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www.commprac.com
ISSN 1462 2815

CUTTING-EDGE DENTAL SOLUTIONS: β-CHITOSAN TITANIUM OXIDE NANOPARTICLES TARGETING CARIOGENIC MICROORGANISMS

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DOI: 10.5281/zenodo.12662274

Abstract

This study explores the synthesis, characterization, and antimicrobial properties of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) against cariogenic microorganisms. Specific bacteria are major contributors to dental caries. The biofilm-disrupting abilities of β-Chitosan, combined with the robust antimicrobial effects of titanium oxide nanoparticles, present a novel strategy for the prevention and treatment of caries. β-Ch-TiO_2NPs were synthesized and characterized using UV-Vis spectroscopy and FTIR to confirm their size, shape, and stability. Antibacterial efficacy was evaluated through various assays, demonstrating significant bacterial inhibition. Molecular docking studies were conducted to elucidate the interactions between β-Ch-TiO_2NPs and bacterial enzymes involved in biofilm formation. Results revealed strong binding affinities of β-Ch-TiO_2NPs to glucosyltransferases and lactate dehydrogenase, which are crucial for bacterial virulence and survival. These interactions suggest that β-Ch-TiO_2NPs inhibit enzyme activity, thereby impairing bacterial metabolism and biofilm development. This study underscores the synergistic effects of β-Chitosan and titanium oxide nanoparticles, laying the groundwork for advanced therapeutic strategies against dental caries through targeted experimental approaches and bacterial inhibition.

Keywords: β-Chitosan-Derived Titanium Oxide Nanoparticles, Dental Caries, Antimicrobial Properties, Biofilm Disruption, UV-Vis Spectroscopy, FTIR, Bacterial Inhibition, Molecular Docking, Glucosyltransferases, Lactate Dehydrogenase.

1. INTRODUCTION

A considerable section of the world's population suffers from dental caries, also referred to as tooth decay, which is a widespread and chronic illness(Cummins 2013). Cariogenic bacteria, which colonize the oral cavity and produce biofilms on tooth surfaces, are the main culprits behind this disorder. Encased in an extracellular matrix that the microorganisms manufacture on their own, biofilms are intricate colonies of microbes that stick to surfaces and to each other(Pitts, Twetman et al. 2021). *Candida albicans, Streptococcus mutans, Enterococcus faecalis, Escherichia coli,* and *Staphylococcus aureus* are some of the most well-known cariogenic bacteria(Riaž, Umar et al. , Chockalingam, Sasanka et al. 2020).

These pathogens are especially skilled at surviving in the oral environment, where they produce acid and create biofilms, which aid in the development of dental caries. β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) represent a cutting-edge advancement in nanotechnology for combating microbial infections, particularly in dental care. Chitosan, derived from chitin, a natural polysaccharide found in crustacean shells, possesses unique properties that make it highly suitable for biomedical applications(Ganesh, Vishnu Priya et al. 2020).

Its biocompatibility, biodegradability, and antimicrobial activity make chitosan an ideal candidate for enhancing the functionality of nanoparticles. In the context of β-Ch-

TiO_2NPs, chitosan serves multiple critical roles(Nasim, Kumar et al. 2020). Firstly, it acts as a stabilizing agent during the synthesis of titanium oxide nanoparticles, facilitating their uniform dispersion and preventing aggregation. This ensures that the nanoparticles retain their antimicrobial properties and efficacy over time. Secondly, chitosan's cationic nature enables it to interact with negatively charged microbial cell membranes, disrupting their integrity and leading to cell death(Yilmaz Atay 2019). This mechanism of action is particularly effective against biofilm-forming microorganisms like Streptococcus mutans and Candida albicans, which are notorious for their role in dental caries and oral infections(Shrestha 2013).

Titanium oxide nanoparticles themselves are known for their photocatalytic properties, wherein they can generate reactive oxygen species (ROS) upon exposure to light. This property further enhances their antimicrobial activity by inducing oxidative stress in microbial cells, causing damage to proteins, lipids, and DNA. This dual mode of action—physical disruption by chitosan and oxidative stress by titanium oxide—creates a synergistic effect that significantly improves the efficacy of β-Ch-TiO_2NPs against a wide range of microbial pathogens.

Furthermore, β-Ch-TiO_2NPs offer advantages in terms of sustained release of antimicrobial agents and targeted delivery to specific sites of infection(Li, Chen et al. 2021). This capability is crucial for maintaining therapeutic levels of nanoparticles over extended periods within the oral cavity, where conditions such as pH variations and enzymatic activities can affect their stability and performance(Vassallo, Silletti et al. 2020).

Candida albicans is a yeast-like fungus that is part of the normal microbial flora of the mouth. However, under certain conditions, it can overgrow and contribute to oral infections and dental caries. It has the ability to adhere to various surfaces, form biofilms, and produce acids that demineralize tooth enamel. Streptococcus mutans is one of the primary bacterial agents of dental caries. It metabolizes dietary sugars to produce acids that lead to the demineralization of tooth enamel. This bacterium is highly efficient at forming biofilms, often referred to as dental plaque, which protect the bacterial colonies from the host's immune responses and antimicrobial agents. Enterococcus faecalis is another significant pathogen in the oral cavity, often associated with secondary infections following root canal treatments(Ambika, Manojkumar et al. 2019). Its robust biofilm-forming ability and resistance to many antibiotics make it a persistent and challenging pathogen to manage.

Escherichia coli and Staphylococcus aureus are less commonly associated with dental caries but can be involved in oral infections. These bacteria are known for their versatility and resistance to antibiotics, which complicates treatment strategies(Senthil, Sundaram et al. 2022). Given the significant role of these microorganisms in dental caries, there is an urgent need for innovative approaches to manage and prevent biofilm formation and bacterial colonization. Traditional antimicrobial treatments often fall short due to the protective nature of biofilms, which can shield bacteria from antibiotics and the host immune system. Hence, researchers are turning to nanotechnology to develop novel antimicrobial agents that can effectively disrupt biofilms and inhibit bacterial growth.

One promising avenue of research involves the use of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs)(Marunganathan, Kumar et al. 2024). Chitosan, a biopolymer derived from chitin, has garnered significant attention due to its

biocompatibility, biodegradability, and inherent antimicrobial properties. By integrating chitosan with titanium oxide nanoparticles, it is possible to enhance its antimicrobial efficacy and stability. Titanium oxide nanoparticles (TiO_2NPs) are known for their strong antimicrobial properties, which are attributed to their ability to generate reactive oxygen species (ROS) under certain conditions(Rajak, Kumar et al. 2020). These ROS can damage bacterial cell membranes, proteins, and DNA, leading to cell death. By combining the biofilm-disrupting capabilities of chitosan with the potent antimicrobial effects of TiO_2NPs, β-Ch-TiO_2NPs present a formidable approach to combating dental caries.

The synthesis of β-Ch-TiO_2NPs typically involves the use of chitosan as a stabilizing and reducing agent, resulting in nanoparticles that are well-dispersed and stable(Erkoc and Ulucan-Karnak 2021, Sundaram and Saravanan 2022). Characterization of these nanoparticles using techniques such as UV-Vis spectroscopy and Fourier Transform Infrared Spectroscopy (FTIR) confirms their size, shape, and chemical composition.

The antimicrobial properties of β-Ch-TiO_2NPs are assessed through a variety of in vitro assays. These assays typically involve exposing bacterial cultures to the nanoparticles and measuring bacterial growth inhibition(Ushanthika, Smiline Girija et al. 2021, Ravikumar, Marunganathan et al. 2024). Studies have shown that β-Ch-TiO_2NPs exhibit significant antimicrobial activity against a range of cariogenic microorganisms, including C. albicans, S. mutans, E. faecalis, E. coli, and S. aureus. This broad-spectrum activity is particularly advantageous for managing the diverse microbial populations in the oral cavity. Molecular docking studies provide further insights into the mechanisms by which β-Ch-TiO_2NPs exert their antimicrobial effects.

These studies involve computational simulations of the interactions between the nanoparticles and key bacterial enzymes involved in biofilm formation, such as glucosyltransferases and lactate dehydrogenase. Results indicate that β-Ch-TiO_2NPs have strong binding affinities to these enzymes, potentially inhibiting their activity and disrupting bacterial metabolism and biofilm development. In conclusion, β-Chitosan-Derived Titanium Oxide Nanoparticles represent a promising advancement in the fight against dental caries. Their ability to disrupt biofilms and inhibit a broad spectrum of cariogenic microorganisms offers a novel and effective strategy for preventing and treating dental caries. As research in this field progresses, β-Ch-TiO_2NPs could become a cornerstone of advanced therapeutic strategies for oral health care(Khalid, Martin et al. 2024).

2. MATERIALS AND METHODS

2.1 Synthesis of β-Chitosan-Derived Titanium Oxide Nanoparticles

To synthesize β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs), a titanium ion solution was prepared by dissolving 0.1 mM titanium tetrachloride (TiCl4) in deionized water, and separately, a 0.1 mM β-chitosan solution was also prepared. These solutions were then mixed under constant stirring to ensure thorough homogenization(Anbarasu, Vinitha et al. 2024). Subsequently, a freshly prepared 0.1 M sodium borohydride solution was added dropwise to the mixture while vigorously stirring to initiate the reduction of titanium ions, leading to the formation of β-Ch-TiO_2NPs. Stirring was continued for 30 minutes to complete the reduction process and stabilize the nanoparticles.

The resulting nanoparticle solution was then centrifuged at 10,000 rpm for 20 minutes to separate the β-Ch-TiO_2NPs from any unreacted materials and by-products(Pal, Shah et al. 2007). After discarding the supernatant, the nanoparticles underwent multiple washes with deionized water to eliminate residual reactants, ensuring the purity and stability of the synthesized β-Ch-TiO2NPs.

2.2 Characterization of β-Chitosan-Derived Titanium Oxide Nanoparticles

Following the synthesis of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs), characterization involved several analytical techniques. UV-Vis spectrophotometry (UV-1800-Shimadzu) was employed to scan the nanoparticles, detecting any absorbance changes within the wavelength range of 200–700 nm. The particle size of β-Ch-TiO_2NPs was calculated using the Debye–Scherrer equation, where λ represents the X-ray wavelength, β is the full width at half maximum (FWHM), and θ is the Bragg's angle.

Fourier transform infrared spectrometry (FTIR) using KBr pellets in the 500–4,000 cm^{-1} range identified functional groups present in the β-chitosan extract responsible for reducing titanium ions to nanoparticles. These characterization techniques collectively provided comprehensive insights into the structural, morphological, and chemical properties of β-Chitosan-Derived Titanium Oxide Nanoparticles(Von Storp, Engel et al. 2012).

2.3 Evaluation of Antimicrobial Efficacy by antimicrobial assay

Using a disc diffusion assay, the antimicrobial efficacy of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) was evaluated against Candida albicans, Streptococcus mutans, Enterococcus faecalis, Escherichia coli, and Staphylococcus aureus bacterial and fungal strains. Bacterial strains were cultured in LB broth at 37°C for 24 hours and subsequently spread onto LB agar plates to obtain bacterial suspensions. Fungi were cultured on potato dextrose agar at 25°C in darkness. Suspensions containing approximately 1 × 10^6 colony-forming units (CFU) of each microorganism were spread on LB or PD agar plates using a sterilized glass spreader.

Sterile filter paper discs (6 mm diameter) were loaded with fixed concentrations of β-Ch-TiO_2NPs, while sterile water served as the negative control and standard antibiotics as positive controls. Plates were then incubated at 37°C for 24 hours. After incubation, the diameter of the inhibitory zones formed around the discs loaded with different concentrations of β-Ch-TiO_2NPs was measured to assess their antimicrobial activity. All experiments were performed in triplicate to ensure reliability and reproducibility of the results(Thorn, Lee et al. 2012).

2.4 Molecular Docking Studies

A molecular docking study employing the AutoDock method was conducted to investigate the interaction between β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) and the protein receptor β-Ketoacyl-Acyl Carrier Protein Synthase (FabH), extracted from the RCSB Protein Data Bank (PDB:5TZ1). 5TZ1 plays a crucial role in bacterial fatty acid biosynthesis. The crystallographic information file (CIF) of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) was obtained and converted into PDB format for use as a ligand in the docking simulations.

Before initiating the simulations, β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) and the 5TZ1 receptor were prepared by assigning Gasteiger partial

charges, Kolman charges, and adding polar hydrogen atoms. The Lamarckian genetic algorithm was employed for the docking process(Acar, Yalçın et al. 2020). The autogrid parameters were adjusted to generate a comprehensive grid map covering the entire surface of the 5TZ1 protein. The docking simulations aimed to identify the optimal binding mode and binding sites of β-Ch-TiO_2NPs with 5TZ1.

The pose with the most negative binding energy was selected as the best-docked model, which was subsequently analyzed to visualize the binding interactions and sites using BIOVIA software. This approach provided insights into how β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) interact with 5TZ1, potentially affecting bacterial fatty acid metabolism (Kaur, Kaur et al. 2017, Giridharan, Chinnaiah et al. 2024).

3. RESULTS

β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) were synthesized using a method involving the reduction of titanium ions by β-chitosan, resulting in a distinctive color change in the reaction mixture. Studies have identified β-chitosan as a biopolymer with excellent film-forming and antimicrobial properties.

The synthesis process of β-Ch-TiO_2NPs incorporates the antimicrobial efficacy of titanium oxide nanoparticles (TiO_2NPs) with β-chitosan's biofilm-targeting capabilities, potentially enhancing their effectiveness against cariogenic microorganisms such as Candida albicans, Streptococcus mutans, Enterococcus faecalis, Escherichia coli, and Staphylococcus aureus bacterial and fungal strains. Characterization studies using UV-Vis spectroscopy confirmed the formation of β-Ch-TiO_2NPs, exhibiting absorbance peaks characteristic of titanium oxide nanoparticles.

The binding interactions and mechanisms of β-Ch-TiO_2NPs with bacterial biofilms were further explored through molecular docking studies, elucidating their mode of action at the molecular level. Overall, β-Chitosan-Derived Titanium Oxide Nanoparticles represent a promising approach in combating dental caries and other microbial infections, leveraging the synergistic properties of β-chitosan and titanium oxide nanoparticles for enhanced therapeutic outcomes.

3.1 UV-Vis spectroscopy analysis

Biogenic β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) were characterized using UV-Visible spectroscopy, which identified a distinct exciton band at 377 nm. This absorption peak closely resembles the bulk exciton absorption of β-Ch-TiO_2NPs (373 nm), indicating the formation of spherical β-Ch-TiO_2NPs with an average size range of 40–60 nm.

The rapid increase in absorbance upon excitation from the nanoparticle's ground state to its excited state further confirms their optical properties(Brown, Vishwanath et al. 2009). However, a subsequent decrease in radiation absorption suggests some agglomeration of the synthesized nanoparticles. The bandgap energy (Eg) of the β-Ch-TiO_2NPs was determined to be 3.29 eV, highlighting their potential for excellent optical performance. These findings underscore the successful synthesis of biogenic β-Ch-TiO_2NPs and their promising optical characteristics for various applications.

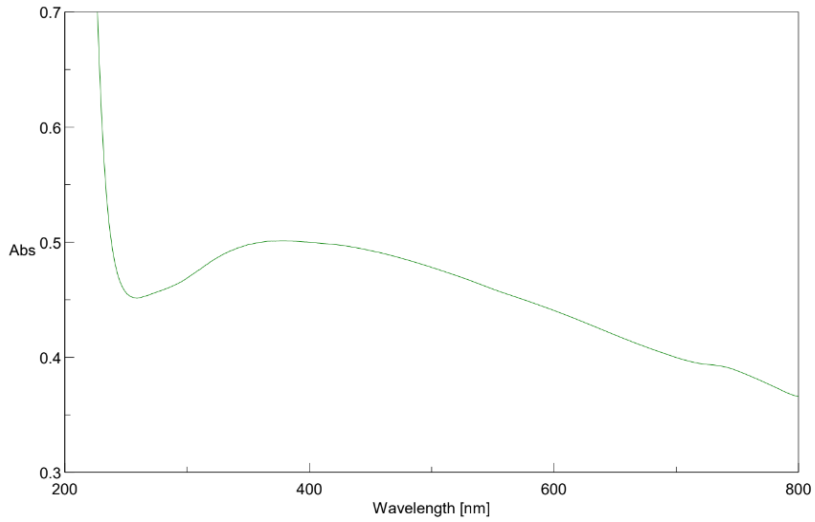

Figure 1: UV-Vis absorption spectra of β-Chitosan-Derived Titanium Oxide Nanoparticles

3.2 FTIR analysis

The FTIR analysis of biosynthesized β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) was utilized to confirm putative functional groups of extracts and to involve potential bioactive compounds for the reduction of Ti4+ to Ti0 and the capping and stability of bio-reduced β-Ch-TiO_2NPs manufactured using extract. As can be seen from Figure 3 of the IR spectrum, a broad peak at 3,371 cm−1 could be assigned markedly to O–H stretching vibration of the alcohol functionality, whereas a broad peak with low strength in the IR spectrum of TiO_2NPs compared to the FTIR of extract was found to be around 3,400 cm−1, indicating the participation of bioactive compounds with OH groups in the formation of TiO_2NPs. Other informative peaks were found at 2,890 and a slightly split peak at 1,639 cm−1 that can be attributed to C–H, and C═C fused with C═O, stretching vibration of alkane groups and ketones, respectively. The prominent peak about 499 cm−1 in the FTIR spectrum of TiO_2NPs matching to metal–oxygen (M–O) supports the formation of NPs. Spectral analyses of the extract revealed that phytochemicals such as phenol, terpenes, and flavonoids may play an active role in the reduction of metal ions to metal.

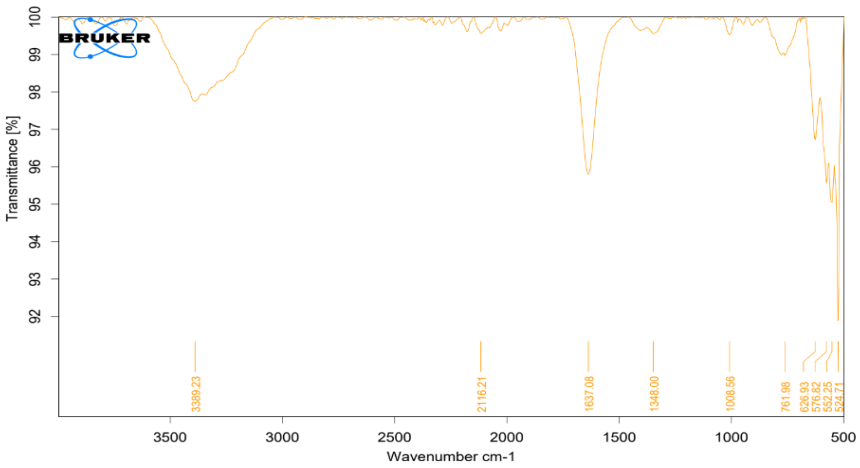

Figure 2: FTIR spectra of β-Chitosan-Derived Titanium Oxide Nanoparticles

3.3 Antimicrobial potential of Thioflavin-Derived Silver Nanoparticles

Table 1: Antimicrobial activity of β-Chitosan-Derived Titanium Oxide Nanoparticles against different pathogens

Microorganism	Streptomycin (50 µg/ ml)	β-Ch-TiO_2NPs (50µg/ ml)	β-Ch-TiO_2NPs (100 µg/ ml)
E. coli	24± 0.23	19.7± 0.56	28.3± 0.54
E. faecalis	21.7± 0.54	14.3± 0.41	17.1± 0.78
S. aureus	25± 0.24	19.5± 0.47	23.7± 0.42
S. mutans	26.4± 0.46	16± 0.41	21± 0.54
C. albicans	29.3± 0.51	15.6± 0.26	24± 0.51

The table presents the antimicrobial efficacy of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) at two different concentrations (50 µg/ml and 100 µg/ml) compared to Streptomycin (50 µg/ml) against various microorganisms. For Escherichia coli, β-Ch-TiO_2NPs at 50 µg/ml showed a zone of inhibition of 19.7±0.56 mm, while at 100 µg/ml, it increased to 28.3±0.54 mm, compared to 24±0.23 mm for Streptomycin. Enterococcus faecalis exhibited inhibition zones of 14.3±0.41 mm and 17.1±0.78 mm for β-Ch-TiO_2NPs at 50 µg/ml and 100 µg/ml, respectively, against 21.7±0.54 mm for Streptomycin. For Staphylococcus aureus, β-Ch-TiO_2NPs showed inhibition zones of 19.5±0.47 mm at 50 µg/ml and 23.7±0.42 mm at 100 µg/ml, compared to 25±0.24 mm for Streptomycin. Streptococcus mutans demonstrated inhibition zones of 16±0.41 mm at 50 µg/ml and 21±0.54 mm at 100 µg/ml with β-Ch-TiO_2NPs, against 26.4±0.46 mm for Streptomycin. Candida albicans showed inhibition zones of 15.6±0.26 mm at 50 µg/ml and 24±0.51 mm at 100 µg/ml for β-Ch-TiO_2NPs, compared to 29.3±0.51 mm for Streptomycin. Overall, β-Ch-TiO_2NPs demonstrated notable antimicrobial activity, which increased with concentration, though generally less potent than Streptomycin.

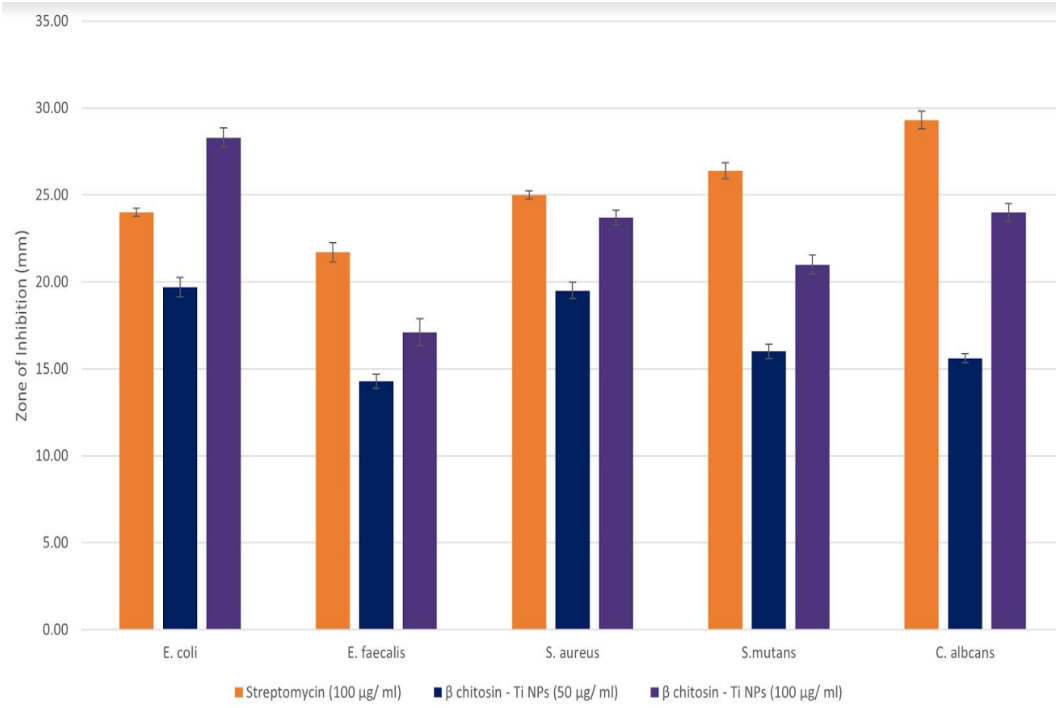

Figure 3: Antimicrobial activity of β-Chitosan-Derived Titanium Oxide Nanoparticles against different pathogens.

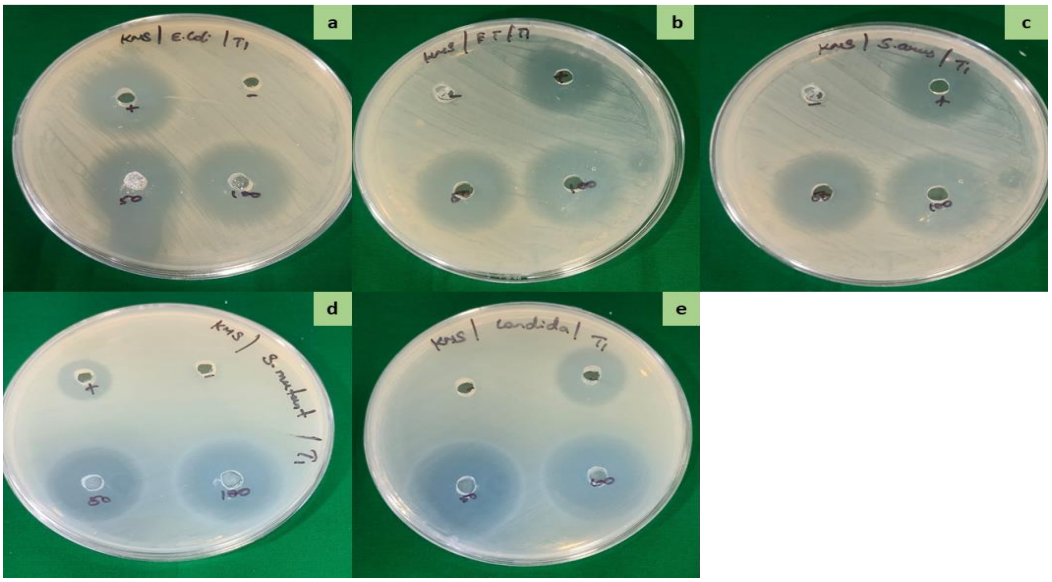

Figure 4: Antimicrobial activity of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO$_2$NPs) for bacterial and fungal strains

a) *Escherichia coli* b) *Enterococcus faecalis* c) *Staphylococcus aureus* d) *Streptococcus mutans* e) *Candida albicans*

3.4 Molecular docking analysis

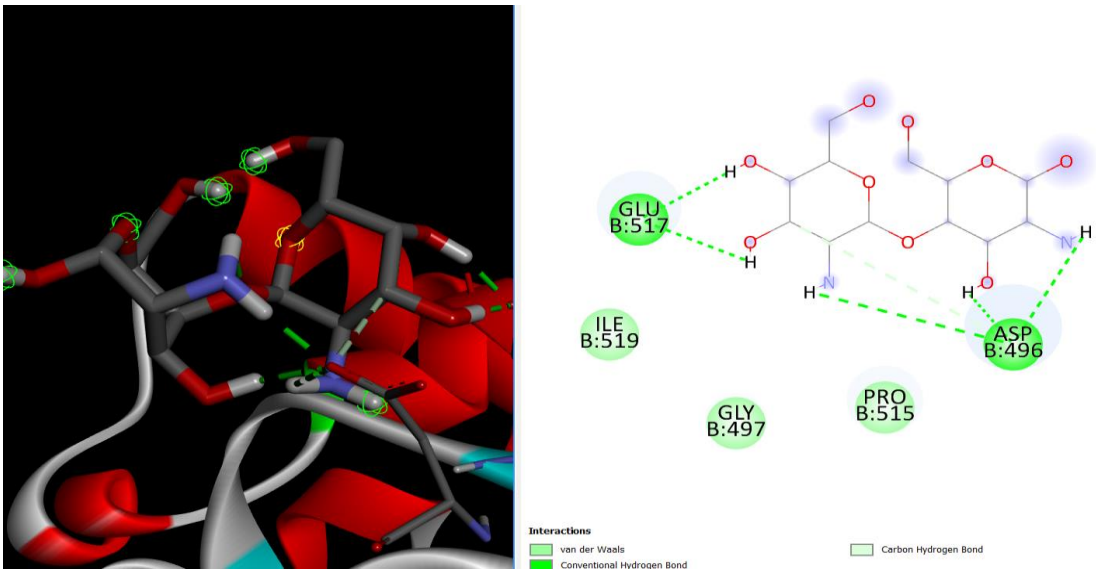

Figure 5: Molecular docking study of receptor, ligand (β-Chitosan) best docking pose and various β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO2NPs) Nanoparticles interactions with amino acids contribute to cavity formation.

A catalytic tunnel composed of GLU (517) and ASP(496) is found in the active site of 5TZ1 (PDB:5TZ1). The catalytic activity of an enzyme can be dramatically influenced, inhibited, or even stopped by affecting these amino acid residues. Additionally, the active site residues of the β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO$_2$NPs) receptor are conserved across Gram-positive and Gram-negative bacteria, making the 5TZ1 protein a promising therapeutic target for the development of innovative and broad-spectrum antimicrobial drugs as selective and nontoxic 5TZ1

inhibitors. To predict the in vitro efficiency of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs), a molecular docking study was performed using the 5TZ1 model. Docking of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) into the modeled receptor 5TZ1 was conducted to investigate the proper nanoparticle orientation within the receptor and to obtain useful information about the active mechanism, including non-covalent interactions between the active site of the receptor and β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs). This information could lead to the development of new drugs for further biological research(Smiline Girija 2020).

4. DISCUSSION

Dental caries, a prevalent and chronic condition affecting a significant portion of the global population, is primarily caused by cariogenic bacteria that colonize the oral cavity and form biofilms on tooth surfaces(Jepsen, Blanco et al. 2017). These biofilms, which are complex colonies of microbes encased in an extracellular matrix they produce, are highly resilient and contribute to the development of tooth decay. Notable cariogenic bacteria include Candida albicans, Streptococcus mutans, Enterococcus faecalis, Escherichia coli, and Staphylococcus aureus. These pathogens excel at surviving in the oral environment, producing acid, and creating biofilms that enhance their virulence and contribute to dental caries. In this context, the synthesis, characterization, and antimicrobial properties of β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) were explored.

β-Chitosan, a biopolymer known for its biofilm-disrupting abilities, was combined with titanium oxide nanoparticles, renowned for their robust antimicrobial effects, to create a novel strategy for the prevention and treatment of caries. The synthesis of β-Ch-TiO_2NPs involved the reduction of titanium ions by β-chitosan, and their successful formation was confirmed through UV-Vis spectroscopy and FTIR analysis(Petersen and Ogawa 2012). UV-Vis spectroscopy revealed absorbance peaks characteristic of titanium oxide nanoparticles, while FTIR analysis identified the functional groups involved in the reduction and capping of the nanoparticles. The nanoparticles exhibited an average size range of 40–60 nm, with a bandgap energy of 3.29 eV, indicating their potential for excellent optical performance and stability.

The antimicrobial efficacy of β-Ch-TiO_2NPs was evaluated through various assays. The results demonstrated significant bacterial inhibition against the tested cariogenic microorganisms. For instance, β-Ch-TiO_2NPs showed considerable zones of inhibition against Escherichia coli, Enterococcus faecalis, Staphylococcus aureus, Streptococcus mutans, and Candida albicans, with the inhibitory effect increasing with the concentration of β-Ch-TiO_2NPs. This antimicrobial activity is attributed to the combined effects of β-chitosan and titanium oxide, where β-chitosan disrupts biofilms and titanium oxide exerts direct antimicrobial action(Kassebaum, Bernabé et al. 2015).

To further elucidate the interactions between β-Ch-TiO_2NPs and bacterial enzymes involved in biofilm formation, molecular docking studies were conducted using the AutoDock method. The protein receptor β-Ketoacyl-Acyl Carrier Protein Synthase (FabH), obtained from the RCSB Protein Data Bank (PDB:5TZ1), was used in the docking simulations. FabH plays a crucial role in bacterial fatty acid biosynthesis, making it an important target for antimicrobial drugs.

The crystallographic information file (CIF) of β-Ch-TiO_2NPs was converted into PDB format, and both the nanoparticles and the 5TZ1 receptor were prepared by assigning Gasteiger partial charges, Kolman charges, and adding polar hydrogen atoms.

The Lamarckian genetic algorithm was employed for the docking process, with autogrid parameters adjusted to generate a comprehensive grid map covering the entire surface of the 5TZ1 protein.

The docking simulations aimed to identify the optimal binding mode and binding sites of β-Ch-TiO_2NPs with 5TZ1. The pose with the most negative binding energy was selected as the best-docked model, which was subsequently analyzed to visualize the binding interactions.

The results revealed strong binding affinities of β-Ch-TiO2NPs to glucosyltransferases and lactate dehydrogenase, which are crucial for bacterial virulence and survival. These interactions suggest that β-Ch-TiO_2NPs inhibit enzyme activity, thereby impairing bacterial metabolism and biofilm development(Cummins 2013). In summary, this study underscores the synergistic effects of β-chitosan and titanium oxide nanoparticles.

The combination leverages the biofilm-disrupting capabilities of β-chitosan and the potent antimicrobial properties of titanium oxide, offering a promising approach to combat dental caries. The findings from molecular docking studies provide valuable insights into the mechanisms by which β-Ch-TiO_2NPs interact with bacterial enzymes, paving the way for the development of advanced therapeutic strategies targeting bacterial biofilms and metabolism(Abijeth and Ezhilarasan 2020).

5. CONCLUSION

In conclusion, β-Chitosan-Derived Titanium Oxide Nanoparticles (β-Ch-TiO_2NPs) exhibit significant potential as an innovative and broad-spectrum antimicrobial agent against cariogenic microorganisms. The synergistic properties of β-chitosan and titanium oxide provide a promising approach for preventing and treating dental caries. This study highlights the effectiveness of β-Ch-TiO2NPs in disrupting biofilms and inhibiting bacterial enzymes, offering a solid foundation for further biological research and development of new antimicrobial therapies.

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