REVIEW ARTICLE

CLINICAL APPLICATIONS OF VARIOUS STEM CELLS IN DENTISTRY: A LITERATURE REVIEW

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ABSTRACT

Regenerative dentistry plays a major role in treatment planning of various dental diseases. Teeth are the most natural, noninvasive source of stem cells. Dental stem cells, which are easy, convenient and affordable to collect, hold promise for a range of very potential therapeutic applications. Present review article discusses history of stem cells, different stem cells relevant for dentistry, their isolation approaches, collection, and preservation of dental stem cells along with the current status of dental and medical applications.

Keywords: Dental Pulp, Regeneration, SHED, Stem Cells, Transplantation

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INTRODUCTION

Stem cells are undifferentiated biological cells that can differentiate into specialized cells and can divide (through mitosis) to produce extra stem cells.[1] Stem cells are the master cells of the body that meet the two conditions of self-replication and the ability to differentiate into at least two different type of cells. The term stem cell was proposed for scientific use by Russian histologist Alexander Maksimov in1908.[2] Therapeutic application of stem cells has created an increasing interest in the study of undifferentiated cell types that constitute the ability to proliferate and differentiate into various tissues. They have the capacity to renew themselves by cell division even after long periods of inactivity.[3] In present scenario various studies concentrate on the regenerative ability of cells as various tissues in the body (such as blood, skin, and gastrointestinal tract) undergo rapid renewal and have regenerative ability which form an important part of treatment plan.[4]

DISCUSSION

Types of stem cells

Two main types of stem cells are being investigated for their use in medicine research. They differ in the ability to self-renew and their degree of differentiation.[5]

- Embryonic Stem Cells
- Adult Stem Cells

Dental Stem Cells

In dentistry, interest in tissue engineering researches on different types of dental stem cells done in vivo and in vitro, increased rapidly among researchers and institutes.[6] Various types of tooth derived stem cells have been utilized in the field of regeneration medicine.[7]

Dental sources of adult stem cells[2]

- Dental pulp dental pulp stem cells (DPSCs)
- Exfoliated deciduous teeth- SHED
- Dental follicle progenitor cells- DFPCs
- Stem cells from root apical papilla- SCAP
- Periodontal ligament stem cells- (PDLSCs)

Dental Pulp Stem Cells (DPSCs)

These are the highly proliferative and clonogenic cells that have been derived from enzymatically disaggregated adult human dental pulp, that form sporadic, but densely calcified nodules in vitro.[8]

The applications related to oral health care included regeneration of an immature tooth with extensive coronal and pulp damage, regeneration of resorbed roots, cervical or apical dentin, whole tooth regeneration, repair and replacement of bone in craniofacial defects can facilitate restoring the physiologic structural integrity.[9] Suitable stem cells for tissue engineering should be able to differentiate into the target tissue/organ and should be easily collected and prepared to provide a further benefit to ensure the safety of the patient. DPSCs hold great clinical procedure due to their differentiation capacity and easy accessibility.[10]

1) De novo pulp regeneration: When the entire pulptissue is lost, regeneration requires the de novo creation of pulp. To create functional pulp for clinical application, three issues must be considered: first, regenerated pulp tissue must be vascularised; second, newly differentiated odontoblasts should form on the existing dentinal wall of the root canal space; finally, new dentin must be produced by differentiated odontoblasts on the existing dentin.[5]

2) Tooth reconstruction: It may be possible to generate a method to biologically replace lost teeth with the help of stem cells. A functional biological replacement tooth must include generation of a root and periodontal ligament with nerve and blood supplies. The crown, is less important since replacement of crowns with synthetic functioning is possible.[10]

Stem Cells From Human Exfoliated Deciduous Teeth (SHED)

Dr. Songtao Shi, isolated stem cells using the deciduous teeth of his 6 year - old daughter and named them as stem cells from human exfoliated deciduous teeth (SHED).[11] These cells have the ability of high proliferation potency and are multipotent mesenchymal stem cells. These cells not only differentiate into dental pulp-

related cells, but also, other cell types such as osteoblasts, adipocytes, neuronal-like cells and endothelial cells.[12]

Applications in Dentistry

Based on the basic tissue engineering principles, Peter Murray et al. identified several major areas of research that might have applications in the development of these techniques.

Root canal revascularization via blood clotting: Revascularization of the necrotic root canal systems by disinfection followed by establishing bleeding into the canal system via over instrumentation. Use of intracanal irrigants (NaOCl and chlorhexdine) along with the placement of antibiotics (e.g., a mixture of ciprofloxacin, metronidazole, and minocycline paste), for several weeks, is a critical step, as it effectively disinfects the root canal systems and increases revascularization of the avulsed and necrotic teeth. The revascularization process offers negligible chances of immune rejection and pathogen transmission, as regeneration of the tissue takes place by the patient's own blood cells.[13]

Postnatal stem cell therapy: The process comprises of postnatal stem cells (derived from skin, buccal mucosa, fat, and bone) being injected into disinfected root canal systems after the apex is opened. This process has many advantages like the harvesting and delivery of autogenous stem cells by syringe, being relatively easy; and the potential of these cells to induce new pulp regeneration. However, there are several disadvantages, like the cells may have a low survival rate and they may migrate to different locations within the body. Instead, all three elements (cells, growth factors, and scaffold) must be considered, to maximize the potential for success of pulp regeneration.[11]

Pulp implantataion: The pulp cells can be grown on biodegradable membrane filters to transform two dimensional into three dimensional cell cultures. The ease of growing these cells on filters in the laboratory, for evaluation of cytotoxicity of test materials, is recognized as the main advantage of this delivery system. Specialized procedures for proper adherance to the root canal walls is required, which is the major problem associated with implantation. As sheets of cells lack vascularity, only the apical portion of the canal systems will receive these cellular constructs, with coronal canal systems filled with scaffolds capable of supporting cellular proliferation.[14]

Scaffold implantation and delivery: A scaffold should contain growth factors, Bone Morphogenic Protein (BMP), fibroblast growth factors, and Vascular endothelial growth factors, to aid stem cell proliferation and differentiation, apart from having nutrients promoting cell survival and growth as well as antibiotics to prevent any bacterial in growth in the canal systems. The scaffold materials may be natural or synthetic, biodegradable or permanent. The synthetic materials like polylactic acid, polyglycolic acid and polycaprolactone degrade within the human body and have been succesfully used for tissue engineering purposes. Limitations consist of difficulties of obtaining high porosity and regular pore size.[11]

Three-dimensional printing: The three dimensional cell printing technique can be used to precisely position cells so that they have the potential to create tissue constructs that mimic the natural tooth pulp tissue structure. Careful orientation of the pulp tissue construct during placement into the cleaned and shaped root canal systems in accordance with its apical and coronal asymmetry is the prime requisite for the success of the technique.[15]

Huang, et al. explored in mice that pulp-like tissue can be regenerated de novo in an emptied root canal space by stem cells from apical papilla and dental pulp that give rise to odontoblast-like cells, producing dentin-like tissue on the existing dentinal walls via stem/progenitor cell-based approaches and tissue engineering technologies.[16]

Periodontal Ligament Stem Cells (PDLSCs)

The concept that stem cells reside in periodontal tissues was first proposed by Melcher.[6] PDLSCs in defined culture conditions differentiate into cementoblasts, adipocytes and collagen forming cells. These cells when transplanted generate a cementum/ PDL-like

structure that contribute periodontal tissue repair.17 However, PDLSCs obtained from mature periodontal ligaments possess stem cell properties similar to MSCs rather than neural crest cells.[18]

Osteogenic Potential of PDLSC

PDLSC are shown to have osteogenic potential. PDLSC isolates to have lower osteogenic potential than BMSC and also dental pulp derived stem cells. Kim et al, who reported new bone formation by PDLSC in a periimplant defect model, albeit at lower levels than BMSC. Although the potential use of PDLSC for generating graft biomaterials for bone tissue engineering in regenerative dentistry can be envisioned, as these cells are more routinely accessible, it is however necessary to delineate more refined isolates of pluripotent progenitors using genomic and proteomic marker characterization.[19]

Periodontal Regeneration by PDLSC

Seo et al. demonstrated a cementum/PDL-like complex generated in surgically created periodontal defects by transplanting in vitro expanded human PDLSCs in a ceramic particle scaffold. Porcine model study reports transplanting autologous swine PDLSCs, which lead to the generation of a root/periodontal complex capable of supporting a porcelain crown, resulting in normal tooth function.[19] Cementum and PDL- like structures adjacent to the surface of scaffolds is formed due to subcutaneous injection of PDLSC with hydroxyapetite or beta- tricalcium phosphate scaffolds. Besides periodontal regeneration, another potential application of PDLSCs is in the area of hybrid tooth engineering in combination with other stem and progenitor cell populations and scaffolds.[20]

Stem Cells From Apical Papilla (SCAP)

A population of stem cells isolated from human teeth was found at the tooth root apex. These cells are called stem cells from apical papilla (SCAP).[21] SCAPs were initially isolated from third molars and incisors of swine by Sonoyama et al. and obtained from humans in 2008.[22]

Clinical Applications of SCAP

Continued Root Formation: Root apical papilla is likely to play a pivotal role in root formation. Despite the fact that pulp tissue is intact root development is halted as apical papilla is removed surgically at an early stage. Further research is needed to verify that this halted root development was not due to damage of Hertwig's epithelial root sheath (HERS) during the removal of the apical papilla of that particular root apex.[23]

Pulp Healing and Regeneration: Immature teeth that presented with radiolucent lesions and non- vital pulp underwent remarkable apexogenesis after conservative treatment suggest that vital pulp tissue must have remained in the canals. Periapical disease can occur while the pulp is only partially necrotic and infected as open apex provided a good communication from the pulp space to the periapical tissues. Along the same line of reasoning, stem cells in pulp tissue and in apical papilla may also have survived the infection and allowed regeneration of pulp and root maturation to occur.[23]

Replantation and Transplantation: Changes in pulp tissue after replantation showed that various hard tissues including dentin, cementum, and bone may form in the pulp space depending on the level of pulp recovery. If pulp and apical papilla are totally lost, then the root canal space may be occupied by cementum, PDL and bone.

Autotransplantation is one of the clinical treatment options for missing teeth. The process often involves extraction of a supernumerary tooth or third molar and implantation into a recipient site. Based on current available information, it is likely that odonto blast lineages are derived from stem cells in pulp tissue or apical papilla. Both SCAP and HERS appear to be important for the continued root development after transplantation. SCAP are also highly probable to survive after transplantation because minimal vascularity is found in apical papilla based on preliminary findings. The reason that transplanting a tooth with little or no root formation results in almost no further root development is unclear.[5]

Bioroot Engineering: Dental implants have recently gained momentum as a preferred option for replacing missing teeth instead of bridges or removable dentures but it requires a direct integration with bone onto its surface as the prerequisite for success. Due to lack of natural contours and its structural interaction with the alveolar bone make dental implants a temporary option until a better alternative is available. SCAP and PDLSCs form a bioroot. Using a minipig model, autologous SCAP and PDLSCs were loaded onto HA/TCP and gelfoam scaffolds, respectively, and implanted into sockets of the lower jaw. A post channel was precreated to leave space for post insertion. Three months later, the bioroot was exposed, and a porcelain crown was inserted. This approach is relatively a quick way of creating a root onto which an artificial crown can be installed. The bioroot is different from a natural root in that the root structure is developed by SCAP in a random manner. Nevertheless, the bioroot is encircled with periodontal ligament tissue and appears to have a natural relationship with the surrounding bone. The mechanical strength of the bioroot, which is approximately two thirds of a natural tooth.[23]

Dental Follicle Progenitor Cells (DFPCs)

Several studies have reported the isolation of progenitor/stem cells from Dental Follicle in different species, using an enzymatic digestion of the Dental Follicle to release cells, followed by a culture of the cells in a stem cell growth medium.[24] In 2005 & 2007, Morsczeck et al. and Kemoun et al., respectively have identified unique undifferentiated lineage committed cells possessing mesenchymal progenitor features in the human dental follicle The cells were referred to as 'dental follicle precursor cells'(DFPCs).[25]

Clinical Applications of DFPCs[5,26]

Periodontal Regeneration: Dental follicle stem cells (DFSCs) have been developed to regenerate periodontium), which could become an alternative cell source for periodontal regeneration therapy. It is also confirmed by Hasegawa et al 2005, that periodontal defects can be managed by reimplantation of these cells.

Repair of Craniofacial defects: Craniofacial defects results from post-cancer ablative surgery, craniofacial osseous deficiencies can also arise from infection, trauma, congenital malformations and progressively deforming skeletal diseases. Although autologous bone graft is considered the best option, it has the limitation of donor sites. Stem cells can be used to treat degenerative bone diseases including TMJ defects. Cells from various sources like articular cartilage cells, fibroblasts, mesenchymal stem cells have been used to reconstruct TMJ. Bone tissue engineering endeavours to repair large bone losses using three dimensional scaffolds to deliver vital cells to the defective site.

Gingival Mesenchymal Stem Cells (GMSCs)

The mesenchymal stem/progenitor cells (MSC) isolated from the gingival lamina propria have been termed variously researchers including GMSCs, gingival tissue derived SCs, gingival multipotent PCs and gingival margin derived stem/PCs human oral mucosa SCs and oral mucosa lamina propria PCs.[27]

Clinical Applications of GMCs

These cells can gain numerous applications in cell and regenerative therapies. Possible areas being aimed to include skin wound repair, tendon regeneration, bone defect regeneration, periodontal regeneration, peri implanatitis, antitumor effect, oral mucositis, collagen-induced arthritis and contact hypersensitivity.[27]

Potental clinical applications in the Orofacial complex

Craniofacial tissue engineering promises the regeneration or de novo formation of dental, oral and craniofacial structures lost due to congenital anomalies, trauma and diseases. Virtually all the craniofacial structures are the derivatives of mesenchymal stem cells. Cells with characteristics of adult stem cells have been isolated from the dental pulp, the deciduous tooth and the periodontium. Mesenchymal cells are used for regeneration of several craniofacial structures such as mandibular condyle, calvarial bone, cranial suture and subcutaneous adipose tissue.[28]

Alveolar Ridge Augmentation: Restoration of

alveolar ridge height is of utmost concern to dentists in trying to prevent the loss of a tooth due to bone destruction induced by periodontal disease and in maintaining the ability of edentulous patients to wear dentures.[29] Appropriate ridge height is also essential for the placement and long-term retention of dental implants. Standard practice involves the use of autologous or allogenic bone grafts oe ceramics, both with and without growth factors, but the outcomes are variable. In animal models, BMSCs are used in conjuction with HA/TCP. They have successful in building alveolar bone[30] and a number of small studies in human patients have used BMSCs along with allogenic bone fragments or with platelet-rich plasma, with another ceramic scaffold, beta-calcium phosphate etc. With further refinement, these types of procedures would mark a major advancement in dental reconstruction.[31]

Tissue Engineering of Temporomandibular Joint from the Stem Cells: Temporomandibular disorders (TMD) manifest as pain, myalgia, headaches and structural destruction collectively known as degenerative joint disease (Okeson1996).[32] The temporomandibular joint (TMJ) like other synovial joints is also prone to rheumatoid arthritis, injuries and congenital anomalies (Stohler 1999).33 The severe form of TMJ disorders necessitates surgical replacement of the mandibular condyle (Sarnat and Lakin, 1992).[34]

In the past few years, we have reported the tissue engineering of a mandibular condyle exhibiting the shape and dimensions of a human cadaver TMJ. The engineered mandibular condyle had stratified layers of cartilage and bone from a single population of bone-marrow derived mesenchymal stem cells (MSCs) and was moulded into the shape and dimensions of a human cadaver mandibular condyle.[34]

Challenges

Stem cell research has undergone huge advancements in the past couple of years. This does not mean, however, that researchers have not faced their share of problems. It has proved particularly challenging for scientists to ensure the long term proliferative ability and pluripotency of embryonic stem and germ cells. These are important characteristics to maintain, as accurate models are necessary to understand the unique genetic and molecular basis by which these cells are able to replicate indefinitely. In addition to providing accurate models, culturing stem cells in vitro is also necessary in order to ensure that sufficient quantities of stem cells are available to treat specific diseases.[35] Teratoma formation has also produced a hurdle that needs to be overcome. Formation of these tumor like masses of cells at injection sites significantly limit the therapeutic potential applications of embryonic stem cells.36Immune challenges also prove a significant barrier to the application of stem cell therapies. If the stem cells are recognized as non-self, they will be rejected and destroyed.[35]

SUMMARY AND CONCLUSION

Due to advancements in the prevention, diagnosis and treatment of human diseases, the inability of most tissues and organs, to repair and regenerate after damage is a problem that needs to be solved. Stem cell research is being pursued in the hope of achieving major medical breakthroughs. Scientists are striving to create therapies that rebuild or replace damaged cells with tissues grown from stem cells and offer hope to people suffering from various ailments.

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