman 1981).

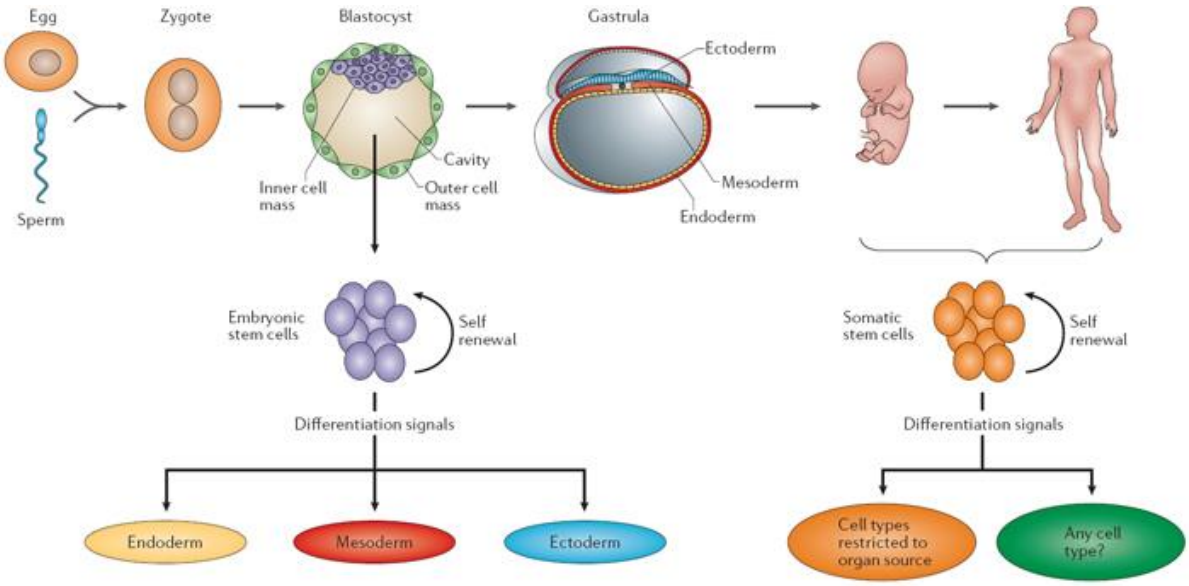

Figure 1 Comparison between embryonic stem cells and adult stem cells. They are both pluripotent and self-renewable, but, unlike adult stem cells, embryonic stem cells can differentiate into all three types of cell layer.
http://www.nature.com/scitable/content/embryonic-and-somatic-stem-cells-as-a-27447

Embryonic stem cells have characteristics that are derived from the preimplantation, which is the prolonged undifferentiated proliferation and stable developmental potential for all three-germ types (Thomson 1998). Embryonic stem cell research has enhanced medicine in many different ways. It has allowed researchers to study early human embryonic development, in order to determine the

characteristics of embryonic stem cells (which have been used to help to generate induced pluripotent cells), and has provided medical professionals with the tools required to develop more effective methods of transplant therapy (Bernstein and others 2006).

Human embryonic stem cells can be used to cure some diseases by replacing damaged cells (Thomson 1998). They provide donor sources by creating clones of the cells or organs for transplantation therapies for diseases such as juvenile diabetes, Parkinson's disease, and heart failure.

Embryonic stem cells also can be used to identify drug targets, to test potential therapies, to study cell differentiation, to understand prevention and treatment of birth defects, and to study early human development. This can be done because of the special characteristic of embryonic stem cells that allows them to undergo replication in vitro (Reubinoff and others 2000). Then, researchers differentiate the cells into what they are interested in for in vitro electrophysiological drug testing to test their therapies, treatment, or preventions (Caspi and others 2009).

However, embryonic stem cell research is experiencing setbacks because the number of embryos available is not sufficient. The source of human embryos is from in vitro fertilization for clinical purposes (for those who have trouble to fertilize naturally, in vivo). Usually the parents have more than 2 or 3 frozen embryos left after they get their baby using in vitro fertilization. There are not that many people who end up donating those for research purposes. The reason why there are not

that many people donating is because there is an ethical controversy regarding the use of human embryos in research (Thomson 1998).

The use of human embryos causes ethical controversies because many people consider the embryo to be the earliest stage of human life. In addition, generating embryonic stem cells for disease-specific or patient-specific treatment is difficult (Yamanaka 2007). In addition to these issues, there are some problems of tissue rejection followed by transplantation in patients. In order to avoid all these disadvantages, a new advanced technology which involves inducing pluripotent cells from the patients' own cell, has been introduced (Takahashi and Yamanaka 2006).

Patient- or disease-specific induced pluripotent stem cells

Limitations of treating patients with embryonic stem cells caused researchers to search for other technologies to replace embryonic stem cells. Inducing pluripotent stem cells from patients' somatic cells is an ongoing research technology that has been developed in order to prevent or treat some diseases. Induced pluripotent stem (iPS) cells are made by the successful reprogramming of the patients' somatic cells into a pluripotent state. The somatic cells can be reprogrammed with some modification to become embryonic stem cell-like iPS cells. Unlike embryonic stem cells, iPS cells guarantee a higher chance of generation of successful patient- and disease-specific stem cells (Okita, Ichisaka, Yamanaka 2007).

The concept behind the iPS cells is using reprogramming. Reprogramming is essentially the reverse process of cell differentiation. This process occurs by modifying the genetic information to express a pluripotency in the patients' somatic

cells; this causes the cells to forget what kinds of cells they were, and thus the cells are 'put back' into an undifferentiated, pluripotent stage (Yamanaka 2007).

Treatment using iPS cells is similar to treatments involving the use of embryonic stem cells, but it is more efficient in the sense that the iPS cells have almost the same characteristics that embryonic stem cells have – namely, they are pluripotent and self-renewable - but they also have characteristics of being patient- and disease-specific. The biggest challenge with iPS cells remains the question of how to create certain cells (Yu and others 2007).

Figure 2 shows the four popular currently available techniques to generate iPS cells. iPS cells can be made by nuclear transfer from somatic cells to an Oocyte, nuclear fusion between somatic cells and embryonic stem cells, culturing germline stem cell (GS cells) and parthenogenetic embryos bone marrow cells, and putting some defined factors into somatic cells(Yamanaka 2007). Additionally, generation using recombinant proteins has been reported recently (Zhou and others 2009). Because of the complexity of generating those cells, some of the techniques are not yet applied to humans, but to mice only. (Yamanaka 2007).

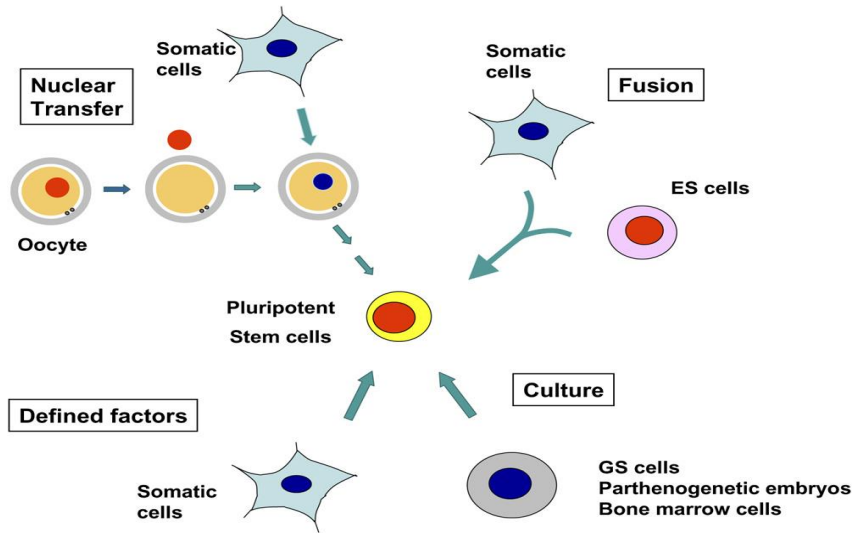

Figure 2 Currently available methods to generate iPS cells
iPS cells can be generated using nuclear transfer, fusion, defined factors, and culturing. These are the techniques to create pluripotent stem cells from adult somatic cells.

Generation of induced pluripotent stem cells

Pluripotent stem cells can be created by long-term culture of special types of cells. For example, germline stem cells, from the testes, can be reprogrammed into multipotent germline stem cells through long-term culture. However the efficiency of creation of these cells is very low and the process requires germline stem cells from over 30 mouse testes and excessive amounts of time. Nevertheless, the stem cells created by long-term culture can differentiate into various kinds of cell types like embryonic stem cells (Yamanaka 2007).

Successful nuclear transfer of embryonic donor cell nuclei involves the transfer of nuclei from the cell that the researcher is interested in generating pluripotency in to the embryonic stem cells derived from human oocytes. One of the famous examples of this process occurred in 1996, with "Dolly", a sheep produced by the nuclear transfer of embryonic stem cells. Subsequently, somatic cloning in other animals like cows and mice has been successfully performed. However, the process has not yet been tried in humans because of the ethical issues surrounding the acquisition of an oocyte and the fact that a higher quality of technique is required in this procedure (Takahashi and others 2007).

After fusion, it was observed that cells exhibited the characteristic of pluripotency. The exact mechanisms and whether somatic genomes reprogramming is fully from the fusion or is not known, but after the fusion, the embryonic hybrid cells share some promoter regions of several genes that are essential for the pluripotency, histone acetylation and methylation patterns with embryonic stem

cells (Kimura and others 2004). After the fusion, to get pure induced pluripotent cells, all of the cell-derived chromosomes and the chromosomes containing Nanog, which encodes the proteins essential for the pluripotency, have to be removed. After the completion of removal of all the embryonic stem cell-derived information, the hybrid cells that remain exhibit pluripotency. Still, the selective removal of genes in chromosomes is a remaining challenge (Yamanaka 2007).

Figure 3 shows Nanog-overexpressing in mouse embryonic stem cells showed a strong positive correlation with the number of embryonic stem cell-like hybrid colonies. If the Nanog gene is present, the more embryonic stem (ES) cell-like hybrid colonies the researchers obtained. If the Nanog gene is present in the ES cell, researchers get more ES-like hybrid colonies. This shows the important role of Nanog in pluripotency of certain cells.

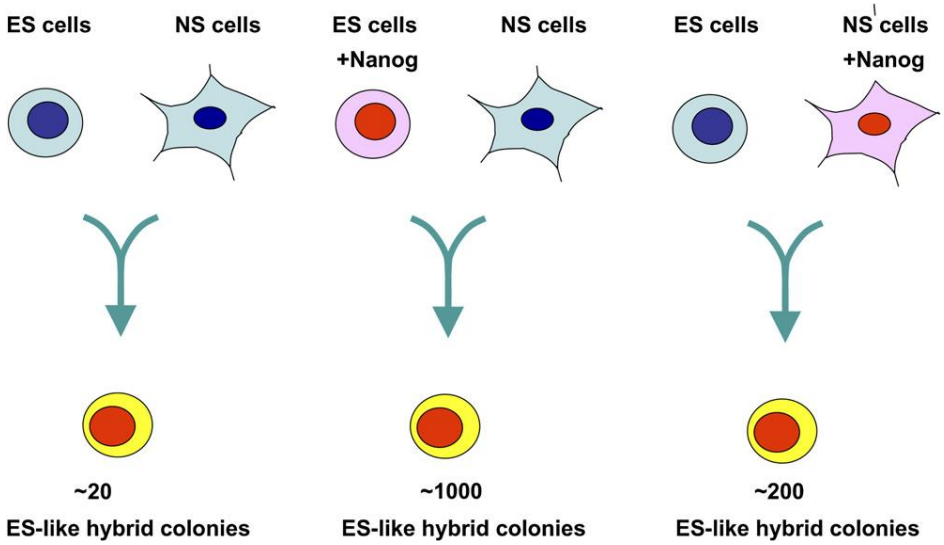

Figure 3 Nanog-mediated engancement of generating by fusion with embryonic stem cells
Fusion of somatic cells with nanog-overexpressing mouse embryonic stem cells shows the highest rate of reprogramming process.

Generating pluripotent cells without embryo

Following the fusion of the somatic cell with the embryonic stem cell, scientists have observed that the somatic cell exhibited the characteristic of pluripotency. Therefore, researchers hypothesize that the successful reprogramming of the somatic cell must be due to some pluripotency-inducing factor within the embryonic stem cell. They examined 24 different factors that might have induced the pluripotency of iPS cells. As it turns out, introducing four transcription factors, Oct-3/4, Sox2, c-Myc, and Klf4, into adult somatic cells can induce the pluripotency of the cells (Takahashi and Yamanaka 2006). Associating the transcription factors with a selection marker, Nanog, resulted in creating iPS cells. The selection marker does not have to be Nanog, but Nanog-selected iPS cells are almost identical to embryonic stem cells in gene expression, DNA methylation, and histone modification patterns (Okita, Ichisaka, Yamanaka 2007).

Generating pluripotent cells by retrovirus-mediated introduction of the transcription factors into adult somatic cells involves the possibility of producing tumor cells. This is because generating iPS cells involves cancer-associated transcription factors, c-Myc and Klf4. Figure 4 shows how four factors work to induce pluripotent stem cells. C-Myc induces two properties of iPS cells, immortality and open and active chromatin structures. However, after these properties are induced, the cell now has the potential to go through senescence and apoptosis. The apoptosis of the cells can be suppressed by another transcription factor, Klf4. Thus, expression of c-Myc and Klf4 confers the possibility of creating a

tumor cell rather than an iPS cell as the final product. Oct-3/4 changes the cell fate from a tumour cell to an embryonic stem cell-like cell, but not yet a pluripotent cell. Finally, the expression of the Sox2 gene induces the pluripotency of the cell (Yamanaka 2007). Other than tumorigenicity, extremely low efficiency of iPS cell induction is a limitation. Less than 1 % of the cells that put genes transcribing four transcription factors by retroviral insertion can be reprogrammed into iPS cells (Hwang and others 2005).

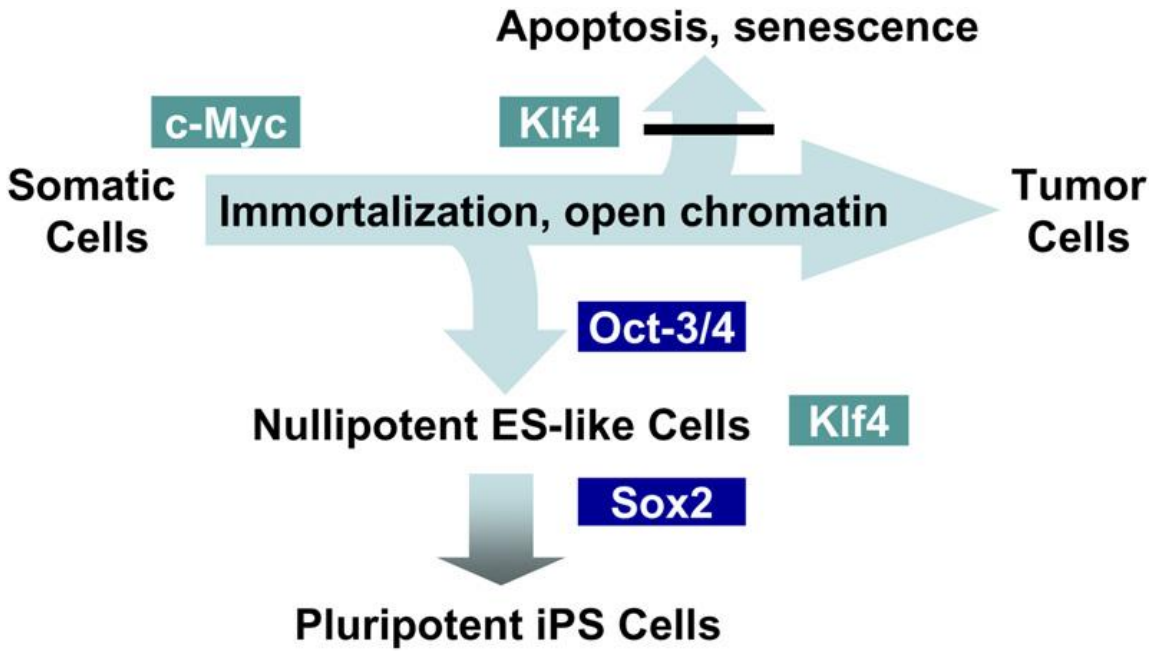

Figure 4 How four transcription factors induce iPS cells
Activation of all four transcription factors induce the iPS cells. C-Myc allows open chromatin and immortalization, Klf4 suppresses apoptosis, and Oct-3/4 and Sox2 together, creates pluripotent iPScells.

Future of iPS cells

Recently, protein-induced pluripotent stem (piPS) cells have been generated using recombinant proteins. It is basically the same technique as retroviral insertion of four transcription factors, Oct-3/4, Sox2, c-Myc, and Klf4, but it is more advanced. The main difference is that piPS cells are generated by delivering the

reprogramming proteins directly into adult somatic cells, rather than delivering genes by retroviral insertion. This transportation is done by the microinjection of proteins into the cell cytoplasm. This protein transduction method reduces the possibility of tumorigenicity, and is a simpler and faster approach that can save some time(Zhou and others 2009).

iPS cells are an appealing choice for future medicine, since there's no ethical issues involved in their use in research and disease treatment. Currently, many researchers are working on developing more efficient methods of reprogramming adult somatic cells to generate iPS cells. As mentioned previously, the currently available methods are by nuclear transfer, fusion with embryonic stem cells, defined factors, culture, and recombinant proteins. There's no best method since each method has advantages and disadvantages over one another. Tumorigenicity and ethical issues are concerns for the methods involved, and thus researchers are trying to find ways to avoid the certain disadvantages of pluripotent cells (Yamanaka 2007).

Conclusion

Adult somatic stem cells are cells that can replace damaged cells. They are undifferentiated cells that are not performing specific jobs yet. Instead, they can differentiate into cells belonging to particular parts of the body in order to replace the damaged cells. But, compared to embryonic stem cells, adult somatic stem cells do present certain disadvantages are they are restricted to only certain type of cells,

rather than all type of germ layers as is the case with embryonic stem cells. stem cell therapy using adult somatic stem cells was not quite successful because of the absence of pluripotency.

Unlike adult somatic stem cells, embryonic stem cells can differentiate into all types of cell in the host's body, which is the characteristic termed pluripotent. These embryonic stem cells can be used in transplant therapy to cure some untreatable diseases. However, because the process of making these embryonic stem cells occurs in vitro, in addition to the fact that this technology involves the use of human zygotes, there have arisen many ethical controversies associated with the generation and use of embryonic stem cells.

Because of the advancements that embryonic stem cells have given rise to in medicine, scientists have researched alternative methods to generate pluripotent cells. iPS cells are embryonic stem cell-like cells that have the characteristic of pluripotency. The generation of iPS cells is being researched worldwide, as iPS cells have the potential to replace embryonic stem cells in stem cell therapy. However, generating certain iPS cells is challenging and research must continue in order to find a method that is able to to generate iPS cells more efficiently.

Among a lot of currently available methods of stem cell therapy, there is no single technique that is better than the others. Some of the techniques would not be desirable for use in humans. Despite such disadvantages associated with the current methods, stem cell therapy is very important because the concept behind using stem cells is thought to be the ultimate cure of a full range of diseases.

However, challenges are encountered when one factors in the ethical controversies surrounding stem cell research and difficulties generating perfect pluripotent cells for therapeutic purposes. Finding methods to reprogram and to generate such perfect iPS cells is still an on-going research topic as currently available methods also have some limitations. As techniques continue to improve, the resultant iPS cells that are generated are observed to be more efficient and display fewer adverse characteristics. Because of the importance of iPS cells in modern research and medicine, these technologies will continue to be researched and improved upon until researchers find the optimum stem cell therapy treatment, which will involve few limitations and be surrounded by no ethical controversies.

Generations of medical researchers have attempted to find the optimal solution to the prevention and treatment of many diseases. A recently emerged advanced technique, known as stem cell therapy, will provide a huge step forward in modern medicine. It is likely that in subsequent years, some incurable diseases that target the human population will be eradicated by stem cell therapy.

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