Tissue Engineering in Urology

Essay

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INTRODUCTION

The genitourinary systems exposed to a variety of possible injuries from the time the fetus develops. Aside from congenital abnormalities, individuals may also suffer from other disorders such as cancer, trauma, infection, inflammation, iatrogenic injuries, or other conditions that may lead to genitourinary organ damage or loss, requiring eventual reconstruction ***(Rackley and Atala, 2007).***

The type of tissue chosen for replacement depends on which organ requires reconstruction. Bladder and ureteral reconstruction may be performed with gastrointestinal tissues. Urethral reconstruction is performed with skin, mucosal grafts from the bladder, rectum, or oral cavity ***(Rackley and Atala, 2007).***

However, a shortage of donor tissue may limit these types of reconstructions and there is a degree of morbidity associated with the harvest procedure. In addition, these approaches rarely replace the entire function of the original organ. The tissues used for reconstruction may lead to complications due to their inherently different functional parameters. In most cases, the replacement of lost or deficient tissues with functionally equivalent tissues would improve the outcome for these patients. This goal may be attainable with the use of tissue engineering techniques ***(Rackley and Atala, 2007).***

Tissue engineering integrates cells, scaffolds, and specific signals to create new functional tissues. In this approach, cells isolated from a small biopsy and expanded in vitro can be seeded onto a suitable scaffold. They are either allowed to develop into new tissue in vitro or transplanted into patients to create new functional tissue that is structurally integrated with the body. Various types of cells can be isolated from tissues and greatly expanded in vitro, potentially providing an unlimited supply of therapeutic cells. The scaffolds provide an appropriate three-dimensional environment, and guide the development of the desired structure from the cells by providing mechanical support until the newly formed tissues are structurally stabilized ***(Kim et al., 2000).***

During the last decade, reconstructive urology improved surgical outcomes through the use of specialized surgical equipment and the primary use of autologous tissue. Tissue engineering (TE), with its subspecialty of stem cells, aims to regenerate urological structures and organs so that full physiological function is restored. The success of cell cultures depends on cell isolation, separation, and selection, as well as optimized conditions for the proliferation of single cell types. Cell culturing is tailored to each individual cell type, and if stem cells are used, their differentiation to the targeted cell type is required ***(Sievert et al., 2007).***

Probably the most controversial area of TE is stem cells. Initially, embryonic stem cells were used due to their totipotency, however, because of their source, their use has raised moral and ethical concerns. Today, pluripotent stem cells, or differentiable adult stem cells, can be separated from many different tissues, including bone marrow striated muscle, fat, skin, synovial membrane, testicles, and amniotic fluid. They can then be differentiated into a variety of cells. In addition to resolving all these specific basic research aspects of TE, legal issues must support the movement of TE from theory to application in clinical trials and in standard "manufacturing" procedures ***(Sievert et al., 2007).***

Application of specific signals (e.g., growth factors, cell adhesion molecules, and mechanical strain) during the process of tissue development may induce the appropriate pattern of gene expression in the cells, and may allow the engineered tissues to maintain proper specific functions. One of the challenges in genitourinary tissue engineering is to expand a small number of genitourinary-associated cells to a clinically useful cell mass.

Although cell transplantation has been proposed for the replacement of a variety of tissues, including skin, pancreas, and cartilage, the concept of transplanting urothelial cells, which line most of the urinary tract, had not been approached in the laboratory setting until the early 1990s, because of the inherent difficulties encountered in expanding urothelial cells in large quantities. Rackley, Atala laboratories has been successful in greatly expanding urothelial cells from small biopsy specimens. The specifics of cell culture must be optimized for every cell type. Smooth muscle cells of ureter, bladder, urethra, and corporal cavernosum have also been successfully harvested and expanded ***(Kim et al., 2000).***

AIM OF THE WORK

Aim of this study is to review the literature discussing the role and applications of tissue engineering in urology.

BASICS OF TISSUE ENGINEERING

Definition

Tissue engineering is defined as an interdisciplinary field which applies the principles of engineering and life sciences towards the development of biological substitutes that aim to maintain, restore or improve tissue function ***(Atala and Lanza, 2001).***

In the last two decades, scientists have attempted to engineer virtually every tissue of the human body. Native tissues are usually preferable for reconstruction. In most cases, the replacement of lost or deficient tissues with functionally equivalent tissues would improve the outcome for these patients. This goal may be attainable with the use of tissue engineering technique ***(Wein et al., 2007).***

Tissue engineering follows the principles of cell transplantation, materials science, and engineering toward the development of biologic substitutes that can restore and maintain normal function. Tissue engineering may involve matrices alone, the body's natural ability to regenerate is used to orient or direct new tissue growth, or it may use matrices with cells ***(Wein et al., 2007)***.

Tissue engineering (TE) may offer new treatment option for patients who need replacement or repair of an organ. The principle is to dissociate cells from a tissue biopsy, to expand these cells in culture, and to seed them onto the scaffold material *in vitro* in order to form a live tissue construct prior to reimplantation into the recipient's organism. In the appropriate biochemical and biomechanical environment these tissues will achieve their full functional potential and serve as native tissue equivalents ***(Horst et al., 2010).***

The TE approach has major advantages over traditional organ transplantation. Tissues that closely match the patient's needs can be reconstructed from a generally readily obtainable biopsy. Moreover the engineered tissue can be transplanted into the patient's body without donor site morbidity and with minimal or no immunogenicity. This eventually conquers several limitations, encountered in tissue transplantation approaches ***(Horst et al., 2010).***

The basic bricks of living organisms, the cells, are a predominant factor for successful TE. Tissue renewal requires an adequate number of regeneration-competent cells that do not elicit immune response. Therefore autologous cells are the ideal choice, as their use circumvents many of the inflammatory and rejection issues associated with non autologous approach ***(Atala, 2008).***

Cells for use in cell therapy and tissue engineering

Native cells: When native cells are used for tissue engineering, a small piece of donor tissue is dissociated into-individual cells. These cells are expanded in culture and either injected directly back into the host or attached to a support matrix and then reimplanted. The source of donor tissue can be heterologous (such as bovine), allogeneic (same species, different individual), or autologous ***(Jennifer et al., 2011).***

The preferred cells to use are autologous cells, where a biopsy of tissue is obtained from the host, the cells are dissociated and expanded in culture, and the expanded cells are implanted into the same host. The use of autologous cells, although it may cause an inflammatory response, avoids rejection, and thus the deleterious side effects of immuno-suppressive medications can be avoided. Ideally, both structural and functional tissue replacement will occur with minimal complications when autologous native cells are used. However, one of the limitations of applying cell-based regenerative medicine techniques to organ replacement has been the inherent difficulty of growing specific cell types in large quantities. Even when some organs, such as the liver, have a high regenerative capacity *in vivo*, cell growth and expansion *in vitro* may be difficult ***(Jennifer et al., 2011).***

With the use of these methods of cell culture, it is now possible to expand a urothelial strain from a single specimen that initially covered a surface area of 1 cm^2 to one covering a surface area of 4,202 m^2 (the equivalent of one football field) within 8 weeks ***(Cilento et al.,1994).***

These studies indicated that it should be possible to collect autologous bladder cells from human patients, expand them in culture, and return them to the donor in sufficient quantities for reconstructive purposes ***(Cilento et al., 1994).***

Major advances have been achieved within the past decade on the possible expansion of a variety of primary human cells, with specific techniques that make the use of autologous cells for clinical application possible. Most current strategies for tissue engineering depend on a sample of autologous cells from the diseased organ of the host. However, for many patients with extensive end-stage organ failure, a tissue biopsy may not yield enough normal cells for expansion and transplantation. In other instances, primary autologous human cells cannot be expanded from a particular organ, such as the pancreas. In these situations, stem cells are envisioned as being an alternative source of cells from which the desired tissue can be derived. Stem cells can be derived from discarded human embryos(human embryonic stem cells), from fetal tissue, or from adult sources (bone marrow, fat, skin) ***(Jennifer et al., 2011).***

Stem Cells

Stem cells are defined by three important properties:

1. The ability to self-renew.
2. The ability to differentiate into a number of different specialized cell types "potency" (Fig. 1)· ***(Harley et al., 2001).***
3. The ability to easily form clonal populations.***(Aboushwareb et al., 2008).***

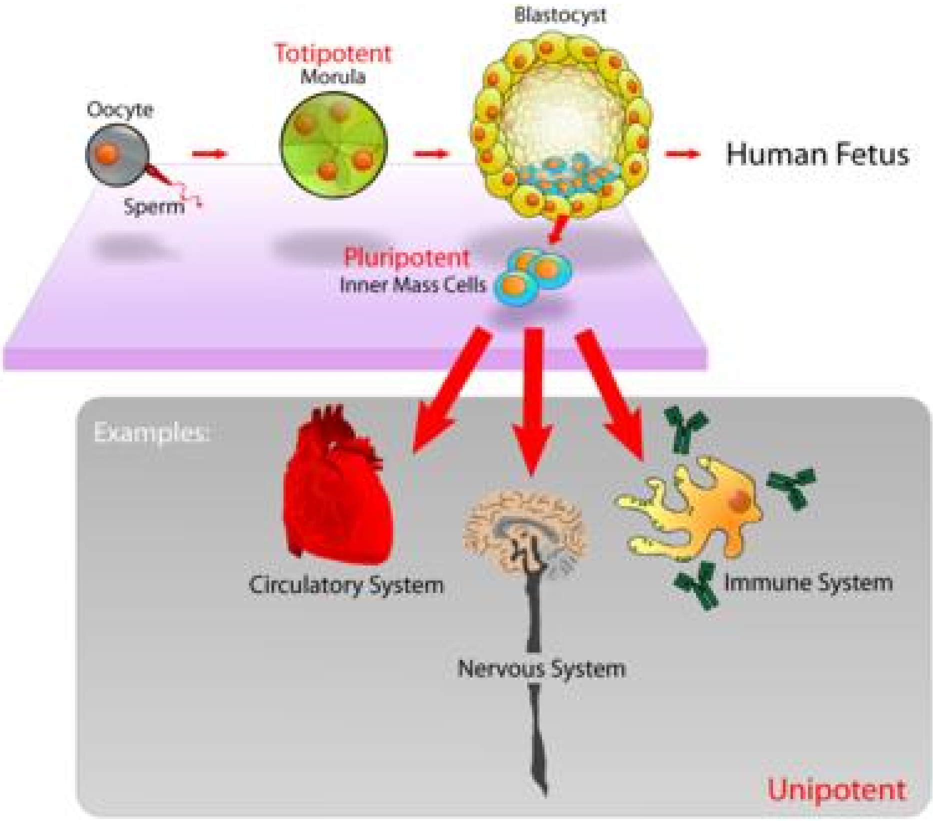


Figure (1): Potency of embryonic stem cells ***(Harley et al.,2001).***

Types of stem cells:

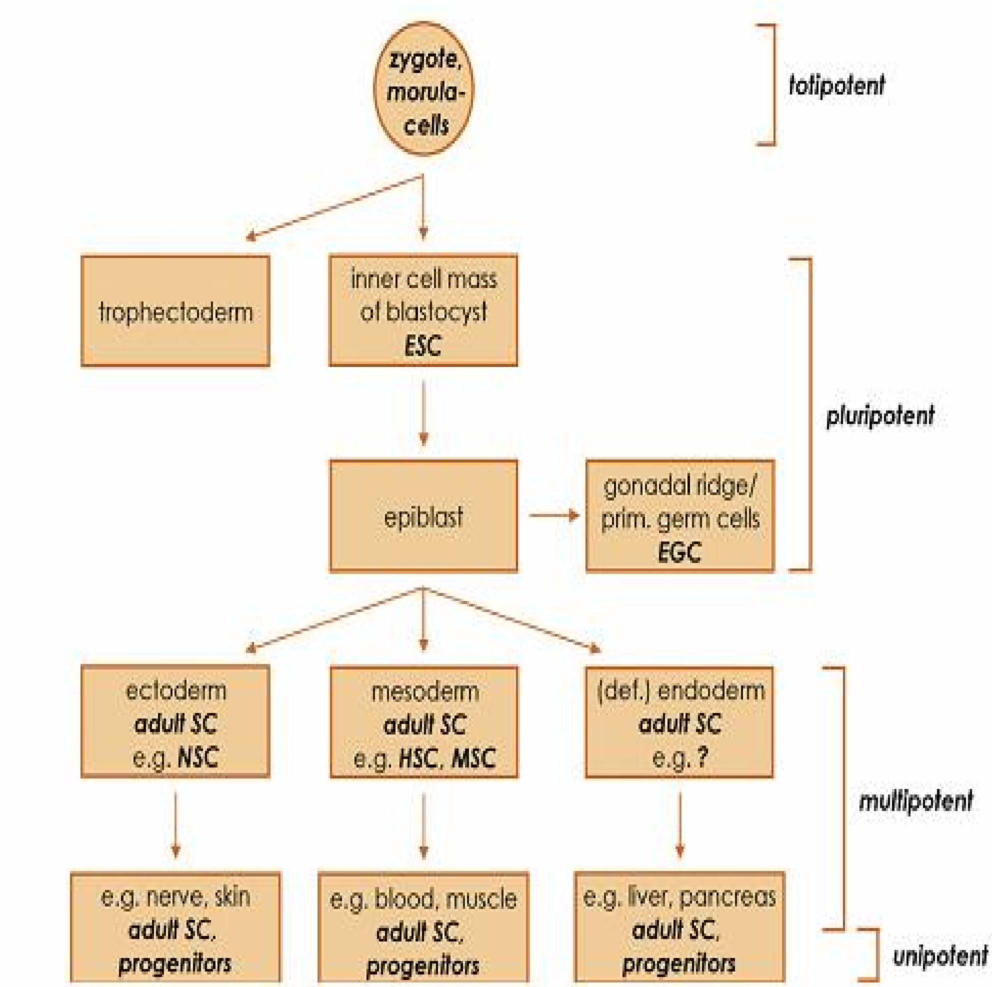
Stem cells to be either totipotent, pluripotent, multipotent or unipotent progenitor cells (Fig. 2) ***(Becker and Jakse, 2007)***.

Totipotent stem cells are produced from the fusion of an egg and sperm cell. Cells produced by the first few divisions of the fertilized egg are also totipotent. These cells can differentiate into embryonic and extraembryonic cell types. Totipotent stem cells are capable of forming cells of the ectoderm, mesoderm, and (definitive) endoderm layers and the gonadal ridge, and they are also capable of forming the supporting trophoblast required for the survival of the developing embryo. They include the zygote and its offspring cells of the morula ***(Becker and Jakse, 2007).***

Pluripotent stem cells are the descendants of totipotent cells and can differentiate into cells derived from any of the three germ layers. Pluripotent stem cells are embryonic stem cells (ESCs) isolated from the inner cell mass of the blastocyst and embryonic germ cells (EGCs) derived from primordial germ cells of an early embryo. They can give rise to cells of the three germ layers and to those of the gonadal ridge, but not to extra-embryonic tissues ***(Becker and Jakse, 2007).***

Multipotent stem cells are isolated from the developing germ layers and/or its descended adult organs are capable of self-renewal and differentiate into multiple organ-specific cell types. They can produce only cells of a closely related family of cells. Examples of such multipotent adult stem cells include haematopoietic stem cells (HSC), mesenchymal stem cells (MSC), and neural stem cells (NSC) ***(Becker and Jakse, 2007).***

Unipotent stem cells can produce only one cell type, but have the property of self-renewal which distinguishes them from non-stem cells. Unipotent cells are progenitor cells or precursor cells that have been reported to exhibit limited or no capacity for self renewal and differentiate into only one defined cell type, such as epithelial cells ***(Becker and Jakse, 2007).***



Def=definitive   SC=Stem cells   ESC=Embryonic stem cells
EGC=Embryonic germ cells   MSC= Mesenchymal stem cells
HSC=Hemopoetic stem cells   NSC=Nerve stem cells

Figure (2): A simplified scheme of stem cell populations Particular stem cell types are classified based on their differentiation capacity. The zygote and cells of the morula stage can give rise to both embryonic and extra-embryonic tissues and hence can generate a complete embryo. The three germ layers, as well as embryonic germ cells, originate from embryonic stem cells from the inner cell mass of the blastocyst. Adult stem cells produce progenitor cells and differentiated tissue ***(Becker and Jakse, 2007).***

Sources of stem cells for use in tissue engineering:

1) Embryonic stem cells: The term ‘embryonic stem cell’ was coined in 1981 after pluripotent cells were found in the inner cell mass of the mouse embryo ***(Martin, 1981).***

Embryonic stem cells exhibit two remarkable properties: the ability to proliferate in an undifferentiated, but still pluripotent state (self-renewal), and the ability to differentiate into a large number of specified cells. As their name implies, embryonic stem cells are derived from the early stage embryo. Although embryonic stem cells research is thought to have much greater potential than adult stem cells, several ethical and legal controversies still exist concerning their use in humans ***(Horst et al., 2010).***

Furthermore, embryonic stem cells have been shown to transdifferentiate into a malignant phenotype forming teratomas ***(Horst et al., 2010).***

They can be isolated by aspirating the inner cell mass from the embryo during the blastocyst stage (5 days post-fertilization) and are usually grown on feeder layers consisting of mouse embryonic fibroblasts or human feeder cells, more recent reports have shown that these cells can be grown without the use of a feeder layer and thus avoid the exposure of these human cells to mouse viruses and proteins. These cells have demonstrated longevity in culture by maintaining their