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Stem Cell Based Tooth Regeneration- An Alternative Approach to Implant Dentistry?

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Abstract:- Implant dentistry is the branch of dentistry that deals with replacing lost teeth and the structures that support them with artificial prostheses that are attached (osteointegrated) to the jaw bone. The regeneration of hard dental tissues has become a reality and a top goal in contemporary dentistry with the development of the current idea of tissue engineering approach and the discovery of the potential of stem cells in dentistry. The current review summarises recent developments in stem-cell-based regeneration techniques for hard dental tissues and assesses the viability of using growth factors and stem cells in scaffold-based or scaffold-free methods to stimulate the regeneration of the entire tooth or just some of its component parts.

Keywords:- *Stem Cell, Regeneration, Scaffold- Based Approach, Scaffold- Free Approach, Growth Factor.*

I. INTRODUCTION

Enamel, dentin, and cementum are the three different types of highly mineralized tissues that make up a tooth. The tooth has a separate developing mechanism even though it is a mineralized hard structure like bones overall. In mature enamel, 95 weight percent of the material is carbonated hydroxyapatite, 4 weight percent of the material is water, and 1 weight percent of the material is a soft organic matrix. Human teeth are mostly made of dentin, which shares the same biochemical makeup as bones (70% hydroxyapatite, 18% collagen, 10% body fluid, and 2% non-collagenous proteins in weight volume). The matrix of demineralized dentin is composed of type I collagen, bone morphogenetic proteins, and growth factors for fibroblasts[2].

Millions of people worldwide suffer from tooth defects, which are a very prevalent medical disease. Currently utilised dental repair procedures include dental implants to replace lost teeth, endodontic therapy to treat pulp necrosis, and fillings for cavities, all of which rely on the use of synthetic materials. Contrarily, the medical and dental areas of tissue engineering and regenerative medicine and dentistry (TERMD) employ biologically based treatment procedures for essential tissue regeneration, and as a result, have the capacity to regenerate live tissues. Understanding the molecular signalling networks that control the formation of natural teeth is essential for developing bioengineered replacement tooth techniques[3].

II. THE MOUTH AS THE KEY TO SYSTEMIC HEALTH OVERALL

Healthy microbes that support the health and vitality of dental tissues in the mouth as well as all other tissues and organs in the rest of the body make up the natural balance of microbiota in healthy persons' mouths. On the other hand, severe periodontitis is frequently accompanied by persistent bacterial infections that can weaken the oral flora's natural defences, causing excruciating pain, swelling, and tooth loss. An unhealthy oral environment can seriously harm distant parts of the body, including the heart, which can accumulate harmful biofilms on the heart valves, as well as stomach and intestinal disorders, including systemic sepsis in severe cases, because the oral cavity is intimately connected to the rest of the body through swallowed saliva, food, and breathing. As a result, dental health is crucial to general health, particularly in those who also have other medical disorders including diabetes, heart disease, high blood pressure, ulcers, and immunocompromised states. The general health of communities across the world depends critically on our capacity to practise good oral hygiene and to quickly and efficiently repair damaged oral and dental tissues.

Current treatments for tooth loss and damage focus on synthetic materials to correct structural flaws but do not perform any biological activities like enhancing blood and nerve supply. As was already said, significant oral tissue damage can undermine a person's self-esteem, which can result in substantial psychosocial and mental health problems as well as malnutrition brought on by challenges with eating and chewing. As with many other ectodermal organs, including hair, sweat glands, finger- and toenails, and teeth, the development of teeth, also known as odontogenesis, is started by interactions between two types of dental tissues, the dental epithelium (DE) and the dental mesenchyme (DM)[4].

- *Crucial Dental Developmental Features*

The number and configuration of tooth kinds (incisors, molars, premolars, etc.) differ significantly between species [5]. We now have a better understanding of how teeth first develop and then precisely occlude (meet) adjacent teeth in a developing jaw, on account of several studies utilising a range of animal models [6]. The molar teeth of rodents like mice and rats as well as those of humans are classified as brachydont teeth, which means they have comparable characteristics including a low crown-to-root ratio and no

further crown development following tooth eruption [7]. The bulk of studies on the molecular pathways governing tooth formation have been carried out in mice because of the remarkable parallels between tooth development in mice and humans [8]. The capacity to quickly create targeted mutant and transgenic mice lines to better understand the roles of various molecular signalling pathways in tooth creation is another benefit of the mouse model [9]. Additionally, unlike molars, rats have continually erupting incisors that serve as important research tools for understanding the DSC niche, or the environment where dental progenitor cells stay before being recruited to regenerate dental tissues [10].

III. COMPONENTS OF THE REGENERATION PROCESS IN DENTAL TISSUE

➢ *Stem Cells*

Dental pulp stem cells (DPSCs), stem cells found in human exfoliated deciduous teeth (SHEDs), periodontal ligament stem cells (PDLSCs), dental follicle precursor cells (DFPCs), stem cells from the apical papilla (SCAPs), and gingival fibroblast stem cells (GFSCs) are the six categories of dental stem cells that have so far been identified in the field of tooth regeneration[11,12]. Stem cells can be divided into totipotent (able to form all types of cells), pluripotent (able to give rise to any type of cell), oligopotent (able to differentiate into a small number of cell types), multipotent (able to form cells of their origin tissue), and unipotent (able to yield one cell type) stem cells based on their ability to differentiate [13].

Whereas embryonic and induced pluripotent stem cells are pluripotent, adult stem cells are multipotent. Depending on the signals acquired either intrinsically (from within the cell) or extrinsically (from outside the cell—either mechanically or chemically), the differentiation process enables stem cells to acquire various roles and traits. A wide range of tissues, including bone marrow, dental, nervous, and adipose tissues, bone, endometrium, muscle, blood, umbilical cord, amniotic fluid, and Wharton's jelly, can be used to collect multipotent human mesenchymal stem cells (hMSCs) [14]. The capacity of hMSCs to develop into non-mesodermal (ectodermal-neurocytes, endodermal-pancreocytes, and hepatocytes) and mesodermal (chondrocytes, osteocytes, and adipocytes) lineages has been demonstrated [15].

Dental stem cells produced from the neural crest are one of the most reliable and accessible sources of autologous stem cells (DSCs). The development of dental hard tissues involves three main cell types: I odontoblasts, which are tall columnar cells found at the edge of the dental pulp and are derived from mesenchymal cells responsible for dentin development; (ii) ameloblasts, which are derived from epithelial cells and are in charge of producing enamel; and (iii) cementoblasts, which have their roots in follicular cells and are located close to a tooth root. Regarding the dental stem cells produced from the neural crest, SHEDs and SCAPs can develop into odontoblasts, DPSCs and PDLSCs can develop into osteoblasts, and PDLSCs can develop into chondrogenic cells and adipocytes [17]. In addition to DPSCs' role in the regeneration of the dentin-pulp complex and PDLSCs' role in the regeneration of periodontal tissues and cementum, SHEDs have the capacity to create dentin-like tissues. Moreover, DFPCs show the ability to regenerate cementum and dentin [18].

➢ *Growth Factors*

Due to their capacity to create a channel of communication between cells and tissues, growth factors are essential proteins involved in the maturation, preservation, and repair processes of dental tissues [19]. Many signalling pathways, such as transforming growth factor beta (TGF-), sonic hedgehog (SHH), bone morphogenetic protein (BMP), and fibroblast growth factors (FGF), control cell proliferation and differentiation. Hedgehog (Shh) proteins are linked to the control of dentogenesis during development and adolescence [20–22].

Signaling centres that regulate tissue connections as well as the ultimate form and size of a single tooth have an impact on tooth morphogenesis. The multiple phases of dental tissue development allow for the detection of the unique functionalities brought on by the activation of these pathways. Several of these phases are beneficial for cell stemness and Shh and FGF proliferation. Inducing polarisation, migration, and calcification at the same time as postnatal differentiation stages include TGF-, BMPs, and Wnt[23–25].

➢ *Scaffolds for the Regeneration of Hard Dental Tissues*

Two primary techniques, namely top-down and bottom-up, are typically utilised in the tissue engineering of dental tissues in order to develop scaffold materials that allow the stem cells and/or growth factors to generate desired tissues [26]. The stem cells are implanted in 3D scaffolds consisting of polymers, natural porous materials, or both in the top-down method.

Natural extracellular matrix that is decellularized. The stem cell aggregates are being used as building blocks by various techniques, such as cell printing, cell sheets, microwells, or self-assembled hydrogels, to create the required tissues [26].

Although a variety of materials, including organic and synthetic polymers, ceramics, composites, metals embedded in porous scaffolds, nanofibers, microparticles, meshes, sponges, and/or gels, have been used in dental tissue engineering approaches, not all of them were appropriate for the regeneration of dental hard tissue. Dental scaffolds commonly consist ofbioactive ceramics, composites, or polymeric biomaterials [27].

➢ *Polymers-Based Scaffolds*

Both natural and synthetic polymers may be employed to build scaffolds for the regeneration of hard dental tissues. Type I collagen, alginate, fibrin, methacrylated gelatin, and platelet-rich plasma (PRP) are among the most effective natural polymers employed in tooth regeneration, whereas poly (lacticco-glycolic acid) and poly-"-caprolactone stand out among synthetic polymers.

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➢ *Bioactive Ceramic Scaffolds*

The group of materials with the greatest research on bone regeneration includes calcium phosphates, hydroxyapatite, biphasic, and tricalcium phosphate [28,29]. Granules made of 3D calcium phosphate offered the following conditions for the growth like the potential of bone to connect to surrounding tissues, osteoconductivity, and odontogenic development of human dental pulp stem cells [30,31]. ZnO and SiO2 dopants were added to tricalcium phosphate scaffolds to boost the mechanical strength and cellular proliferation of the bioceramics [32]. After 6 weeks after implantation, new hard tissues formed on a scaffold made of porous hydroxyapatite, -tricalcium phosphate, and polygricolide fibres, with dentin-like layers on the inner wall and odontoblasts nearby aligned to the hard tissues [33].

Glass ceramics and bioactive glasses are composed of a variety of oxides, including SiO2, CaO, Na2O, Fe2O3, P2O5, and MgO [34]. Variable crystallinity (between 30 and 90%), biocompatibility, opacity or translucency, and resorbability are characteristics of glass ceramics [35]. Although their brittleness, high density, and poor dimensional stability prevent the use of bioactive and ceramic glasses as scaffolds in tissue regeneration, Mechanical strength, 3D bioactive scaffolds seeded with human dental pulp stromal cells, and the emergence of sporadic calcified tissues all caused osteogenic gene expression[36].

➢ *Composite Scaffolds*

3D dental scaffolds with superior mechanical characteristics have been created using composites that combine fibrin with synthetic polymers or other inorganic materials [37,38]. Other examples include composites made for the formation of dentin, such as those containing PLGA porous polymers and various ceramics, such as calcium carbonate, tricalcium phosphate, and hydroxyapatite, as well as PLA-based scaffolds doped with calcium silicate and dicalcium phosphate [40] and PCL with biodentine [41].

➢ *Tooth Regeneration*

The formation of artificial teeth is a complicated process that involves repeatable molecular communication between the dental mesenchyme and dental epithelium (DE), similar to the development of natural teeth (DM). Early embryonic DE has the ability to start tooth development, according to previous studies [42], however later in tooth development, the DM takes on the odontogenic capability, giving it the ability to start tooth development when recombined with epithelium [43,44]. Since the DE is no longer present in erupted teeth, adult teeth in humans have lost their capacity to regenerate [45].

Thus, two potential theoretical methods for human entire tooth regeneration have been suggested in order to address this problem. One method involves creating a tooth, tooth bud, or tooth root in a dish before transplanting it back into the area of the defect. An artificial crown might be supported by a bioengineered tooth root once it has been firmly fixed in the jaw bone. Making a bioengineered replacement tooth bud in vivo at the location of the defect is a second strategy.

➢ *Bioengineered Teeth Generated from Mouse Embryonic Tooth Buds*

Many studies have been conducted using the mouse embryonic tooth bud model to investigate potential approaches to tooth regeneration. To produce an artificial tooth bud, mouse DE and DM cell layers were formed and subsequently merged [46]. The crucial element of this model was the use of DE tissue from the early embryonic stage, either whole or dissociated, to guarantee effective tooth development. When the DE layer was mixed with embryonic bone marrow cells, a combination of embryonic neural stem cells taken from embryonic spinal cords, or dissociated DM cells, tooth development was shown [47,48]. The first time that totally functioning teeth were successfully regenerated was in 2007 using artificial tooth buds that were once more made from embryonic tooth bud cells [49]. The procedure was successfully used to create fully functional teeth again in the years that followed [50,51], making it by far the most sophisticated model for functional tooth regeneration from embryonic tissues. In this model, single-cell suspensions of DE and DM from mouse embryonic tooth germs were mixed within a collagen drop, in vitro cultivated to reach the early bell stage, and then transplanted into a host mouse jaw at the age of 8 weeks. Unexpectedly, this investigation found that full-sized teeth formed and erupted and occluded similarly to normal teeth [50].

➢ *Adult Postnatal Teeth Bioengineered from Tooth Buds*

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Using natural tooth bud extracellular matrix (ECM) scaffolds made from decellularized postnatal tooth buds (dTBs) to direct the production of bioengineered teeth of a certain size and shape was one potential approach [56]. This strategy was developed in light of earlier research that demonstrated the safety of using moderate decellularization procedures to safely remove immunogenic components from

whole organs and tissues while preserving the natural ECM and its signalling components [57]. Based on the successful use of decellularized extracellular matrix (dECM) scaffolds for applications in regenerative medicine, postnatal porcine tooth buds were used to create dTB-ECM scaffolds, which were then reseeded with porcine DE cells, human DPSCs, and human umbilical vein endothelial cells (HUVECs) to facilitate revascularization.

After transplantation into adult minipig tooth extraction sockets, these dTB-ECM constructions were allowed to develop for 1 or 3 months [56]. These dTB-ECM constructions reliably controlled the creation of ordered, bioengineered teeth that were equal in size to real human teeth, as was discovered after harvest and analysis [56]. The dTB-ECM biomimetic tooth bud model is a potentially effective treatment for tooth regeneration in people [59] due to the recent approval of decellularized ECM scaffolds for therapeutic purposes in humans by the US FDA [58].

➢ *Currently Existing Obstacles to Teeth Regeneration*

There are still certain difficulties even if the methodologies outlined have a lot of potential for use in regenerative translational dentistry. One is how to design bioengineered teeth that are the exact size and form that are desired. Even though it is well known that attaining proper tooth morphology and occlusion with opposing teeth is essential for maintaining healthy teeth for a lifetime [60], a thorough mechanistic understanding of the signalling networks that control natural tooth formation or of how these networks could be altered to control the precise size and shape of bioengineered teeth is still unknown [61]. Also, there is a growing understanding of the significance of mechanical forces in controlling tooth morphogenesis [62]. To illustrate the significance of jaw-tooth interactions for typical tooth morphology, a recent research found that tooth buds cultivated in vitro without the presence of the surrounding jaw bone had dental cusp offsets[63]. The absence of suitable human DE cell sources is a significant obstacle to the development of therapeutically applicable bioengineered tooth buds. The most promising cell source at the moment may be autologous iPS cells, which might then be differentiated to produce DE cells capable of contributing to tooth creation, as was stated in the paragraph that came before. Cell homing has been suggested as a potential strategy for bioengineered replacement tooth creation in situ, in the jaw at the location of past tooth loss. This cutting-edge method depends on biomaterials that have the ability to produce bioactive chemicals to entice stem cells from the surrounding tissues to move to the scaffold, fill it, and eventually aid in the formation of new tissues [64].

IV. CONCLUSION

Stem cell treatments for dental tissue regeneration are now an intriguing possibility because to the current and substantial body of research, which has considerably increased understanding of how normal tooth development may direct dental tissue and entire tooth tissue engineering techniques. For a number of reasons, including the requirement to standardise and control techniques for DSC isolation, validation, expansion, handling, storage, and shipping, the utilisation of DSCs for clinical applications in dental tissue regeneration remains difficult. A significant issue is the exorbitant expenditures connected to ex vivo cell manipulation, not to mention the intrinsic risks connected to cell transplantation, such as contamination, pathogen transmission, and carcinogenesis [65]. However, we foresee a future with numerous applications, including the replacement of whole teeth lost to disease and/or trauma as well as the repair of significant craniomaxillofacial defects brought on by birth defects or trauma, as well as the repair of common tooth defects like dental caries and dental pulp regeneration. It is evident that continuing research into the biology of DSCs, dental tissue regeneration, and dental tissue development will lay a strong basis for future clinical treatments and regenerative methods in translational dental medicine.

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