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## REVIEW ARTICLE

# Dental Tissue Engineering by Neural Differentiation of Dental Stem Cells and Nano-systems: A Review

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### Abstract:

#### Background:

Pulpitis is a pulpal inflammation. It generally occurs when there is inflammation within a tooth as a result of anything like grinding or decay. After dental inflammation, microcirculation and sensory nerve activity seem to play the most critical role in reducing inflammation. Therefore, researchers emphasize the study of dental nerve activity, especially in acute clinical problems in inflamed teeth and pulp regeneration. This review aims to investigate the possibility of using dental stem cells to regenerate dental nerves in order to repair dentin-pulp complexes for maintaining and restoring tooth structure and function, which nanosystems can help in this matter.

#### Materials and Methods:

In this paper, we review the literature regarding the theory of dental tissue engineering by neural differentiation of dental stem cells and nano-systems, and the comprehensive search on PubMed, Scopus, and Web of Science was conducted up to July 2022.

#### Results:

According to recent studies, dental soft and hard tissue healing also includes nerve fibers. A deeper understanding of how dental nerves are implicated in pulpitis may assist endodontic treatment. Stem cell-based treatments may be used to regenerate dental nerves to repair dentin-pulp complexes to maintain and restore tooth structure and function.

#### Conclusion:

The emphasis on dental nerve regeneration appears to be a critical stage in fostering spontaneous tooth regeneration as well as a sustainable tooth regeneration method. It is essential to further investigate dental tissue engineering by neural differentiation of dental stem cells.

**Keywords:** Cell differentiation, Dental stem cells, Dental nerve, Dental restoration, Neural differentiation, Dental tissue engineering.

### Article History

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## 1. INTRODUCTION

One of the most serious public health concerns is tooth decay and pulp inflammation, often known as pulpitis. Pulpitis may be diagnosed clinically as a toothache caused by an accumulation of polymorpholeukocytes in the pulp area [1, 2]. Toothache, which varies in intensity depending on the stimulus which stimulates the damaged tooth, is sensed by a cascade of sensory nerve impulses to the brain [3]. One of the functions of dental nerves is to provide sensory signals to the central nervous system (CNS), which causes a variety of responses in

the body. Primary sensory fibers detect stimuli in the pulp of the tooth and initiate signal transmission [4]. According to sensory signals, dental nerve fibers are thought to be only responsible for delivering pain impulses in pulpitis [5 - 7]. However, recent studies have shown that they have also been linked to the regulation of inflammatory reactions.

It was proven that after tooth inflammation, microcirculation and sensory nerve activation diminish inflammation [8 - 11]. Microcirculation is affected by pulp nerve activity. Therefore, researchers emphasize upon the study of dental nerve activity, especially in acute clinical problems in inflamed teeth and pulp regeneration [12, 13]. Also, nerve fibers are an important component of the dental pulp, especially during

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inflammation, which directs the tooth's essential functions, such as angiogenesis, rousing immune cells, preserving pulp structure, and strengthening defense mechanisms. In addition, neuropeptides derived from nerve regeneration may aid in repairing injured teeth [14, 15]. In other words, the regenerated dental nerve stimulates the soft tooth component (pulp) and aids in the formation of the hard tooth component [16].

Stem cell-based treatments may be used to regenerate dental nerves to repair dentin-pulp complexes to maintain and restore tooth structure and function [17, 18]. Dental stem cells (DSCs) are found in the dental pulp and help to regenerate and repair damaged teeth by reducing inflammation caused by caries. To rebuild dental pulp, all neural crest-derived DSCs are pluripotent and grow into endothelium, adipocyte, odontoblast, chondrocyte, myocyte, neural, and osteoblast cells [19 - 22]. In addition, all DSCs were shown to be able to differentiate into neuron-like cells in tissue engineering of various target organs, including the brain and spinal cord, as defined *in vitro* situations [23 - 26].

Apexification and dental pulp stem cells (DPSCs) implantation were explored by Xuan and colleagues, to treat pulp necrosis in immature teeth. After 12 months, DPSCs implantation restored three-dimensional pulp tissues with blood channels and sensory neurons. In comparison to apexification, DPSCs implantation enhanced root length and reduced apical foramen width. DPSCs also regenerate sensory nerves in the tooth pulp. According to the results, DPSCs implantation may be able to restore entire dental pulp and repair trauma-related tooth damage [27].

Tooth regenerative therapy *via* DSCs mainly depends on tissue engineering by utilizing nano-structured biomaterials [28, 29]. Using nano-designed materials, it can now concentrate numerous diverse activities in a small volume and improve targeting quality while lowering the cost and delivery of active molecules. In addition, multi-active therapies and

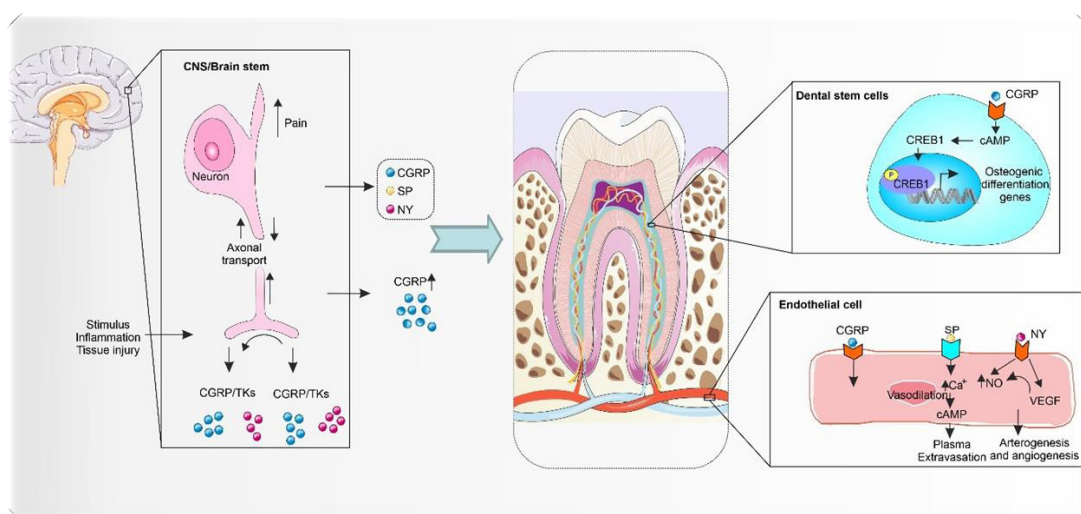
nanomaterials with extracellular mimetic nanostructures are critical for resolving infection and inflammation while guiding pulp cell colonization and differentiation [30, 31].

It has been observed that human neural tissue's extracellular matrix has a nano-to-micro hierarchical structure. Today, nanotechnology is used in all medical fields [32 - 36]. Nano-systems are the only option to reconstruct human neural tissue extracellular matrices with outstanding biomimetic properties and physicochemical properties, which makes significant advances in neural tissue engineering [37]. Nano-system is produced from a variety of natural and synthetic biomaterials [38 - 40], such as collagen [41, 42], chitosan [43], gelatin [44], poly- $\epsilon$ -caprolactone (PCL) [45], poly (lactic-co-glycolic acid) (PLGA) [46], *etc.* that promote neural tissue formation. Unfortunately, the number of studies related to tooth reconstruction, pertaining neural regeneration is very limited. Most studies focused on DSCs regenerating the central/peripheral nervous system [47 - 50].

This review first discussed dental tissue engineering by neural differentiation of dental stem cells and the role of nano-system in the regeneration of nerve tissue in the system of teeth.

## 2. SEARCH STRATEGY

To include related articles and studies of neurogenerative in relevance to teeth restoration, narrative research was conducted on PubMed, Scopus, and Web of Science up to July 2022. The research only involved full-text articles published in English. For instance, the research syntax used on PubMed included “dental stem cells,” “dental Stem cells,” DSC or DSCs, Neurogenesis, “Neurogenesis,” or “tooth's nerves,” “tooth nerve” or “dental nerve,” and “tooth regeneration,” or “dental regeneration,” “tooth restoration,” “dental restoration,” and, “nano-systems,” or “nano-system,” “nanosystem”, “nanosystems,” “nanocomposite”, “nanocomposites,” and, “nerve differentiation,” and, “nanotechnology,” or “nanotech.”



**Fig. (1).** The function of dental pulp neuronal-like cells for the regeneration of tooth.

### 3. ROLE OF DENTAL NERVE IN TOOTH DEVELOPMENT

The dental pulp is a neural network composed of the dental nerve of the fifth branch of the cranial nerve, which includes both sensory and sympathetic nerves. The sensory nerves of the pulp are situated in the trigeminal ganglion, where thousands of axons enter the tooth through the apical foramen and branch in the whole pulp. Most neural packets enter the dentin and form the parietal network of nerves (plexus of Raschkow). The end of the free nerve is on the odontoblast cell layer. The nerves continue to the coronal region, create a plexus in the vicinity of the odontoblasts and eventually join the dentinal tubules. They have diverse functions, positions, and associations with the pulp, dentin, vasculature, and immune cells. The dental pulp nerve secretes some neuropeptides such as neuropeptide Y (NY), substance P (SP), calcitonin gene-related peptide (CGRP), vasoactive intestinal polypeptide (VIP), galanin, and pituitary adenylate cyclase-activating polypeptide (PACAP), neurokinin A (NKA), *etc.* which have a vital role in regulating some function in teeth such as dentinogenesis, vascularization, immune reaction, and bone regeneration [51 - 54] (Fig. 1).

#### 3.1. Dentinogenesis (the formation of dentin)

Nerve fibers are involved in about 30 to 70% of the odontoblastic process, called intra-tubular nerves. Dentinal tubules include multiple nerve endings in the pre-dentin and dentin located at a distance of 100-150 micrometers from the pulp, which accompanies the odontoblast process [55]. CGRP is secreted from dental pulp nerves and is effective in dentin calcification [56]. According to another research, various concentrations of CGRP were studied on bone colonies. The results suggest that the higher concentration of this neuropeptide raises the ossification level [57]. The molecular mechanism of this reaction, CGRP, binds to target cell surface receptors that cause the expression of ossification-related genes [58]. Furthermore, the research found that CGRP plays a vital role in the creation of the dentin bridge during pulpotomy healing [59]. A post-pulpotomy study of first maxillary molars in 56-day-old Wistar rats showed that after seven days, large numbers of newly germinated CGRP-IR nerve fibers appeared in the remaining pulp, and some terminated in differentiating odontoblast layer and initial matrix layer of the dentin bridge were completed. These findings suggest that sensory neuropeptides such as CGRP may be involved in the formation of dentin bridges in the rat molar [59].

#### 3.2. Regulation of Vascularization

Preservation of pulp function is due to microcirculation, which provides tissues with nutrients and removes metabolites and wastes. During this circulation, the blood pressure in the pulp vascular system is coordinated with the interstitial tissue, and sufficient blood pressure is maintained in the pulp vascular system. The intensity of blood flow in the pulp vessels varies throughout life, including growth, puberty, and especially inflammation. In the first stage of inflammation, the intensity of blood flow and vascular permeability increases. Then, leukocytes migrate to the venular network [60, 61]. Under these conditions, the human pulp can see sufficient oxygen and nutrients for damaged tissue. Observations have shown that

pulp blood vessels are functionally related to nerve fibers [62]. Studies have shown that the rate of blood flow in the anterior or molar teeth of rats exposed to electrical stimulation increases, whereas if the inferior alveolar nerve is denervated, the rate of blood flow to the stimulus decreases [63 - 65]. NY is released by the dental pulp nerve, which is situated near blood vessels and reduces blood flow from the pulp *via* a calcium-dependent process.

In contrast, increased expression of NY regulates angiogenesis factors and repairs pulpal inflammation. Besides, interdental sensory nerves provide neuropeptides, such as CGRP and SP, that facilitate blood flow from the pulp [66 - 68]. In addition to neuropeptides secreted by sensory nerves, glutamate can regulate blood flow in vasodilation. Glutamate transporter-expressing neurons release glutamate into the pulp [69], which binds to the pulp's metabotropic glutamate receptors (GluRs), which regulate pulp blood flow [70].

As a result, neuropeptides secreted from the dental nerve have a critical role in regulating tooth function. Loss of nerve in the tooth stops blood flow to the tooth pulp and causes the dentin to change color (yellow, brown, even black), sometimes referred to as pulp necrosis, leading to tooth death. Different trauma studies in animals and humans have revealed that nerve fibers respond to dentin and pulp damage by germinating their terminal branches in the adjacent residual pulp and altering their neuropeptides. Therefore, a few days after injury, the undamaged nerve fibers return close to their original shape and function. Its feature has the role of restoring dental function [71 - 74]. El Karim *et al.* discovered that culturing human pulp fibroblasts in the presence of neuropeptides such as calcitonin gene-related peptides increased the expression of angiogenin and VEGF proteins. This is the first study to show that dental neuropeptides play a role in regulating angiogenic growth factor expression, implying that neuropeptides may play an important role in pulpal inflammation and repair in the future [66].

#### 3.3. Regulation of Immune Reaction

Dendritic cells (DCs) expressing class II antigens in the periodontoblastic region of the dental pulp (OX6<sup>+</sup>) are closely related to the nerve section of the tooth [75]. When tooth decay progresses to the dentin area and is not treated, DC cells and nerve cells in the tooth decay area move precisely at the pulp-dentin border [76]. At the level of myelinated nerve fibers in the dental pulp, there is a toll-like receptor (TLR)-4 and cluster of differentiation 14 (CD14), which are known as the first line of defense against infection and critical molecules in the innate immune system, respectively [77, 78]. The spatial similarity of the nerve endings to the DC cells indicates an almost similar function, suggesting the possibility of paracrine nerve interaction and immunity in the early stages of carious pulpitis. Therefore, the dental nerve is crucial for the dental immune system. A recent bioinformatics study was conducted to identify possible pulpitis biomarkers. Immune system signaling pathways such as the IL-7 signaling pathway, the Toll-like receptor signaling pathway, the NF-kappa B signaling pathway, and the TNF signaling pathway are activated during dental pulp inflammation, as are neuroactive ligand-receptor interactions [79].

Observations show that the activation of trigeminal sensory

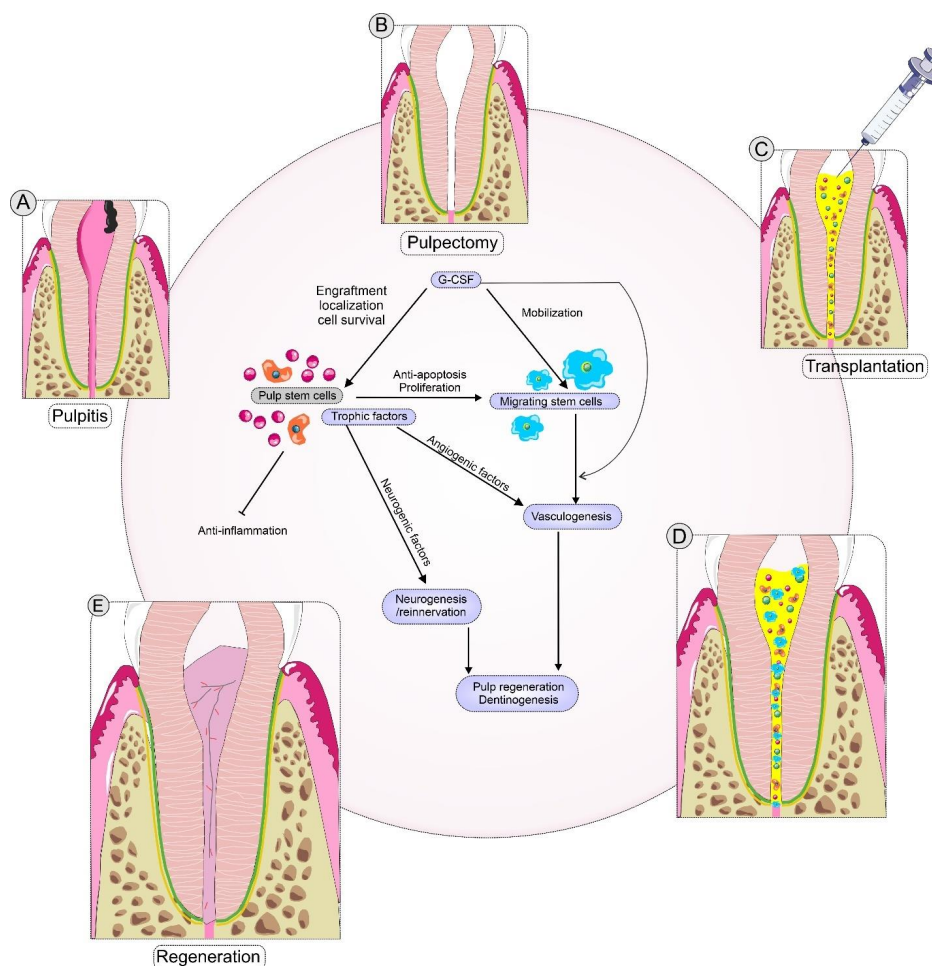
neurons combined with the sensitivity of transient receptor potential vanilloid subtype 1 (TRPV1) is mediated by the activation of the TLR4-mediated mechanism by lipopolysaccharides (LPS) microorganisms [80, 81]. This response lowers the activation threshold of nerve cells and leads to a hyperalgesic state. As previously described, neuropeptides are involved in regulating localized microcirculation in pulpitis. It promotes and directs immune cells to local tissues by increasing blood flow and vascular permeability. It has been observed that the denervated molar pulps of mice weaken the immune system in the inflammatory tissue of the pulp compared to healthy teeth [82, 83]. Ho *et al.* showed that at the surface of immune cells, such as monocytes/macrophages, there are receptors for receiving neuronal signals [84]; as a result, the tooth nerves can recruit immune cells directly. A recent study showed that macrophages, as inflammatory and immunologic mediators in the trigeminal ganglion, play a major part in the development of pulpitis and hyperalgesia according to the processes of CNS underpinning ectopic pain after peripheral inflammation [85]. Various studies have shown that the neuropeptides SP and CGRP interact with human monocytes and T lymphocytes

through chemical mechanisms [86, 87].

Following dental pulp injury, neuropeptides enhance vascular permeability and promote immune cell chemotaxis into the tooth pulp by direct contact with immune cells. If the dental nerve is destroyed, there is no longer a stimulus to invoke immune cells [82, 83]. Although, in healthy teeth, neuropeptide production and secretion decrease (35), it can be concluded that the main factor in maintaining the number of immune cells in healthy tooth pulp is not nerve cells.

### 3.4. The Role of Nerves in Apical Periodontitis

Inflammation of the teeth and pulpitis can affect periapical tissue and eventually lead to acute/chronic apical periodontitis. Reports have shown that the sensory nerves of the tooth gather in the periapical lesions and recruit macrophages and dendritic cells in periapical tissue. Thus, Dental nerves are essential in initiating the immune response and tissue repair in apical periodontitis [88, 89]. Apical periodontitis is associated with bone destruction in the periapical region, in which the sprouting of dental nerves for bone regeneration is observed [72, 90].



**Fig. (2).** Diagrams illustrating how the pulp regenerates following the implantation of pulp stem cells with G-CSF into permanent adult teeth of the dog pulpitis model, in which the stages are (A) teeth with pulpitis, (B) pulpectomy, (C, D) Transplantation, and (E) regeneration.

### 3.5. Reconstruction of the Dental Nerve in the Tooth Regeneration

Pulp/dentin reconstruction is one of the most critical challenges and the ultimate goal for functional tooth restoration. Tooth innervation can maintain dental pulp homeostasis. Tooth sensory nerves can stimulate the pulp's defense system by invoking immune and inflammatory cells to the injury site [65]. In the sensory denervation of damaged teeth, blood flow and immune cell recruitment are disrupted, resulting in necrosis of the tooth pulp [91, 92]. Fristad *et al.*, by intermittent stimulation of the rat pulp, observed that re-innervation/neurogenesis improves coronal dentin [65]. Ihara *et al.* examined pulp stem cells with granulocyte stimulating factor (G-CSF) in dog teeth to regenerate tooth pulp. The results showed that the nerves labeled DiI to the trigeminal ganglion responded positively to the electrical pulp test, and the sensory signals were felt as pain. Neurogenesis axons can aid in the process of angiogenesis, the release of immune cells, and the regulation of inflammation in damaged tissue to strengthen the pulp's defense mechanism (Fig. 2) [93]. Also, the blood flow in the regenerated pulp after two months was similar to that of a normal pulp, which could play an essential role in regenerating the pulp and dentin by controlling inflammation. The interaction of nerve fibers and blood flow maintains pulp tissue homeostasis, which can cause the enlarged apical portion to fade after pulpectomy and the formation of lateral dentin, which is a benefit in inhibiting tooth fractures. This restoration prevents secondary tooth decay and prolongs tooth life [93].

### 3.6. DSCs and Differentiated Neuronal Markers

Depending on their origin, DSCs inherit some of the typical features of nerve cells [94, 95]. DSCs inherit some of the typical features of nerve cells, depending on their origin. DSCs have been displayed to be able to express specific neuronal markers, for example, glial fibrillary acidic protein (GFAP), S100 calcium-binding protein B (S100B), and nestin. In addition, DSCs express more neuronal markers than bone marrow mesenchymal stem cells (BMSCs), indicating that DSCs can be utilized as perfect cells for neural differentiation and regeneration. Along with neuronal markers, the expression of neurotrophic factors is essential. Neurotrophic factors such as glial cell line-derived neurotrophic factor (GDNF), brain-derived neurotrophic factor (BDNF), and nerve growth factor (NGF) can be expressed through various DSCs. The prepared conditioning media from DSCs can induce neurite outgrowth and improve the growth rate of Schwann cells *in vitro* [96]. In addition, DSCs, unlike other somatic stem cells, display expression of embryonic stem cells (ESC) pluripotency markers, such as stage-specific embryonic antigen-4 (SSEA-4), octamer-binding transcription factor 4 (Oct-4), tumor rejection antigen (TRA)-1-60 and, Nanog, when derived from the immature dental pulp during the development of the tooth.

DSCs have neuron-like characteristics, which makes *in vitro* differentiation easier. Several methods for DSC-to-neuron differentiation have been developed. In summary, inductive protocols need to DSCs versatile differentiation. DSCs can be differentiated into neurons, Schwann cells, oligodendrocytes,

and dopaminergic-like cells. Therefore, DSCs are suitable stem cell sources to treat nervous diseases in stem cell therapy. The efficiency of nervous disease therapies with stem cells is strongly influenced by trophic factors such as GDNF, BDNF, NT-3, NGF, platelet-derived growth factor (PDGF), and vascular endothelial growth factor (VEGF) [97, 98]. The DSCs express these trophic factors more than MSCs derived from adipose tissue (ADSCs) and bone marrow (BMSCs) [97, 99]. Furthermore, DSCs display more neural supportive and neuroprotective properties than other MSCs in response to nervous system damage. DSCs possess the capability to decrease neurodegeneration by the secretion of NGF and BDNF in the neuronal apoptosis in early stages and stimulate the survival of motor and sensory neurons in spinal cord injury (SCI) [100, 101].

Additionally, secreted trophic factors *via* DSCs promoted the regeneration of axons despite the attendance of inhibitors of axon growth in the entirely transected spinal cord model of SCI [102, 103]. DSCs also secreted cytoprotective factors, providing direct and indirect protection against cell death in an ischemic astrocyte model of injury [104, 105]. DPSCs have displayed a higher cytokine expression facilitating neural differentiation compared with other stem cells, such as stem cells from the apical papilla (SCAP), Dental follicle stem cells (DFSCs), and BMSCs [106]. SCAP showed neural differentiation activity. During tooth development, the dental papilla converts to dental pulp tissue. Findings suggest that SCAP characteristics in terms of maturity are more similar to ectomesenchyme than DPSCs [107].

SCAP was derived from the apical papilla tissue of immature permanent human teeth, whose proliferation activity is two or three times more than DPSCs. These SCAPs express several neuronal markers such as  $\beta$ III-tubulin, nestin, GFAP, and neurofilament M upon stimulation of neural differentiation with EGF and bFGF [108]. DFSCs are isolated from wisdom teeth and have this multilineage differentiation capability [109]. Morszeck *et al.* reported that DFSCs could form neurosphere-like structures in serum-free media comprising FGF-2 and EGF plated onto low-attachment cell culture plates. Their study also showed that DFSCs-derived neural cells grown on gelatin and laminin show a relatively long axon-like cell extension, showing that the morphology of neural cells and axon-like cell extensions were related to cell culture substrates. In addition, they showed that the time-dependent addition of supplements to neural stem cells, such as retinoic acid, FGF-2, and EGF, affected the neuronal cell lineage differentiation into  $\gamma$ -aminobutyric acid -ergic (GABAergic) or dopaminergic neurons [109].

Inflammatory cytokines may both aid and impede the management of neurogenesis. IL-6 is a neuromodulator that affects brain stem cell renewal, progenitor cell division, and differentiation [110]. IL-4, IL-11, and IFN-g all promote increased neuronal differentiation [111]. SHED displayed a predominance expression of IL-6, IFN-g, and IL-4 in this study, but DPSCs revealed a 1.34-fold greater expression of IL-11 than SHED [112].

SHED showed substantially greater amounts of cytokines related to neuroprotective factor synthesis than DPSCs. Among



these cytokines are NT-3, BDNF, and GDNF. There is evidence that genetically altered cells may live longer and operate better, and that overexpressing neuroprotective proteins or neurotransmitters in cells might allow more rapid neural regeneration [113 - 115].

Among the neurogenesis-related cytokines, b-NGF expression was 6.85 times higher in DPSCs than in SHED [112]. NGF, a member of the protein family known as neurotrophins, is an important regulator of neurodevelopment and cell proliferation. In previous studies, it was found to be able to repair nerve injury in clinical therapy; Phyto *et al.* reported that increased levels of NGF expression supported reinnervation in pulp regeneration in a rat molar [116]. When employed as paracrine factors, NT-3, GDNF, and BDNF, which were shown to be more abundant in SHED than in DPSCs, were found to stimulate neuronal differentiation in both SHED and DPSCs. This enabled SHED to provide a more immunologically tolerant environment for stem cell transplantation, as well as superior circumstances for cell proliferation and neurogenesis. DPSCs, on the other hand, have been promoted as beneficial for angiogenesis.

The PDLSC secretome may increase the functioning of the PI3K/Akt/mTOR axis, leading to the restoration of BDNF production and the reduction of oxidative stress and inflammation in injured neurons. Furthermore, the presence of neuroprotective chemicals such as NT-3, IL-10, growth factors, and immunomodulatory cytokines in the conditioned medium provides a neuroprotective effect [117]. Furthermore, prior research indicated that the SCAP had much greater amounts of BDNF and GDNF than the other DSCs [103].

### 3.7. Nano-system Involved in Dental Nerve Differentiation and other Neural Tissue

One of the promising new therapies in the field of endodontics is dentin–pulp complex tissue engineering. The materials employed in the field of regenerative endodontics now, for instance, mineral trioxide and calcium hydroxide aggregates, cannot guarantee the entire dentin–pulp complex regeneration, particularly about neurovascular induction. A water-soluble graphene oxide–copper (GO-Cu) nanocomposite was fabricated by Li *et al.* [118], who studied its neurovascularization and odontogenic-inducing potentials *via* DSCs. Low doses ( $\leq 10 \mu\text{g/mL}$ ) of graphene oxide–copper was ineffective on DPSCs' promoted adhesion, proliferation, and viability. The treatment of DSCs with graphene oxide–copper led them to differentiate into odontoblasts and secrete higher quantities of GDNF and VEGF at the time when cultured on a scaffold made of calcium phosphate cement (CPC/GO-Cu) coated with graphene oxide–copper. The treatment of human umbilical vein endothelial cells (HUVECS) with graphene oxide–copper led to more substantial migratory potential, tube formation, and higher vascular endothelial growth factor expression. Additionally, using the graphene oxide–copper treatment, the immunomodulatory genes, such as hepatocyte growth factor (HGF), 2,3-dioxygenase (IDO), and human leukocyte antigen G (HLA-G), were upregulated in DPSCs. The subcutaneous transplantation of calcium phosphate cement/graphene oxide–copper into nude mice led to the

formation of dentin–pulp complex–like structures expressing CD31, growth-associated protein 43 (GAP43), and dentin sialophosphoprotein (DSPP), and the blood vessel numbers and the mineralized area were lower in the CPC-alone group than the CPC/GO-Cu group. These results reflect graphene oxide–copper's neurovascularization and odontogenic-inducing potentials and its promising applicability for regenerative endodontics purposes [118].

They reflected the graphene oxide–copper biocompatibility and its neurovascularization-inducing and odontogenic behaviors toward DPSCs. As a Cell Counting Kit-8 (CCK-8) assay, EdU incorporation and live/dead staining have affirmed that at low doses of GO-Cu, DSCs proliferate and survive actively. As the formation of the calcified nodule and odontogenic genes' protein and mRNA expression have shown, graphene oxide–copper may lead to promoted odontogenic differentiation of DPSCs. Improved migration, greater VEGF expression of HUVECs, higher tubular-structure formation, and higher expression of GDNF and VEGF in DSCs confirm the neurovascularization-inducing potential. In addition, graphene oxide–copper leads to increased expression of immunomodulatory genes HGF, IDO, and HLA-G in DPSCs. Eight weeks after the subcutaneous transplantation of CPC/GO-Cu loaded with DPSCs, dentin–pulp complex–structures are recreated in the defect region, and immunofluorescence staining reflects CD31, GAP43, and DSPP expression within the newly regenerated tissues. This research showed graphene oxide–copper's neurovascularization and odontogenic-inducing effects on DSCs, a new promising dental material used in regenerative endodontics [118].

The aligned electrospun PCL/PLGA material could be employed as a scaffold for spinal cord restoration. The inserted DFCSs onto PCL/PLGA can cause differentiation into neural cells in lesions of SCI. There is a hypothesis that aligned electrospun fibers have the potential to aid the spinal cord structure as well as cause the differentiation of neural cells [119]. A brand-new biomaterial called chitosan scaffolds has proved to facilitate inducing DSCs to turn into neural cells and can possibly be utilized in SCI therapy in the future [120]. Heparin-polyoxamer (HP), can simply create a gel at body temperature as a thermosensitive hydrogel; therefore, it is appropriate for *in vivo* treatments. It is revealed that nerve lesions could be repaired when HP is loaded with growth factors, namely FGF and NGF. This is achieved through activation of the Janus kinase (JAK)-signal transducer and activator of transcription (STAT), the mitogen-activated protein kinase (MAPK)/extracellular signal-regulated kinase 1/2 (ERK), The phosphatidylinositol 3-kinase (PI3K)/protein kinase B (AKT) and signaling pathways [121]. The combination of bFGF and DPSCs remarkably affected neuronal reproduction, recovery of function, and spinal cord tissue reconstruction [122]. According to statistical analyses, HP-bFGF-DPSCs outperform HP alone or HP-bFGF in nerve reconstruction [122]. DSC-based treatment will have a promising future for the treatment of SCI. Various studies showed that collagen scaffolds combined with neural-induced DPSCs could foster axonal outgrowth and myelination. The DPSCs could also be induced to differentiate into functional oligodendrocytes by increasing the exogenous gene expression

of the oligodendrocyte lineage transcription factor 2 (OLIG2). This presented a therapeutic capability in sciatic nerve injury [123].

#### 4. LIMITATIONS IN DENTAL TISSUE ENGINEERING AFTER IMPLANTING

Painful post-traumatic trigeminal neuropathy (PTTN) is a known complication of dental implant therapy. PTTN is a painful condition that may result from injury to the sensory division of the trigeminal nerve [124, 125]. Treatment of this condition is challenging and there is presently no established standard of treatment for managing neuropathic pain. Another challenge in tissue engineering is rejection. When the immune system recognizes the transplant as foreign, it rejects it, triggering a chain reaction that finally causes the transplanted organ or tissue to be destroyed. Cytokines, soluble molecules generated by many types of immunocompetent cells that impact each other, play an important role in the immune response [126]. The immunological responses were studied from two perspectives: cellular and humoral.

##### 4.1. Cellular Immunity

When the body is exposed to implant material, macrophages are the first phagocytes to get activated, resulting in the early stages of inflammation [127, 128]. The presence of macrophages in rejected implants was connected to the formation of granulation tissue, suggesting a connection between the two processes. Titanium metal particles were found in the cytoplasm of macrophages around the site of granulation tissue [129]. Moreover, several studies have shown that the presence of M2 macrophages in peri-implant tissue is associated with decreased inflammation, improved wound healing, and effective implant osseointegration [130, 131].

Dendritic cells are involved in the cellular immune response. However, more study is needed to discover the particular influence of DCs on dental implant rejection in order to achieve a positive outcome. Langerhans cells (LC), a kind of DC, were found in the stratified epithelium, such as the skin's epidermis and the mucosa lining the mouth. They have the ability to modify the immune milieu of the oral mucosa as well as preserve oral tissues during infection [132]. Gooty *et al.* also found that the number of LCs (factor CD1a) in the epithelium and lamina propria of healthy mucosa collected prior to implant placement was considerably greater than healthy peri-implant mucosa acquired at the time of gingival former implantation. The researchers concluded that diminished immune responses were caused by a smaller amount of LCs CD1a in the peri-implant tissue [133]. According to research, the quantity of mature LCs is decreasing as a result of dental implant installation. The implants, on the other hand, have a negative impact on precursor LCs and may increase their quantity in peri-implant tissue.

##### 4.2. Humoral Immunity

Cytokines, growth factors, and hormones are only a handful of the molecules that govern immune system cell communication [134]. Elevated IL-1 levels have been associated with dental implant failure, presenting a full picture

of host reactivity in patients with peri-implantitis collapse [135]. Furthermore, IL-1 is acknowledged as an important factor in differentiating between healthy implants and peri-implantitis [136, 137]. TNF- activates the significant mediators of inflammation when coupled with IL-1 [138]. Chronic inflammation, such as peri-implantitis and periodontitis, raises tumor necrosis factor-alpha levels. Extreme TNF- production, as shown by our findings [139], is a significant clinical issue for acute and chronic inflammatory diseases. Patients with failed implants exhibited considerably greater TNF- levels than those with successful implants. This TNF increase has been linked to the failure of dental implants [126, 136, 140 - 145].

Researchers discovered that titanium dental implants boost the synthesis and release of proinflammatory cytokines such as interleukin (IL)-1, tumor necrosis factor-alpha, interleukin (IL)-17, interleukin (IL)-6, interleukin (IL)-8, and interleukin (IL)-2. These upregulations may be linked to implant failure and osseointegration loss. However, various studies have shown contradictory results about the rise in these cytokines. More study is needed to identify the real role of these substances in dental implant rejection [146].

Interleukin-10 (IL-10) is a versatile cytokine that may affect a variety of hematopoietic cells. This cytokine's principal role is to inhibit or stop the inflammatory response. Because of its favorable *in vivo* regulatory influence, capacity to reduce acute inflammatory feedback, and ability to produce a negative feedback loop that lowers the release of inflammatory mediators [147], IL-10 is a crucial immune modulator [148]. Inhibiting the production of inflammatory cytokines such as IL-6 and TNF- may reduce macrophage and Th17 response activity [149]. TGF- $\beta$ 1 may be involved in the control of inflammation and immune suppression [150]. Important in wound healing and the local inflammatory response [127].

#### CONCLUSION

In this review, the complex role of the dental nerve in regenerating dentin and regulating the uptake of immune cells in pulp inflammation and tooth decay was discussed, which goes beyond the traditional view of nerve function only to transmit pain signals. These advances can provide abundant information about nerve and pulp function. Therefore, accurate knowledge of dental pulp neurology and regulatory mechanisms of pulp nerve network can be new therapeutic perspectives in endodontics and the development of approaches to target dental pulp neurons.

Nanotechnology for tissue engineering applications has made significant advances in the regeneration of bone, cartilage, arteries, nerves, and bladder tissue. However, the number of nanotechnology studies in pulp nerve regeneration has been minimal. Furthermore, the use of the unique properties of synthetic nanomaterials in extracellular matrix simulation (mimicking *in vivo* situations) has been neglected. Therefore, we propose that future studies focus on the cellular and molecular mechanisms of the nervous system in the context of nano-systems like nanocomposite to identify unknown parts of the role of the pulp nerve in tooth regeneration.

## LIST OF ABBREVIATIONS

- CNS = Central nervous system  
 DSCs = Dental stem cells  
 DPSCs = Dental pulp stem cells  
 NY = Neuropeptide Y  
 TRPV1 = Transient receptor potential vanilloid subtype 1

## CONSENT FOR PUBLICATION

Not applicable.

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## CONFLICT OF INTEREST

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## REFERENCES

- [1] Kim S, Liu M, Simchon S, Dörscher-Kim JE. Effects of selected inflammatory mediators on blood flow and vascular permeability in the dental pulp. *Proc Finn Dent Soc* 1992; 88(1): 387-92. [PMID: 1508895]
- [2] Zanini M, Meyer E, Simon S. Pulp inflammation diagnosis from clinical to inflammatory mediators: A systematic review. *J Endod* 2017; 43(7): 1033-51. [http://dx.doi.org/10.1016/j.joen.2017.02.009] [PMID: 28527838]
- [3] Hossain M, Bakri M, Yahya F, Ando H, Unno S, Kitagawa J. The role of transient receptor potential (TRP) channels in the transduction of dental pain. *Int J Mol Sci* 2019; 20(3): 526. [http://dx.doi.org/10.3390/ijms20030526] [PMID: 30691193]
- [4] Barnett EM, Evans GD, Sun N, Perlman S, Cassell MD. Anterograde tracing of trigeminal afferent pathways from the murine tooth pulp to cortex using herpes simplex virus type 1. *J Neurosci* 1995; 15(4): 2972-84. [http://dx.doi.org/10.1523/JNEUROSCI.15-04-02972.1995] [PMID: 7536824]
- [5] Renton T, Wilson NHF. Understanding and managing dental and orofacial pain in general practice. *Br J Gen Pract* 2016; 66(646): 236-7. [http://dx.doi.org/10.3399/bjgp16X684901] [PMID: 27127274]
- [6] Rodd HD, Boissonade FM. Innervation of human tooth pulp in relation to caries and dentition type. *J Dent Res* 2001; 80(1): 389-93. [http://dx.doi.org/10.1177/00220345010800011601] [PMID: 11269734]
- [7] Fukuda K. Diagnosis and treatment of abnormal dental pain. *J Dent Anesth Pain Med* 2016; 16(1): 1-8. [http://dx.doi.org/10.17245/jdapm.2016.16.1.1] [PMID: 28879289]
- [8] Edwall L, Jr DS. Influence of changes in microcirculation on the excitability of the sensory unit in the tooth of the cat. *Acta Physiol Scand* 1971; 82(4): 555-66. [http://dx.doi.org/10.1111/j.1748-1716.1971.tb05002.x] [PMID: 5098568]
- [9] Matthews B. Responses of intradental nerves to electrical and thermal stimulation of teeth in dogs. *J Physiol* 1977; 264(3): 641-64. [http://dx.doi.org/10.1113/jphysiol.1977.sp011687] [PMID: 557544]
- [10] Närhi MVO, Hirvonen TJ, Hakumäki MOK. Responses of intradental nerve fibres to stimulation of dentine and pulp. *Acta Physiol Scand* 1982; 115(2): 173-8. [http://dx.doi.org/10.1111/j.1748-1716.1982.tb07062.x] [PMID: 7136809]
- [11] Pagano S, Lombardo G, Costanzi E, et al. Morpho-functional effects of different universal dental adhesives on human gingival fibroblasts: An *in vitro* study. *Odontology* 2021; 109(2): 524-39. [http://dx.doi.org/10.1007/s10266-020-00569-x] [PMID: 33211211]
- [12] Kim S. Neurovascular interactions in the dental pulp in health and inflammation. *J Endod* 1990; 16(2): 48-53. [http://dx.doi.org/10.1016/S0099-2399(06)81563-3] [PMID: 2201743]
- [13] Heyeraas Tønder K, Naess G. Nervous control of blood flow in the dental pulp in dogs. *Acta Physiol Scand* 1978; 104(1): 13-23. [http://dx.doi.org/10.1111/j.1748-1716.1978.tb06246.x] [PMID: 696351]
- [14] Bhandi S, Alkahtani A, Mashyakhly M, et al. Effect of ascorbic acid on differentiation, secretome and stemness of stem cells from human exfoliated deciduous tooth (SHEDs). *J Pers Med* 2021; 11(7): 589. [http://dx.doi.org/10.3390/jpm11070589] [PMID: 34206203]
- [15] Patil S, Reda R, Boreak N, et al. Adipogenic stimulation and pyrrolidine dithiocarbamate induced osteogenic inhibition of dental pulp stem cells is countered by cordycepin. *J Pers Med* 2021; 11(9): 915. [http://dx.doi.org/10.3390/jpm11090915] [PMID: 34575692]
- [16] Zhan C, Huang M, Yang X, Hou J. Dental nerves: A neglected mediator of pulpitis. *Int Endod J* 2021; 54(1): 85-99. [http://dx.doi.org/10.1111/iej.13400] [PMID: 32880979]
- [17] Volponi AA, Pang Y, Sharpe PT. Stem cell-based biological tooth repair and regeneration. *Trends Cell Biol* 2010; 20(12): 715-22. [http://dx.doi.org/10.1016/j.tcb.2010.09.012] [PMID: 21035344]
- [18] Yamada Y, Nakamura-Yamada S, Konoki R, Baba S. Promising advances in clinical trials of dental tissue-derived cell-based regenerative medicine. *Stem Cell Res Ther* 2020; 11(1): 175. [http://dx.doi.org/10.1186/s13287-020-01683-x] [PMID: 32398041]
- [19] Lee SM, Zhang Q, Le AD. Dental stem cells: Sources and potential applications. *Curr Oral Health Rep* 2014; 1(1): 34-42. [http://dx.doi.org/10.1007/s40496-014-0012-0]
- [20] Yoshida S, Tomokiyo A, Hasegawa D, Hamano S, Sugii H, Maeda H. Insight into the role of dental pulp stem cells in regenerative therapy. *Biology* 2020; 9(7): 160. [http://dx.doi.org/10.3390/biology9070160] [PMID: 32659896]
- [21] Miran S, Mitsiadis TA, Pagella P. Innovative dental stem cell-based research approaches: The future of dentistry Stem cells int 2016; 2016 [http://dx.doi.org/10.1155/2016/7231038]
- [22] Hughes D, Song B. Dental and nondental stem cell based regeneration of the craniofacial region: A tissue based approach. *Stem cells int* 2016; 2016 [http://dx.doi.org/10.1155/2016/8307195]
- [23] Kandalam S, De Berdt P, Ucakar B, et al. Human dental stem cells of the apical papilla associated to BDNF-loaded pharmacologically active microcarriers (PAMs) enhance locomotor function after spinal cord injury. *Int J Pharm* 2020; 587: 119685. [http://dx.doi.org/10.1016/j.ijpharm.2020.119685] [PMID: 32712253]
- [24] De Berdt P, Vanacker J, Ucakar B, et al. Dental apical papilla as therapy for spinal cord injury. *J Dent Res* 2015; 94(11): 1575-81. [http://dx.doi.org/10.1177/0022034515604612] [PMID: 26341974]
- [25] Wu T, Xu W, Chen H, et al. Comparison of the differentiation of dental pulp stem cells and periodontal ligament stem cells into neuron-like cells and their effects on focal cerebral ischemia. *Acta Biochim Biophys Sin* 2020; 52(9): 1016-29. [http://dx.doi.org/10.1093/abbs/gmaa082] [PMID: 32845287]
- [26] Rajan TS, Giacoppo S, Diomedea F, et al. The secretome of periodontal ligament stem cells from MS patients protects against EAE. *Sci Rep* 2016; 6(1): 38743. [http://dx.doi.org/10.1038/srep38743] [PMID: 27924938]
- [27] Xuan K, Li B, Guo H, et al. Deciduous autologous tooth stem cells regenerate dental pulp after implantation into injured teeth. *Sci Transl Med* 2018; 10(455): eaaf3227. [http://dx.doi.org/10.1126/scitranslmed.aaf3227] [PMID: 30135248]
- [28] Samiei M, et al. Early osteogenic differentiation stimulation of dental pulp stem cells by calcitriol and curcumin. *Stem cells int* 2021; 2021. [http://dx.doi.org/10.1155/2021/9980137]
- [29] Sharifi S, Zaheri Khosroshahi A, Maleki Dizaj S, Rezaei Y. Preparation, physicochemical assessment and the antimicrobial action of hydroxyapatite-gelatin/curcumin nanofibrous composites as a dental biomaterial. *Biomimetics* 2021; 7(1): 4. [http://dx.doi.org/10.3390/biomimetics7010004] [PMID: 35076470]
- [30] Qu T, Liu X. Nano-structured gelatin/bioactive glass hybrid scaffolds for the enhancement of odontogenic differentiation of human dental pulp stem cells. *J Mater Chem B Mater Biol Med* 2013; 1(37): 4764-72. [http://dx.doi.org/10.1039/c3tb21002b] [PMID: 24098854]
- [31] Qu T, Jing J, Ren Y, et al. Complete pulpodentin complex



- regeneration by modulating the stiffness of biomimetic matrix. *Acta Biomater* 2015; 16: 60-70.  
[<http://dx.doi.org/10.1016/j.actbio.2015.01.029>] [PMID: 25644448]
- [32] Hamidi-Asl E, Raouf JB, Hejazi MS, *et al.* A genosensor for point mutation detection of P53 gene PCR product using magnetic particles. *Electroanalysis* 2015; 27(6): 1378-86.  
[<http://dx.doi.org/10.1002/elan.201400660>]
- [33] Hamidi A, Sharifi S, Davaran S, Ghasemi S, Omid Y, Rashidi MR. Novel aldehyde-terminated dendrimers; synthesis and cytotoxicity assay. *Bioimpacts* 2012; 2(2): 97-103.  
[PMID: 23678447]
- [34] Maleki Dizaj S, Sharifi S, Jahangiri A. Electrospun nanofibers as versatile platform in antimicrobial delivery: current state and perspectives. *Pharm Dev Technol* 2019; 24(10): 1187-99.  
[<http://dx.doi.org/10.1080/10837450.2019.1656238>] [PMID: 31424308]
- [35] Sharifi S, Samani AA, Ahmadian E, *et al.* Oral delivery of proteins and peptides by mucoadhesive nanoparticles. *Biointerface Res Appl Chem* 2019; 9(2): 3849-52.  
[<http://dx.doi.org/10.33263/BRIAC92.849852>]
- [36] Maleki Dizaj S, Alipour M, Dalir Abdollahinia E, *et al.* Curcumin nanoformulations: Beneficial nanomedicine against cancer. *Phytother Res* 2022; 36(3): 1156-81.  
[<http://dx.doi.org/10.1002/ptr.7389>] [PMID: 35129230]
- [37] Zhang L, Webster TJ. Nanotechnology and nanomaterials: Promises for improved tissue regeneration. *Nano Today* 2009; 4(1): 66-80.  
[<http://dx.doi.org/10.1016/j.nantod.2008.10.014>]
- [38] Shahi S, Özcan M, Maleki Dizaj S, *et al.* A review on potential toxicity of dental material and screening their biocompatibility. *Toxicol Mech Methods* 2019; 29(5): 368-77.  
[<http://dx.doi.org/10.1080/15376516.2019.1566424>] [PMID: 30642212]
- [39] Ahmadian E, Shahi S, Yazdani J, Maleki Dizaj S, Sharifi S. Local treatment of the dental caries using nanomaterials. *Biomed Pharmacother* 2018; 108: 443-7.  
[<http://dx.doi.org/10.1016/j.biopha.2018.09.026>] [PMID: 30241047]
- [40] Khezri K, Dijaz SM, Saadat YR, *et al.* Osteogenic differentiation of mesenchymal stem cells via curcumin-containing nanoscaffolds. *Stem cells int* 2021; 2021 Article ID: 1520052  
[<http://dx.doi.org/10.1155/2021/1520052>]
- [41] Li X, Fan C, Xiao Z, *et al.* A collagen microchannel scaffold carrying paclitaxel-liposomes induces neuronal differentiation of neural stem cells through Wnt/ $\beta$ -catenin signaling for spinal cord injury repair. *Biomaterials* 2018; 183: 114-27.  
[<http://dx.doi.org/10.1016/j.biomaterials.2018.08.037>] [PMID: 30153562]
- [42] Sharifi S, Dizaj SM, Samiei M, *et al.* Application of collagen and mesenchymal stem cells in regenerative dentistry. *Curr Stem Cell Res Ther* 2022; 17(7): 606-20.  
[<http://dx.doi.org/10.2174/1574888X17666211220100521>] [PMID: 34931969]
- [43] Wu D, Zhang Y, Xu X, *et al.* RGD/TAT-functionalized chitosan-graft-PEI-PEG gene nanovector for sustained delivery of NT-3 for potential application in neural regeneration. *Acta Biomater* 2018; 72: 266-77.  
[<http://dx.doi.org/10.1016/j.actbio.2018.03.030>] [PMID: 29578088]
- [44] Wang S, Guan S, Li W, *et al.* 3D culture of neural stem cells within conductive PEDOT layer-assembled chitosan/gelatin scaffolds for neural tissue engineering. *Mater Sci Eng C* 2018; 93: 890-901.  
[<http://dx.doi.org/10.1016/j.msec.2018.08.054>] [PMID: 30274126]
- [45] Zhang S, Wang XJ, Li WS, *et al.* Polycaprolactone/polysialic acid hybrid, multifunctional nanofiber scaffolds for treatment of spinal cord injury. *Acta Biomater* 2018; 77: 15-27.  
[<http://dx.doi.org/10.1016/j.actbio.2018.06.038>] [PMID: 30126591]
- [46] Rao J, Cheng Y, Liu Y, *et al.* A multi-walled silk fibroin/silk sericin nerve conduit coated with poly(lactic-co-glycolic acid) sheath for peripheral nerve regeneration. *Mater Sci Eng C* 2017; 73: 319-32.  
[<http://dx.doi.org/10.1016/j.msec.2016.12.085>] [PMID: 28183615]
- [47] Das S, Bellare JR. Dental pulp stem cells in customized 3D nanofibrous scaffolds for regeneration of peripheral nervous system. *Methods Mol Biol* 2018; 2125: 157-66.  
[[http://dx.doi.org/10.1007/9781\\_2018\\_194](http://dx.doi.org/10.1007/9781_2018_194)] [PMID: 30294747]
- [48] Li J, Diao S, Yang H, Cao Y, Du J, Yang D. IGFBP5 promotes angiogenic and neurogenic differentiation potential of dental pulp stem cells. *Dev Growth Differ* 2019; 61(9): 457-65.  
[<http://dx.doi.org/10.1111/dgd.12632>] [PMID: 31599466]
- [49] Vizoso F, Eiro N, Cid S, Schneider J, Perez-Fernandez R. Mesenchymal stem cell secretome: Toward cell-free therapeutic strategies in regenerative medicine. *Int J Mol Sci* 2017; 18(9): 1852.  
[<http://dx.doi.org/10.3390/ijms18091852>] [PMID: 28841158]
- [50] Ibarretxe G, Crende O, Aurrekoetxea M, Garcia-Murga V, Etxaniz J, Uda F. Neural crest stem cells from dental tissues: A new hope for dental and neural regeneration. *Stem Cells Int* 2012; 2012: 1-12.  
[<http://dx.doi.org/10.1155/2012/103503>] [PMID: 23093977]
- [51] Hildebrand C, Fried K, Tuisku F, Johansson CS. Teeth and tooth nerves. *Prog Neurobiol* 1995; 45(3): 165-222.  
[[http://dx.doi.org/10.1016/0301-0082\(94\)00045-J](http://dx.doi.org/10.1016/0301-0082(94)00045-J)] [PMID: 7777672]
- [52] Norevall LI, Forsgren S. NPY/sympathetic and NPY/VIP innervation of the blood vessels supplying rat tooth-related structures: Effects of sympathectomy. *Neuropeptides* 1999; 33(3): 216-26.  
[<http://dx.doi.org/10.1054/npep.1999.0758>] [PMID: 10657495]
- [53] Swanson WB, Gong T, Zhang Z, *et al.* Controlled release of odontogenic exosomes from a biodegradable vehicle mediates dentinogenesis as a novel biomimetic pulp capping therapy. *J Control Release* 2020; 324: 679-94.  
[<http://dx.doi.org/10.1016/j.jconrel.2020.06.006>] [PMID: 32534011]
- [54] Batih V, Khabadze Z, Ahmad W, Nazarova D, *et al.* Treatment of chronic apical periodontitis in patients with prevalent parasympathetic vegetative nervous system. *Deutscher Wissenschaftsberohd German Science Herald* 2017; p. 69.
- [55] Boaz K. Orban's Oral Histology and Embryology. Elsevier 2015.
- [56] Silverman JD, Kruger L. An interpretation of dental innervation based upon the pattern of calcitonin gene-related peptide (CGRP)-immunoreactive thin sensory axons. *Somatosens Res* 1987; 5(2): 157-75.  
[<http://dx.doi.org/10.3109/07367228709144624>] [PMID: 3501146]
- [57] Bernard GW, Shih C. The osteogenic stimulating effect of neuroactive calcitonin gene-related peptide. *Peptides* 1990; 11(4): 625-32.  
[[http://dx.doi.org/10.1016/0196-9781\(90\)90171-Z](http://dx.doi.org/10.1016/0196-9781(90)90171-Z)] [PMID: 2235673]
- [58] Zhang Y, Xu J, Ruan YC, *et al.* Implant-derived magnesium induces local neuronal production of CGRP to improve bone-fracture healing in rats. *Nat Med* 2016; 22(10): 1160-9.  
[<http://dx.doi.org/10.1038/nm.4162>] [PMID: 27571347]
- [59] Zhang M, Fukuyama H. CGRP immunohistochemistry in wound healing and dentin bridge formation following rat molar pulpotomy. *Histochem Cell Biol* 1999; 112(5): 325-33.  
[<http://dx.doi.org/10.1007/s004180050413>] [PMID: 10603071]
- [60] Tonder KJH. Vascular reactions in the dental pulp during inflammation. *Acta Odontol Scand* 1983; 41(4): 247-56.  
[<http://dx.doi.org/10.3109/00016358309162331>] [PMID: 6578666]
- [61] Takahashi K. Changes in the pulpal vasculature during inflammation. *J Endod* 1990; 16(2): 92-7.  
[[http://dx.doi.org/10.1016/S0099-2399\(06\)81570-0](http://dx.doi.org/10.1016/S0099-2399(06)81570-0)] [PMID: 2388023]
- [62] Rodd HD, Boissonade FM. Immunocytochemical investigation of neurovascular relationships in human tooth pulp. *J Anat* 2003; 202(2): 195-203.  
[<http://dx.doi.org/10.1046/j.1469-7580.2003.00153.x>] [PMID: 12647869]
- [63] Kerezoudis NP, Fried K, Olgart L. Haemodynamic and immunohistochemical studies of rat incisor pulp after denervation and subsequent re-innervation. *Arch Oral Biol* 1995; 40(9): 815-23.  
[[http://dx.doi.org/10.1016/0003-9969\(95\)00048-T](http://dx.doi.org/10.1016/0003-9969(95)00048-T)] [PMID: 8651885]
- [64] Kerezoudis NP, Nomikos GG, Olgart LM, Svensson TH. Serotonin in rat oral tissues: Role of 5-HT1 receptors in sympathetic vascular control. *Eur J Pharmacol* 1995; 275(2): 191-8.  
[[http://dx.doi.org/10.1016/0014-2999\(94\)00772-Y](http://dx.doi.org/10.1016/0014-2999(94)00772-Y)] [PMID: 7540979]
- [65] Fristad I, Kvinnsland IH, Jonsson R, Heyeraas KJ. Effect of intermittent long-lasting electrical tooth stimulation on pulpal blood flow and immunocompetent cells: A hemodynamic and immunohistochemical study in young rat molars. *Exp Neurol* 1997; 146(1): 230-9.  
[<http://dx.doi.org/10.1006/exnr.1997.6523>] [PMID: 9225756]
- [66] El karim IA, Linden GJ, Irwin CR, Lundy FT. Neuropeptides regulate expression of angiogenic growth factors in human dental pulp fibroblasts. *J Endod* 2009; 35(6): 829-33.  
[<http://dx.doi.org/10.1016/j.joen.2009.03.005>] [PMID: 19482180]
- [67] Kerezoudis NP, Olgart L, Edwall L. Involvement of substance P but not nitric oxide or calcitonin gene-related peptide in neurogenic plasma extravasation in rat incisor pulp and lip. *Arch Oral Biol* 1994; 39(9): 769-74.  
[[http://dx.doi.org/10.1016/0003-9969\(94\)90006-X](http://dx.doi.org/10.1016/0003-9969(94)90006-X)] [PMID: 7528497]
- [68] Heyeraas KJ, Kim S, Raab WHM, Byers MR, Liu M. Effect of electrical tooth stimulation on blood flow, interstitial fluid pressure and substance P and CGRP-immunoreactive nerve fibers in the low compliant cat dental pulp. *Microvasc Res* 1994; 47(3): 329-43.

- [69] [\[http://dx.doi.org/10.1006/mvre.1994.1026\]](http://dx.doi.org/10.1006/mvre.1994.1026) [PMID: 7521929]  
Zerari-Mailly F, Braud A, Davido N, Touré B, Azérad J, Boucher Y. Glutamate control of pulpal blood flow in the incisor dental pulp of the rat. *Eur J Oral Sci* 2012; 120(5): 402-7.  
[\[http://dx.doi.org/10.1111/j.1600-0722.2012.00989.x\]](http://dx.doi.org/10.1111/j.1600-0722.2012.00989.x) [PMID: 22984997]
- [70] Hofman S, Azérad J, Boucher Y. Effects of excitatory amino acid receptor antagonists on pulpal blood flow of the rat mandibular incisor. *J Dent Res* 2002; 81(4): 253-8.  
[\[http://dx.doi.org/10.1177/154405910208100406\]](http://dx.doi.org/10.1177/154405910208100406) [PMID: 12097309]
- [71] Brännström M. Etiology of dentin hypersensitivity. *Proc Finn Dent Soc* 1992; 88(1): 7-13.  
[PMID: 1508916]
- [72] Byers MR, Närhi MVO. Dental injury models: Experimental tools for understanding neuroinflammatory interactions and polymodal nociceptor functions. *Crit Rev Oral Biol Med* 1999; 10(1): 4-39.  
[\[http://dx.doi.org/10.1177/10454411990100010101\]](http://dx.doi.org/10.1177/10454411990100010101) [PMID: 10759425]
- [73] Oswald RJ, Byers MR. The injury response of pulpal NPY-IR sympathetic fibers differs from that of sensory afferent fibers. *Neurosci Lett* 1993; 164(1-2): 190-4.  
[\[http://dx.doi.org/10.1016/0304-3940\(93\)90889-S\]](http://dx.doi.org/10.1016/0304-3940(93)90889-S) [PMID: 7512246]
- [74] Okiji T, Jontell M, Belichenko P, Dahlgren U, Bergenholtz G, Dahlström A. Structural and functional association between substance P- and calcitonin gene-related peptide-immunoreactive nerves and accessory cells in the rat dental pulp. *J Dent Res* 1997; 76(12): 1818-24.  
[\[http://dx.doi.org/10.1177/00220345970760120301\]](http://dx.doi.org/10.1177/00220345970760120301) [PMID: 9390474]
- [75] Sakurai K, Okiji T, Suda H. Co-increase of nerve fibers and HLA-DR- and/or factor-XIIIa-expressing dendritic cells in dentinal caries-affected regions of the human dental pulp: an immunohistochemical study. *J Dent Res* 1999; 78(10): 1596-608.  
[\[http://dx.doi.org/10.1177/00220345990780100401\]](http://dx.doi.org/10.1177/00220345990780100401) [PMID: 10520964]
- [76] Wadachi R, Hargreaves KM. Trigeminal nociceptors express TLR-4 and CD14: A mechanism for pain due to infection. *J Dent Res* 2006; 85(1): 49-53.  
[\[http://dx.doi.org/10.1177/154405910608500108\]](http://dx.doi.org/10.1177/154405910608500108) [PMID: 16373680]
- [77] Couve E, Osorio R, Schmachtenberg O. Reactionary dentinogenesis and neuroimmune response in dental caries. *J Dent Res* 2014; 93(8): 788-93.  
[\[http://dx.doi.org/10.1177/0022034514539507\]](http://dx.doi.org/10.1177/0022034514539507) [PMID: 24928097]
- [78] Xin B, Lin Y, Tian H, Song J, Zhang L, Lv J. Identification of pulpitis-related potential biomarkers using bioinformatics approach. *Comput Math Methods Med* 2021; 2021: 1-12.  
[\[http://dx.doi.org/10.1155/2021/1808361\]](http://dx.doi.org/10.1155/2021/1808361) [PMID: 34630628]
- [79] Diogenes A, Ferraz CCR, Akopian AN, Henry MA, Hargreaves KM. LPS sensitizes TRPV1 via activation of TLR4 in trigeminal sensory neurons. *J Dent Res* 2011; 90(6): 759-64.  
[\[http://dx.doi.org/10.1177/0022034511400225\]](http://dx.doi.org/10.1177/0022034511400225) [PMID: 21393555]
- [80] Ferraz CCR, Henry MA, Hargreaves KM, Diogenes A. Lipopolysaccharide from porphyromonas gingivalis sensitizes capsaicin-sensitive nociceptors. *J Endod* 2011; 37(1): 45-8.  
[\[http://dx.doi.org/10.1016/j.joen.2007.07.001\]](http://dx.doi.org/10.1016/j.joen.2007.07.001) [PMID: 21146075]
- [81] Fristad I, Heyeraas KJ, Jonsson R, Kvinnsland IH. Effect of inferior alveolar nerve axotomy on immune cells and nerve fibres in young rat molars. *Arch Oral Biol* 1995; 40(11): 1053-62.  
[\[http://dx.doi.org/10.1016/0003-9969\(95\)00065-W\]](http://dx.doi.org/10.1016/0003-9969(95)00065-W) [PMID: 8670024]
- [82] Fristad I, Heyeraas KJ, Kvinnsland IH, Jonsson R. Recruitment of immunocompetent cells after dentinal injuries in innervated and denervated young rat molars: An immunohistochemical study. *J Histochem Cytochem* 1995; 43(9): 871-9.  
[\[http://dx.doi.org/10.1177/43.9.7543913\]](http://dx.doi.org/10.1177/43.9.7543913) [PMID: 7543913]
- [83] Ho WZ, Lai JP, Zhu XH, Uvaydova M, Douglas SD. Human monocytes and macrophages express substance P and neurokinin-1 receptor. *J Immunol* 1997; 159(11): 5654-60.  
[\[http://dx.doi.org/10.4049/jimmunol.159.11.5654\]](http://dx.doi.org/10.4049/jimmunol.159.11.5654) [PMID: 9548509]
- [84] Gao L, Fan F, Wang L, et al. Polarization of macrophages in the trigeminal ganglion of rats with pulpitis. *J Oral Rehabil* 2022; 49(2): 228-36.  
[\[http://dx.doi.org/10.1111/joor.13245\]](http://dx.doi.org/10.1111/joor.13245) [PMID: 34398484]
- [85] Ruff MR, Wahl SM, Pert CB. Substance P receptor-mediated chemotaxis of human monocytes. *Peptides* 1985; 6(S2): 107-11.  
[\[http://dx.doi.org/10.1016/0196-9781\(85\)90142-1\]](http://dx.doi.org/10.1016/0196-9781(85)90142-1) [PMID: 2417206]
- [86] Foster CA, Mandak B, Kromer E, Rot A. Calcitonin gene-related peptide is chemotactic for human T lymphocytes. *Ann N Y Acad Sci* 1992; 657(1 Calcitonin Ge): 397-404.  
[\[http://dx.doi.org/10.1111/j.1749-6632.1992.tb22785.x\]](http://dx.doi.org/10.1111/j.1749-6632.1992.tb22785.x) [PMID: 1637095]
- [87] Toriya Y, Hashiguchi I, Maeda K. Immunohistochemical examination of the distribution of macrophages and CGRP-immunoreactive nerve fibers in induced rat periapical lesions. *Dent Traumatol* 1997; 13(1): 6-12.  
[\[http://dx.doi.org/10.1111/j.1600-9657.1997.tb00002.x\]](http://dx.doi.org/10.1111/j.1600-9657.1997.tb00002.x) [PMID: 9206382]
- [88] Yang G, Kawashima N, Kaneko T, Suzuki N, Okiji T, Suda H. Kinetic study of immunohistochemical colocalization of antigen-presenting cells and nerve fibers in rat periapical lesions. *J Endod* 2007; 33(2): 132-6.  
[\[http://dx.doi.org/10.1016/j.joen.2006.08.014\]](http://dx.doi.org/10.1016/j.joen.2006.08.014) [PMID: 17258630]
- [89] Kimberly CL, Byers MR. Inflammation of rat molar pulp and periodontium causes increased calcitonin gene-related peptide and axonal sprouting. *Anat Rec* 1988; 222(3): 289-300.  
[\[http://dx.doi.org/10.1002/ar.1092220310\]](http://dx.doi.org/10.1002/ar.1092220310) [PMID: 3265042]
- [90] Haug SR, Heyeraas KJ. Effects of sympathectomy on experimentally induced pulpal inflammation and periapical lesions in rats. *Neuroscience* 2003; 120(3): 827-36.  
[\[http://dx.doi.org/10.1016/S0306-4522\(03\)00269-0\]](http://dx.doi.org/10.1016/S0306-4522(03)00269-0) [PMID: 12895522]
- [91] Byers MR, Taylor PE. Effect of sensory denervation on the response of rat molar pulp to exposure injury. *J Dent Res* 1993; 72(3): 613-8.  
[\[http://dx.doi.org/10.1177/00220345930720031001\]](http://dx.doi.org/10.1177/00220345930720031001) [PMID: 8450121]
- [92] Olgart L, Kostouros GD, Edwall L. Local actions of acetylcholine on vasomotor regulation in rat incisor pulp. *Acta Physiol Scand* 1996; 158(4): 311-6.  
[\[http://dx.doi.org/10.1046/j.1365-201X.1996.69319000.x\]](http://dx.doi.org/10.1046/j.1365-201X.1996.69319000.x) [PMID: 8971251]
- [93] Iohara K, Murakami M, Takeuchi N, et al. A novel combinatorial therapy with pulp stem cells and granulocyte colony-stimulating factor for total pulp regeneration. *Stem Cells Transl Med* 2013; 2(7): 521-33.  
[\[http://dx.doi.org/10.5966/sctm.2012-0132\]](http://dx.doi.org/10.5966/sctm.2012-0132) [PMID: 23761108]
- [94] Gronthos S, Mankani M, Brahimi J, Robey PG, Shi S. Postnatal human dental pulp stem cells (DPSCs) *in vitro* and *in vivo*. *Proc Natl Acad Sci* 2000; 97(25): 13625-30.  
[\[http://dx.doi.org/10.1073/pnas.240309797\]](http://dx.doi.org/10.1073/pnas.240309797) [PMID: 11087820]
- [95] Miura M, Gronthos S, Zhao M, et al. SHED: Stem cells from human exfoliated deciduous teeth. *Proc Natl Acad Sci* 2003; 100(10): 5807-12.  
[\[http://dx.doi.org/10.1073/pnas.0937635100\]](http://dx.doi.org/10.1073/pnas.0937635100) [PMID: 12716973]
- [96] Wang D, Wang Y, Tian W, Pan J. Advances of tooth-derived stem cells in neural diseases treatments and nerve tissue regeneration. *Cell Prolif* 2019; 52(3): e12572.  
[\[http://dx.doi.org/10.1111/cpr.12572\]](http://dx.doi.org/10.1111/cpr.12572) [PMID: 30714230]
- [97] Mead B, Logan A, Berry M, Leadbeater W, Scheven BA. Paracrine-mediated neuroprotection and neurogenesis of axotomized retinal ganglion cells by human dental pulp stem cells: Comparison with human bone marrow and adipose-derived mesenchymal stem cells. *PLoS One* 2014; 9(10): e109305.  
[\[http://dx.doi.org/10.1371/journal.pone.0109305\]](http://dx.doi.org/10.1371/journal.pone.0109305) [PMID: 25290916]
- [98] Sakai K, Yamamoto A, Matsubara K, et al. Human dental pulp-derived stem cells promote locomotor recovery after complete transection of the rat spinal cord by multiple neuro-regenerative mechanisms. *J Clin Invest* 2012; 122(1): 80-90.  
[PMID: 22133879]
- [99] Caseiro AR, et al. Neuromuscular regeneration: Perspective on the application of mesenchymal stem cells and their secretion products. *Stem cells int* 2016; 2016  
[\[http://dx.doi.org/10.1155/2016/9756973\]](http://dx.doi.org/10.1155/2016/9756973)
- [100] Mead B, Logan A, Berry M, Leadbeater W, Scheven BA. Intravitreally transplanted dental pulp stem cells promote neuroprotection and axon regeneration of retinal ganglion cells after optic nerve injury. *Invest Ophthalmol Vis Sci* 2013; 54(12): 7544-56.  
[\[http://dx.doi.org/10.1167/iovs.13-13045\]](http://dx.doi.org/10.1167/iovs.13-13045) [PMID: 24150755]
- [101] Nosrat IV, Widenfalk J, Olson L, Nosrat CA. Dental pulp cells produce neurotrophic factors, interact with trigeminal neurons *in vitro*, and rescue motoneurons after spinal cord injury. *Dev Biol* 2001; 238(1): 120-32.  
[\[http://dx.doi.org/10.1006/dbio.2001.0400\]](http://dx.doi.org/10.1006/dbio.2001.0400) [PMID: 11783998]
- [102] Arthur A, Shi S, Zannettino ACW, Fujii N, Gronthos S, Koblar SA. Implanted adult human dental pulp stem cells induce endogenous axon guidance. *Stem Cells* 2009; 27(9): 2229-37.  
[\[http://dx.doi.org/10.1002/stem.138\]](http://dx.doi.org/10.1002/stem.138) [PMID: 19544412]
- [103] Kolar MK, Itte VN, Kingham PJ, Novikov LN, Wiberg M, Kelk P. The neurotrophic effects of different human dental mesenchymal stem

- cells. *Sci Rep* 2017; 7(1): 12605.  
[<http://dx.doi.org/10.1038/s41598-017-12969-1>] [PMID: 28974767]
- [104] Song M, Jue SS, Cho YA, Kim EC. Comparison of the effects of human dental pulp stem cells and human bone marrow-derived mesenchymal stem cells on ischemic human astrocytes *in vitro*. *J Neurosci Res* 2015; 93(6): 973-83.  
[<http://dx.doi.org/10.1002/jnr.23569>] [PMID: 25663284]
- [105] Song M, Lee JH, Bae J, Bu Y, Kim EC. Human dental pulp stem cells are more effective than human bone marrow-derived mesenchymal stem cells in cerebral ischemic injury. *Cell Transplant* 2017; 26(6): 1001-16.  
[<http://dx.doi.org/10.3727/096368916X694391>] [PMID: 28105979]
- [106] Kumar A, Kumar V, Rattan V, Jha V, Bhattacharyya S. Secretome cues modulate the neurogenic potential of bone marrow and dental stem cells. *Mol Neurobiol* 2017; 54(6): 4672-82.  
[<http://dx.doi.org/10.1007/s12035-016-0011-3>] [PMID: 27422132]
- [107] Linde A, Goldberg M. Dentinogenesis. *Crit Rev Oral Biol Med* 1993; 4(5): 679-728.  
[<http://dx.doi.org/10.1177/10454411930040050301>] [PMID: 8292714]
- [108] Sonoyama W, Liu Y, Yamaza T, *et al*. Characterization of the apical papilla and its residing stem cells from human immature permanent teeth: A pilot study. *J Endod* 2008; 34(2): 166-71.  
[<http://dx.doi.org/10.1016/j.joen.2007.11.021>] [PMID: 18215674]
- [109] Morszeck C, Völlner F, Saugspier M, *et al*. Comparison of human dental follicle cells (DFCs) and stem cells from human exfoliated deciduous teeth (SHED) after neural differentiation *in vitro*. *Clin Oral Invest* 2010; 14(4): 433-40.  
[<http://dx.doi.org/10.1007/s00784-009-0310-4>] [PMID: 19590907]
- [110] Taga T, Fukuda S. Role of IL-6 in the neural stem cell differentiation. *Clin Rev Allergy Immunol* 2005; 28(3): 249-56.  
[<http://dx.doi.org/10.1385/CRIAI.28:3:249>] [PMID: 16129909]
- [111] Borsini A, Zunszain PA, Thuret S, Pariante CM. The role of inflammatory cytokines as key modulators of neurogenesis. *Trends Neurosci* 2015; 38(3): 145-57.  
[<http://dx.doi.org/10.1016/j.tins.2014.12.006>] [PMID: 25579391]
- [112] Kang CM, Shin MK, Jeon M, Lee YH, Song JS, Lee JH. Distinctive cytokine profiles of stem cells from human exfoliated deciduous teeth and dental pulp stem cells. *J Dent Sci* 2022; 17(1): 276-83.  
[<http://dx.doi.org/10.1016/j.jds.2021.03.019>] [PMID: 35028048]
- [113] Bakshi A, Shimizu S, Keck CA, *et al*. Neural progenitor cells engineered to secrete GDNF show enhanced survival, neuronal differentiation and improve cognitive function following traumatic brain injury. *Eur J Neurosci* 2006; 23(8): 2119-34.  
[<http://dx.doi.org/10.1111/j.1460-9568.2006.04743.x>] [PMID: 16630059]
- [114] Oudega M, Hao P, Shang J, *et al*. Validation study of neurotrophin-3-releasing chitosan facilitation of neural tissue generation in the severely injured adult rat spinal cord. *Exp Neurol* 2019; 312: 51-62.  
[<http://dx.doi.org/10.1016/j.expneurol.2018.11.003>] [PMID: 30471251]
- [115] Descamps B, Saif J, Benest AV, *et al*. BDNF (Brain-Derived Neurotrophic Factor) Promotes embryonic stem cells differentiation to endothelial cells *via* a molecular pathway, including MicroRNA-214, EZH2 (Enhancer of Zeste Homolog 2), and eNOS (Endothelial Nitric Oxide Synthase). *Arterioscler Thromb Vasc Biol* 2018; 38(9): 2117-25.  
[<http://dx.doi.org/10.1161/ATVBAHA.118.311400>] [PMID: 30354255]
- [116] Sone PP, Kaneko T, Zaw SYM, *et al*. Neural regeneration/remodeling in engineered coronal pulp tissue in the rat molar. *J Endod* 2020; 46(7): 943-9.  
[<http://dx.doi.org/10.1016/j.joen.2020.04.002>] [PMID: 32467021]
- [117] Pizzicannella J, Gugliandolo A, Orsini T, *et al*. Engineered extracellular vesicles from human periodontal-ligament stem cells increase VEGF/VEGFR2 expression during bone regeneration. *Front Physiol* 2019; 10: 512.  
[<http://dx.doi.org/10.3389/fphys.2019.00512>] [PMID: 31114512]
- [118] Li W, Mao M, Hu N, *et al*. A graphene oxide-copper nanocomposite for the regeneration of the dentin-pulp complex: An odontogenic and neurovascularization-inducing material. *Chem Eng J* 2021; 417: 129299.  
[<http://dx.doi.org/10.1016/j.cej.2021.129299>]
- [119] Li X, *et al*. A therapeutic strategy for spinal cord defect: Human dental follicle cells combined with aligned PCL/PLGA electrospun material. *BioMed res int* 2015; 2015.
- [120] Zhang J, Lu X, Feng G, *et al*. Chitosan scaffolds induce human dental pulp stem cells to neural differentiation: Potential roles for spinal cord injury therapy. *Cell Tissue Res* 2016; 366(1): 129-42.  
[<http://dx.doi.org/10.1007/s00441-016-2402-1>] [PMID: 27147262]
- [121] Li R, Li Y, Wu Y, *et al*. Heparin-polyoxamer thermosensitive hydrogel loaded with bFGF and NGF enhances peripheral nerve regeneration in diabetic rats. *Biomaterials* 2018; 168: 24-37.  
[<http://dx.doi.org/10.1016/j.biomaterials.2018.03.044>] [PMID: 29609091]
- [122] Luo L, *et al*. Effects of transplanted heparin-polyoxamer hydrogel combining dental pulp stem cells and bFGF on spinal cord injury repair. *Stem cells int* 2018; 2018.  
[<http://dx.doi.org/10.1155/2018/2398521>]
- [123] Askari N, Yaghoobi MM, Shamsara M, Esmaili-Mahani S. Tetracycline-regulated expression of OLIG2 gene in human dental pulp stem cells lead to mouse sciatic nerve regeneration upon transplantation. *Neuroscience* 2015; 305: 197-208.  
[<http://dx.doi.org/10.1016/j.neuroscience.2015.07.088>] [PMID: 26254831]
- [124] Headache Classification Committee of the International Headache Society (IHS) The International Classification of Headache Disorders. 3<sup>rd</sup>. *Cephalalgia* 2018; 38: pp. (1)1-211.
- [125] Amadou-Diaw N, Braud A, Boucher Y. Persistent, neuropathic-like trigeminal pain after dental implant loading. *J Clin Exp Dent* 2022; 14(2): e185-91.  
[PMID: 35173902]
- [126] Podzimek S, Tomka M, Nemeth T, Himmlöva L, Matucha P, Prochazkova J. Influence of metals on cytokines production in connection with successful implantation therapy in dentistry. *Neuroendocrinol Lett* 2010; 31(5): 657-62.  
[PMID: 21173747]
- [127] Chen X, Li HS, Yin Y, Feng Y, Tan XW. Macrophage proinflammatory response to the titanium alloy equipment in dental implantation. *Genet Mol Res* 2015; 14(3): 9155-62.  
[<http://dx.doi.org/10.4238/2015.August.7.25>] [PMID: 26345848]
- [128] Negahdari R, Ghavimi MA, Barzegar A, *et al*. Antibacterial effect of nano-curcumin inside the implant fixture: An *in vitro* study. *Clin Exp Dent Res* 2021; 7(2): 163-9.  
[<http://dx.doi.org/10.1002/cre2.348>] [PMID: 33210463]
- [129] Olmedo D, Fernández MM, Guglielmotti MB, Cabrini RL. Macrophages related to dental implant failure. *Implant Dent* 2003; 12(1): 75-80.  
[<http://dx.doi.org/10.1097/01.ID.0000041425.36813.A9>] [PMID: 12704960]
- [130] Pajarinen J, Kouri VP, Jämsen E, Li TF, Mandelin J, Kontinen YT. The response of macrophages to titanium particles is determined by macrophage polarization. *Acta Biomater* 2013; 9(11): 9229-40.  
[<http://dx.doi.org/10.1016/j.actbio.2013.06.027>] [PMID: 23827094]
- [131] Robson MC, Dubay DA, Wang X, Franz MG. Effect of cytokine growth factors on the prevention of acute wound failure. *Wound Repair Regen* 2004; 12(1): 38-43.  
[<http://dx.doi.org/10.1111/j.1067-1927.2004.012109.x>] [PMID: 14974963]
- [132] Arizon M, Nudel I, Segev H, *et al*. Langerhans cells down-regulate inflammation-driven alveolar bone loss. *Proc Natl Acad Sci USA* 2012; 109(18): 7043-8.  
[<http://dx.doi.org/10.1073/pnas.1116770109>] [PMID: 22509018]
- [133] Gooty J, Kannam D, Guntakala V, Palaparthy R. Distribution of dendritic cells and langerhans cells in peri-implant mucosa. *Contemp Clin Dent* 2018; 9(4): 548-53.  
[[http://dx.doi.org/10.4103/ccd.ccd\\_688\\_18](http://dx.doi.org/10.4103/ccd.ccd_688_18)] [PMID: 31772461]
- [134] Iocca O. Introduction to evidence-based implant dentistry. *Evidence-Based Implant Dentistry*. Springer 2016; pp. 1-18.  
[<http://dx.doi.org/10.1007/978-3-319-26872-9>]
- [135] Al Duboni G, *et al*. The potential role of inflammatory cytokines as useful diagnostic indicators in osseointegrated dental implants. *World Appl Sci J* 2012; 20(5): 649-55.
- [136] Corrêa MG, Pimentel SP, Ribeiro FV, Cirano FR, Casati MZ. Host response and peri-implantitis. *Braz Oral Res* 2019; 33(1): e066.  
[<http://dx.doi.org/10.1590/1807-3107bor-2019.vol33.0066>] [PMID: 31576950]
- [137] Dodo CG, Meirelles L, Aviles-Reyes A, Ruiz KGS, Abranches J, Cury AADB. Pro-inflammatory analysis of macrophages in contact with titanium particles and porphyromonas gingivalis. *Braz Dent J* 2017; 28(4): 428-34.  
[<http://dx.doi.org/10.1590/0103-6440201701382>] [PMID: 29160393]
- [138] Duarte PM, Serrão CR, Miranda TS, *et al*. Could cytokine levels in the peri-implant crevicular fluid be used to distinguish between healthy implants and implants with peri-implantitis? A systematic review. *J*

- Periodontal Res 2016; 51(6): 689-98.  
[<http://dx.doi.org/10.1111/jre.12354>] [PMID: 26774043]
- [139] Bielemann AM, Marcello-Machado RM, Leite FRM, *et al.* Comparison between inflammation-related markers in peri-implant crevicular fluid and clinical parameters during osseointegration in edentulous jaws. *Clin Oral Investig* 2018; 22(1): 531-43.  
[<http://dx.doi.org/10.1007/s00784-017-2169-0>] [PMID: 28710652]
- [140] Jacobi-Gresser E, Huesker K, Schütt S. Genetic and immunological markers predict titanium implant failure: A retrospective study. *Int J Oral Maxillofac Surg* 2013; 42(4): 537-43.  
[<http://dx.doi.org/10.1016/j.ijom.2012.07.018>] [PMID: 22925444]
- [141] Petković AB, Matic SM, Stamatović NV, *et al.* Proinflammatory cytokines (IL-1 $\beta$  and TNF- $\alpha$ ) and chemokines (IL-8 and MIP-1 $\alpha$ ) as markers of peri-implant tissue condition. *Int J Oral Maxillofac Surg* 2010; 39(5): 478-85.  
[<http://dx.doi.org/10.1016/j.ijom.2010.01.014>] [PMID: 20207110]
- [142] Ata-Ali J, Flichy-Fernández AJ, Alegre-Domingo T, Ata-Ali F, Palacio J, Peñarrocha-Diago M. Clinical, microbiological, and immunological aspects of healthy *versus* peri-implantitis tissue in full arch reconstruction patients: A prospective cross-sectional study. *BMC Oral Health* 2015; 15(1): 43.  
[<http://dx.doi.org/10.1186/s12903-015-0031-9>] [PMID: 25888355]
- [143] Kontinen YT, Lappalainen R, Laine P, Kitti U, Santavirta S, Teronen O. Immunohistochemical evaluation of inflammatory mediators in failing implants. *Int J Periodontics Restorative Dent* 2006; 26(2): 135-41.  
[PMID: 16642902]
- [144] Darabi E, Kadkhoda Z, Amirzargar A. Comparison of the levels of tumor necrosis factor- $\alpha$  and interleukin-17 in gingival crevicular fluid of patients with peri-implantitis and a control group with healthy implants. *Iran J Allergy Asthma Immunol* 2013; 12(1): 75-80.  
[PMID: 23454782]
- [145] Duarte PM, de Mendonça AC, Máximo MBB, Santos VR, Bastos MF, Nociti Júnior FH. Differential cytokine expressions affect the severity of peri-implant disease. *Clin Oral Implants Res* 2009; 20(5): 514-20.  
[<http://dx.doi.org/10.1111/j.1600-0501.2008.01680.x>] [PMID: 19302394]
- [146] Baseri M, Radmand F, Hamed R, Yousefi M, Kafil HS. Immunological aspects of dental implant rejection. *BioMed Res Int* 2020; 2020: 1-12.  
[<http://dx.doi.org/10.1155/2020/7279509>] [PMID: 33376734]
- [147] França FL, Honorio-França AC, Honorio MS, *et al.* Dental implant surfaces treated with phosphoric acid can modulate cytokine production by blood MN cells. *Braz Oral Res* 2019; 33: e040.  
[<http://dx.doi.org/10.1590/1807-3107bor-2019.vol33.0040>] [PMID: 31508724]
- [148] Medzhitov R, Shevach EM, Trinchieri G, *et al.* Highlights of 10 years of immunology in nature reviews immunology. *Nat Rev Immunol* 2011; 11(10): 693-702.  
[<http://dx.doi.org/10.1038/nri3063>] [PMID: 21941295]
- [149] Atoum M. ACC interleukin-10 gene promoter haplotype as a breast cancer risk factor predictor among Jordanian females. *OncoTargets Ther* 2016; 9: 3353-7.  
[<http://dx.doi.org/10.2147/OTT.S101628>] [PMID: 27330315]
- [150] Yang X, Wei H, Qin L, *et al.* Reciprocal interaction between fish TGF- $\beta$ 1 and IL-1 $\beta$  is responsible for restraining IL-1 $\beta$  signaling activity in grass carp head kidney leukocytes. *Dev Comp Immunol* 2014; 47(2): 197-204.  
[<http://dx.doi.org/10.1016/j.dci.2014.07.023>] [PMID: 25092146]

