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Review Article

Stem Cells Advancement and Applications: A Regenerative Medicines

Priyanka Thakur *[1], Nirmala [1], Jaspreet Kaur [1], Piyush Kaushal [1], Abhimanyu Bhardwaj [1], Vinay Pandit [2]

[1] Rayat Bahra University, University School of Pharmaceutical Sciences, Mohali, Punjab, India-140103

[2] Department of Pharmaceutics, Laureate Institute of Pharmacy, Kathog, Kangra, HP, India- 176029

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*Address for Correspondence:

Priyanka Thakur, Rayat Bahra University, University School of Pharmaceutical Sciences, Mohali, Punjab, India-140103

Abstract

Innovative advancements in stem cell research have led to the development of organoids that serve as in vitro models for human organ development and disease studies. Developments in the culture of human pluripotent stem cells (hPSCs) have facilitated the creation of made tailored differentiation approaches, which have important uses in regenerative medicine. These advancements have enabled the implantation of hPSC-derived cell therapy products into patients, and the results of numerous ongoing clinical trials have been encouraging. A novel strategy for customized cell-based treatments for a range of human illnesses is ectopic expression of reprogramming factors, which allows adult somatic cells to be reprogrammed into induced pluripotent stem cells (IPSCs). The IPSCs technology is a useful tool for drug development and disease modelling, in addition to providing possible remedies. Similar to embryonic stem cells (ESCs), induced pluripotent stem cells (iPSCs) are capable of genetic correction and can develop into any type of cell in the body. These features offer IPSCs a possible path way for the development of long-term treatments for a wide range of diseases that are currently incurable. Additionally, we review the potential uses of IPSCs and clinical examination of future cell culture strategies for large-scale production to improve patient accessibility.

Keywords: Induced pluripotent stem cells (IPSCs), regenerative medicine, stem cell reprogramming, embryonic stem cells, and human pluripotent stem cell.

Introduction:

The discovery of self-renewal cells by living cells was one of the major breakthroughs, and was reported by Till and Mc Culloch in 1961. Pluripotent stem cells have an incredible ability to transform into any cell type, including brain and muscle cells. They could also maintain copying. This makes them extremely helpful to researchers studying diseases, generating new treatments, and even expecting to repair damaged tissue. There are certain ethical questions regarding the types of PSCs, but scientists are working on ways to avoid these obstacles. Induced pluripotent stem cells (IPSCs) are similar to pluripotent stem cells. They are formed from normal adult cells and can differentiate into other cell types. This makes them very intriguing for repairing objects in our bodies via regenerative medicine. [1] These stem cells are like master builders in our bodies, capable of developing into whatever cells we require. We have discovered a means to overcome major obstacles using induced stem cells, unlocking the door to promising new treatments, such as discovering entirely new world healing possibilities , since the first report on iPSCs by Yamanaka et al. in 2006 . Human pre-implantation embryos, such as morula or blastocyst stage embryos, were the only source of human IPSC in 2007.[2] Human IPSC scientists initially created mouse-induced pluripotent stem cells by introducing four critical genes, Oct4, SOX2, Klf4, and c-Myc, into a retrovirus. This breakthrough has paved the way for comparable methods. Following this achievement, human IPSCs were created by virally delivering reprogramming elements identical to adult human fibroblast cells. Scientists have transformed ordinary skin cells into a specific type of cell called induced pluripotent stem cells by introducing certain genes into them. Induced pluripotent stem cells can be produced from somatic cells. This process involves reprogramming mature, differentiated cells back into a pluripotent state, where they have the potential to develop into any cell type in the body. This technology holds significant promise for regenerative medicine, disease modeling, and drug discovery. The diversity of the resulting cell population is a problem with IPSC production, which may result from several variables, including changes in the initial somatic cell's genetic and epigenetic states, variances in reprogramming efficiency, and the random character of the reprogramming process itself. Because of this, distinct IPSCs in a culture might have distinct pluripotent

potentials or reactions to differentiation signals. They are produced from somatic cells by co-expression of a certain pluripotency-associated factors. These cells also give rise the gametes. Human ESCs are obtained using extra in vitro fertilization embryos and have specialized epigenetic landscapes that are important for pluripotency maintenance. There are several important characteristics of induced pluripotent stem cells (IPSCs) and embryonic stem cells (ESCs), including their capacity to differentiate into any type of cell and their application in the generation of chimaeras and teratomas. Induced pluripotent stem cells (IPSCs) exhibit full pluripotency and may develop into complete embryos and extraembryonic cell. These properties make IPSCs useful for drug testing, disease modeling, toxicity research, and cell therapy. These characteristics are extremely helpful in fundamental biomedical research. The transcription factor-mediated generation of IPSCs requires global changes in somatic cells.[3] However the limitations of using human tissue samples have been overcome by human IPSC-derived organoids, which preserve distinct cell types in an extended in vivo-like environment. Developments in the biology of the extracellular matrix (ECM) especially with respect to 3D laminin-rich matrices, have made it possible to observe in vitro morphology. Human IPSCs cultivated on ECM-based hydro gels have been successfully used to create diverse organ systems utilizing this culture approach. Methods for creating a variety of organoids, including the intestine, optic-cup, liver, cerebral, gastric, lung, nephron, ductal

, inner ear, cardiac, skin, and blood vessel organoids, have been established since the publication of this concept. Organoids are acknowledged as an important drug screening platform and have the potential to be used in regenerative medicine research to examine the effects of long-term alternative therapy.[4] Organoid technology has applications in many fields of study, including genome editing with tools called clustered regularly interspaced short palindromic repeats (CRISPR-Cas9), which offers important insights into genetic abnormalities . Organoids can also be employed in cancer modelling to learn more about the etiology, growth, and progression of cancer as well as coculture with viruses to research infectious diseases. Organoids are extremely useful for investigating the mechanisms underlying illnesses and human development. However, despite tremendous progress, many organoids produced from human induced pluripotent stem cells (IPSCs) remain immature in culture because of the absence of critical adult tissue characteristics. Consequently, it is critical to create reliable in vitro maturation techniques that consider the cellular and environmental elements required to create mature and functional organoids.[5]

Generation of Organoids

The self-organizing and self-patterning properties of homogeneous cell populations were leveraged to construct organoids. The completely differentiated cell types and spatial organization of the parent organ are replicated in these ex-vivo “mini-organs.” Proper consideration of various elements is necessary for the successful generation of organoids, including the extracellular matrix (ECM), which influences the final characteristics of the organoids, specific cell types of the organ or tissue, suitable culture conditions, and signaling pathways that govern differentiation.

Development of IPSCS-Derived Organoids

Numerous studies published since 2010 have detailed the creation of different IPSC-derived organoids that mimic human tissues. These organoids can be divided into three groups according to their differentiation mechanisms, which lead to their formation: ectoderm, mesoderm, and endoderm. Organoids that mimic the skin, inner ear, brain, and eyes are ectoderm-representing organoids. The Lancaster group demonstrated the potential to reproduce brain development by developing the first cerebral organoids culture system derived from IPSCs.[6] New avenues for research on early neural development and associated disorders have been opened up by cerebral organoids, which are composed of neurons and several glial cell types, including oligodendrocytes. Furthermore, an organoids of the human choroid plexus that can secrete cerebrospinal fluid has been created to forecast the permeability of pharmacological molecules in the central nervous system, which may be useful for brain homeostasis studies. Numerous organoids have been effectively created to replicate particular tissues and organs, demonstrating encouraging outcomes in medical uses. For example, retinal and corneal organoids containing light-responsive photoreceptor cells have been created, showing promising results in cell treatment. It has also been possible to generate ear organoids with cochlear hair cells and sensory neurons.[7] Furthermore, skin organoids that accurately mimic the intricate structure of the human skin have been created. Organoids that depict the kidney, heart, and blood vessels are created as we move on the mesoderm, one of the three germ layers. Extracellular matrix (ECM) and suspension culture techniques have been used to create and vascularise renal organoids, including nephrons, demonstrating breakthroughs in this field. Furthermore, new and innovative multi-organoids systems have been introduced, including hepatobiliary-pancreatic organoids and boundary organoids.[8] IPSC-derived mesenchyme- free organoids have also been created.

In Vitro Models for Developmental Study, Disease Modelling, and Testing

The creation of IPSC-derived organoids should, in basic terms, precisely match the development of human organs. Precise spatiotemporal signals and correct constructions are required for tissue construction and cell differentiation. Our understanding of human tissue and organ biology is greatly improved by organoids that function as 3D models. Compared with conventional two-dimensional (2D) culture and mammalian models, this technology offers a more realistic approach. Moreover, organoids have transformed the study of human development biology by providing a simple method for determining organ creation. Organoid systems, as opposed to traditional 2D models, can replicate histopathological features more faithfully by

assembling a variety of cell types. They also make it possible to use genome-editing technology to recapitulate hereditary diseases. Organoids can be directly co-cultured with pathogens in infectious disease research, thereby providing models for understanding the pathophysiology and processes of the disease. Organoid systems have a number of benefits that have been utilised in the investigation of numerous illnesses, including malignancies, host-pathogen interactions and genetic abnormalities.[9] For example, organoids made from induced pluripotent stem cells (IPSCs) specific to a patient accurately mimic the pathophysiology of humans. Owing to this fidelity, drug toxicity and efficacy at the tissue and organ levels can be predicted with greater accuracy, accounting for the wide range of clinical reactions observed in individual patients. For these investigations, disease-specific bio banks are essential sources of samples that enable the testing of new drug screening methods and precision medicine technique. Organoids, for instance, have aided in the development of tailored medicines for cystic fibrosis, testing of medications for Zika virus infection, and advancement of colorectal cancer treatments.[10]

Table No. 01 represents the clinical Trial Report of pluripotent stem cell's development of various cell line for the indication of different diseases in culture medium which was the published data by the researchers. Several disorders, such as age-related macular degeneration (AMD), Parkinson's disease, spinal cord injury, and heart failure, are now being treated in clinical trials using cells produced from human pluripotent stem cells as given in Table 1.

Table 1: Pluripotent Stem Cells Clinical Trials Report.

ID Number	Year	Cell Line	Derived Cell Type	Indication	Culture Medium	Scaffold	Ref
NCT01217008	2010	ESC(H7)	Oligodendrocyte progenitor cells	Spinal cord injury	Serum- free (KOSR)	Feeder cells (MEF)	[11]
NCT01345006	2011	ESC(MA0 9)	Retinal pigmented epithelial cells	Stargardt's macular dystrophy	Serum- free (KOSR)	Feeder cells (MEF)	[12]
UMIN000011929	2013	iPSC(autologous, episomal)	Retinal pigmented epithelial cells sheet	Exudative AMD	XF medium (Prime ES)	Feeder cells (autologou s)	[13] [14]
NCT02286089	2014	ESC	Retinal pigmented epithelial cells	Advanced dry AMD	Serum- free (KOSR)	Feeder cells (human fibroblast)	[15]
CT02239354	2014	ESC(CyT 49)	Pancreatic beta-cell precursors	Type 1 diabetes mellitus	XF medium (XF-KOSR)	Feeder- free (human serum)	[16]
NCT02057900	2014	ESC(16)	Cardiomyocytes	Severe heart failure	XF medium (Nutristem)	Feeder cells (human fibroblast)	[17]
NCT02923375	2016	iPSC(episomal)	Mesemchymal stem cells	Steriod-resistant Acute GVHD	XF medium (E8)	Feeder- free (vitronecti n)	[18]
NCT03119636	2017	ESC(Q- CTS-hESC-1)	Neural precursor cells	Parkinson's disease	XF medium (E8)	Feeder- free (vitronecti n)	[19]
UMIN000033564	2018	iPSC(QHJI01s04, episomal)	Dopaminergic progenitors	Parkinson's disease	AOF medium (StemFit)	Feeder- free (Laminin5 11E8)	[20]
UMIN000036539 (jRCTa050190084)	2019	iPSC(YZWJs524, episomal)	Corneal epithelial cell sheet	Limbal stem-cell deficiency	AOF medium (StemFit)	Feeder- free (Laminin5 11E8)	[21]

NCT04161 67	2019	iPSC(allo geneic, episomal)	Natural killer cells	Cancer	XF medium (FRM-FMM)	Feeder- free (vitronecti n)	[22]
UMIN0000 35074 (jRCTa031	2020	iPSC(YZ WJs513, episomal)	Neural stem/ progenitor cells	Spinal cord injury	AOF medium (StemFit)	Feeder- free (Laminin5	[23]
190228						11E8)	[24]
jRCTa5019 0104	2020	iPSC(QHJ I01s04, episomal)	Cartilage	Articular cartilage damage	AOF medium (StemFit)	Feeder- free (Laminin5 11E8)	[25]
NCT04945 018	2021	iPSC(QHJ I14s04, episomal)	Cardiomyocytes spheroits	Heart failure	AOF medium (StemFit)	Feeder- free (Laminin5 11E8)	[26]
NCT04696 328 (UMIN000 32989)	2021	iPSC(QHJ I14s04, episomal)	Cardiomyocytes sheet	Ischemic cardiomyop athy	AOF medium (StemFit)	Feeder- free (Laminin5 11E8)	[27] [28]

Human Pluripotent Stem Cell (hPSC) culture for Regenerative medicine:

Human pluripotent stem cells (hPSCs) can differentiate into cells from any of the three germ layers and can self-renew indefinitely. These cells are known as human embryonic stem cells (hESCs) and induced pluripotent stem cells. Human pluripotent stem cells can be directed to develop into desired cell lineages by adding particular growth factors and small compounds. A significant development in this area is the ability to generate multiple types of functionally differentiated cells, which are currently used as alternative cell sources for cell-based therapies, drug discovery, and disease modelling. Several disorders, such as age-related macular degeneration (AMD), Parkinson's disease, spinal cord injury, and heart failure, are now being treated in clinical trials using cells produced from human pluripotent stem cells (Table 1).[29]

The technique used to generate high-quality cells is a major factor determining the effectiveness of cell therapy. Allogeneic and autologous cells are the two main types of cell transplantation products. With scale-up procedures facilitating the creation of standardized treatments that guarantee consistent outcomes across a wide patient population, the allogeneic strategy concentrates on producing a large and stable output from uniform raw materials to treat multiple patients. By customizing the therapy for each patient, autologous transplantation, on the other hand, adopts a personalized medicine strategy. To provide the most individualized treatment possible, cells and tissues taken from a patient at a hospital are cultivated and then given back to the same patient at the same facility. The upstream and downstream processes are involved in the cell manufacturing process. Cell amplification, induction of differentiation, and live cell creation are examples of upstream processes. Cell isolation, purification, aliquoting, freezing, and packaging were all performed by downstream procedures. For cell treatment to be both safe and successful, standardized process technologies must be established. The most widely used of these procedures is cell culture, and improvements in this field should improve the safety and effectiveness of treatment. Previously, the early phases of hPSC production involved the use of feeder cells and serum-derived components. Successful pre-clinical and clinical research utilizing human pluripotent stem cells depends on ancillary materials (Ams). These components, which are not included in the formation of finished products during processing, include culture media, growth factors, cytokines, buffered solutions, and coated plates. Furthermore, human pluripotent stem cell lines meant for clinical applications require rigorous safety evaluations and comprehensive characterization through well-organized quality control processes for a more productive and economical process. Under current good manufacturing practice (cGMP)- compliant settings, several clinical-grade hPSC lines have been generated for use in clinical trials.[30]

Methods of Generation and Biomedical Applications of Human iPSCs:

A number of diseases that are presently incurable, such as heart infections, lung and kidney problems, diabetes, neurological diseases, and heart infections, may be treated in the future with the help of induced pluripotent stem cells (IPSCs). There are various biological uses for these cells to generate induced pluripotent stem cells (IPSCs), adult cells can be reprogrammed into pluripotent stem cells by the use of retroviral or lentiviral vectors. The biomedical applications of iPSCs are still in their early stages, but the potential of this technology is enormous. Induced pluripotent stem cells (IPSCs) originate from several technological applications. These technologies and applications are essential for furthering medical research and creating new therapeutics:

Integrating method*:* Integrating methods are the most common method for generating iPSCs; they are relatively efficient, with success rates of up to 50%. However, integrating methods cause safety concerns because the viral vector can integrate into the host cell's genome at random, potentially causing mutations. This can lead to cancer and other health problems. These techniques require the insertion of specific genes into the DNA of the body via a virus. The majority of the inserted genes are crucial for maintaining the ability to enter various cell types. Integrating methods are relatively efficient, but they have safety concerns because the viral vector can integrate into the host cell's genome at random, potentially causing mutations. To make iPSC-based treatments safer, significant efforts have been made to create cells without incorporating an external sequence into their genomes. These alternatives include proteins, or mRNA, rather than foreign DNA.[31]

Non-integration method: Kyto University CiRA frequently uses a secure episomal vector-based method to produce iPSCs. Because integrating techniques do not require introducing genes into the host cell genome, they are typically regarded as safer, producing induced pluripotent stem cells (iPSCs). However, these techniques have low success rates of up to 10%. Non-integrating techniques do not reduce the number of genes in the genome of somatic cells. Instead, genes are introduced into the cell using a non-viral vector, such as an mRNA or liposome. Non-integrating methods are generally considered safer than integrating methods; however, they are also less efficient. The Center for iPSCs Research and Application (CiRA) was founded. The first center is devoted to iPSC research and therapeutic development.[32]

Regenerative medicines*:* To develop new treatments for conditions such as heart disease, Parkinson's disease, and spinal cord injury, IPSCs can be used to generate various cell types. IPSCs have the potential to differentiate into diverse cell types including liver, heart, and neuronal cells. This versatility makes them a promising source for cell transplantation therapies targeting diseases such as Parkinson's, heart, and liver diseases. Sharing many regenerative properties with embryonic stem cells (ESCs), IPSCs are crucial for regenerative medicine and show significant potential for addressing numerous severe and life-threatening conditions. Recently, IPSCs-based clinical trials have been initiated to treat macular degeneration, Parkinson's disease, and heart disease.[33]

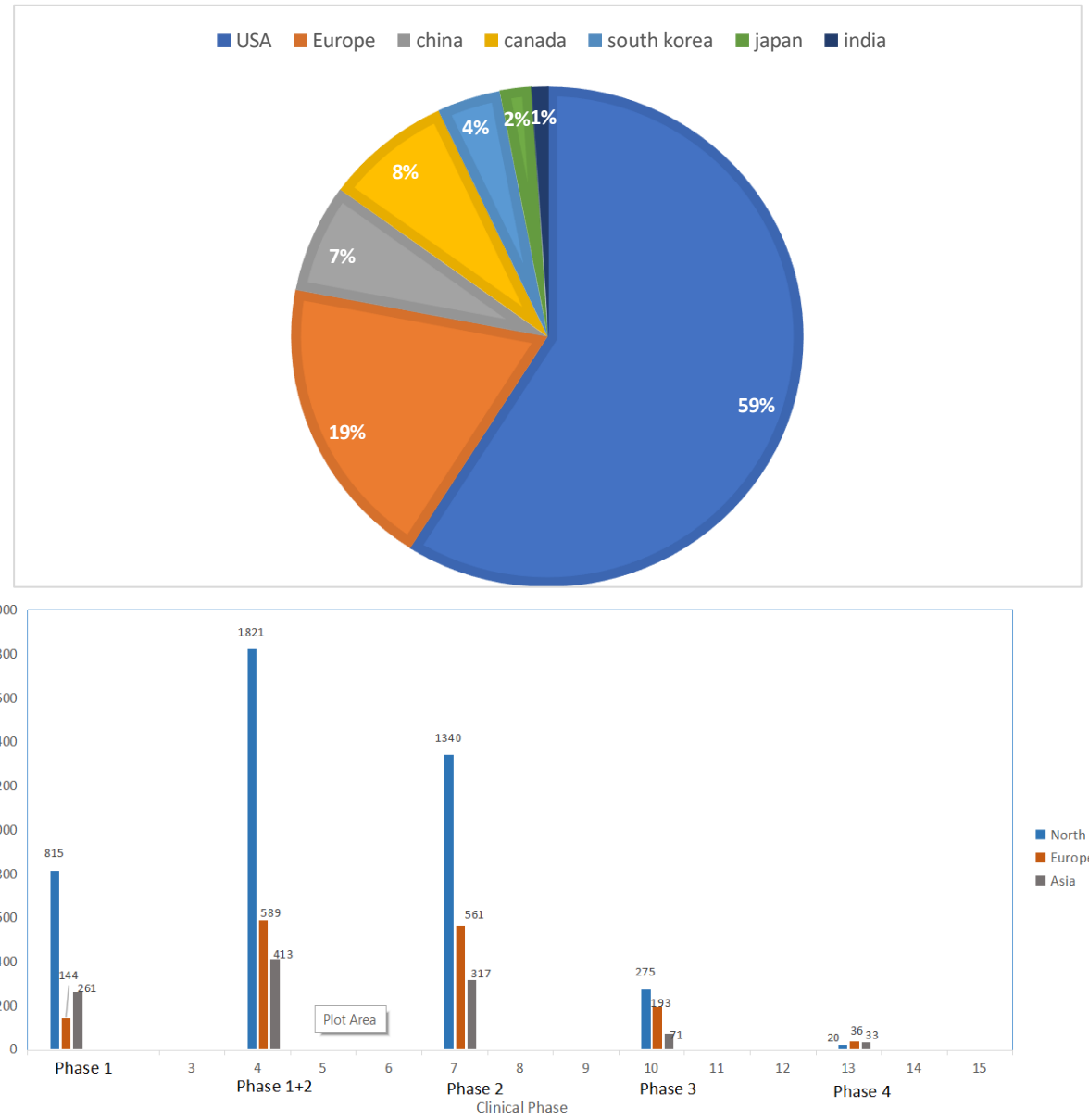
Graphical Representation of the relative number of Stem Cell Clinical Tails:

Table 02 represent the data on the number of clinical trials that include the term "stem cell" in major geographical regions were sourced on December 14.12.2017, from two primary databases: https://clinicaltrails.gov for North America and Asia, and the EU https://www.clinicaltrialsregister.euctr-search/search Clinical Trials Region for Europe.

Table 2: Clinical Trials of Stem cells of various Geographical Region

Country/ region	USA	Canada	Europe	China	Korea	Japan	India
Stem cell clinical trials up to 2018*	2360	277	788	326	160	27	70
Phase 1	766	49	144	153	50	11	47
Phase 1+2	1671	150	589	228	107	16	62
Phase 2	1211	129	561	183	82	8	44
Phase 3	184	91	193	39	17	8	7
Phase 4	18	2	36	24	9	0	0
Ongoing	908	117	574	141	54	11	12
Suspended	18	2	28	2	0	0	1
Terminated	255	22	108	2	9	1	4
Completed	1020	118	231	48	59	12	27
With results	418	51	107	4	6	6	2
Without results	1942	226	678	322	154	21	68

These figures encompass clinical studies, regardless of whether their phase or status was specified, and they also included studies that were terminated or prematurely ended.



(a) (b)

Figure 1: Clinical Trials Report in Phases of various Regions.

Figure 1 represents the relative number of stem cell clinical trials. (a) Geographic Region: Graph compares the number of stem cell clinical trials conducted in various regions around the world, such as North America, Europe, Asia, and other regions that are leaders in stem cell research. (b) Clinical Phase: This shows the number of studies at each stage of clinical trial process, including Phase I, Phase II, Phase III, and Phase IV.

Clinical Applications

IPSCs strategy has given us a capable apparatus to think about instruments of cell destiny choices and to show human infection. IPSCs have been employed in recent clinical studies to treat conditions such as Parkinson's disease, heart disease, and macular degeneration.[34] To fully realize the therapeutic potential of IPSCs in a wide range of diseases, ongoing advancements in these technologies are essential. This includes the development efficient cell replacement therapies for diseases affecting the kidneys, heart, and the nervous system.

Cell Replacement

The potential to differentiate into different cell types including neurons, heart cells, liver cells, and blood cells, is possessed by induced pluripotent stem cells (iPSCs). They can replace lost cells as a result of illness, trauma, or aging. As a result, iPSCs have great potential in regenerative medicine, especially for the replacement of diseased or damaged cells in certain organs. With continued research and development, cell replacement using iPSCs holds immense promise for the treatment of a wide range of currently incurable diseases. The ability of scientists to effectively transfer therapeutic cells and iPSCs or progenitor cells produced from iPSCs to target tissues while maintaining their complete functionality and viability presents a substantial hurdle in bringing this promise to practical reality. While challenges remain, the field is rapidly advancing, holding monstrous potential for revolutionizing healthcare in the future.[35]

Certain compounds that can substitute conventional reprogramming factors have made it possible to manufacture induced pluripotent stem cells in recent years. Furthermore, reprogramming factors possessing the ability to penetrate cells can be used to create IPSCs, enabling these proteins to cross the cell membrane effectively. The length of the reprogramming procedure affects the properties resulting IPSCs. In the process of being cultivated, IPSCs often take on characteristics that are closer to those of embryonic stem cells (ESCs) than to early-stage reprogramming cells. This implies that even after IPSCs are first generated, reprogramming occurs.

Cell delivery:

There are currently two basic ways to transport IPSCs and their progeny to internal organs: (a) injecting the cells intravenously and hoping they travel to the target region or (b) administering the cells locally, either by placing a catheter or by performing open surgery. This involves administering differentiated iPSCs and their derivatives to the target site in the body to treat various diseases or conditions. In contrast, cells are usually injected directly into the buffer for systemic administration. However, this approach frequently results in a considerable percentage of cell death. Cell delivery methods for differentiated PSCs vary depending on the target tissue and desired therapeutic effect; some common routes include:

(a) Injection: directly injecting differentiated cells into the affected tissue (e.g., intra cerebral injection for neurodegenerative diseases)

(b) Encapsulation: encapsulating cells in biocompatible scaffolds or hydrogels for targeted delivery and sustained release (e.g., cartilage regeneration)

(c) Micro-fluidic: utilising micro-fluidic devices for precise and controlled delivery of cell suspensions to specific tissue locations.

Cell Survival and Function:

Induced pluripotent stem cells, or IPSCs, can be produced from a variety of cell types, each of which has a distinct reprogramming efficiency. They consist of stomach epithelial cells, hepatocytes, blood cells, melanocytes, and keratinocytes. However, because they are readily available and easy to cultivate, fibroblasts are the most commonly used cell type, and they undergo a systemic reprogramming process that starts with the down regulation of somatic expression. To decrease stem cell apoptosis and promote the survival of iPSC- derived cells after transplantation, it is critical to create a supportive environment that attenuates the inflammatory response. It is essential to address the variables that affect cell survival and function to successfully design and apply IPSC-based treatments. Understanding and addressing the factors influencing cell survival and function are crucial for the successful development and implementation of PSC-based therapies. By optimizing culture conditions, manipulating signalling pathways, and employing genetic and epigenetic tools, researchers are continuously refining strategies to harness the therapeutic potential of these versatile cells while mitigating the associated risks. Alternatively, by altering the delivery structure with appropriate signalling molecules, it may be possible to mimic the crucial roles that specialized cells play in a particular environment.[36]

Dermatology

Skin is the perfect tissue for testing new iPSC-based treatments, and it is easily monitorable, highly proliferative, and accessible. IPSCs have been used to generate many skin cell lineages, including keratinocytes, melanocytes, fibroblasts, and ectodermal precursor cells. Researchers have shown that Mouse IPSC-derived keratinocytes can be transplanted to regenerate different epidermis and skin appendages with properties resembling those of normal keratinocytes by using mouse fibroblasts in athymic mice. Furthermore, it has been demonstrated that human IPSC-derived keratinocytes may generate functional organotypic skin in 3D models and cultures. These studies demonstrate that IPSCs can generate autologous donor cells for use in cell-based therapies for skin conditions. In vitro 3D skin mimics have also been prepared using IPSC-derived components, which is an important step towards their eventual clinical application. These opponents may be employed for drug screening because they exhibit typical skin morphology, stratification, and terminal differentiation. Genetically correct iPSC-derived cells have the potential to cure several of the most extreme mutation-induced hereditary skin disorders, including epidermolysis bullosa (EB). Genetic blistering, known as EB, may cause severe blistering and scarring of the skin. To generate biologically corrected pluripotent stem cells, particularly for each patient, precise gene editing methods can be employed, such as those based on CRISPR/Cas-associated systems. After modification, patients who require induced pluripotent stem cells (IPSCs) can receive them again after they develop into skin cells. Numerous illnesses affecting other organs can be treated using comparable therapies (Fig. 2).[37]

The Cell treatment techniques that use IPSC-derived cells entail extracting patient- specific primary cells from diverse organs, growing them *in vitro*, and reprogramming them into induced pluripotent stem cells as given in Figure 02.

These IPSCs can be genetically altered if necessary. The repair of unaltered IPSCs clones are subsequently differentiated into the specific cell types. These differentiated cells can be used for autologous transplantation or to create novel treatment techniques, such as evaluating new medication modalities.

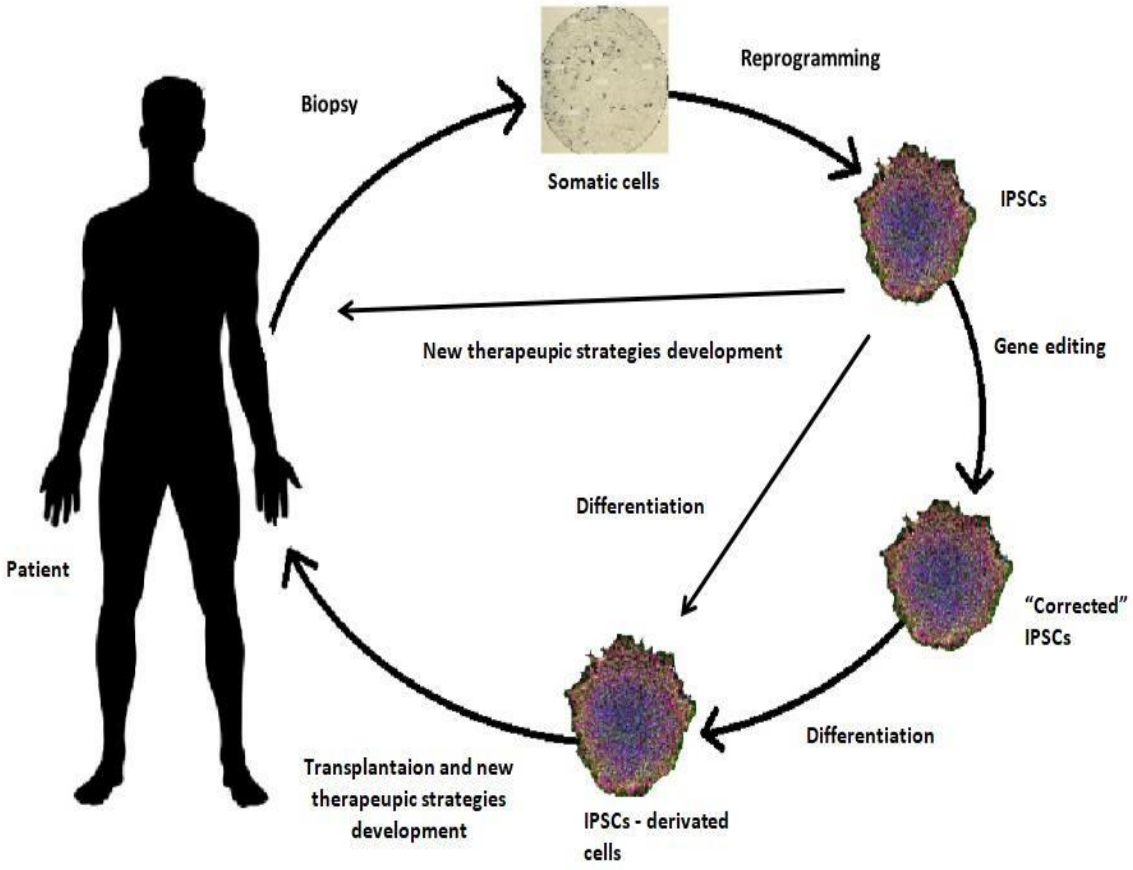


Figure 2: Cell Treatment Technique (IPSC derived Cell).

Cardiology

Novel strategies for cardiac repair have been made possible by recent advancements in the reprogramming of induced pluripotent stem cells (IPSCs). The ability to convert IPSCs into cardiomyocytes and other cardiac cell types presents a promising avenue for the treatment of heart diseases. Cardiomyocytes, similar to cells generated from iPSCs, are spontaneously contractile and resemble cardiomyocytes in both structure and molecular makeup. The application of induced pluripotent stem cells (IPSCs) to restore damaged heart tissue following a heart attack has been studied by researchers. This is significant because heart disease ranks among the world's leading causes of illness and mortality. Overall, a billion cardiomyocytes can be destroyed in patients with major myocardial infections beyond the ability of the heart to heal it-self. Heart failure is the result of Significant cell loss in the myocardium, which initiates the replacement of cardiomyocytes with fibrous tissue. Researchers have established overcoming cardiomyocytes from induced pluripotent stem cells (IPSCs) obtained from individuals with hypertrophic cardiomyopathy associated with diastolic dysfunction to conduct research on the molecular causes and possible therapeutic targets of diastolic failure. Additionally patient-specific iPSC-derived cardiomyocytes have been created and used to simulate and research the genetic and molecular mechanisms underlying diabetic cardiomyopathy in individuals with different types of diabetes. Human IPSC-derived cardiomyocytes, endothelial cells, and cardiac fibroblasts have been used to create three-dimensional (3D) cardiac microtissues that mimic various cardiovascular diseases. Few clinical trials have been conducted to determine the effectiveness of cardiomyocytes produced from iPSCs. Numerous clinical trials of IPSC- derived cardiomyocytes have been conducted to assess their efficacy. For instance, a clinical trial (NCT04696328) was launched by Cuorips, Inc., an Osaka University spin-off, to evaluate the safety and effectiveness of human allogeneic IPSC-derived cardiomyocyte sheets for the treatment of patients with ischemic cardiomyopathy. Heart seed, Inc., is currently conducting phase 1 and 2 trials (NCT04945018) in which patients with serious cardiac failure are administered iPSC-derived cardiomyocyte spheroids for testing. As both inquiries are still in progress, no outcomes have been revealed. Moreover, human iPSCs have been created using cells obtained from patients to examine ventricular and atrial arrhythmias, which frequently result in abrupt cardiac death.[38]

Urology and Nephrology

Renal failure is a serious health concern worldwide that has a substantial impact on both mortality and chronic illness. Through iPSC-based therapy, a kinetic alternative to kidney transplantation can be provided. IPSCs have been shown to effectively differentiate into renal progenitor cells and nephrogenic intermediate mesoderm. Many cell types are involved, and kidney organoids resembling nephrogenesis have been produced using induced pluripotent stem cells. These cells can differentiate into multiple cell types that make

up the adult kidney, in addition to the nephric duct, ureteric bud, proximal tubular cells, and mature glomerular podocytes. There are multiple cell types, and nephrogenesis-like kidney organoids obtained from induced pluripotent stem cells have been generated. These organoids hold promise for regenerative medicine and customized treatment. Using iPSC-derived cells, recellularization of decellularized kidney scaffolds is an additional technique for creating human replacement kidneys. Scientists have used induced pluripotent stem cells (IPSCs) from patients with certain kidney diseases to create disease models.[39]

Neurology

Induced pluripotent stem cells (IPSCs) from humans and mice have been used to generate multipotent neuronal progenitors as well as a variety of well-differentiated neural cell types. Studies using these progenitors for cell replacement therapy in mouse models have yield encouraging results. Together with brain microvascular endothelial cells that are also produced from human IPSCs, the spinal cord chip system simulates vascularized human motor neuron tissue in vitro. Similarly, the development of blood-barrier chip technology has enabled drug screening and the modelling of neurological diseases.[40] An isogenic in vitro model can be produced by generating cells in both systems from the same iPSC donor source. In vitro developmental therapies and preclinical approaches for Neurodegenerative research are being increasingly developed using iPSCs. Parkinson's disease is a common neurological disorder that causes the death of dopamine neurons in the substantia nigra. Transplanting brain progenitors produced from induced pluripotent stem cells (IPSCs) is a potential treatment strategy. Clinical research in Japan is exploring the use of dopaminergic neurons generated from IPSCs as a treatment for Parkinson's. In 2018, a patient showed improved clinical symptoms 18-24 months after receiving implantation of autologous IPSC-derived midbrain dopaminergic progenitor cells. Aspen Neuroscience is developing two iPSCs based therapeutics: a gene-corrected dopaminergic neuron therapy obtained from autologous iPSCs for hereditary Parkinson's disease, and a dopaminergic neuron treatment developed from autologous iPSCs for idiopathic PD.[41]

Future Perspectives:

Human induced pluripotent stem cells have high potential for personalized treatment via autologous cell transplantation because they can be produced from any individual.[42] However, experiences with autologous CAR-T cell production have highlighted problems such as high manufacturing costs and the need to develop a stable supply chain. Consequently, the widespread adoption of autologous IPSC-derived cell transplantation may be time-consuming. A more viable strategy would be multiple organizations to establish HLA-matched IPSC banks. Despite this, creating a sufficient number of HLA homozygous lines requires significant donor support and resources. Efforts are currently underway to commercialize universal allogeneic transplantation, which has the potential to treat multiple people using cells from a single donor source.[43] The primary impediment to successful allogeneic translation is host rejection, which is predominantly caused by the immunological response of T cells. Recent research has focused on overcoming T-cell-mediated immune responses by targeting certain genetic pathways.[44] Studies have examined techniques such as knocking down beta-2 micro-globulin (B2M) and class II Trans activator (CIITA) genes. The B2M gene is critical for displaying HLA class I molecules on the cell membrane surface. These molecules are responsible for presenting antigens to CD8+T cells, which triggers an immunological response against non-self-cells.[45, 46] Donor cells may avoid being recognized and attacked by the recipient CD8+T cells by removing B2M gene expression. However, through a process called "missing self," donor cells lacking HLA class I molecules can activate natural killer (NK) cells, including innate immunological responses and ultimately eliminate these cells.[42] With less immunological rejection and better donor-recipient cell compatibility, these developments hope to increase the viability and success of allogeneic cell. NK cell activation can be suppressed by CD47, which donor cells have used to produce HLA-E-modified genes to lessen the "missing self" response. Furthermore, methods utilizing CD64 over expression to elicit antibodies have demonstrated potential for surmounting immunological rejection directed against small antigens.[47] It is still essential to ensure that these techniques are safe, and more clinical studies are needed to shed light on this issue. To achieve allogeneic transplantation at a reasonable cost in the future, scalability issues and strict quality control of manufacturing procedures must be addressed.[48, 49] A large number of clinical-grade stem cells, usually between 10^8 and 10^11 cells, must be cultivated following cGMP guidelines for use in human clinical therapy. However, approximately 10^10 cells are required for manufacturing on a commercial scale. Platforms that are stable, manageable, and expandable have been suggested by recent developments in cultural systems. Because they do not require micro carriers and streaming downstream procedures, suspension culture-based techniques have received special attention for their effective cell production. However, some issues must be resolved, such as unchecked spheroid aggregation and possible shear stress-related cell quality erosion. These problems may result in apoptosis, inhibition of cell development and variations in cell purity and quality.[50]

Conclusion

Rapid developments in cell reprogramming technology have opened up new avenues for drug screening, disease modelling, and regenerative medicine. Advancements in stem cell research hold tremendous potential for regenerative medicine, paving the way for new treatment and cures for various diseases and injuries. These include the creation of personalized therapies for heart disease, diabetes, and neurodegenerative disorders, as well as the ability to grow organs and tissues for transplantation. Only a small number of clinical trials involving the transplantation of iPSC-derived cells have been conducted thus far, and the development of iPSC-based

therapeutics is still in its early phase. Despite this promise, challenges persist, such as ethical concerns, the risk of immune rejection, and ensuring the safety and effectiveness of treatments. Continued research and cooperation among scientists, clinicians, and policymakers are crucial to overcoming these hurdles and integrating stem cell therapies into standard medical practice. In summary, stem cell advancement are revolutionizing regenerative medicine, providing hope for previously untreatable conditions and greatly enhancing patient's quality of life. Ongoing innovation and diligent research are essential to fully unlock the potential of these therapies.

Abbreviations

ESCs: embryonic stem cells

IPSCs: induced pluripotent stem cells hPSC: human pluripotent stem cell

hiPSCs: human induced pluripotent stem cells ECM: extra cellular matrix

hESCs: human embryonic stem cells AMD: age-related macular degeneration PD: Parkinson's disease

Ams: ancillary material EB: epidermal bullosa 3D: three dimensional

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