Review Paper

Bioethical Issues in Stem Cell Research

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ABSTRACT

A rapid growth and progress in the field of biotechnology have also added a number of ethical and policy issues particularly related to stem cell research. The stem cells have the potential to differentiate into different types of specialized cells and tissues in our body. The human derived stem cells are pluripotent having a great potential to be used as regenerative medicine and cell replacement therapies useful for the treatment of many diseases. This possibility is more effective when these stem cells are derived from human embryos. It is important that the embryonic stem cells must be obtained from early human embryos. It means that a potential human life needs to be destroyed for saving the life of another person. This raises a number of ethical issues and as we proceed towards the future in stem cell research closer to clinical use. There will be more ethical issues arising related to accessibility to the patients and its efficiency. In this article we have discussed the earlier and the current status covering the most significant bioethical issues related to stem cell research.

Keywords: Bioethics, human embryonic stem cell, rregenerative medicine, stem cell research, ethical issues.

INTRODUCTION

Ethics and Bioethics

Ethics is the idea of right, and wrong behaviour and accepted moral standards. It is applicable to all sorts of business, accounting and customer service [1]. Bioethics is the word made up of two parts. The word, "Ethics" deals with ethical issues and questions like, "What should we do? Are all things considered?" and the word, "bio" refers to life. In general, it deals with the application and conclusion that can be drawn from the health-related life sciences. Bioethics is not restricted only to local affairs that can be solved by a local society, but are global issues which shows its effect worldwide [2].

Stem Cells

Stem cells are the cells of the body that divide and differentiate into other types of cells and take specific functions. All stem cells have three general properties, (i) they are unspecialized, (ii) have self-renewal ability for longer periods and (iii) give rise to specialized cell types during cell division. [3]. they exist both in embryos and adult cells. The stem cells of embryos are called embryonic stem cells, and the stem cells of adult tissues are called adult stem cells [4]. Stem cell therapy is commonly known as regenerative medicine. It deals with the repair mechanism of diseased, dysfunctional or injured tissue with the help of stem cells or their derivatives. As per the Recent research human stem cells can differentiate into muscle cells, nerve cells, heart cells, blood cells and other types of cells. It has widened applications in the field of healthcare. For example, it can be used to produce replacement cells and tissues to cure several diseases and conditions, like Parkinson's disease, Alzheimer's disease, leukemia, stroke, heart disease, diabetes, multiple sclerosis, rheumatoid arthritis, spinal cord injury and skin conditions, including burns, etc. Prior to their human trials, certain noble drugs were being tested on the cultured liver and skin cells for their safety and efficacy. It also improves our knowledge of normal human development [5].

Classification of stem cells

Earlier stem cells were classified under three broad categories namely embryonic stem cells and adult stem cells as described below.

A) **Embryonic stem cells (ESC)**: These cells are derived from inner cell mass of an embryo at the blastula stage.

B) **Adult stem cells**: These types of cells are derived from particular tissues like:

(i) Mesenchymal stem cells: present in many tissues. In the bone marrow, these cells differentiate mainly into the bone, cartilage, and fat cells. As stem cells, they are multipotent but exceptionally they act as pluripotent cells and can specialize in the cells of any germ layer.

(ii) Neural stem cells: give rise to nerve cells and their supporting cells—oligodendrocytes and astrocytes.

(iii) Hematopoietic stem cells: form all kinds of blood cells- red, white, and platelets.

(iv) Skin stem cells: form all types of skin cells e.g., Keratinocyte, which form a protective layer of skin.

C) **Induced Pluripotent Stem Cells**: These are genetically reprogrammed adult somatic cells converted into ESC's like cells [6]. (Figure 1)

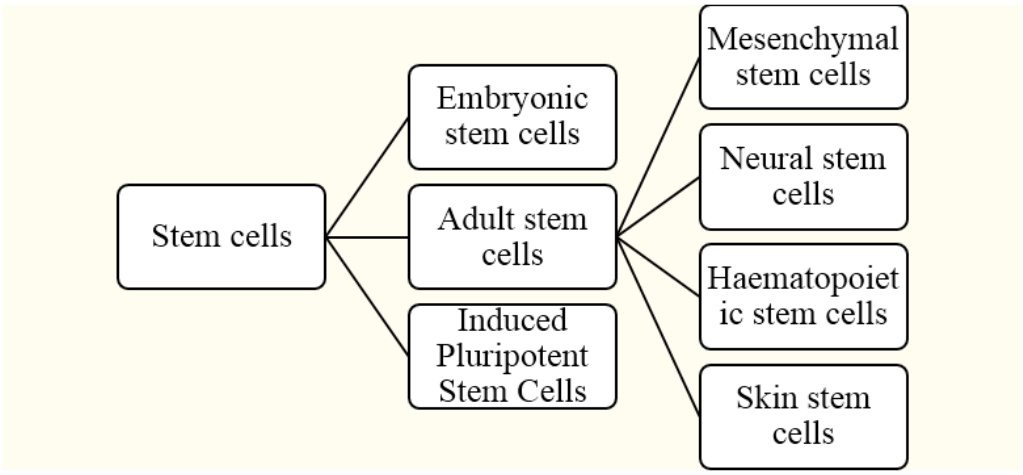

Figure.1. The types of stem cells based on their origin (Ilic, D., & Polak, J. M. (2011). *Stem cells in regenerative medicine: introduction. British Medical Bulletin, 98(1),*

This classification was considered insufficient as new research has discovered how to turn fully differentiated adult cells back into embryonic stem cells and vice versa.

Based on their differential potential, a new classification had been proposed to study the stem cells systematically. These classifications include five categories of stem cells [6-7]. (Figure 2):

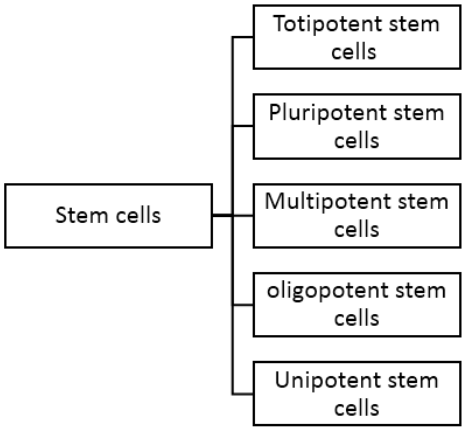

Figure.2. The flow chart showing Classification of stem cells on the basis of their differential potential (Ilic, D., & Polak, J. M. (2011). *Stem cells in regenerative medicine: introduction. British Medical Bulletin, 98(1), 117–126.*) (Smith, A. A glossary for stem-cell biology. *Nature* **441,** 1060 (2006)

a) **Totipotent stem cells**: These are the undifferentiated cells and are found in early development. These are omnipotent in nature and have the capacity to divide and differentiate into cells of the whole organism.
b) **Pluripotent stem cells (PSCs)**: Pluripotent stem cells are able to differentiate into cells that arise from the 3 germ layers — ectoderm, endoderm, and mesoderm – from which all tissues and organs develop. These form the cells of all germ layers, but not extra embryonic structures, such as the placenta. [6]
c) **Multipotent stem cells**: Multipotent stem cells are found in most tissues and differentiate into cells from a single germ layer. These can differentiate into specialized cells of specific cell lineages. For example- Hematopoietic stem cells, which can develop into several types of blood cells. [3]
d) **Oligopotent stem cells**: Oligopotent stem cells are able to self-renew and form 2 or more lineages within a specific tissue. For example- A neural stem cell that can create a subset of neurons in the brain. [7]
e) **Unipotent stem cells**: These cells are only able to differentiate into one cell type. For example- Spermatogonial stem cells [7].

History and Evolution of stem cells

Researchers have discovered number of ways to derive embryonic stem cells from early mouse embryos nearly three decades ago and were tried for treating infertility in human beings by way of in vitro fertilization procedures. The details of evolution of stem cells and their utility is presented in table 3 [8].

Table 3. Showing history and evolution of stem cell research

No.	Year	Scientist Name	Event
1.	1981 - Mouse beginnings	Martin Evans	Identification of embryonic stem cells in mice
2.	1997 - Dolly the sheep	Ian Wilmut	First artificial animal clone – Dolly the sheep
3.	1998 - Stem cells in human	James Thomson and John Gearhart	Isolation of human embryonic stem cells.

4.	2001 - Bush controversy	US President George W. Bush	Limits the funding on human embryonic stem cell research.
5.	2006 - Cells reprogrammed	Shinya Yamanaka	Creation of embryonic-like cells from adult cells forming "Induced pluripotent stem cells"
6.	2007 - Nobel prize	Evans, Mario Capecchi, and Oliver Smithies	Shares the Nobel prize for the work on genetics and embryonic stem cells.
7.	2009 - Obama	President Barak Obama	Lifts 2001 restrictions on funding for human embryonic stem cell research.
8.	2010 - Spinal injury	Geron	Spinal injury treatment by the use of human embryonic stem cells as a part of the trial.
9.	2012 - Blindness treated		Promising the role of Human embryonic stem cells in the treatment of blindness.
10.	2012 - Another Nobel	Shinya Yamanaka and John Gurdon	Shares Nobel prize for creating induced pluripotent stem cells.
11.	2013 - Therapeutic cloning	Shoukhrat Mitalipov	Produced human embryonic stem cells from foetal cells using therapeutic cloning.
12.	2014 Pre-embryonic state	Charles Vacanti and Haruko Obokata	Discovery for the conversion of any cell into the pre-embryonic state.
13.	2014 Therapeutic cloning – with adult cells	Dieter Egli	Production of human embryonic stem cells from adult cells, using therapeutic cloning.
14.	2014 Human trials	Masayo Takahashi	To treat age-related blindness using induced pluripotent stem cells.
15.	2015 Dental Stem Cells	Ophir Klein [9]	Stem cells extracted from fossiled rodent teeth to grow fully functional new teeth
16.	2016 PSC-derived brain organoids	Otani et al. [10]	3D brain organoids from human embryonic Stem cells, induced pluripotent stem cells and nonhuman primates used to identify species specific differences
17.	2018 clonal evolution (CE) model and the cancer stem cell (CSC) model	Lucei Laplane [11]	Cancer stem cells modulate patterns and processes of evolution in cancers
18.	2019 Co-evolutionary and systemic study on the evolution of emerging stem cell-based therapies	Ávila-Robinson, Alfonso & Islam, Nazrul & Sengoku, Shintaro [12]	Therapeutic technologies understanding, diagnosis, and treatment of human diseases

Applications of stem cells in Regenerative Medicine

The application of stem cells in clinical research for the treatment of various disorders in humans is progressing rapidly, and a number of stem cell-based therapies are already in clinical use. It suggests that stem cell transplantation also has potential as a therapy for neurodegenerative diseases. Clinical

trials are in progress involving grafting brain tissue from aborted foetuses into patients with Parkinson's disease, and Huntington's disease [13]. However, the application of stem cells in the treatment of certain disorders such as Alzheimer disease, Multiple sclerosis, diabetes, heart diseases and others are quite promising, which improves the possibility of treating such diseases in humans [14]. In regenerative medicine, it deals with the functional re-modeling of particular type of tissue or organ from the patients encountered with severe injuries or chronic diseases because of the ability of stem cells to differentiate into other cell types and indefinite cell division potential. One such example is Heart scars formed after heart attack, and liver cirrhosis which can be treated from Mesenchymal stem cells (MSC's). Krabbes disease are also known to be cured by transplanting umbilical cord stem cells which can give rise to myelin tissues. The patients with neuroblastoma are reported to be treated by maintaining tissue homeostasis. Various tissues such as craniofacial tissue, brain tissue, diaphragm tissue, and liver tissue have been reported to be regenerated by using bone marrow stem cells [15].

Merits of stem cells

- They can be self-renewed indefinitely, therefore can be used in laboratories.
- Stem cell can provide unlimited source of new human tissue.
- A million of cells can be produced in lab from a small starting number of cells.
- To use stem cell for treatment of patients with inherited genetic disease (e.g.: cystic fibrosis).
- Useful in finding out the mechanism of disease development.
- Useful in understanding and curing complicated diseases like cancer.

Demerits of stem cells

- 90% of the drugs fails during the clinical trials, and not approved for use, making this lengthy process costly and inefficient.
- Cell culture models used are not very similar to the tissue of interest that is the tissue that the disease affects.
- Tissue generated this way would be genetically different from that of a patient. The risk of tissue rejection by the patient immune system would be high, requiring other drugs to suppress their immune system after transportation.
- Embryos are still being harmed and killed as the majority of ES cells research is privately funded.
- Life begins at fertilization and any research that facilitates embryo destruction considering human stem cell research as unethical.

Recent Advances in the field of stem cell research

The following advancements in treatment of incurable diseases using different types of stem cells have been reported.

(i) Stem cell in cancer treatment

Scientists have employed different strategies to make use of stem cells in combating cancer. They were modified to express anti-cancer effector proteins such as pro-apoptotic and anti-proliferative proteins or express anti-angiogenesis factors to limit cancer cells' blood supply and create a non-supportive micro environment for the tumour. Another way of treating cancer is by the modification of stem cells such as Mesenchymal Stem Cells that show tumour-tropic properties that stimulate immune response against cancer. For example, human MSCs were modified to produce IL-12 and IL-18, which was reported to have an immune response against cervical cancer, renal cell carcinoma and glioblastoma through the activation of natural killer cells and tumour-specific T cells in mice [16].

(ii) Stem cell-mediated suicide gene therapy

Inactive forms of anticancer drugs are converted into active forms only at the tumour site by genetically engineered stem cells, this process is known as stem cell-mediated suicide gene therapy.

Various suicide gene systems utilized are:

a) Toxic anti-metabolite 5-fluorouracil produced from conversion of 5-fluorocytosine via cytosine deaminase enzyme.

b) Inactive irinotecan drug converted into topoisomerase inhibitor through cytosine deaminase enzyme.

c) Ganciclovir drug is converted to its phosphorylated forms which blocks the DNA synthesis via thymidine kinase enzyme.

These systems in mouse model has shown to be efficient in the treatment of brain, breast, ovarian, and prostate cancer [16].

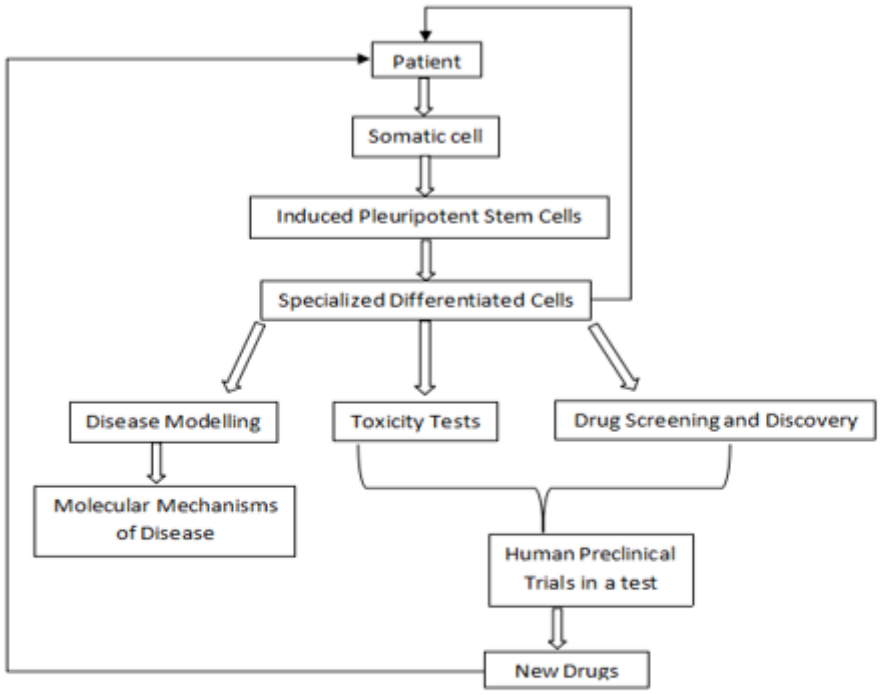

Figure.3. Showing conversion of human somatic cells to differentiated stem cells and their utilization for understanding the molecular mechanism of the disease and developing into new drug cure.(Beckmann, A., Schubert, M., Hainz, N., Haase, A., **Martin, U.,** Tschernig, T., and Meier, C. 2016. Ultrastructural demonstration of Cx43 gap junctions in induced pluripotent stem cells from human cord blood. *Histochem Cell Biol* 146, no. 5:529.)

(iii) Stem cells as delivery vehicles for oncolytic viruses:

Oncolytic viruses have displayed selective growth on tumours, since these viruses replicate within the cancer cells. The therapeutic efficacy of these viruses for the lysis of cancer cells might get reduced due to host immune system. The use of stem cells as a delivery vehicle for these viruses showed promising results in pre-clinical studies e.g. the mesenchymal stem cells can serve as effective carrier of oncolytic viruses to hepatocarcinoma in mice and ovarian tumour in humans. In 2014 oncolytic herpes virus was injected in mesenchymal stem cells which increased the anti-tumour efficacy of virus when compared to direct injection of these oncolytic herpes simplex virus in a mouse model. Talimogene laherparepvec virus was reported to be the first accepted oncolytic virus by US FDA, for the treatment of melanoma. A recent clinical study revealed that the combination of using T-VEC oncolytic virotherapy with immunotherapy has an increased drift of T cells towards tumour vicinity. A group of researchers managed to edit HLA genes in human Embryonic Stem Cells (hESCs) in vitro using zinc-finger nucleases, enabling the ESCs to evade the HLA-restricted cytotoxic T-lymphocytes [16].

(iv) Stem cell-based therapy in fighting cancer
In 2013, HLA gene editing was successfully done in human embryonic stem cells by using zinc-finger nucleases, which enabled the embryonic stem cells to escape the HLA-restricted cytotoxic T-lymphocytes, indicating the way towards generating universal cells from allogenic donors [16].

(v) Stem cells against neurodegenerative disorders.
Clinical trials for the treatment of neurodegenerative diseases like Parkinson's disease by the use of stem cell has witnessed a new era for stem cell therapy. The Dopamine neurons which have been used to treat Parkinson's disease could now be derived from pluripotent stem cells. Neural stem cells derived from embryonic stem cells are used to restore the function of damaged nervous system. There are experimental evidences for the derivation of neural stem cells from the neonatal rat's hippocampus which were implanted in the brain of Alzheimer's disease rat models. This neural stem cells were found to be able to differentiate into new cholinergic neurons with improved ability of spatial learning and memory to counter the Alzheimer's disease [16].

Ethical Issues in Stem Cell Research

Stem cell research is surrounded by many ethical questions because it uses human embryonic stem cells which require the destruction of the embryo, but it is not particularly related to embryo only, as the production of chimeras has raised many ethical issues. Embryos do have moral status which is critical but not completely problematic [17]. Another ethical aspect of embryonic stem cell is the safety and informed consent of both donor and recipient during oocyte harvesting and stem cell products respectively. The responsible conduct, justice of research, and also, its marketing process is a concern to ethicists to minimize ethical issues, distress, pain and suffering, various guidelines for ethical conducts have been made by professional groups [17-18].

Controversy related to use of embryonic stem cells

Controversy related to use of embryonic stem cells Stem cell research is a controversial topic considering it both as ethical and also unethical. It is considered ethical because great advances could be made toward the goal of relieving human suffering, but on the other side it is unethical as there is major use of human embryos for achieving the goal and the use of adult stem cells, and umbilical cord blood stem cells are beyond any ethical issues [13-14]. As human embryonic stem cell (hESCs) research involves destruction of human embryos making it the point of political and ethical controversy. Different people have different views regarding embryo's morality like in further stage of development embryo develops into a normal human being, whereas, for others embryo or blastocyst is just a cluster of cells that can be used for research purposes [18].

There are Opponents of Embryonic Stem Cell Research having various arguments against the use of human embryonic stem cell (hESCs) for their use for research and as therapeutics declaring it as unethical. Some strong arguments against its use are:

- Adult human beings are developed from continuous growth of an embryo. Some scientists believe that the isolation of embryonic stem cells from an embryo is unethical, because its isolation causes the death of embryo, which is considered as loss of life.
- The research on embryonic stem cells is considered as illegal by some bioethicist because its isolation causes the destruction, discard and risk of injury or death of an embryo. In 2001, the president of United States banned the use of embryonic stem cells for research and therapeutic purposes.

CONCLUSION

Stem cells are the cells of the body that divide and differentiate into other types of cells and take specific functions. Bioethics is a special branch of ethical science which deals with the moral aspects of biological and life sciences. The use of stem cells in stem cell therapy helps in the treatment of various neurodegenerative diseases like Parkinson's, Alzheimer's, Huntington's disease, and other

diseases like diabetes and cancer for which there are no effective treatments available. There are several ethical issues involved in the stem cell research like their origins and method of production; the availability of stem cells is very less from human body. The sources of stem cells considered to be best are found in human foetus and embryos which can be further differentiated into different tissues and organs giving new life to the human being. To avoid the ethical concerns the foetal stem cell lines could be cultured in the lab from the cells isolated from the aborted foetuses. The other option could be isolation of stem cells from 5-7 days old blastocyst if not considered as unethical. The extra embryos generated during IVF only one is used in implantation process rest can be further grown in test tube culture and can be used for isolation of stem cell in research purpose. After delivery the stem cells can also be isolated from placenta. All these can be deposited into stem cell banks from where it can be donated for the treatment of incurable diseases after differentiation into required diseased tissues and organs of the human body. By this way the ethical issues concerned with the human embryonic stem cells could be solved paving the way towards minimizing the human sufferings, avoiding the critical ethical issues.

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