INTRODUCTION

Tissue engineering has been a topic of extensive research over the last years, the ability of human body to regenerate tissue loss such as bone, cartilage, nerves, skin and muscle is limited, the isolation of mesenchymal stem cells (MSCs) and later the embryonic stem cells in conjunction with the advances made in cellular biology, tissue engineering, genetics and recombinant technology has initiated the development of new techniques and new therapeutic strategies allowing treatment of many pathological conditions providing restoration of tissue continuity and function ***(Pountos et al., 2006)***.

Mesenchymal stem cells are present in many human tissues and serve as a readily available source of undifferentiated cells being capable to form specific tissues like bone, cartilage, fat, muscle and tendon; they represent an attractive and promising field in tissue regeneration and engineering for treatment applications in a wide range of trauma and neurosurgical conditions ***(Pountos et al., 2006)***.

The bone marrow serves as a reservoir for different classes of stem cells, in addition to haemopoietic stem cells, the bone

marrow comprises a population of marrow stromal cells and these cells exhibit multi-lineage differentiation capacity, and are able to generate progenitors with restricted developmental potential, including fibroblasts, osteoblasts, adipocytes and chondrocyte progenitors, in addition, MSCs have been shown to possess immunosuppressive activity in vitro and in vivo, clinical trials are underway to test whether MSCs are beneficial in patients undergoing allogeneic bone marrow transplantation, with the expanding role of stem cell transplants in different areas of medicine, including neurosurgery, Cardiology, and orthopedics, MSCs become very important in the next few years ***(Pountos et al., 2006)***.

AIM OF THE WORK

This essay tries to make a spotlight on the ongoing researches concerning stem cell transplantation as a line of treatment in spinal cord injury on basic of stem cell biology including methods including of preparation, isolation, types of stem cells used in spinal cord as well as displaying the ethical debate surrounding stem cell research.

STEM CELL BIOLOGY

Stem cell is a cell that has the ability to divide for indefinite periods often throughout the life of an organism. The stem cells, when provided with the right signals, have the potential to differentiate into different types of cells that constitute an organism, these cells when be differentiated can have a characteristic shape and specialized functions, such as heart cells, skin cells or nerve cells.

Stem cells have two distinctive properties:

1. They can make identical copies of themselves for a long period of time (self renewal).
2. Give rise to mature cells that have a characteristic morphology.

(Sommer et al., 2009)

Stem cells proliferate, migrate, and differentiate to form organisms during embryogenesis. During adulthood, stem cells are present within tissues/ organs including the CNS where they may differentiate into neurons ***(Coutts and Keirstead, 2008)***.

Since the identification and characterization of stem cells, a great deal of interest has been given to their potential for treatment of spinal cord injury (SCI), traumatic brain

injury, and degenerative brain diseases. Considering their characteristic abilities to self-renew and differentiate into any cell type in the body, the therapeutic promise of stem cells is justified, before effective therapies can be developed, several issues need to be addressed and resolved, these issues range from increasing our basic knowledge about the stem cell's biology to prevailing over moral concerns fueled by religious and/or political ideas ***(Hardy et al., 2008)***.

Stem cell definitions:

A stem cell is defined by its ability of self-renewal and its Totipotency. self-renewal is characterized by the ability to undergo an asymmetric division in which one of the resulting cells remains a ''stem cell,'' without signs of aging, and the other (daughter) cell becomes restricted to one of the germ layers. A stem cell may become quiescent and at later stages re-enter the cycle of cell division ***(Orford and Scadden, 2008)***.

A true stem cell is a totipotent cell; it can become any cell type present in an organism. many consider the zygote to be the only true totipotent (stem) cell because it is able to differentiate into either a placenta cell or an embryonic cell, others define the cells of the inner cell mass within the blastocyst as embryonic stem cells (ESCs), these cells are pluripotent because they can not become a placenta cell besides ESCs, undifferentiated cells can be found among differentiated

cells of a specific tissue after birth, these cells are known as adult stem cells, although a better term would be ‘‘somatic stem cell’’ because they are also present in children and umbilical cords. there is ample evidence that adult stem cells are not restricted to a particular germ layer and can transdifferentiated, an important advantage of adult stem cells over ESCs is that they can be harvested without destruction of an embryo as a result, adult stem cells have gained ample interest for their application in a variety of disorders ***(Zietlow et al., 2008)***.

Differentiation:

The pluripotent stem cell differentiates into a multipotent cell of the three germ layers these three layers are:

1. The ectodermal layer (from which skin and neural tissue originate),
2. The mesodermal layer (connective tissue, muscle, bone, and blood cells),
3. The endodermal layer (gastrointestinal tract and internal glandular organs) the multipotent cell differentiates into a unipotent cell of a particular cell lineage within its own germ layer the unipotent cell is capable of becoming a cell type within that particular cell lineage at the successive phases of differentiation (or determination), the resulting progeny are known as

progenitor cells; ‘‘stem cell-like’’ cells capable of self-renewal within the central nervous system, unipotent neural progenitors become the neurons and glial cells present in brain and spinal cord.

Transdifferentiation:

In classic embryology, the totipotent stem cell becomes unipotent through successive phases of fate restriction the steps in this process were thought to be irreversible, however, recently it was shown in vitro that the fate of multipotent cells can be changed to another germ layer ***(Park et al., 2008)***.

This process is known as transdifferentiation the unlimited potential of transdifferentiation prompted many investigators to obtain cells that normally derive from stem cells that are more difficult to harvest from stem cells that are easier to harvest for instance, it is less complicated to harvest stem cells from skin or bone marrow than from the brain, thus, it would be more efficient to obtain neural cells from skin or bone marrow-derived stem cells through transdifferentiation ***(Park et al., 2008)***.

Transdifferentiation has often been shown using nonspecific markers and ignoring possible artifacts caused by culturing methods, therefore, the existence of transdifferentiation is still debated; it should be kept in mind that forced differentiation into a cell from a lineage within an

unnatural germ layer could result in abnormal phenotypes that, after grafting, could induce carcinogenesis ***(Park et al., 2008)***.

Potential for spinal cord repair:

After SCI, endogenous regenerative events occur, indicating that the spinal cord attempts to repair itself. **Schwann** cells, the myelinating and regeneration-promoting cell in the peripheral nervous system, migrate from spinal roots into the damaged tissue and myelinate spinal cord axons, the expression of regeneration-associated genes is increased in damaged neurons, there is a surge in proliferation of local adult stem cells and progenitor cells. however, axonal growth is thwarted by growth inhibitors present on oligodendrocyte myelin debris and on cells that form scar tissue, also, the newborn stem cells and progenitor cells do not integrate functionally into the injured spinal cord tissue, thus the endogenous regenerative events that occur after injury fail to repair the spinal cord ***(Mathews et al., 2008)***.

Improved functional outcome after SCI may be elicited by neuroprotective approaches that limit secondary tissue loss and thus the loss of function. alternatively, functional recovery could be elicited by axon growth promoting approaches that result in restoration of damaged and/or formation of new axon circuits that could become involved in function, there is little doubt that stem cells and neural progenitor cells could become

invaluable components of repair strategies for the spinal cord, they can become neural cells that may support anatomical/functional recovery. alternatively, they may secrete growth factors that could support neuroprotection and/or axon regeneration the potential of stem cells or progenitor cells to support spinal cord repair has been studied extensively ***(Nishikawa et al., 2008)***.

Their short comings for repair are also understood over the last decade, stem cells have often been studied without implementing explicit criteria that would define the used cells as such. consequently, the therapeutic potential of true stem/progenitor cells is still unknown, other matters related to the use of stem/progenitor cells for SCI also need to be resolved before effective therapies can be developed ***(Park et al., 2008)***.

Cell Replacement in the Injured Spinal Cord

Considering the ability of stem cells to become any cell type, their potential use for cell replacement strategies is common sense with the appropriate combination of growth factors (induction cocktail), ESCs can be used to obtain neurons and glial cells. ES-derived neurons can survive and integrate after injection into the injured rat spinal cord ***(Yu and Thomson, 2008)***.

It was shown that transplanted mouse ESCs myelinate axons in the myelin-deficient shiverer rat spinal cord also, mouse ESCs grafted into the injured (normal) rat spinal cord result in improved functional recovery; importantly, ESCs were found to survive well within the injured spinal cord, suggesting that long-term treatments could be achieved using this approach ***(Lowry et al., 2008)***.

Human ESC can be directed toward multipotent neural precursors, motor neurons, and oligodendrocyte progenitor cells, the latter were found to differentiate into mature oligodendrocytes in vitro and in vivo. moreover, these cells are able to myelinate axons after transplantation into the spinal cord of myelin-deficient shiverer mice and adult rats ***(Mathews et al., 2008)***.

Neural progenitor cells (i.e., multipotent cells from which the cells of the central nervous system arise) often aggregate into neurospheres, the neural progenitor cells transplanted into the injured rat spinal cord favored differentiation into astrocytes, these results indicated the need for differentiation protocols before grafting. fetal neural precursor cells genetically modified to express noggin, an antagonist of bone morphogenetic protein, differentiate preferably into neurons and oligodendrocytes. transplantation of these cells into the injured mouse spinal cord resulted in

improved functional outcome; however, this result could not be shown by others using the same approach ***(Lowry et al., 2008)***.

Human neural progenitor cells can be harvested from blastocyst-stage embryos and manipulated to generate functional neurons and glia; when human neural progenitor cells were grafted into the injured rat spinal cord, some of them were found to differentiate into oligodendrocytes; moreover, this finding was accompanied by improved functional outcome ***(Park et al., 2008)***.

Mesenchymal stem cells from bone marrow may also have therapeutic promise for SCI; although still debated, these particular adult stem cells have been shown to differentiate into bone, fat, tendon, and cartilage cells; it has been published that these cells can also transdifferentiate in vitro into liver, skeletal and cardiac muscle cells and into CNS cells ***(Orford and Scadden, 2008)***.

This makes mesenchymal bone marrow stromal stem cells interesting for strategies for repair of the injured spinal cord; many medical fields are exploring mesenchymal stem cells, for instance, for repair of the heart after myocardial infarction, osteogenesis imperfecta in orthopedics, organogenesis in internal medicine, intervertebral disc disease

in neurosurgery and stroke/neurodegenerative diseases in neurology ***(Orford and Scadden, 2008)***.

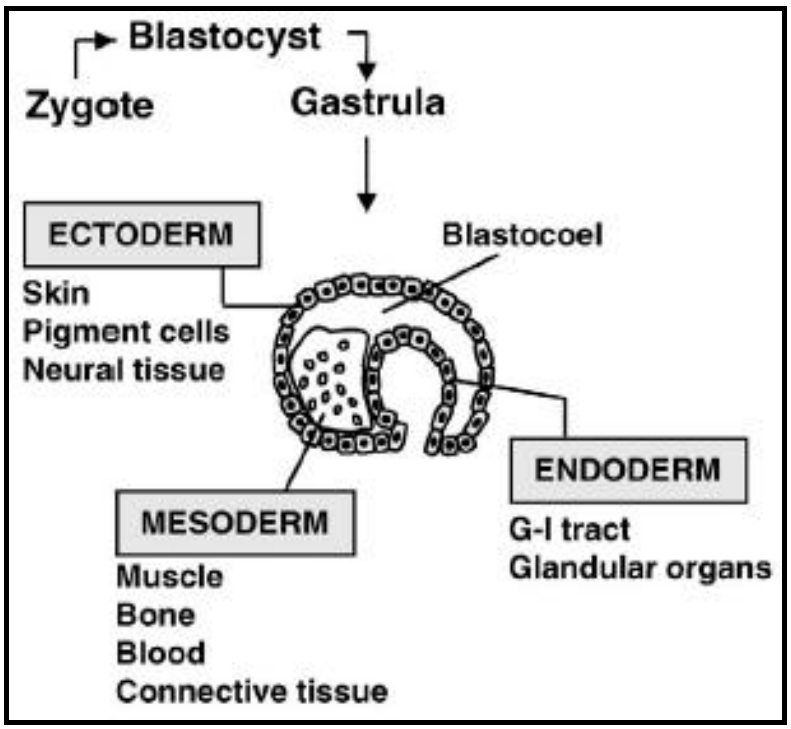

Fig. (1): All tissues in an organism originate from the 3 germ layers: the ectoderm, endoderm, and mesoderm layers. Neural cells that form the central and peripheral nervous system derive from the ectoderm ***(Orford and Scadden, 2008)***.

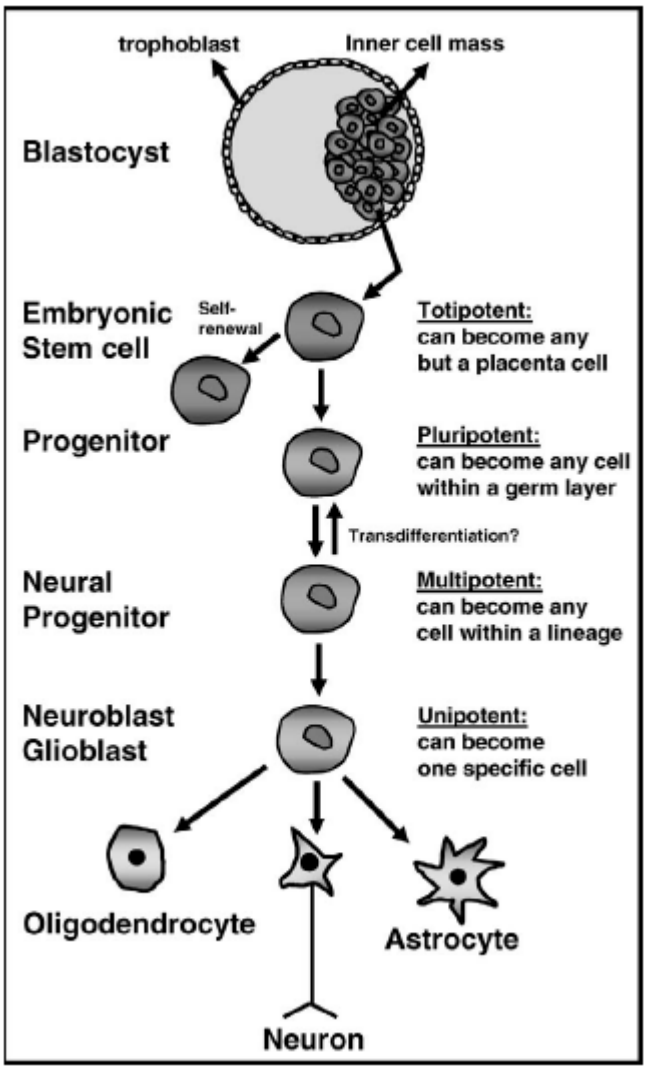

Fig. (2): From embryonic stem cell to differentiated neural cell. Embryonic stem cells from the inner cell mass of the blastocyst ***(Orford and Scadden, 2008)***.

Injury to the spinal cord initiates a series of biochemical events that are associated with a progressive decline in blood flow to the injured spinal cord and that exacerbate the extent of tissue damage. this problem is compounded by the poor

regenerative response shown by axons in CNS and as a result, white matter tracts are permanently interrupted, causing paralysis enhancing the regenerative response of the CNS is a formidable challenge and requires an understanding of the barriers to repair at both the molecular and cellular levels. ***(Orford and Scadden, 2008)***.

These obstacles have been assiduously outlined over many years and come in a variety of forms the four greatest obstacles are:

1. Proliferation of fibroblasts, astrocytes, microglia and endothelial cells at the lesion site forming a neuroglial scar that acts as a physical and /or chemical barrier,

2. An absence of Schwann cells, which help in guiding any regenerating axons,

3. The absence of neurotrophic factors to enhance axonal growth,

4. Inhibition of axonal growth by post-injury myelin-associated proteins such as Nogo-A,1 (MAG) and oligodendrocyte myelin glycoprotein, a variety of cell transplantation approaches have been tested in an effort to replace lost tissue by grafting cells to repair areas with damaged myelin or provide a tissue bridge for nerve fiber growth. 2-4 Mesenchymal stem cells (MSCs) have the

capability for self-renewal and differentiation into various lineages of mesenchymal tissues, 5-10 including osteocyte, chondrocyte, neurocyte.

Moreover, these cells should not elicit graft versus host disease when transplanted into the injury site; these features of MSCs attract a lot of attention from investigators in the context of cell based therapies of several human diseases. despite the fact that bone marrow represents the major source of MSCs, the use of bone marrow derived cells is not always acceptable due to the high degree of viral infection and the significant drop in cell number and proliferative capacity with age ***(Orford and Scadden, 2008)***.

Furthermore, MSCs content of human marrow is scarce, up to 10 cells in every million monocytes.

Umbilical cord blood mesenchymal stem cell

The blood remaining in the umbilical cord following birth contains haematopoietic precursors and this has become an important source for transplantation of hematopoietic stem cells ***(Rowland et al., 2008)***.

The presence of mesenchymal stem/progenitor cells in cord blood has recently been identified; however, there is controversy as to whether umbilical cord blood (UCB) contains MSCs that are capable of differentiating into cells of