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IPSCS FOR ENHANCING STEM CELL TRANSPLANTS

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ABSTRACT

Induced pluripotent stem cells (iPSCs) have the potential to pave the way for personalized therapies in stem cell research. Their combination of abilities to differentiate into all three germ layers and circumvent the issue of graft rejection renders them a potential candidate for stem cell transplants. Research on iPSCs is relatively scarce and few studies get to the clinical trial stage. To encourage more studies on iPSCs, specifically regarding stem cell transplants, this article makes the comparison of iPSCs to adult stem cells and embryonic stem cells to highlight their superior attributes.

KEYWORDS: Stem Cell Transplants, Pluripotency, Patient-Specific, Epigenetic Memory, Mutations of Induced Pluripotent Stem Cells

INTRODUCTION

In stem cell transplants, adult stem cells and embryonic stem cells are typically used, since there is extensive research and many clinical trials supporting their effectiveness. However, a novel type of stem cell - iPSCs - could potentially revamp this field as they are pluripotent, patient-specific, and self-renewing. Induced pluripotent stem cells can be programmed to attain the characteristics of any cell. Discovered by Nobel Prize winners, Shinya Yamanaka and Sir John B. Gurdon, induced pluripotent stem cells are cells derived from anyone through a virus that introduces four genes (Oct ¾, Klf4, c-Myc, Sox2). Due to their pluripotent ability, they have revolutionized the way scientists observe and understand diseases. Thus, scientists are positive about its potential applications for disease modeling, drug discovery or validation, and cell or gene therapy.

Adult Stem Cells (ASC) and Embryonic Stem Cells (ESC) are used in a variety of cell therapies. The first adult stem cell transplantation was performed by Edward Donnall Thomas in 1957; since then, this method of treatment has greatly evolved. Adult Stem Cells (or Somatic Stem Cells) are cells derived from one's body which can then be cultured with desired growth factors and transplanted back into the patient. They can be found in bone marrow, fat, or blood but aren't accessible and are limited in number. As ASCs are multipotent, they are limited to replacing a small number of nearby cells similar to the organ source (US National Research Council & US Institute of Medicine Committee on the Biological and Biomedical Applications of Stem Cell Research, 2002). Embryonic stem cells were first cultured in vivo in 1998 by Professor James Alexander Thomson and his team. These stem cells are the inner cell mass from embryos, obtained in the blastocyst stage.

Cell transplants are used in cases where patients have damaged/diseased bone marrow and cannot make cells or lost a substantial amount of cells (e.g., due to chemotherapy or radiation therapy). Such diseases include sickle cell anemia, leukemia, multiple myeloma, certain lymphomas, etc.. There are various methods of transplanting cells in cell therapy. Autologous cell transplant refers to cells derived from the patient (e.g., adult stem cells or induced pluripotent stem cells) then cultured and transplanted back into the patient to avoid immune rejection. Allogeneic transplants rely on a donor's cells (e.g., adult stem cells or embryonic stem cells) hence many issues with the graft can occur. The histocompatibility antigens (human leukocyte antigen, HLA) of the donor and recipient must match to decrease the likelihood of immune rejection.

Several studies show the strengths of induced pluripotent stem cells (iPSCs) in their ability to divide into all three germ layers, but there is a lack of studies done specifically on their applications in human stem cell transplants. There is a necessity for progress in stem cell therapies as the survival rate one year after allogeneic adult stem cell transplants performed between 2013 and 2016 was low at 72% (Penack et al., 2020). Scientists found that a form of immune rejection as well as unintended side effects of the transplant are the main causes (e.g. graft-vs-host disease or infections).

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This article delves into a comprehensive analysis of prior studies on induced pluripotent stem cells, aiming to shift the trajectory of research endeavors. Employing a comparative approach with an analytical focus, the intention is to underscore specific areas that warrant further exploration and investigation. The overarching goal is to contribute to the expansion of knowledge and encourage a more nuanced understanding within the scientific community.

LITERATURE REVIEW

iPSCs have drawn in researchers because of their distinctive characteristics, there is a wide body of literature supporting their advantages. However many concerns arose over the method of deriving these cells and other strands of literature explored ways to address those.

One concern that emerged was the method of lentiviral transduction, which may activate unnecessary gene expression (e.g., cancer expression). As a remedy, researchers have found a virus-free technique where plasmids are used instead to introduce the factors of reprogramming (they used LIN28 instead of c-Myc). Without the need to integrate DNA and the lack of packaging vectors, plasmid transfection iPS cells expressed pluripotent markers and had the same morphology as lentivirus iPSCs. This method had around the same efficiency as adenovirus transduction but had lower efficiency than using lentiviruses (Si-Tayeb et al., 2010). As plasmids still have the chance to enter the genome and accidentally activate cancer genes, researchers found a way to remove the doxycycline-regulated lentiviral vectors with cre-recombinase after the cells become pluripotent. They could successfully reprogram fibroblast cells from patients with sporadic Parkinson's Disease into dopaminergic neurons. The process was efficient and used non-integrating plasmids or vectors without using virus-carrying factors (Soldner et al., 2009).

It is important to note that researchers expressed concerns over the use of the c-Myc retrovirus as it can encourage cancer growth. However, Nakagawa et al. derived iPS cells from mouse and human fibroblasts without using the oncogene (2007). They found that c-Myc wasn't needed to create iPSCs and the cells had high quality but its removal slowed the process of culturing. Li et al. also showed that iPS cells could be derived from mice with only three factors (without c-Myc) (2011). The iPS cell line could aid in the full-term development of tetraploid-complemented mice. Thus, induced pluripotent stem cells have many characteristics that could lead to and improve stem cell therapies.

The alternative to iPSCs, embryonic stem cells, have their own issues. As the destruction of an embryo is required, there are some ethical concerns in its use. It is important to note that these cells are also pluripotent but are usually from a donor, so immune rejection is likely to develop in the patient. To avoid this issue, scientists have developed a method called somatic cell nuclear transfer (SCNF). The patient's DNA replaces the donor's DNA in the donor's egg, a form of cloning. Animal embryos have been cloned already, such as the renowned sheep, Dolly. Dolly was created from an adult epithelial cell (from a mammal). However, many issues arose in her health. For example, her telomeres (usually indicate age) were 20% shorter than those of other sheep her age. Moreover, she developed premature arthritis in her left leg at 5 and a half years old whereas the average sheep develops such a condition at 10 years. As this procedure has a low reprogramming efficiency, created many abnormalities, and has only been used in animals, there are still many developments needed in SCNF (Ogura et al., 2013).

iPSCs' pluripotency attribute

Pluripotency is characterized by the stem cell's ability to differentiate; induced pluripotent stem cells are versatile as they can show the characteristics of a range of cells. In fact, they can differentiate into all three embryonic germ layers (ectoderm, endoderm, and mesoderm). In regular cells, telomeres, which protect DNA and are located at the end of the chromosomes, shorten each time cell division occurs, hence there is a limit to the number of replications (Zvereva et al., 2010). In iPSCs, the telomerase enzyme maintains the length of the telomeres and allows the stem cells to divide infinitely.

In 2022, researchers reprogrammed bone marrow cells from a patient with multiple myeloma (MM) into iPS cells. After creating an embryo body, an immunofluorescence analysis of the reprogrammed cells revealed that iPS cells could differentiate into all three germ layers. They confirmed the identity of the cell line via short tandem repeat analysis (with the MM cells from the patient) (Hong et al., 2022).

Amyotrophic lateral sclerosis is a neurodegenerative disorder that targets the motor neurons and other neurons, leading to their decline. Without neurons to transmit signals, the weakened muscles often lead to paralysis. With no known cure and the only treatment option being a moderately effective drug, Riluzole, researchers have opted for animal models to better understand it. Researchers could differentiate iPS cells into motor neurons, oligodendrocytes, and astrocytes in an attempt to model the disease (Richard et al., 2015). iPSCs can divide into a variety of cells and have the potential to be used in stem cell transplants.

Adult stem cells are known to be multipotent and can only differentiate into cells related to their lineage. Due to this limitation, scientists were drawn towards autologous transplants. Autologous adult stem cells could potentially be used in cell transplants but in diseases such as cancer where cells are destroyed, acquiring cells from an already weakened area may worsen the patient's condition. Therefore, autologous stem cell transplants may not be ideal as patients may not have the optimal cell quality or quantity. Tsukasaki et al. reported that the eight patients with adult T-cell leukemia who have undergone autologous adult stem cell transplants all died, either because of issues with the transplant or a relapse of the disease (Tsukasaki et al., 1998).

Patient-specific quality of iPSCs

Graft-versus-host-disease (GVHD) is the main concern with embryonic stem cells and allogeneic adult stem cell transplants.

Graft refers to the transplanted tissue and the host refers to the donor's tissue. There are two variations of this disease: either the white blood cells from the donor attack the cells of the patient or the patient's immune system attacks the donor's cells. Induced pluripotent stem cells, on the other hand, are patient-specific. As they are mostly derived directly from a patient's cells and reprogrammed, they don't put the patients at risk for this disease.

Sickle cell anemia is a single-point mutation that causes the sickling of red blood cells (RBC). There is a genetic mutation in β-globin which creates sickled hemoglobin; they can lead to anemia, vaso-occlusion, and premature cell death. Researchers used a knock-in mice model to test whether they could reprogram mutant cells and rectify them before cell transplantation (Nakagawa et al., 2007). They replaced the mouse β-globin genes with human sickle globin genes and replaced the mouse β-globin genes with the human α-globin genes. Nakagawa et al. obtained fibroblasts from the tail tip of the mice and reprogrammed them to iPS cells via transcription factors (Oct4, Sox2, Kl4, and a lentivirus encoding a 2-lox c-Myc cDNA). To repair the cells, the researchers electroporated them with a targeting vector. The cells that had undergone homologous recombination were then exposed to radiation before finally being transplanted into three mice. Morphological analysis of red blood cells revealed that mice treated with iPSCs had a decrease in poikilocytosis, anisocytosis, polychromasia, and reticulocyte. 12 weeks after the transplant the conditions of the mice improved. There was an increased RBC count, hemoglobin, and packed cell volume levels. It is important to note that there was no tumor formation, despite a concern for it as the method to reprogram the cells was through oncogenes (c-Myc).

Another study that demonstrates iPSCs' characteristics of patient-specificity was conducted by the RIKEN Institute (Mandai et al., 2017). Age-related macular degeneration (AMD) affects central vision. One form of AMD is neovascular AMD ("wet" AMD), which is caused by abnormal blood cell growth under the retina. The vessels may leak fluid which damages the macula. There are existing treatments for patients with AMD such as anti-vascular endothelial growth factor (VEGF) and photodynamic therapy. However, there are various issues with both treatments. Many patients who received anti-VEGF drugs/therapies have had recurrences of AMD and photodynamic therapy has been linked to developing skin carcinogenesis (Borgia et al., 2018; Rofagha et al., 2013). Consequently, stem cell transplants have piqued the interest of researchers. The researchers in this study tried to create grafts of retinal pigment epithelial cells (RPE) as AMD is identified by the degradation of these cells. Embryonic stem cell and allogeneic adult stem cell transplants were unsuccessful in many patients as their immune systems rejected the graft (Algvere et al., 1999). Indeed, when immunosuppressants were used, many patients had adverse reactions.

The RIKEN Institute group reprogrammed induced pluripotent stem cells to slow the progression of the disease in a 70-year-old patient with neovascular AMD. She had received previous treatment from the anti-VEGF injections whereby her vision continually declined. Hence, researchers cultured cells derived from the subject's skin into retinal pigment epithelium cells (RPE cells). They created sheets of iPS-RPE cells, which have undergone several quality checks, and were then transplanted into the patient. The 70-year-old patient didn't show signs of graft rejection or worsening of vision. As iPSCs are reprogrammed in a patient-specific manner, there is no risk of developing GVHD or need for immunosuppressants.

Unexplored characteristics of iPSCs

Epigenetic memory

Induced pluripotent stem cells have their shortcomings, they may have an epigenetic memory and may develop mutations. Several studies have shown that iPS cells may harbor epigenetic memory as they could more easily be reprogrammed to cells similar to the parent cell. Kim et al. (2010) reprogrammed blood-derived iPSCs and fibroblast-derived iPSCs into osteoblasts. Through staining with alizarin, researchers found that fibroblast-derived iPS cells had more calcium deposition and expressed more Bglap, Sp7, and Runx2 (genes associated with osteoblasts). This suggests that fibroblast iPS cells have more potential to be reprogrammed into osteoblasts because of their epigenetic memory. Researchers postulate that residual methylation may be a cause for an enhanced differentiation of iPS cells to cells similar to their parent type. There is a need for a method to reprogram cells to a fully pluripotent state.

However, many researchers have found that repeated culturing of iPS cells can remove the effect of epigenetic memory (Hu et al., 2016; Polo et al., 2010; Rizzi et al., 2012). Hu et al. showed that human iPSCs from endothelial cells, fibroblast, or cardiac progenitor cells had different reprogramming efficiencies. Endothelial-derived iPS cells could be differentiated more efficiently in the early stages of differentiation relative to fibroblast or cardiac progenitor-derived cells. Though, in later passages, the distinction between the cells' reprogramming efficiency diminished. This suggests that epigenetic memory isn't permanent. More research is needed to understand the mechanisms behind such memory.

Mutations

Another study has revealed that differentiating iPS cells for a long time puts the cells at risk of developing mutations. They noted heterozygosity loss and genetic variability (Xavier Doss & Sachinidis, 2019).

Researchers from RIKEN, as mentioned above, tried to treat a patient with AMD via transplanting retinal iPS cells. However, they decided not to treat the second patient as there were genetic mutations in the iPSCs that were supposed to be transplanted (and also because his neovascular membrane showed signs of response to the anti-VEGF drugs) (Mandai et al., 2017). A system of quality controls for iPS cells is needed before transplantation can happen on a larger scale. As this is a relatively new type of stem cell, there is little research done and more is needed to understand their mechanisms.

CONCLUSION

Induced pluripotent stem cells appear to be a better candidate for stem cell transplants than adult or embryonic stem cells as they can differentiate into a wide range of cells, create an infinite number of cell lines, and exhibit patient-specificity. They should, however, be used with caution and under strict regulations (e.g. quality controls) as their epigenetic memory and the formation of mutations remain yet to be fully understood. More research must be conducted to better understand the mechanisms of iPSCs and their potential to aid in the recovery of patients with damaged or lost cells.

REFERENCE

1. Algvere, P. V., Gouras, P., & Dafgård Kopp, E. (1999). Long-term outcome of RPE allografts in non-immunosuppressed patients with AMD. European Journal of Ophthalmology, 9(3), 217–230. https://doi.org/10.1177/112067219900900310
2. Borgia, F., Giuffrida, R., Caradonna, E., Vaccaro, M., Guarneri, F., & Cannavò, S. (2018). Early and late onset side effects of photodynamic therapy. Biomedicines, 6(1), 12. https://doi.org/10.3390/biomedicines6010012
3. Deinsberger, J., Reisinger, D., & Weber, B. (2020). Global trends in clinical trials involving pluripotent stem cells: A systematic multi-database analysis. Npj Regenerative Medicine, 5(1). https://doi.org/10.1038/s41536-020-00100-4
4. Doss, M. X., & Sachinidis, A. (2019). Current challenges of IPSC-based disease modeling and therapeutic implications. Cells, 8(5), 403. https://doi.org/10.3390/cells8050403
5. Hong, J.-H., Kim, J.-H., Kang, K.-W., Lee, B.-H., Park, Y., & Kim, B.-S. (2022). Generation of a human induced pluripotent stem cell line kumi006 from a patient with multiple myeloma. Stem Cell Research, 61. https://doi.org/10.1016/j.scr.2022.102767
6. Hu, S., Zhao, M.-T., Jahanbani, F., Shao, N.-Y., Lee, W. H., Chen, H., Snyder, M. P., & Wu, J. C. (2016). Effects of cellular origin on differentiation of human induced pluripotent stem cell–derived endothelial cells. JCI Insight, 1(8). https://doi.org/10.1172/jci.insight.85558
7. Kim, K., Doi, A., Wen, B., Ng, K., Zhao, R., Cahan, P., Kim, J., Aryee, M. J., Ji, H., Ehrlich, L. I., Yabuuchi, A., Takeuchi, A., Cunniff, K. C., Hongguang, H., Mckinney-Freeman, S., Naveiras, O., Yoon, T. J., Irizarry, R. A., Jung, N., … Daley, G. Q. (2010). Epigenetic memory in induced pluripotent stem cells. Nature, 467(7313), 285–290. https://doi.org/10.1038/nature09342
8. Kobold, S., Bultjer, N., Stacey, G., Mueller, S. C., Kurtz, A., & Mah, N. (2023). History and current status of clinical studies using human pluripotent stem cells. Stem Cell Reports, 18(8), 1592–1598. https://doi.org/10.1016/j.stemcr.2023.03.005
9. Li, W., Zhao, X., Wan, H., Zhang, Y., Liu, L., Lv, Z., Wang, X.-J., Wang, L., & Zhou, Q. (2011). IPS cells generated without C-myc have active DLK1-dio3 region and are capable of producing full-term mice through tetraploid complementation. Nature News. https://www.nature.com/articles/cr201125
10. Liang, G., & Zhang, Y. (2012). Embryonic stem cell and induced pluripotent stem cell: An epigenetic perspective. Cell Research, 23(1), 49–69. https://doi.org/10.1038/cr.2012.175
11. Mandai, M., Watanabe, A., Kurimoto, Y., Hirami, Y., Morinaga, C., Daimon, T., Fujihara, M., Akimaru, H., Sakai, N., Shibata, Y., Terada, M., Nomiya, Y., Tanishima, S., Nakamura, M., Kamao, H., Sugita, S., Onishi, A., Ito, T., Fujita, K., … Takahashi, M. (2017). Autologous induced stem-cell–derived retinal cells for macular degeneration. New England Journal of Medicine, 376(11), 1038–1046. https://doi.org/10.1056/nejmoa1608368
12. Nakagawa, M., Koyanagi, M., Tanabe, K., Takahashi, K., Ichisaka, T., Aoi, T., Okita, K., Mochiduki, Y., Takizawa, N., & Yamanaka, S. (2007). Generation of induced pluripotent stem cells without Myc from mouse and human fibroblasts. Nature Biotechnology, 26(1), 101–106. https://doi.org/10.1038/nbt1374
13. National Research Council (US) and Institute of Medicine (US) Committee on the Biological and Biomedical Applications of Stem Cell Research. (2002). Adult stem cells - stem cells and the future of regenerative medicine ... National Library of Medecine. https://www.ncbi.nlm.nih.gov/books/NBK223693/
14. Ogura, A., Inoue, K., & Wakayama, T. (2013). Recent advancements in cloning by somatic cell nuclear transfer. Philosophical transactions of the Royal Society of London. Series B, Biological sciences. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3539358/
15. Ouyang, X., Liu, Y., Zhou, Y., Guo, J., Wei, T.-T., Liu, C., Lee, B., Chen, B., Zhang, A., Casey, K. M., Wang, L., Kooreman, N. G., Habtezion, A., Engleman, E. G., & Wu, J. C. (2021). Antitumor effects of IPSC-based cancer vaccine in pancreatic cancer. Stem Cell Reports, 16(6), 1468–1477. https://doi.org/10.1016/j.stemcr.2021.04.004
16. Parrotta, E., De Angelis, M. T., Scalise, S., Candeloro, P., Santamaria, G., Paonessa, M., Coluccio, M. L., Perozziello, G., De Vitis, S., Sgura, A., Coluzzi, E., Mollace, V., Di Fabrizio, E. M., & Cuda, G. (2017). Two sides of the same coin? unraveling subtle differences between human embryonic and induced pluripotent stem cells by Raman spectroscopy. Stem Cell Research & Therapy, 8(1). https://doi.org/10.1186/s13287-017-0720-1
17. Penack, O., Peczynski, C., Mohty, M., Yakoub-Agha, I., Styczynski, J., Montoto, S., Duarte, R. F., Kröger, N., Schoemans, H., Koenecke, C., Peric, Z., & Basak, G. W. (2020). How much has allogeneic stem cell transplant–related mortality improved since the 1980s? A retrospective analysis from the EBMT. Blood Advances, 4(24), 6283–6290. https://doi.org/10.1182/bloodadvances.2020003418
18. Polo, J. M., Liu, S., Figueroa, M. E., Kulalert, W., Eminli, S., Tan, K. Y., Apostolou, E., Stadtfeld, M., Li, Y., Shioda, T., Natesan, S., Wagers, A. J., Melnick, A., Evans, T., & Hochedlinger, K. (2010). Cell type of origin influences the molecular and functional properties of mouse induced pluripotent stem cells. Nature Biotechnology, 28(8), 848–855. https://doi.org/10.1038/nbt.1667
19. Richard, J.-P., & Maragakis, N. J. (2015). Induced pluripotent stem cells from ALS patients for Disease Modeling. Brain Research, 1607, 15–25. https://doi.org/10.1016/j.brainres.2014.09.017
20. Rizzi, R., Di Pasquale, E., Portararo, P., Papait, R., Cattaneo, P., Latronico, M. V., Altomare, C., Sala, L., Zaza, A., Hirsch, E., Naldini, L., Condorelli, G., & Bearzi, C. (2012). Post-natal cardiomyocytes can generate IPS cells with an enhanced capacity toward cardiomyogenic re-differentation. Cell Death & Differentiation, 19(7), 1162–1174. https://doi.org/10.1038/cdd.2011.205
21. Rofagha, S., Bhisitkul, R. B., Boyer, D. S., Sadda, S. R., & Zhang, K. (2013). Seven-year outcomes in ranibizumab-treated patients in anchor, Marina, and Horizon. Ophthalmology, 120(11), 2292–2299. https://doi.org/10.1016/j.ophtha.2013.03.046
22. Scesa, G., Adami, R., & Bottai, D. (2021). IPSC preparation and epigenetic memory: Does the tissue origin matter? Cells, 10(6), 1470. https://doi.org/10.3390/cells10061470
23. Si-Tayeb, K., Noto, F. K., Sepac, A., Sedlic, F., Bosnjak, Z. J., Lough, J. W., & Duncan, S. A. (2010). Generation of human induced pluripotent stem cells by simple transient transfection of plasmid DNA encoding reprogramming factors. BMC developmental biology. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2923111/
24. Soldner, F., Hockemeyer, D., Beard, C., Gao, Q., Bell, G. W., Cook, E. G., Hargus, G., Blak, A., Cooper, O., Mitalipova, M., Isacson, O., & Jaenisch, R. (2009). Parkinson's disease

patient-derived induced pluripotent stem cells free of viral reprogramming factors. Cell, 136(5), 964–977. https://doi.org/10.1016/j.cell.2009.02.013

25. Tsukasaki, K., Maeda, T., Arimura, K., Taguchi, J., Fukushima, T., Miyazaki, Y., Moriuchi, Y., Kuriyama, K., Yamada, Y., & Tomonaga, M. (1998). Poor outcome of autologous stem cell transplantation for adult T cell leukemia/lymphoma: A case report and review of the literature. Bone Marrow Transplantation, 23(1), 87–89. https://doi.org/10.1038/sj.bmt.1701533
26. Zvereva, M. I., Shcherbakova, D. M., & Dontsova, O. A. (2010). Telomerase: Structure, functions, and Activity Regulation. Biochemistry (Moscow), 75(13), 1563–1583. https://doi.org/10.1134/s0006297910130055