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Biofunctional Applications of Chitosan in Dentistry - An Overview

Venkateshwaran Krishnaswami[✉,1,*] **Selvakumar Muruganantham**[✉,2] **Jacob Raja**[✉,3] **Kumar Janakiraman**[✉,4] **Vaidevi Sethuraman**[✉,4] **Lakshmi Kanakaraj**[✉,5]

1 Department of Pharmaceutics, S. A. Raja Pharmacy College, Vadakangulam, Tirunelveli 627116, Tamil Nadu, India
2 Department of Pharmaceutics, Vivekanandha Pharmacy College for Women, Sankari, Salem-637303, Tamil Nadu, India
3 Department of Periodontology, Rajas Dental College and Hospital, Kavalkinaru, Tirunelveli 627105, Tamil Nadu, India
4 Department of Biotechnology, Rathinam Technical Campus, Eachanari, Coimbatore 641021, India
5 Chettinad School of Pharmaceutical Sciences, Chettinad Hospital and Research Institute, Chettinad Academy of Research and Education, Kelambakkam 603103, Tamil Nadu, India

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Abstract
Chitosan, with its unique properties, has garnered the interest of materials scientists worldwide, leading to exploration in bio-dental applications. In dentistry, chitosan has shown a wide range of applications. The creation of effective biomaterials for the prevention and treatment of dental disorders is a challenge for researchers in the present scenario. The natural semi-synthetic polysaccharide-based biomaterial Chitosan is derived from chitin, which is sourced from marine organisms. Due to its peculiar characteristics, chitosan offers benefits in biomedicine and can be utilized in the development of capsules (micro and nanoparticles), powders, scaffolds, films, beads, hydrogels, and bandages. Chitosan gives numerous benefits, inculcating its antimicrobial properties, mucoadhesiveness, and biomimetic mineralization, which contribute to its teeth-hardening effects due to its remineralizing capabilities. Such benefits have directed research and interest toward its dental applications Translation of research to clinical applications is better suited for chitosan due to its multifactorial activities. Therefore, this article provides an overview of chitosan, which mainly covers the basic information of chitosan, with a focus on different techniques of preparation of chitosan-based formulations. It also explores the usage of chitosan in the treatment of various dental disorders.

Keywords:
chitosan; dental applications; tissue engineering; osseointegration; nanoformulations

1. Introduction

The linear cationic polysaccharide-based polymer chitosan is obtained by deacetylation of chitin. It is also observed in fungal cell walls, cuticles of insects, etc [1–3]. Chitosan has significant potential in Pharmaceutical and related industries. Further, it holds anti-bacterial, anti-fungal, mucoadhesive, and gelling properties. Chitosan is also nontoxic, biodegradable and biocompatible. It may interact with various negatively charged moieties such as enzymes, proteins, etc which makes it suitable for various drug delivery applications. The copolymer units of N-acetyl-glucosamine and glucosamine are present in Chitosan. The functional groups associated with chitosan support electrochemical interaction at molecular and cellular levels [4–6]. The poor water solubility of chitosan leads to the development of various tailor-made forms of chitosan derivatives (carboxy methyl/thiolated). Various drug loaded nanoformulations were reported using chitosan with the aid of sustaining/targeting the release of encapsulated drug. Chitosan based copolymers are reported for stimuli-responsive drug delivery systems [7]. The release of encapsulated drug from the chitosan is governed by various mechanisms such as swelling, ad-

* **Corresponding Author:**
Venkateshwaran Krishnaswami, Department of Pharmaceutics, S. A. Raja Pharmacy College, Vadakangulam, Tirunelveli, Tamil Nadu, India, venkpharm1@gmail.com; Tel.: +91-9943564836

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sorption, diffusion, erosion or degradation, and a combination of erosion and degradation. The functional properties of chitosan are due to the prevalence of the amine group in its structure [8–10]. Chitosan toxicity is based on its degree of deacetylation in the aspects of molecular weight. Chitosan is composed of different elements which include Carbon (44.11%), Hydrogen (6.84%), and Nitrogen (7.97%). Chitosan exhibits low solubility in neutral and alkaline pH environments.It plays an imperative role in different multifactorial drug delivery applications inculcating parenteral drug delivery, ocular drug delivery, mucosal drug delivery, per-oral drug delivery, gene delivery, vaccine delivery, cancer therapy, pulmonary drug delivery, and intranasal drug delivery, as shown in Figure 1 [11,12].

2. Chitosan Based Nanoformulation for Dentistry

The research into the biology of mineralized hard tissues, especially dental tissues like dentin and enamel, has been greatly impacted by nanotechnology. Dietary acids are the primary cause of enamel demineralization, a common clinical issue. Antimicrobial and dental restorative materials are influenced by nanoparticle technology, and environmentally friendly approaches to synthesizing nanomaterials are required to reduce their impact. In order to demonstrate the superior performance of dental nanoparticles, future advances should concentrate on actual difficulties, genuine advancements, and real clinical scenarios [13]. Particular physiological and chemical characteristics of these nanomaterials include their nano size, increased surface-to-volume ratio for improved bonding qualities, greater reactivity, and chemical wettability. These characteristics have been applied to the identification and treatment of oral malignancies as well as dentinal hypersensitivity and oral biofilm elimination. Nanoparticles are being investigated as drug-delivery vehicles for treating dental caries, dentinal hypersensitivity, tooth remineralization, oral biofilm management, local anaesthesia, root canal disinfection, and periodontal infections One of the important applications in dentistry is the selective delivery of these agents to specific areas or cells, with periodontal disease being one such instance. Due to their regulated rates of release and durability in periodontal pockets, these nanoparticles can reach deep pockets that may be inaccessible to other types of drugs. The most important type of nanoparticles is polymeric ones since they are biodegradable and easily customizable. Newer nano-biomaterials, such as chitosan, which is derived from marine materials, have been developed as a result of advancements in biomedicine and are finding increased application in the dentistry and medical domains. Full-bodied drug delivery carrier systems with high mechanical strength, good contact time, and sustained drug release when in close contact with the oral mucosa are made possible by chitosan-based composites [14–16]. Chitosan possesses a range of biological activities, such as hemostatic, antifungal, mucoadhesive, and antibacterial qualities. Because of its cationic properties, it interacts electrostatically with negatively charged materials like sialic acid and epithelium surfaces [17]. Tissue engineering, gene therapy, drug delivery systems, and other applications may be impacted by these interactions. Furthermore, positively charged chitosan forms a bond with cell DNA, which prevents the synthesis of microbial RNA and makes it easier for active agents to penetrate [18].

As chitosan has high antibacterial qualities, and gelling properties, and doesn't require preservatives, researchers have looked into using it in dentifrices. The development of polyherbal toothpaste based on chitosan has inhibited the growth of *Porphyromonas gingivalis* and *Streptococcus mutans* bacteria, hence enhancing the antimicrobial and anti-inflammatory properties of toothpaste's active components. Although the results have been varied, attempts have also been made to enhance the antibacterial capabilities of chitosan-based adhesives. The potential of chitosan as dentin collagen to enhance the mechanical and biological characteristics of collagen constructions has been investigated. The toughness, tensile strength, and chemical stability of dentin collagen can all be markedly increased by chemically/photodynamically crosslinking the collagen matrix with carboxy methyl cellulose, according to recent studies [19].

In 2023, Hoveizi et al. studied the effects of titanium oxide nanoparticles (TiO NPs) and human endometrial stem cells (EnSCs) on dental pulp regeneration and repair in male Wistar rats. In this study, EnSCs were positioned on a three-dimensional scaffold made of chitosan and TiO NPs, exposed to pulps, and then covered with the scaffolds. The CS/EnSCs/TiO group had more dentin overall and in better quality at 8 weeks than the other groups. Dentin formation was accelerated and improved in quality when EnSCs with TiO NPs and 3D chitosan scaffolds were combined [20]. A method of remineralizing human carious-like enamel by the use of chimeric peptide-mediated carboxymethyl chitosan/amorphous calcium phosphate nano complexes was developed by Xiao et al. in 2017. The developed nanocomplexes were synthesized, amorphous calcium phosphate nanoparticles were broken down, and their morphology was examined. The purpose of the peptides was to bind the amorphous calcium phosphate nanoparticles to the demineralized enamel surface and direct their arrangement. Strong mechanical

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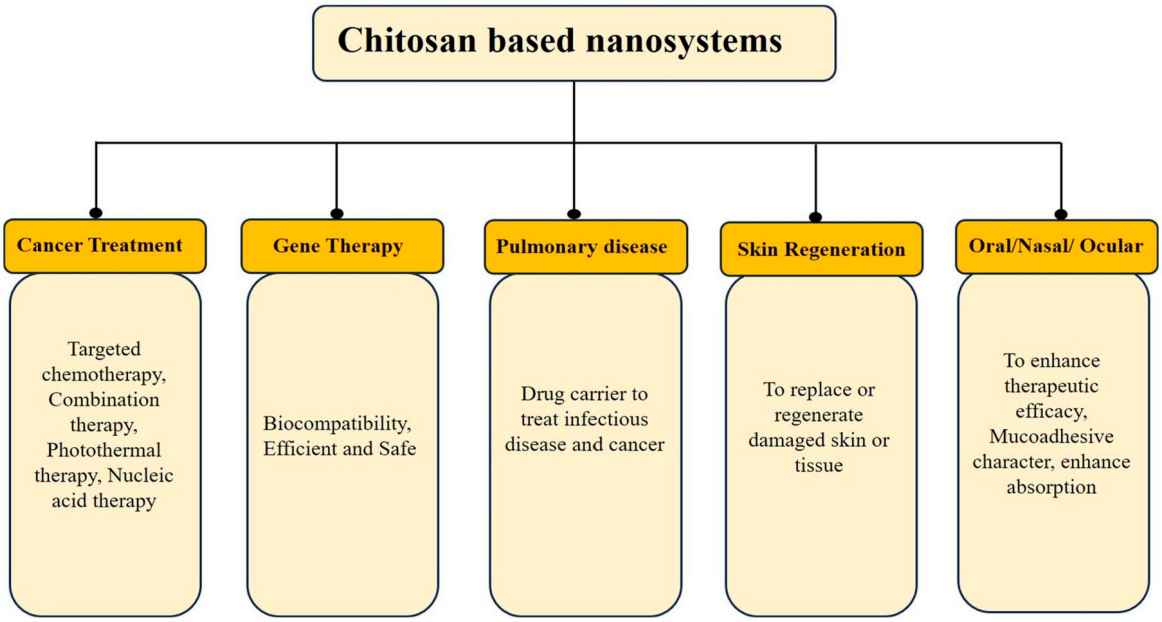


Figure 1: Chitosan based drug delivery systems for multifactorial applications.

characteristics and stability characterized the produced carboxymethyl chitosan/amorphous calcium phosphate nano complexes were exhibited [21]. A new bio nanocomposite comprising chitosan, carboxymethyl starch, and montmorillonite was developed in 2017 by Jahanizadeh et al. for the delivery of curcumin. Various ratios of carboxymethyl starch, montmorillonite, and Chitosan were employed to increase the effectiveness of trapping. With a 91% entrapment efficiency and an average particle size of 35.9 nm, the ideal formulation was used. Along with showing antibacterial action against *Streptococcus mutans*, the nanocomposite also successfully stopped biofilm formation in dental models [22]. In 2021, Karthik et al. produced benzodioxane-coupled piperazine-decorated chitosan silver nanoparticles (Bcp*C@AgNPs) and tested their antibacterial and anti-biofilm capabilities against MRSA (methicillin-resistant Staphylococcus aureus). The nanoparticles had a minimum inhibitory concentration of 10 ± 0.03 mm at 200 μg/mL and unique anti-biofilm capabilities. The toxicity studies revealed decreased harmful effects in the myoblast cell line (L6) study. This study focuses on the biocidal efficiency of Bcp*C@AgNPs and their prospective clinical trial targets [23].

For its structural qualities and biocompatibility, A dental cement nanocomposite including chitosan and dicalcium phosphate was reported. With a 30% (w/w) dicalcium phosphate content, the nanocement's compressive strength was 4.37 ± 0.67 MPa, which is similar to cancellous bone. In a salt-buffered sand (SBF) environment for 14 days, the nanocement displayed a rough and porous shape. All cement composites exhibited bioactivity; however, the needle-shaped crystal calcium sulfate hemihydrate (CSH) phase's growth was reduced by the addition of chitosan. The nucleation and crystallization of hydroxyapatite were accelerated by the addition of calcium and the reduction of apatite phase solubility, which raised the pH of the SBF solution. The study concludes that Portland clinker can be effectively modified to include chitosan and dicalcium phosphate for use in root-end dentistry applications [24]. In 2020, Ma et al. developed a drug delivery system based on chitosan nanoparticles to improve Cur's therapeutic impact on polymicrobial biofilms. Curcumin was loaded onto a positively charged chitosan nanoparticle known as CSNP-Cur, and its antibiofilm properties against Staphylococcus aureus and Candida albicans were evaluated. Excellent antibiofilm action against preformed biofilms, fungus, and planktonic bacteria was demonstrated by the CSNP-Cur. On silicone surfaces, it decreased the thickness of the biofilm and eliminated microbial cells embedded in it, indicating its potential as a novel therapeutic approach for infections linked to polymicrobial biofilms [25]. In another study, Ashraf et al. developed *Mentha piperita* essential oils loaded with chitosan nanogel to be used as an antibiofilm agent against *Streptococcus mutans* and shield its dental plaque. The addition of *Mentha piperita* essential oils increased the z-average while decreasing the nanoparticles' monodispersity. Over 360 h at room temperature in a hydroalcoholic solvent, the maximal release of *Mentha piperita* essential oils was approximately 50%. As

significant enzymes involved in extracellular polymers, the study showed that chitosan nanogels provided an effective nanoformulation with the most inhibitory efficacy against specific glycosyltransferase genes (gtfB, C as well as D). According to the study's findings, the developed nanoparticles may be used in toothpaste or mouthwash formulations as antibiofilm agents [26].

In 2019, Hu et al. developed pH-responsive quaternary ammonium chitosan-liposome nanoparticles using chitosan (N, N, N-trimethyl chitosan), and doxycycline, and these nanoparticles showed good antibacterial activity and cytocompatibility with human periodontal ligament cells (hPDLCs). They demonstrated the ability to cure periodontal disease by inhibiting biofilm growth and preventing alveolar bone absorption in animal tests [27]. To overcome the drawbacks in the treatment of periodontitis, another study developed a tinidazole functionalized biodegradable chitosan/poly (ε-caprolactone) mucoadhesive hybrid nanofiber membrane. The nanofiber membrane, measuring 143.55 ± 8.5 nm in diameter and 83.25 ± 1.8% in entrapment efficiency, exhibited effective drug transport and antibacterial activity. In early clinical trials, the nanofiber membrane significantly reduced clinical periodontitis indicators and was shown to be non-cytotoxic on mouse fibroblasts [28]. Recently, Sun et al. used a chitosan-based biomaterial reinforced with calcium zirconium nanoparticles (CZNP) to study the adaptive strategies of dental pulp stem cells (DPSCs) in response to oral disease situations. The study analyzed the performance of the chitosan-CZNP biomaterial and measured the vitality and proliferation of DPSCs under conditions of oral illness. The findings demonstrated that while maintaining or improving fracture toughness, tensile strength, and apatite formation, increasing the weight percentage of CZNP resulted in increased porosity and drug release. The biomaterial with the highest levels of apatite formation, fracture toughness, and drug release was the one containing 7.5wt% CZNP. This study demonstrates the potential of chitosan-CZNP biomaterials in oral tissue engineering and regenerative medicine and advances our knowledge of the coping mechanisms of DPSCs [29].

In 2024, Hu et al. developed new chitosan nanoparticles loaded with the peptide QP5, which is derived from amelogenin (TMC-QP5/NPs). Their potential for remineralization and their capacity to inhibit endogenous matrix metalloproteinases were examined. Several techniques were used to investigate the loading and encapsulation effectiveness of TMC-QP5/NPs. Dentin bonding was dramatically improved when TMC-QP5/NPs-induced remineralized dentin displayed increased μSBS and decreased interfacial microleakage. Dentin remineralization was aided by the peptide-loaded chitosan nanoparticles, which also effectively inactivated endogenous MMPs, which points to a potentially effective method for improving dentin adhesive repairs. The results of the study imply that TMC-QP5/NPs may be a viable tactic for improving dentin adhesive restorations. Although cationic nanoparticles play a critical role in biofilm removal, serious concerns have been raised regarding their possible detrimental effects on normal cells [30]. In another study, pH-triggered charge-switching nanoparticles have been developed to remove biofilms. Stable at pH 7.4, these nanoparticles can target specific bacteria in affected biofilm areas without causing harm to healthy cells. Additionally, because they encapsulate bioactive compounds with high loading efficiency and release them as needed, they are efficient delivery systems for these substances. These nanoparticles' antibacterial and anti-biofilm properties were greatly enhanced upon encapsulation. It is further anticipated that this new approach to functional foods will be more efficient and safer [31].

3. Chitosan for Dental Application as Implants

Dental implants, which are generally placed via surgery or specialized equipment, are long-term substitutes for natural teeth in oral rehabilitation and maxillary reconstruction. Modern dental implantology has advanced, making implants more dependable and palatable to patients [32]. Osseointegrated dental implants are becoming more popular, and because of their stability, excellent resistance to corrosion, and biocompatibility, metallic materials are now the most commonly used. Even after therapy, insufficient osseointegration between the implant surface and the transplanted site remains a significant concern. It is difficult to create the perfect dental implant that inhibits the growth of bacterial biofilms while promoting osseointegration. In implantation, controlling the structural qualities of a surface is crucial, and many changes have been made for better bone healing, appropriate blood clot maintenance, and mechanical fixing [33]. Additionally, biochemical substances such as proteins, medications, and biomolecules are being utilized to enhance local distribution on a metallic implant's surface.

The interaction between the tissue and implant material is typically impacted by the absence of appropriate cell-binding sites and antibacterial characteristics in medical implants. Peri-implant disorders (PIDs), including peri-implantitis (PI) and peri-implant mucositis (PIM), are the primary issue with implants in dentistry. PIM is a reversible inflammatory disease that targets the soft tissues surrounding implants, resulting in severe clinical and socio-economic consequences as well as implant fail-

ure. This can worsen patients' quality of life considerably and accelerate the deterioration of their dental health [34]. Biomaterials must also control biofilm-related germs, as implant infections can be hazardous and lead to treatment failure. They should perform their intended function in medical therapy, not negatively impact the recipient organism, and elicit the best possible positive reaction [35]. Orthodontic mini-implants are well-liked because of their durability, affordability, and low level of discomfort. On the other hand, they may result in infections such as peri-implantitis and peri-mucositis. According to a study, chitosan gel can minimize irritation by lowering bacterial contamination on mini-implants. The use of chitosan gel reduced the number of bacteria by 26.59%, with the chlorhexidine gel group showing the largest reduction. In the case of *Treponema denticola* and *Tannerella forsythia*, the difference was noteworthy. Red-complex bacteria and total oral bacteria were both decreased by the chitosan-containing gel [36]. Alhazmi et al. assessed the structural and morphological uses of coated micro-implants, hydroxyapatite, and chitosan nanoparticles in 2022 as a means of preventing oral pathogenic germs. The *Salvadora persica* plant was used to synthesize the nanoparticles, which were then tested for antibacterial effectiveness against four common oral bacterial strains. The antibacterial activity of the nanoparticles coated on titanium micro-implants was assessed using the disc diffusion method. The results indicated a minimum bacterial concentration and minimum inhibitory concentrations at 8 μg mL^{-1} and 16 μg mL^{-1} for *Streptococcus salivarius*, *Enterococcus faecalis*, *Streptococcus mutans*, and *Streptococcus sanguinis*, respectively [37]. To find out how coating chitosan on orthodontic mini-implants may prevent *Porphyromonas gingivalis,* an investigation was carried out. Twenty-five mini-implants were used, with five coated in chitosan, five in chitosan-azithromycin, five in azithromycin, and five left uncoated. Following a 24-h incubation period, the azithromycin-treated group showed the greatest suppression of Porphyromonas gingivalis biofilm mass, followed by chitosan along with azithromycin and chitosan alone. Bacterial viability count and biofilm mass were shown to be strongly positively correlated in the study, indicating that Porphyromonas gingivalis biofilm development was successfully reduced by chitosan coating [38].

Dental implants' surface treatment is essential to managing their structural properties. Many processes, including plasma spraying, blast media, acid etching, and oxidation, can sustain blood clots, promote bone healing, and enhance mechanical anchoring. These changes have the potential to accelerate osseointegration and inhibit the growth of biofilm [39–45]. Osteointegration can be enhanced by chemically altering implant surfaces, and chitosan is a highly effective agent for stimulating osteoblasts and forming new bone [46]. Its benign nature has strengthened its usage in implants, where it is frequently employed for tissue regeneration, encouraging mineralization and osteogenesis. Due to its osteoconductive nature, high molecular weight biopolymer qualities, bioactivity, and ease of processing, chitosan has garnered interest in the field of dental implant therapy [47]. Chitosan coatings can modify the surface's mechanical, morphological, and biological properties to produce a bioactive surface. These coatings have an impact on bone health and increase clinical longevity in people with poor health. On dental implants, chitosan coating improves biocompatibility and antibacterial activity by promoting the production of apatite, and cell proliferation, reducing the hydrophilicity and surface roughness, and perhaps incorporating antibiotics for better healing [48]. However, because chitosan lacks surface reactivity, it does not stick to the implant surface. Understanding the mechanisms influencing bioactivity, surface characteristics, and bonding strength to titanium implants is necessary for optimizing bioactive chitosan coatings [49].

In 2021, Del Olmo et al. created a titanium implant surface covered with a catechol anchor group that was either conjugated or nonconjugated with chitosan. On coated surfaces, the antibacterial properties of the substance were assessed against *Staphylococcus aureus* and *Escherichia coli*. After 300 min of contact between the bacterial suspensions and the surfaces, the vitality of the suspensions was measured using epifluorescence. According to a study, catechol-conjugated chitosan was observed to be less bactericidal than unconjugated chitosan. After 300 min, unconjugated surfaces had a higher density of injured bacteria than surfaces coated with catechol-conjugated chitosan, with 70% of S. aureus and 84% of *Escherichia coli*. Two significant issues associated with the use of zirconia-based materials in dental implants are poor osteogenesis and peri-implantitis resulting from bacterial colonization. [50]. To enhance osteogenic differentiation and antimicrobial properties, a functional surface modification technique has been devised. The PEI/HA/CGA-CS (poly (ethylene imine)/hyaluronic acid/chitosan-chlorogenic acid) coating increases the zirconia surface's wettability and keeps the CGA release stable. It stimulates early calcification, growth, division, and adherence of cells. Without affecting osteoblast biological activity, the coating efficiently suppresses *Aggregatibacter actinomycetemcomitans* and *Porphyromonas gingivalis* bacterial growth and activity. Furthermore, it stimulates the Wnt/β-catenin signaling pathway and increases the expression of Nephronectin protein, which both improve osteogenesis in MC3T3-E1 cells [51]. The impact of two degrees of deacetylation (DDA) on chitosan

material as coatings for laser surface microtopographic implants was investigated by Alnufaiy et al. in 2020. In this investigation, three groups of Laser-Lok implant discs were divided, and two of the groups received coatings of either 80 or 95 DDA chitosan. The cell morphology, vitality, and osteogenic potential of the chitosan material were assessed using hMSC-TERT 20 cells. The LL 95 group expressed more osteogenic markers and displayed enhanced cell spreading and attachment, according to the results. According to the study, a high DDA of chitosan encourages osteoblast production and biomineralization, which may improve osseointegration and subsequent dental implant healing processes [52].

Titanium (Ti) is widely utilized because of its biocompatibility, even though implant dentistry has great success rates with this material. It is not, however, immune to bacterial infections, which can result in malfunctions such as inadequate oral hygiene, infection, immobility, mechanical problems, poor osseointegration, and bioinertness. Efforts have been undertaken to cover the implant surface to improve osseointegration and overcome these constraints [53]. In 2007, Bumgardner et al. examined titanium implants coated with chitosan using 16 rabbits, and chitosan-coated pins were inserted in their tibia, and their rehabilitation and growth of bones were assessed histologically. The findings revealed a limited inflammatory response and the usual healing pattern of lamellar bone production after fibrous, woven bone creation. Unfortunately, because of inadequate cortical bone thickness, 31% of the implants migrated into the tibial marrow cavity after implantation. The study reveals the theory that chitosan coatings can cause orthopedic, dental, and craniofacial implants to osseointegrate—a tight bone apposition [54]. According to another study, Ti surfaces were coated with a micro-nanostructured hydroxyapatite layer that was loaded with chitosan. The composite coating strengthened the biological and antibacterial qualities, hastening the creation of the apatite layer and improving cell attachment, propagation, and multiplication. Reduced biological qualities and improved antibacterial qualities were the results of increased chitosan coverage [55]. By using micro-computed tomography (micro-CT), Lopez-Valverde et al. conducted a pilot investigation in 2022 to assess the osseointegration and bone development surrounding chitosan-coated implants in dogs. Five dogs were used in the study, and four implants total—two groups—were placed into their jaws: the ChtG (chitosan-coated implant group) and the control group. Euthanasia was carried out twelve weeks post-surgery, and sectioned bone blocks were taken and subjected to micro-CT scanning. Two bone characteristics were examined: peri-implant bone area and bone-to-implant contact In this study, the main bone characteristics of the peri-implant bone area and the bone in contact with the implant surface were examined. Regarding the control group, statistically significant results were found for the ChtG group. The outcomes showed the value of chitosan coatings; however, more comprehensive experimental models and higher sample sizes are required to validate the findings. The effectiveness of chitosan coatings on titanium surfaces in promoting dental implant osseointegration was also validated by the investigation [56].

A novel technique has been put up for creating chitosan conversion coating on magnesium substrates for orthopedic implants. The coating is the outcome of a chemical reaction that promotes the surface integration of magnesium to form a corrosion-resistant layer. Coordinate-covalent bonding is used to adhere the chitosan deposit to the substrate. A CHI/BG composite film was formed when charged chitosan molecules and BG particles moved toward the magnesium substrate due to the high activity of magnesium. Whereas, the CHI-coated sample displayed both magnesium and magnesium hydroxide, the in vitro bioactivity of the CHI/BG-coated magnesium sample mostly demonstrated magnesium hydrogen phosphate (III) hexahydrate [57]. Ag-chitosan shows great potential as a coating material for dental implants, as it enhances the passivation and corrosion resistance of titanium implants. Using a sol-gel dip coating method, Etrat Anees et al. produced chitosan-hydroxyapatite (Ch-HA) composite coatings on 316L stainless steel in 2024. Electrochemical tests, FTIR, SEM, and X-ray diffraction were used to characterize the coatings. The surface morphology revealed holes and cracks-free dense microstructures. According to electrochemical investigations, 1.5gCh-HA was the ideal coating concentration for improving the corrosion resistance of 316L SS when compared to 316L SS which was left bare. Additionally, the coatings demonstrated suitable adherence to the dental implant-grade 316L SS substrate [58]. In another study, Ag-chitosan nanoparticles were evaluated as a potential coating material for titanium dental implants. The bioactive chitosan that was isolated from *Aspergillus flavus* Af09 slowed down the development of biofilms, prevented QS synthesis, and stopped the growth of S. mutans and P. gingivalis. The absence of cell cytotoxicity in the nanoparticles indicates their biocompatibility [59]. In 2022, Pakawat et al. produced gold nanoparticles covered with chitosan-grafted thymol (CST) as an antibacterial material. CST was used as a capping agent for the synthesis of AuNPs (gold nanoparticles) and was modified for the Mannich process. For AuNP production, a concentration of 0.020%w/v was suitable. Strong surface plasmon resonance was observed in the AuNP solution at 502 nm, suggesting electrostatic repulsion and

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the capping agent function of CST. Cariogenic bacteria in the oral cavity have been successfully controlled by using CST coated on AuNP surface. The antibacterial activity of the nanoparticles against Streptococcus mutans ATCC 25175 and Streptococcus sobrinus ATCC 33402 was greatly affected by the capping agent's tuning during the manufacturing process. Additionally, the study revealed that applying chitosan coatings to titanium surfaces can enhance dental implant osseointegration. Following euthanasia, computed microtomography was used to evaluate the extracted bone blocks, indicating that chitosan coatings on titanium surfaces are suitable for antibacterial applications [60].

A graphene-chitosan hybrid dental implant (GC hybrid implant) was created in 2020 by Sunho Park et al. and showed improved wettability and roughness. The ideal state (1% GC hybrid implant) decreased bacterial activity and biofilm formation while increasing osteoblast development. The results of this study highlight the potential of the GC hybrid implant as a new type of dental implant. In comparison to 3% and 5% GC hybrid implants, the antibacterial qualities and anti-biofilm formation effects of 1% GC hybrid implants were enhanced. The antibacterial properties of graphene are concentration- and time-dependent. According to Alayande et al, high quantities of roughened graphene may provide a deep valley structure that improves bacterial adhesion efficiency. Bacterial attachment to surfaces may be sustained due to the enhanced hydrophobic force and π–π interactions between graphene and bacterial cell membranes, perhaps overcoming electrostatic repulsion [61]. To inhibit periopathogenic microorganisms on titanium dental implants, a recent study focused on the use of sonodynamic antimicrobial chemotherapy (SACT) and antimicrobial photodynamic treatment (aPDT). As a photo-sonosensitizer, chitosan nanoparticles-indocyanine green (CNPs-ICG) were employed. The investigation observed a statistically significant decrease in log CFU/mL of periopathogens among the treatment-treated groups. Comparing PSACT/CNPs-ICG to other groups, the latter had a noticeably greater capacity to remove the biofilm. Microscopic pictures showed that dead and malformed cells made up the majority of the biofilms treated with PSACT. The findings demonstrate PSACT/CNPs-ICG's capability for cleaning dental implant surfaces from the polymicrobial synergism of biofilm-forming periopathogens [62].

4. Chitosan for Dental Application as a Scaffold

Dental tissue engineering uses scaffold-based techniques to create an environment for cell attachment and proliferation, whereas scaffold-free techniques, including cell treatments and micro-tissue, are used to regenerate tissue. Particularly in dental/bone replacement applications, scaffolds are useful in treating dental infections because they remove diseased tissue and allow filling material to get inserted [63]. These scaffolds should be non-toxic and secure, allow cell attachment without interfering with normal function as well as proliferation, and show an adequate biodegradation period throughout the regeneration of new tissue. Mechanically stable scaffolds facilitate manipulation, adjustment, and incorporation into tissue defects [64]. Chitosan based systems have been used for various dental disorders as shown in Figure 2.

Research on chitosan scaffolds for tissue and bone engineering gets well explored. By adding polymers, biomaterials, or bioactive compounds, one might enhance the scaffolds' characteristics. When it comes to chitosan scaffold synthesis, fungal sources are chosen over marine sources because of their superior physico-chemical characteristics [65]. With its diverse features, chitosan as a scaffold biomaterial offers many advantages. It is appealing for use in tissue engineering applications due to its adaptability in surface chemistry and biological characteristics. Chemical cross-linking, composite synthesis with reinforcing agents, synthetic or natural polymers, and adjusting ionic strength and solubility are some methods for enhancing chitosan scaffolds. These uses, however, shouldn't have any harmful impact on cells or change the biological characteristics of the scaffold [66]. Chitosan-based scaffolds have been mentioned in several publications as potentially useful in dental care.

Generally, the binding and growth of osteoblast cells are often facilitated by the interaction of fibrin glue and platelet-rich plasma with chitosan. The effects of activated and fibrin glue on the osteogenic differentiation and proliferation of human dental pulp stem cells (h-DPSCs) were investigated by Sadeghinia et al. in 2019. Porous composite scaffolds based on chitosan-gelatin/nanohydroxyapatite treated with fibrin glue and platelet-rich plasma were studied for their in vitro behavior. To seed h-DPSCs, four groups of composite scaffolds were created, and the scaffolds' surface had an ordered fibrin network, as seen in the 14-day scanning electron microscopy image. When compared to chitosan-gelatin/nanohydroxyapatite, all groups treated with fibrin glue and platelet-rich plasma demonstrated better h-DPSC seeding adhesion. The fibrin network on the composite scaffolds treated with fibrin glue and platelet-rich plasma increased the mineralization and osteoblastic differentiation of the collected cells [67]. In 2024, Anaya-Sampayo et al. created scaffolds from hydroxyapatite, chitosan, gelatin, and lyophilized platelet-rich fibrin with or without alginate. They assessed the characteristics, release of growth factors, and viability of os-

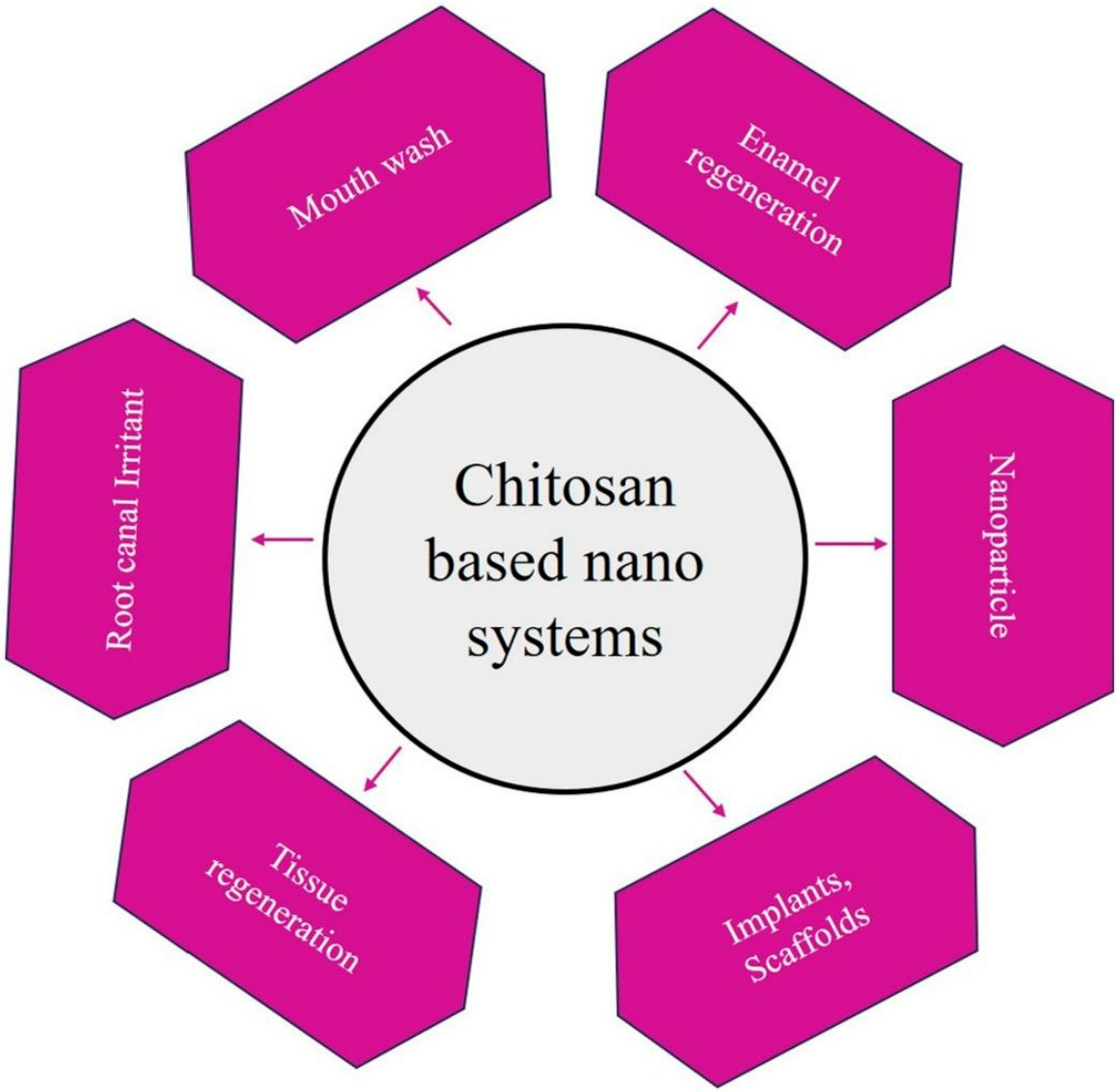


Figure 2: Chitosan for various dental disorders.

teoblasts and DPSC. The scaffolds underwent morphological characterization, swelling profiles, and degradation analysis after being produced by freeze-drying and crosslinking with glutaraldehyde. The outcomes demonstrated that scaffolds supplemented with platelet-rich fibrin improved the viability of DPSC and osteoblast-DPSC. All scaffolds exhibited similar profiles of swelling and degradation, with pore diameters varying between 100 and 250 μm. The chitosan-based scaffold exhibited ideal physical-biological properties to promote the survivability of DPSC and osteoblast-DPSC cells, indicating enhanced scaffold biocompatibility for the regeneration of bone tissue [68].

In 2019, a chitosan and dicarboxylic acid (CS/DA) scaffold was tested in mice by Sukpaita et al. to promote bone regeneration in calvarial deformities. Eighteen mice were used in the investigation, and they were split into three groups: those with empty defects, those with defects and CS/DA scaffold, and those with defects and hPDLCs. After 6 and 12 weeks, in vivo bone regeneration was seen, and micro-CT and histological analysis demonstrated that CS/DA scaffolds greatly enhanced in vitro osteoblast-related gene expression by hPDLCs. According to the research, CS/DA scaffolds have high osteoinductive and osteoconductive qualities and can be employed as a bone-regenerating material [69]. Covarrubias et al. (2018) used a chitosan-gelatin polymer blend and thick bioactive glass (BG) nanoparticles or mesoporous bioactive glass nanospheres to build bone healing nanocomposite scaffolds. In the in-vitro study, the crys-

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tallization of bone-like apatite could be accelerated by the scaffolds, which also demonstrated good cytocompatibility. Higher alkaline phosphatase activity showed that bioactive glass nanoparticles were more effective in encouraging osteogenic differentiation of dental pulp stem cells. Bioactive glass nanoparticles (5%)/chitosan-gelatin bionanocomposite generated the newest bone (∼80%) after 8 weeks of implantation, according to in vivo investigations; this makes them appealing for bone restoration applications [70].

The goal of regenerative dentistry is to provide better biomaterials that support the pulp-dentin complex's regeneration, which is powered by resident cells. A chitosan scaffold (CHSC) that produced bioactive quantities of simvastatin for cell-free tissue engineering was evaluated in 2018 by Soares et al. They performed a dose-response experiment to determine the bioactive dose of simvastatin that could induce an odontoblastic phenotype in dental pulp cells (DPCs). To replicate the cell-free approach in vitro, the biomaterials were integrated into a three-dimensional culture platform, akin to an artificial pulp chamber. The findings demonstrated that simvastatin, at 0.1 μmol/L, significantly induced an odontoblastic phenotype on the DPC/CHSC construct while having no detrimental effects on adhesion or cell viability. DPCs' capacity for chemotaxis and regeneration was enhanced by the CHSC-simvastatin 1.0 scaffold [71]. In 2017, Varoni et al. developed a trilayer porous scaffold based on chitosan for periodontal regeneration Using genipin, they produced two compartments for the regeneration of bone and gingiva and a third compartment for the regeneration of the periodontal ligament (PDL). Compared to the low molecular weight chitosan compartment, the medium molecular weight chitosan compartment deteriorated more gradually and had greater resilience to compression. In cytocompatibility assays, more than 90% of human primary periodontal cell populations survived. In vivo experiments demonstrated scaffold vascularization, tissue ingrowth, and good biocompatibility in wild-type mice. The study discovered a thick mineralized matrix within the medium molecular weight chitosan area and also revealed scaffold compartments including human gingival fibroblasts, osteoblasts, and PDL fibroblasts. According to these findings, the resorbable trilayer scaffold is a viable option for periodontal regeneration [72]. A recent study focused on creating biodegradable nanofibrous scaffolds for periodontal bone repair. The researchers created pure polylactic acid and chitosan/polylactic acid blends using emulsion electrospinning and then examined the mechanical and biological characteristics of each. The outcomes demonstrated that the addition of chitosan nanoparticles improved the mechanical characteristics of pure polylactic acid nanofibers, encouraging bone marrow stem cells' cell adhesion and osteogenic development. On the other hand, it also caused a rise in TLR4 (Toll-like receptor 4) and inflammatory mediator expression in human periodontal ligament cells. The findings imply that the TLR4 pathway may be involved in the regulation of the increased production of inflammatory mediators [73]. To produce individualized bone regeneration structures, In 2019, Bakopoulou et al. combined biomimetic chitosan/gelatin (CS/Gel) scaffolds with oral cells such as DPSCs. Using glutaraldehyde (GTA), two scaffold types, CS/Gel-0.1 and CS/Gel-1, were created and seeded with DPSCs. Both in vitro and in vivo evaluations were carried out. The outcomes demonstrated that both scaffolds-maintained cell viability and generated a nanocrystalline calcium phosphate phase rich in hydroxyapatite. According to this study, scaffolds containing CS/Gel-0.1 (0.1% (v/v)) are more successful in upregulating osteo/odontogenic genes [74].

In 2022, Guilherme Neves et al. used mesenchymal stem cells (MSC) and calcium phosphates (CaP) to construct and analyze polymeric porosity scaffolds for regenerative dentistry. It was discovered that 5% of CaP types, namely HA and Brushite, were linked to Chitosan-Xanthan Scaffolds. Following implantation, the Chitosan-Xanthan scaffolds displayed increased inflammatory cell counts after 7 and 30 days, as well as greater cell viability after 48 h. According to a recent study, 2% chlorhexidine gluconate can be delivered using hematite-doped bioglass/chitosan scaffolds as an alternate implant approach for treating infected root canals. When tested against Enterococcus faecalis, the scaffolds demonstrated both osteoinduction and antibacterial action. After 14 days, the addition of Fe_2O_3 entirely eradicated bacterial growth and improved medication release. The scaffolds' impressive osteoinduction suggests that endodontic therapy may benefit from using them [75]. In 2020, Aksel et al. investigated the antimicrobial efficacy of hydrogel scaffolds loaded with antibiotics against Enterococcus faecalis as well as their capacity to promote the proliferation and mineralization of dental pulp stem cells. They discovered that whilst antibiotic-loaded chitosan-fibrin gels displayed no CFUs (colony-forming units), antibiotic-loaded fibrin, and chitosan-fibrin gels decreased CFUs and cell viability. The chitosan-fibrin gel loaded with two antibiotics demonstrated improved antibacterial qualities without sacrificing increased cell viability, cell spreading, or mineralization activity. The results of the study showed that chitosan-fibrin gels loaded with two antibiotics were superior at encouraging the proliferation and mineralization of dental pulp stem cells [76]. In 2019, Bordini et al. developed a porous scaffold consisting of chitosan, calcium aluminate, and sodium alginate (CH-AlCa) together

with 1,25-dihydroxy vitamin D3 (also known as 1,25VD) to boost the odontogenic capacity of HDPCs. The porous scaffold exhibited improved odontoblastic phenotypic expression on HDPCs, an ordered and connected pore network, and higher porosity. The scaffold supplemented with 1α,25VD increased the cells' capacity to exhibit an odontoblastic phenotype. According to the study's findings, HDPCs' chemotaxis and regeneration capability gets increased by the CH-AlCa scaffold, and the cells' ability to express an odontoblastic phenotype is enhanced when low-dosage 1α,25VD is added to this scaffold. This suggests that low dosages of 1,25VD and calcium-aluminate-enriched chitosan scaffolds have potential as a cell-free tissue engineering strategy for pulp capping [77]. Another study examined the application of bioactive lactose-modified chitosan (CTL)-coated alginate bone scaffolds for the proliferation and differentiation of human dental pulp stem cells. According to the study, when CTL is used as a coating for porous scaffolds, it can raise extracellular matrix deposition and alkaline phosphatase activity. When differentiation stimuli were introduced, the scaffolds also enhanced osteogenic activity and cell adhesion. According to this study, hDPSCs and CTL scaffolds could be employed in concert to speed up bone mending [78].

Bio-based three-dimensional (3D) polymer scaffolds that support cell adhesion and preserve metabolic processes should be non-toxic, biocompatible, and biodegradable. For tissue engineering, they ought to resemble an in vivo milieu where cells or growth factors can be incorporated to restore damaged tissues or organs [79,80]. The 3D scaffolds are commonly prepared by using conventional techniques such as particle leaching, gas foaming, phase separation, freeze drying, melt molding, fiber meshes, and solution casting techniques. In 2024, Paczkowska-Walendowska et al. developed a3D-printed hydrogel scaffold containing an extract of *Scutellariae baicalensis*. The hydrogel, which had an amorphous dispersion and the highest printability, contained 2.5% w/v of chitosan, 2% w/v of gelatin, and 10% w/w of extract. With an initial burst release and a continuous release profile, the hydrogel also demonstrated a considerable increase in baicalin release in vitro. Additionally, the study assessed the capacity of 3D-printed scaffolds to limit the activity of the hyaluronidase enzyme to determine their anti-inflammatory qualities. The material's biocompatibility was demonstrated by cytotoxicity testing, and it sped wound healing by 97.1% after 24 h, suggesting that it may be used to treat periodontal disorders [81].

In pulpectomized root canals, the restoration of functional tooth pulp represents a novel therapeutic approach in dentistry. To do this, a scaffold that promotes tooth pulp tissue neoformation and inhibits the proliferation of remaining endodontic bacteria is required. An inventive cellularized fibrin hydrogel with antibacterial qualities was developed by Ducret et al. and combined with chitosan. The microstructure, antibacterial activity, and viability and spreading of dental pulp-mesenchymal stem/stromal cell formulations were investigated. Comparative investigation revealed that chitosan had a strong antibacterial impact in the fibrin network, comparable vitality of DP-MSCs, fibroblast-like shape, proliferation rate, and capacity to produce type I/III collagen [82].

Several problems, including drug-induced bleeding patients, vascular anomalies, platelet defects, coagulation disorders, and inherited bleeding diseases, make managing bleeding patients after dental surgery difficult [83]. Haemostatic medications based on chitosan can be used to halt bleeding and encourage faster bleeding times. To enhance their performance, continuous research and development is being done. Chitosan is antibacterial, bioactive, harmless, biodegradable, and biocompatible, and also promotes healing. A larger surface area is required for contact with platelets to maximize the haemostatic effects. When gentamicin was added to chitosan scaffolds, chitosan gallium-MBG exhibited enhanced haemostatic ability, increased antibacterial activity, and enhanced biocompatibility [83].

5. Mouthwash

Plaque-induced gingivitis, microbial biofilms on the surfaces of teeth, and poor oral hygiene can be overcome by mouthwashes. A randomized clinical trial compared chitosan mouthwash and chlorhexidine mouthwash regarding their effects on dental plaque accumulation and gingivitis [81]. The results indicated that the chitosan mouthwash significantly reduced plaque accumulation, gingival inflammation, and colony-forming units The antibacterial properties of Fluoridated Chitosan Polymers have been found effective against Oral biofilms. Chitosan chlorhexidine (CH) mouth rinse is effective against microbes and its clinical effects show action on plaque control which indicates the effectiveness of chitosan in plaque control. They observed that both chitosan and chlorhexidine were found to be effective in controlling plaque [80]. However, a combination of both provides even better results.

6. Enamel Regeneration

Changes in lifestyle may occur due to the deterioration of human dental enamel. The regeneration and remineralization of dental enamel is very tough. The carbonate hydroxyapatite nanorods (prisms) are the major component of Dental enamel. Non-invasive methods are highly

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recommended for dental enamel regeneration. Chitosan supports the remineralization of enamel and dentin. Chitosan and agarose in the form of biopolymer-based hydrogel have been reported for the remineralization of an acid-etched native enamel surface. Their developed hydrogels were characterized and observed similar hierarchical HAP structure to the native enamel from nano- to microscale. Chitosan has shown carbonation and moderated the formation of HAP nanorods in addition to providing an extracellular matrix to support growing enamel-like structures. These reports indicate the guiding property of chitosan towards the formation of hard tissues as dental enamel. Amelogenin peptides like LRAP (leucine-rich amelogenin peptide) are effective in enamel repair. These naturally occurring amelogenins as smaller peptide analogs may be a competent, low-cost, and safe strategy for enamel biomimetics to curb the high prevalence of incipient dental caries.

Chitosan effectively inhibits biofilm development and bacterial growth while promoting enamel regeneration. The antibacterial activity of chitosan varies based on its molecular weight and degree of deacetylation. The chitosan amino group is responsible for anti-bacterial action, which permits entry to the bacteria. Chitosan offers superior antibacterial action synergistically with composite materials. Chitosan can act as a reservoir for calcium and phosphorus ion deposition, which aids in the remineralization of enamel caries sites. The remineralization of chitosan pre-treated enamel white spot lesions (WSLs) by bioglass in the presence of the pellicle layer has been reported [28].

7. Nanoparticles

This study reports on the efficacy of antimicrobial photodynamic therapy using aluminum phthalocyanine, a photosensitizer encapsulated in chitosan nanoparticles, against Streptococcus mutans biofilm at three different irradiation times. To evaluate the impact of pellicle layer formation, they created 50 artificial enamel white spot lesions and treated them with various formulation groups. They observed that the Chitosan pre-treatment can enhance white spot lesion remineralization with bioglass biomaterials when a short-term salivary pellicle is present.

8. Limitations or Challenges of Using Chitosan in Dentistry

Chitosan has been widely used in different fields. However, the solubility is the major task of chitosan. In order to create many more advanced dental formulations for clinical utility in humans there are still many unresolved issues and challenges to be resolved. Chitosan due to its unique properties shown potential applications in drug delivery. The effectiveness of chitosan towards dental drug delivery may be enhanced by the utilization of chitosan-based derivatives such as thiolated or carboxy methyl chitosan. However, a systematic approach related to the selectivity of drug delivery system, in vitro and in vivo toxicity and safety issues of chitosan-based biomaterials and their synthesis methods must be investigated very closely before formulation development.

9. Conclusion

The notable range of bio properties of chitosan makes a suitable candidate in dentistry. For the preparation of various formulations such as hydrogels, nanodispersions, nanomicelles, nanocomposites the chitosan has been utilized. The natural existence, biodegradable and non-toxic nature of chitosan also supports its usage in drug delivery applications. Chitosan has to be explored still towards the development of novel targeted/stimuli responsive based drug delivery formulations.

List of Abbreviations

aPDT	Antimicrobial Photodynamic Treatment
AuNPs	Gold Nanoparticles
BG	Bioactive Glass
CFU	Colony-Forming Units
CGA	Chlorogenic Acid
CNPs-ICG	Chitosan Nanoparticles-Indocyanine Green
CS/DA	Chitosan and Dicarboxylic Acid
CSH	Calcium Sulfate Hemihydrate
CST	Chitosan-Grafted Thymol
CZNP	Calcium Zirconium Nanoparticles
DDA	Degrees of Deacetylation
DPSCs	Dental Pulp Stem Cells
GTA	Glutaraldehyde
HA	Hyaluronic Acid
hPDLCs	Human Periodontal Ligament Cells
LRAP	Leucine Rich Amelogenin Peptide
micro-CT	Micro-Computed Tomography
MRSA	Methicillin Resistant Staphylococcus Aureus
MSC	Mesenchymal Stem Cells
PEI	Poly (Ethylene Imine)
PI	Peri-Implantitis
PIDs	Peri-Implant Disorders
PIM	Peri-Implant Mucositis

SACT	Sonodynamic Antimicrobial Chemotherapy
SBF	Salt-Buffered Sand
Ti	Titanium
TLR4	Toll-Like Receptor 4
WSL	White Spot Lesions
ZOI	Zone of Inhibition

Author Contributions

Conceptualization, review, editing and visualization: V.K. and S.M.; Supervision: J.R. and L.K.; Writing-original draft preparation: K.J. and V.S. All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

The authors do not declare any conflict of interest.

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AI Declaration

The authors confirm that no AI tools were used to generate any content of this manuscript. The authors take full responsibility for the accuracy, originality, integrity, and scientific content of the manuscript.

References

[1] Tiyaboonchai, W. Chitosan nanoparticles: A promising system for drug delivery. *Naresuan Univ. J.* **2003**, *11*, 51–66. Available online: https://www.thaiscience.info/journals/Article/NUJ/10440936.pdf.

[2] Pangestuti, R.; Kim, S.-K. Neuroprotective properties of chitosan and its derivatives. *Mar. Drugs* **2010**, *8*, 2117–2128. [CrossRef] [PubMed]

[3] Bhumkar, D.R.; Pokharkar, V.B. Studies on effect of pH on cross-linking of chitosan with sodium tripolyphosphate: A technical note. *AAPS Pharmscitech* **2006**, *7*, E138–E143. [CrossRef] [PubMed]

[4] Agnihotri, S.A.; Mallikarjuna, N.N.; Aminabhavi, T.M. Recent advances on chitosan-based micro- and nanoparticles in drug delivery. *J. Control. Release* **2004**, *100*, 5–28. [CrossRef]

[5] Nagpal, K.; Singh, S.K.; Mishra, D.N. Chitosan nanoparticles: A promising system in novel drug delivery. *Chem. Pharm. Bull.* **2010**, *58*, 1423–1430. [CrossRef]

[6] Soutter, W. *Chitosan Nanoparticles—Properties and Applications*, 2nd ed.; AZoNano: Manchester, UK 2013.

[7] Mohammed, M.A.; Syeda, J.T.M.; Wasan, K.M.; Wasan, E.K. An overview of chitosan nanoparticles and its application in non-parenteral drug delivery. *Pharmaceutics* **2017**, *9*, 53. [CrossRef]

[8] Avadi, M.R.; Sadeghi, A.M.M.; Mohammadpour, N.; Abedin, S.; Atyabi, F.; Dinarvand, R.; Rafiee-Tehrani, M. Preparation and characterization of insulin nanoparticles using chitosan and Arabic gum with ionic gelation method. *Nanomed. Nanotechnol. Biol. Med.* **2009**, *6*, 58–63. [CrossRef]

[9] Prabaharan, M.; Mano, J.F. Chitosan-based particles as controlled drug delivery systems. *Drug Deliv.* **2005**, *12*, 41–57. [CrossRef]

[10] Tokumitsu, H.; Ichikawa, H.; Fukumori, Y. Chitosan-gadopentetic acid complex nanoparticles for gadolinium neutron-capture therapy of cancer: Preparation by novel emulsion-droplet coalescence technique and characterization. *Pharm. Res.* **1999**, *16*, 1830–1835. [CrossRef]

[11] Gupta, H.; Velpandian, T.; Jain, S. Ion- and pH-activated novel in-situ gel system for sustained ocular drug delivery. *J. Drug Target.* **2010**, *18*, 499–505. [CrossRef]

[12] Gupta, S.; Vyas, S.P. Carbopol/chitosan based pH triggered in situ gelling system for ocular delivery of timolol maleate. *Sci. Pharm.* **2010**, *78*, 959–976. [CrossRef] [PubMed]

[13] Bhardwaj, A.; Bhardwaj, A.; Misuriya, A.; Maroli, S.; Manjula, S.; Singh, A.K. Nanotechnology in dentistry: Present and future. *J. Int. Oral Health* **2014**, *6*, 121–126. [PubMed]

[14] Mascarenhas, R.; Hegde, S.; Manaktala, N. Chitosan nanoparticle applications in dentistry: A sustainable biopolymer. *Front. Chem.* **2024**, *12*, 1362482. [CrossRef] [PubMed]

[15] Pragati, S.; Ashok, S.; Kuldeep, S. Recent advances in periodontal drug delivery systems. *Int. J. Drug Deliv.* **2009**, *1*, 1–14. Available online: https://ijdd.arjournals.org/index.php/ijdd/article/view/1.

[16] Sanap, P.; Hegde, V.; Ghunawat, D.; Patil, M.; Nagaonkar, N.; Jagtap, V. Current applications of chitosan nanoparticles in dentistry: A review. *Int. J. Appl. Dent. Sci.* **2020**, *6*, 81–84. [CrossRef]

[17] Agrawal, A.; Reche, A.; Agrawal, S.; Paul, P. Applications of Chitosan Nanoparticles in Dentistry: A Review. *Cureus* **2023**, *15*, e49934. [CrossRef]

[18] Mati-Baouche, N.; Elchinger, P.-H.; de Baynast, H.; Pierre, G.; Delattre, C.; Michaud, P. Chitosan as an adhesive. *Eur. Polym. J.* **2014**, *60*, 198–212. [CrossRef]

[19] Chauhan, S.; Das, S.; Baig, F.A.H.; Hussain Qadri, S.S.; Radhika, D.; Modi, H. Applications of chitosan in dentistry—A review article. *J. Pharm. Negat. Results* **2022**, *13*, 1359–1364. [CrossRef]

[20] Hoveizi, E.; Naddaf, H.; Ahmadianfar, S.; Gutmann, J.L. Encapsulation of human endometrial

stem cells in chitosan hydrogel containing titanium oxide nanoparticles for dental pulp repair and tissue regeneration in male Wistar rats. *J. Biosci. Bioeng.* **2023**, *135*, 331–340. [CrossRef]

[21] Xiao, Z.; Que, K.; Wang, H.; An, R.; Chen, Z.; Qiu, Z.; Lin, M.; Song, J.; Yang, J.; Lu, D.; et al. Rapid biomimetic remineralization of the demineralized enamel surface using nano-particles of amorphous calcium phosphate guided by chimaeric peptides. *Dent. Mater.* **2017**, *33*, 1217–1228. [CrossRef]

[22] Jahanizadeh, S.; Yazdian, F.; Marjani, A.; Omidi, M.; Rashedi, H. Curcumin-loaded chitosan/carboxymethyl starch/montmorillonite bio-nanocomposite for reduction of dental bacterial biofilm formation. *Int. J. Biol. Macromol.* **2017**, *105*, 757–763. [CrossRef] [PubMed]

[23] Karthik, C.S.; Chethana, M.H.; Manukumar, H.M.; Ananda, A.P.; Sandeep, S.; Nagashree, S.; Mallesha, L.; Mallu, P.; Jayanth, H.S.; Dayananda, B.P. Synthesis and characterization of chitosan silver nanoparticle decorated with benzodioxane coupled piperazine as an effective anti-biofilm agent against MRSA: A validation of molecular docking and dynamics. *Int. J. Biol. Macromol.* **2021**, *181*, 540–551. [CrossRef] [PubMed]

[24] Panahi, F.; Rabiee, S.M.; Shidpour, R. Synergic effect of chitosan and dicalcium phosphate on tricalcium silicate-based nanocomposite for root-end dental application. *Mater. Sci. Eng. C-Mater. Biol. Appl.* **2017**, *80*, 631–641. [CrossRef] [PubMed]

[25] Ma, S.; Moser, D.; Han, F.; Leonhard, M.; Schneider-Stickler, B.; Tan, Y. Preparation and antibiofilm studies of curcumin loaded chitosan nanoparticles against polymicrobial biofilms of Candida albicans and Staphylococcus aureus. *Carbohydr. Polym.* **2020**, *241*, 116254. [CrossRef] [PubMed]

[26] Ashrafi, B.; Rashidipour, M.; Marzban, A.; Soroush, S.; Azadpour, M.; Delfani, S.; Ramak, P. Mentha piperita essential oils loaded in a chitosan nanogel with inhibitory effect on biofilm formation against S. mutans on the dental surface. *Carbohydr. Polym.* **2019**, *212*, 142–149. [CrossRef]

[27] Hu, F.; Zhou, Z.; Xu, Q.; Fan, C.; Wang, L.; Ren, H.; Xu, S.; Ji, Q.; Chen, X. A novel pH-responsive quaternary ammonium chitosan-liposome nanoparticles for periodontal treatment. *International J. Biol. Macromol.* **2019**, *129*, 1113–1119. [CrossRef]

[28] Khan, G.; Yadav, S.K.; Patel, R.R.; Kumar, N.; Bansal, M.; Mishra, B. Tinidazole functionalized homogeneous electrospun chitosan/poly (ε-caprolactone) hybrid nanofiber membrane: Development, optimization and its clinical implications. *Int. J. Biol. Macromol.* **2017**, *103*, 1311–1326. [CrossRef]

[29] Sun, X.; Zhang, Y.; Fukumoto, S.; Masuda, K.; Dong, N. Adaptive mechanisms of dental pulp stem cells (DPSCs) in response to oral disease: Evaluating chitosan-calcium zirconium nanoparticle biomaterials for tissue engineering. *Mater. Chem. Phys.* **2024**, *323*, 129610. [CrossRef]

[30] Hu, D.; Tian, T.; Ren, Q.; Han, S.; Li, Z.; Deng, Y.; Lu, Z.; Zhang, L. Novel biomimetic peptide-loaded chitosan nanoparticles improve dentin bonding via promoting dentin remineralization and inhibiting endogenous matrix metalloproteinases. *Dent. Mater.* **2023**, *40*, 160–172. [CrossRef]

[31] Wang, M.; Li, Y.; Zhao, Y.; Gao, H.; Xu, Z.; Chen, L.; Liu, J.; Liang, H. pH-triggered chitosan-sodium caseinate nanocarriers with charge-switching property: Characterization and applications in dental care. *Food Hydrocoll.* **2024**, *152*, 109919. [CrossRef]

[32] Shukla, R.; Mishra, P.; Handa, M.; Hasnain, M.S.; Beg, S. Chitosan as a biomaterial for implantable drug delivery. In *Chitosan in Drug Delivery 2022*; Academic Press: Cambridge, MA, USA, 2022; pp. 133–158. [CrossRef]

[33] Aneksomboonpol, P.; Mahardawi, B.; Nan, P.N.; Laoharungpisit, P.; Kumchai, T.; Wongsirichat, N.; Aimjirakul, N. Surface structure characteristics of dental implants and their potential changes following installation: A literature review. *J. Korean Assoc. Oral. Maxillofac. Surg.* **2023**, *49*, 114–124. [CrossRef] [PubMed]

[34] Hallmann, L.; Gerngroß, M.-D. Chitosan and its application in dental implantology. *J. Stomatol. Oral Maxillofac. Surg.* **2022**, *123*, e701–e707. [CrossRef] [PubMed]

[35] Raut, H.K.; Das, R.; Liu, Z.; Liu, X.; Ramakrishna, S. Biocompatibility of Biomaterials for Tissue Regeneration or Replacement. *Biotechnol. J.* **2020**, *15*, e2000160. [CrossRef] [PubMed]

[36] Anggani, H.S.; Rusli, V.; Bachtiar, E.W. Chitosan gel prevents the growth of Porphyromonas gingivalis, Tannerella forsythia, and Treponema denticola in mini-implant during orthodontic treatment. *Saudi Dent. J.* **2021**, *33*, 1024–1028. [CrossRef] [PubMed]

[37] Alhazmi, A.S.; Syame, S.M.; Mohamed, W.S.; Hakim, A.S. Incorporation of Plant Extracted Hydroxyapatite and Chitosan Nanoparticles on the Surface of Orthodontic Micro-Implants: An In-Vitro Antibacterial Study. *Microorganisms* **2022**, *10*, 581. [CrossRef]

[38] Anggani, H.S.; Perdana, R.G.; Siregar, E.; Bachtiar, E.W. The effect of coating chitosan on Porphyromonas gingivalis biofilm formation in the surface of orthodontic mini-implant. *J. Adv. Pharm. Technol. Res.* **2021**, *12*, 84–88. [CrossRef]

[39] Mahmood, A.; Maher, N.; Amin, F.; Alqutaibi, A.Y.; Kumar, N.; Zafar, M.S. Chitosan-based materials for dental implantology: A comprehensive review. *Int. J. Biol. Macromol.* **2024**, *268*, 131823. [CrossRef]

[40] Polo-Corrales, L.; Latorre-Esteves, M.; Ramirez-Vick, J.E. Scaffold design for bone regeneration. *J. Nanosci. Nanotechnol.* **2014**, *14*, 15–56. [CrossRef]

[41] Kim, Y.; Zharkinbekov, Z.; Raziyeva, K.; Tabyldiyeva, L.; Berikova, K.; Zhumagul, D.; Temirkhanova, K.; Saparov, A. Chitosan-Based Biomaterials for Tis-

sue Regeneration. *Pharmaceutics* **2023**, *15*, 807. [CrossRef]

[42] Leong, K.F.; Chua, C.K.; Sudarmadji, N.; Yeong, W.Y. Engineering functionally graded tissue engineering scaffolds. *J. Mech. Behav. Biomed. Mater.* **2008**, *1*, 140–152. [CrossRef]

[43] Schönweger, F.; Sprecher, C.M.; Milz, S.; Dommann-Scherrer, C.; Meier, C.; Dommann, A.; Neels, A.; Wahl, P. New Insights into Osteointegration and Delamination from a Multidisciplinary Investigation of a Failed Hydroxyapatite-Coated Hip Joint Replacement. *Materials* **2020**, *13*, 4713. [CrossRef] [PubMed]

[44] Jadhav, L.; Madiwal, V.; Rajwade, J.M. The dental implant surface: A review of the past, present and future. *J. Mater. Sci. Mater. Med.* **2026**, *37*, 21. [CrossRef] [PubMed]

[45] Sarvaiya, B.B.; Kumar, S.; Pathan, M.S.H.; Patel, S.; Gupta, V.; Haque, M. The Impact of Implant Surface Modifications on the Osseointegration Process: An Overview. *Cureus* **2025**, *17*, e81576. [CrossRef] [PubMed]

[46] Paradowska-Stolarz, A.; Mikulewicz, M.; Laskowska, J.; Karolewicz, B.; Owczarek, A. The Importance of Chitosan Coatings in Dentistry. *Mar. Drugs* **2023**, *21*, 613. [CrossRef] [PubMed]

[47] Schoenbaum, T.; Moy, P.; Aghaloo, T.; Elashoff, D. Risk Factors for Dental Implant Failure in Private Practice: A Multicenter Survival Analysis. *Int. J. Oral Maxillofac. Implant.* **2021**, *36*, 388–394. [CrossRef]

[48] Xu, Y.; Xia, D.; Han, J.; Yuan, S.; Lin, H.; Zhao, C. Design and fabrication of porous chitosan scaffolds with tunable structures and mechanical properties. *Carbohydr. Polym.* **2017**, *177*, 210–216. [CrossRef] [PubMed]

[49] López-Valverde, N.; Aragoneses, J.; López-Valverde, A.; Rodríguez, C.; Macedo de Sousa, B.; Aragoneses, J.M. Role of chitosan in titanium coatings. trends and new generations of coatings. *Front. Bioeng. Biotechnol.* **2022**, *10*, 907589. [CrossRef] [PubMed]

[50] Del Olmo, J.A.; Pérez-Álvarez, L.; Pacha-Olivenza, M.; Ruiz-Rubio, L.; Gartziandia, O.; Vilas-Vilela, J.L.; Alonso, J.M. Antibacterial catechol-based hyaluronic acid, chitosan and poly (N-vinyl pyrrolidone) coat-ings onto Ti6Al4V surfaces for application as biomedical implant. *Int. J. Biol. Macromol.* **2021**, *183*, 1222–1235. [CrossRef] [PubMed]

[51] Wu, T.; Zhou, Q.; Hong, G.; Bai, Z.; Bian, J.; Xie, H.; Chen, C. A chlorogenic acid-chitosan com-plex bifunctional coating for improving osteogenesis differentiation and bactericidal properties of zirconia implants. *Colloids Surf. B Biointerfaces* **2023**, *230*, 113484. [CrossRef] [PubMed]

[52] Alnufaiy, B.M.; Lambarte, R.N.A.; Al-Hamdan, K.S. The Osteogenetic Potential of Chitosan Coated Implant: An In Vitro Study. *J. Stem Cells Regen. Med.* **2020**, *16*, 44–49. [CrossRef] [PubMed]

[53] Shurbaji, S.; Malki, A.; Rathore, H.A. Surface modifications of titanium dental implants: optimizing antibacterial activity and osteoimmunomodulation. *Front. Bioeng. Biotechnol.* **2026**, *14*, 1795260. [CrossRef] [PubMed]

[54] Bumgardner, J.D.; Chesnutt, B.M.; Yuan, Y.; Yang, Y.; Appleford, M.; Oh, S.; McLaughlin, R.; Elder, S.H.; Ong, J.L. The integration of chitosan-coated titanium in bone: An in vivo study in rabbits. *Implant. Dent.* **2007**, *16*, 66–79. [CrossRef] [PubMed]

[55] Li, B.; Xia, X.; Guo, M.; Jiang, Y.; Li, Y.; Zhang, Z.; Liu, S.; Li, H.; Liang, C.; Wang, H. Biological and antibacterial properties of the micro-nanostructured hydroxyapatite/chitosan coating on titanium. *Sci. Rep.* **2019**, *9*, 14052. [CrossRef] [PubMed]

[56] López-Valverde, N.; López-Valverde, A.; Cortés, M.P.; Rodríguez, C.; De Sousa, B.M.; Aragoneses, J.M. Bone Quantification Around Chitosan-Coated Ti-tanium Dental Implants: A Preliminary Study by Micro-CT Analysis in Jaw of a Canine Model. *Front. Bioeng. Biotechnol.* **2022**, *10*, 858786. [CrossRef] [PubMed]

[57] Francis, A.; Yang, Y.; Boccaccini, A.R. A new strategy for developing chitosan conversion coating on magnesium substrates for orthopedic implants. *Appl. Surf. Sci.* **2019**, *466*, 854–862. [CrossRef]

[58] Anees, E.; Riaz, M.; Imtiaz, H.; Hussain, T. Electrochemical corrosion study of chitosan-hydroxyapatite coated dental implant. *J. Mech. Behav. Biomed. Mater.* **2023**, *150*, 106268. [CrossRef] [PubMed]

[59] Divakar, D.D.; Jastaniyah, N.T.; Altamimi, H.G.; Alnakhli, Y.O.; Muzaheed; Alkheraif, A.A.; Haleem, S. Enhanced antimicrobial activity of naturally derived bioactive molecule chitosan conjugated silver nanoparticle against dental implant pathogens. *Int. J. Biol. Macromol.* **2018**, *108*, 790–797. [CrossRef] [PubMed]

[60] Chittratan, P.; Chalitangkoon, J.; Wongsariya, K.; Mathaweesansurn, A.; Detsri, E.; Monvisade, P. New Chitosan-Grafted Thymol Coated on Gold Nanoparticles for Control of Cariogenic Bacteria in the Oral Cavity. *ACS Omega* **2022**, *7*, 26582–26590. [CrossRef] [PubMed]

[61] Park, S.; Kim, H.; Choi, K.S.; Ji, M.K.; Kim, S.; Gwon, Y.; Park, C.; Kim, J.; Lim, H.-P. Graphene–Chitosan Hybrid Dental Implants with Enhanced Antibacterial and Cell-Proliferation Properties. *Appl. Sci.* **2020**, *10*, 4888. [CrossRef]

[62] Sayar, F.; Karimi, M.R.; Boroujerdi, S. Efficacy of antimicrobial photodynamic therapy with chitosan nanoparticles for decontamination of dental implants contaminated with Aggregatibacter actinomycetemcomitans. *Sci. Rep.* **2025**, *15*, 34213. [CrossRef]

[63] Abdolahinia, E.D.; Golestani, S.; Seif, S.; Afra, N.; Aflatoonian, K.; Jalalian, A.; Valizadeh, N.; Abdollahinia, E.D. A review of the therapeutic potential of dental stem cells as scaffold-free models for tissue engineering application. *Tissue Cell* **2024**, *86*, 102281. [CrossRef] [PubMed]

[64] Sultana, N.; Cole, A.; Strachan, F. Biocomposite Scaffolds for Tissue Engineering: Materials, Fabrication Techniques and Future Directions. *Materials* **2024**, *17*, 5577. [CrossRef] [PubMed]

[65] Iqbal, N.; Ganguly, P.; Yildizbakan, L.; Raif, E.M.; Jones, E.; Giannoudis, P.V.; Jha, A. Chitosan Scaffolds from Crustacean and Fungal Sources: A Comparative Study for Bone-Tissue-Engineering Applications. *Bioengineering* **2024**, *11*, 720. [CrossRef] [PubMed]

[66] Silvestro, I.; Sergi, R.; Scotto d'Abusco, A.; Mariano, A.; Martinelli, A.; Piozzi, A.; Francolini, I. Chitosan scaffolds with enhanced mechanical strength and elastic response by combination of freeze gelation, photo-crosslinking and freeze-drying. *Carbohydr. Polym.* **2021**, *267*, 118156. [CrossRef] [PubMed]

[67] Sadeghinia, A.; Davaran, S.; Salehi, R.; Jamalpoor, Z. Nano-hydroxy apatite/chitosan/gelatin scaffolds enriched by a combination of platelet-rich plasma and fibrin glue enhance proliferation and differentiation of seeded human dental pulp stem cells. *Biomed. Pharmacother.* **2019**, *109*, 1924–1931. [CrossRef] [PubMed]

[68] Anaya-Sampayo, L.M.; García-Robayo, D.A.; Roa, N.S.; Rodriguez-Lorenzo, L.M.; Martínez-Cardozo, C. Platelet-rich fibrin (PRF) modified nano-hydroxyapatite/chitosan/gelatin/alginate scaffolds increase adhesion and viability of human dental pulp stem cells (DPSC) and osteoblasts derived from DPSC. *Int. J. Biol. Macromol.* **2024**, *273*, 133064. [CrossRef] [PubMed]

[69] Sukpaita, T.; Chirachanchai, S.; Suwattanachai, P.; Everts, V.; Pimkhaokham, A.; Ampornaramveth, R.S. In vivo bone regeneration induced by a scaffold of chitosan/dicarboxylic acid seeded with human peri-odontal ligament cells. *Int. J. Mol. Sci.* **2019**, *20*, 4883. [CrossRef] [PubMed]

[70] Covarrubias, C.; Cádiz, M.; Maureira, M.; Celhay, I.; Cuadra, F.; von Marttens, A. Bionanocomposite scaffolds based on chitosan–gelatin and nanodimensional bioactive glass particles: In vitro properties and in vivo bone regeneration. *J. Biomater. Appl.* **2018**, *32*, 1155–1163. [CrossRef] [PubMed]

[71] Soares, D.G.; Anovazzi, G.; Bordini, E.A.F.; Zuta, U.O.; Leite, M.L.A.S.; Basso, F.G.; Hebling, J.; Costa, C.A.d.S. Biological Analysis of Simvastatin-releasing Chitosan Scaffold as a Cell-free System for Pulp-dentin Regeneration. *J. Endod.* **2018**, *44*, 971–976.e1. [CrossRef] [PubMed]

[72] Varoni, E.; Vijayakumar, S.; Canciani, E.; Cochis, A.; De Nardo, L.; Lodi, G.; Rimondini, L.; Cerruti, M. Chitosan-Based Trilayer Scaffold for Multitissue Pe-riodontal Regeneration. *J. Dent. Res.* **2017**, *97*, 303–311. [CrossRef] [PubMed]

[73] Shen, R.; Xu, W.; Xue, Y.; Chen, L.; Ye, H.; Zhong, E.; Ye, Z.; Gao, J.; Yan, Y. The use of chitosan/PLA nanofibers by emulsion eletrospinning for periodontal tissue engineering. *Artif. Cells Nanomed. Biotechnol.* **2018**, *46*, 419–430. [CrossRef] [PubMed]

[74] Bakopoulou, A.; Georgopoulou, A.; Grivas, I.; Bekiari, C.; Prymak, O.; Loza, K.; Epple, M.; Papadopoulos, G.C.; Koidis, P.; Chatzinikolaidou, M. Dental pulp stem cells in chitosan/gelatin scaffolds for enhanced orofacial bone regeneration. *Dent. Mater.* **2018**, *35*, 310–327. [CrossRef] [PubMed]

[75] Neves, J.G.; Rocha, D.N.; Lopes, C.C.; Barbosa, R.M.; Ferreira, L.F.; Westin, C.B.; Moraes, Â.M.; Calsa, B.; Santamaria, M., Jr.; Correr-Sobrinho, L.; Correr, A.B. Calcium Phosphates Chitosan–Xanthan Composite Scaffolds Associated with Mesenchymal Stem Cells for Regenerative Dentistry Application. *Ceram. Int.* **2022**, *48*, 23088–23095. [CrossRef]

[76] Aksel, H.; Mahjour, F.; Bosaid, F.; Calamak, S.; Azim, A.A. Antimicrobial activity and biocompatibility of antibiotic-loaded chitosan hydrogels as a potential scaffold in regenerative endodontic treatment. *J. Endod.* **2020**, *46*, 1867–1875. [CrossRef] [PubMed]

[77] Bordini, E.A.F.; Cassiano, F.B.; Silva, I.S.P.; Usberti, F.R.; Anovazzi, G.; Pacheco, L.E.; Pansani, T.N.; Leite, M.L.; Hebling, J.; Costa, C.A.d.S.; et al. Synergistic potential of 1α,25-dihydroxyvitamin D3 and calcium-aluminatechitosan scaffolds with dental pulp cells. *Clin. Oral Investig.* **2020**, *24*, 663–674. [CrossRef] [PubMed]

[78] Porrelli, D.; Gruppuso, M.; Vecchies, F.; Marsich, E.; Turco, G. Alginate bone scaffolds coated with a bioactive lactose modified chitosan for human dental pulp stem cells proliferation and differentiation. *Carbohydr. Polym.* **2021**, *273*, 118610. [CrossRef] [PubMed]

[79] Chan, B.P.; Leong, K.W. Scaffolding in tissue engineering: General approaches and tissue-specific considerations. *Eur. Spine J.* **2008**, *17*, 467–479. [CrossRef] [PubMed]

[80] Loh, Q.L.; Choong, C. Three-dimensional scaffolds for tissue engineering applications: role of porosity and pore size. *Tissue Eng. Part B Rev.* **2013**, *19*, 485–502. [CrossRef] [PubMed]

[81] Paczkowska-Walendowska, M.; Koumentakou, I.; Lazaridou, M.; Bikiaris, D.; Miklaszewski, A.; Plech, T.; Cielecka-Piontek, J. 3D-Printed Chitosan Based Scaffolds with Scutellariae baicalensis Extract for Dental Applications. *Pharmaceutics* **2024**, *16*, 359. [CrossRef] [PubMed]

[82] Ducret, M.; Montembault, A.; Josse, J.; Pasde-loup, M.; Celle, A.; Benchrih, R.; Mallein-Gerin, F.; Alliot-Licht, B.; David, L.; Farges, J.-C. Design and characterization of a chitosan-enriched fibrin hydro-gel for human dental pulp regeneration. *Dent. Mater.* **2019**, *35*, 523–533. [CrossRef] [PubMed]

[83] Lestari, W.; Yusry, W.N.A.W.; Haris, M.S.; Jaswir, I.; Idrus, E. A glimpse on the function of chitosan as a dental hemostatic agent. *Jpn. Dent. Sci. Rev.* **2020**, *56*, 147–154. [CrossRef] [PubMed]

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