ANTIBACTERIAL EFFICACY EVALUATION OF SILVER NANOPARTICLES AND CHITOSAN COATINGS ON TITANIUM SURFACES OF GRAM-POSITIVE AND GRAM-NEGATIVE BACTERIAL RESISTANCE: A SYSTEMATIC REVIEW AND META-ANALYSIS

Dr. Veena Khandagave[1], Dr. Sounyala Rayannavar[2], Dr. Hitendra Ramesh Jain[3], Dr. Manasi Jadhav[4], Dr. Tatva Keswani[5], Dr. Lisamarie S. L.[6]

[1]*Post graduate student, Department of Prosthodontics crown and bridge, KLE V K Institute of dental sciences, Belagavi.*
[2]*Professor, Department of Prosthodontics crown and bridge, KLE V K Institute of dental sciences, Belagavi.*
[3]*Professor & HOD, Department of Public Health Dentistry, SMBT Dental College & Hospital , Sangamner.*
[4]*Post graduate student, Department of Prosthodontics crown and bridge, KLE V K Institute of dental sciences, Belagavi.*
[5]*Post graduate student, Department of Prosthodontics crown and bridge, KLE V K Institute of dental sciences, Belagavi.*
[6]*Post graduate student, Department of Oral and maxillofacial Medicine and Radiology, Manipal college of Dental sciences, Manipal.*

Corresponding Author: Dr. Veena Khandagave[1]

KEYWORDS

cell viability, chitosan, colony forming units, silver nanoparticles, systematic review, titanium implant

ABSTRACT

Aim: to evaluate the antibacterial efficacy of silver nanoparticles (AgNPs) and chitosan (CH) coating on titanium (Ti) surfaces against gram-positive and gram-negative bacteria

Methods: Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) guidelines were used and registered in PROSPERO- CRDXXXX. Electronic databases were searched from January 2000 till September 2024 for studies evaluating theantibacterial efficacy of silver nanoparticles and chitosan coating on titanium surfaces against gram-positive and gram -negative bacteria in terms of colony forming units (CFU) and cell viability on (Ti) surfaces. Screening process was done by two independent authors. Quality assessment of included studies was evaluated using the Cochrane risk of bias (ROB)-2 tool. The standardized mean difference (SMD) was used as summary statistic measure with random effect model ($p<0.05$).

Results: Four studies were included in review and three studies for meta-analysis. Quality assessment revealed a moderate to low ROB. The antibacterial efficacy was assessed in terms of colony forming units (CFU) for *Staph. Aureus* and cell viability.Meta-analysis showed that reduction in CFU of *Staph. aureus* was (SMD-12.66 (-19.54 – 5.78) more by silver nanoparticles and chitosan coated group ($p<0.05$) while greater cell viability on Ti surface was (SMD - 0.41 (-1.10 – 1.91) more by silver nanoparticles and chitosan coated group ($p>0.05$).Funnel plot did not show any presence of possible publication bias in meta-analysis. More in -vitro studies or comparative analytical studieswith should be conducted to validate the findings of this study.

Conclusion:it was found that antibacterial coating is highly effective in preventing implant associated infections and these coatings showed good anti-bactericidal property by inhibiting surface adhesion of gram positive and gram- negative bacteria with minimal cytotoxicity.

Introduction

The titanium (Ti) and its alloys have been widely used for dental and orthopaedic implants due to their good mechanical properties, corrosion resistance and biocompatibility in the physiological environment.[1] However, the Ti-based implants do not have intrinsic antibacterial property. There is always a risk for the Ti-based implants to suffer from bacterial adhesion, colonization and infection, which would result in insufficient healing, implant failure and repeated surgical intervention.[2] Therefore, construction of anti-infective surfaces on the Ti-based implants to combat implant-associated infections is urgently needed to reduce the post-surgical complications and huge socio-economic repercussions.[3,4]

Titanium (Ti) and its alloys are currently considered to be the most widely used biomaterials for dental implants because of their superior biocompatibility and excellent physicochemical properties.[5] However, implant-associated infection remains one of the most perilous complications of these procedures, leading to the failure of implant surgery along with psychological trauma and economic burden.[6]

The formation of biofilm on the surfaces of implants following by bacteria adhesion is the primary cause of infections of the mucosa and bone adjacent to the implant.[5] The process of biofilm formation involves unicellular organisms coming together to form a contiguous community encompassed in an exopolysaccharide matrix.[6] Thus, the biofilm makes the bacteria more invasive and competitive against the host defenses and presents difficulties for antibacterial treatments.[7]At the same time, the risk to develop a prosthetic implant infection (PII) is enhanced by numerous factors, including diabetes, obesity, and immunocompromised states.[2] PII induces the failure of prosthetic implantations, increasing the costs for healthcare system and resulting in high rates of morbidity and mortality.[3] Staphylococcus species are the major cause of PII and, among them, S. aureus is the most worrisome because of its ability to become resistant to antibiotics, forming biofilms and overtaking the immune response.[8]

Therefore, establishing long-term antibacterial coatings on the surfaces of titanium implants to inhibit biofilm formation is of prime importance in the prevention of implant-associated infections.[9]

Currently, the incorporation of antibacterial drugs into the coatings of dental implant surfaces has attracted increasing attention and is considered an effective strategy to prevent bacterial biofilm formation.[10]

Several techniques have been explored to endow the Ti surfaces with antifouling and antibacterial properties.[8,9] The passive antifouling coatings are able to hinder initial adhesion of bacteria on the Ti-based materials.[10] Various types of antifouling coatings have been proposed, including polymer brushes[9,11,12], thin hydrogel layers[13], polymeric multilayers[14], self-assembled monolayers[15], and textured surfaces. However, the practical application of antifouling coatings is limited by their poor long-term stability and effectiveness upon exposure to the physiological environment.[12] The active bactericidal coatings, able to kill bacteria on contact, have been introduced on the Ti-based materials to achieve long-term anti-infective effect.[13]

Antimicrobials, such as positively-charged polymers, antimicrobial peptides, metallic nanoparticles, antiseptics and antibiotics, have been integrated with the Ti-based materials as the active bactericidal coatings.[12] Among them, silver-based antibacterial agents have long been known and used due to their superior and long-lasting antimicrobial properties against a wide range of microbes.[13]

They can not only integrate with amine-functionalized molecules to form stable and versatile surface coatings, but also act as a reducing agent to reduce metal ions and coordinate with the as-formed metallic NPs through two adjacent hydroxyls.[11] Chitosan is a natural unbranched bio-polysaccharide derived from chitin via a deacetylation reaction, and exhibits high antimicrobial activity against a wide variety of pathogenic microorganisms.[12]

Going through evidences, no study to date, has provided a comprehensive, quantitative analysis on theantibacterial efficacy of silver nanoparticles and chitosan coating on titanium surfaces.

Hence, we improvised our research by including relevant literature and carried out a systematic review aiming to assess and evaluate the antibacterial efficacy of silver nanoparticles and chitosan coating on titanium surfaces against gram-positive and gram-negative bacteria's.

Methodology

Protocol development

This reviewwas conducted and performed in according to the preferred reporting items for systematic review and meta-analysis (PRISMA) 2020 statement[16]and registered in Prospective Registration of Systematic Review (PROSPERO)- CRDXXXX.

Study design

The following focused research question in the Participants (P), Intervention (I), Comparison and Outcome (O) format was proposed "Is there any difference in the antibacterial efficacy of silver nanoparticles and chitosan coating on titanium surfaces against gram-positive and gram -negative bacteria's"?

The PICO criteria for this review were as follows:

P (Population) – titanium disc

I (Intervention) – silver + chitosan -coated titanium discs

C (Comparison) – uncoated titanium discs

O (Outcome) – to assess the better antimicrobial efficacy in terms of colony forming units (CFU) for *Staph. Aureus* and cell viability

S (Study designs) – in vitro studies, analytical studies, randomized controlled trial (RCTs)

Eligibility Criteria: studies were selected based on following criteria's

a) **Inclusion Criteria:** following were the inclusion criteria
   1) Articles published in English language
   2) Articles having sufficient comparative data on antibacterial efficacy of silver nanoparticles and chitosan coating on titanium surfaces against gram-positive and gram -negative bacteria's
   3) Studies published between January 2000 – September 2024 and having relevant data on antibacterial efficacy of silver nanoparticles and chitosan coating in terms of colony forming units (CFU) for *Staph. Aureus* and cell viability
   4) Randomized controlled trials (RCTs), in vitro studies, analytical studieswere taken into consideration
   5) Articles from open access journals
   6) Articles reporting the study outcomes in terms of frequency, mean and standard deviation

b) **Exclusion Criteria:** following were the exclusion criteria
   1) Any studies conducted before 2000
   2) Articles in other than English language
   3) Reviews, abstracts, letter to the editor, editorials, animal studies and in vitro studies were excluded
   4) Articles not from open access journals
   5) Articles not reporting the study outcomes in terms of mean and standard deviation

Search Strategy

A comprehensive electronic search was performed tillSeptember2024 for the studies published within the last 24 years (from 2000 to 2024) using the following databases: PubMed, google scholar and EBSCOhostto retrieve articles in the English language. The searches in the clinical trials database, cross-referencing and grey literature were conducted using Google Scholar, Greylist, and OpenGrey.

Appropriate key words and Medical Subject Heading (MeSH) terms were selected and combined with Boolean operators like AND/OR. The relevant data was searched using the following keywords and their combinations: "surface characterization" (MeSH term) AND "biomaterials" (MeSH term); "titanium coating" (MeSH term) AND "silver nanoparticles" (MeSH term); "chitosan" (MeSH term) AND "nanoparticles" (MeSH term) AND "antimicrobial efficacy" (MeSH term); "bioactive molecules" (MeSH term) AND "*S Aureus* OR *E Coli*" (MeSH term) AND "analytical studies" (MeSH term); "randomized trials" AND "in vitro studies" (MeSH term).

In addition to the electronic search, a hand search was also made, and reference lists of the selected articles were screened. The reference lists of identified studies and relevant reviews on the subject were also scanned for possible additional studies.

Screening process

Search and screening were done by two authors. The process of choosing of articles was divided into two phases. Two reviewers looked over the titles and abstracts of every article in first round. Articles that didn't fit into the inclusion were removed. Phase-two, involved independent screening and review of few full papers by the same reviewers. Discussions were held to settle by any disputes. A third reviewer was bought in to make the ultimate decision when two reviewers could not agree upon something. All three authors came to agreement on choice in the end. When more information was needed, the studies corresponding authors were contacted by email.

Data extraction

For all included studies, following descriptive study details were extractedby two independent reviewing authors and using pilot-tested customized data extraction forms in Microsoft excel sheet with the following headings included in the final analysis: author(s), country of study, year of study, study design, modalities used, outcome evaluated, parameters assessed, estimationmethod and conclusion.

Quality assessment of included studies

The methodological quality among included studies was executed by using Cochrane collaboration risk of bias (ROB) -2 tool[17]. The tool has various domains like random sequence generation (selection bias), allocation concealment (selection bias), blinding of personnel and equipments (performance bias), blinding of outcome assessment (detection bias), incomplete outcome data (attrition bias), selective reporting (reporting bias) and other biases through their signalling questions in Review Manager (RevMan) 5.3 software. The overall risk for individual studies was assessed as low, moderate or high risk based on domains and criteria. The study was assessed to have a low overall risk only if all domains were found to have low risk. High overall risk was assessed if one or more of the six domains were found to be at high risk. A moderate risk assessment was provided to studies when one or more domains were found to be uncertain, with none at high risk.

Statistical analysis

The standardized mean difference (SDM) with 95% CI was calculated for continuous outcomes. A fixed effects model (Mantel-Haenszel method) was used if there was no heterogeneity ($p > 0.05$ or I-squared $\leq 24\%$), otherwise a random effects model (Der Simonian-Laird method) was used.[18] All statistical analyses were performed using the RevMan 5.3 (Cochrane Collaboration, Software Update, Oxford, UK). The significance level was kept at $p<0.05$.

Assessment of heterogeneity

The significance of any discrepancies in the estimates of the treatment effects of the different trials was assessed by means of Cochran's test for heterogeneity and the I^2 statistics, which describes the percentage of the total variation across studies that is due to heterogeneity rather than chance. Heterogeneity was considered statistically significant if $P < 0.1$. A rough guide to the interpretation of I^2 given in the Cochrane handbook is as follows: (1) from 0 to 40%, the heterogeneity might not be important; (2) from 30% to 60%, it may represent moderate heterogeneity; (3) from 50% to 90%, it may represent substantial heterogeneity; (4) from 75% to 100%, there is considerable heterogeneity.[19]

Investigation of publication bias

To test for the presence of publication bias, the relative symmetry of the individual study estimates was assessed around the overall estimates using Begg's funnel plot. A funnel plot

(plot of the effect size versus standard error) was drawn. Asymmetry of the funnel plot may indicate publication bias and other biases related to sample size, although asymmetry may also represent a true relationship between trial size and effect size.[20]

Results

Study Selection

After copies evaluation, reference rundown of all included examinations was screened. Of which 135 examinations were barred. After this full text articles were evaluated for qualification and articles that didn't meet consideration rules were barred. Only fourstudiesfitted into inclusion criteria and were subjected to qualitative analysis and three studies for meta-analysis as shown in in **Figure 1 below**

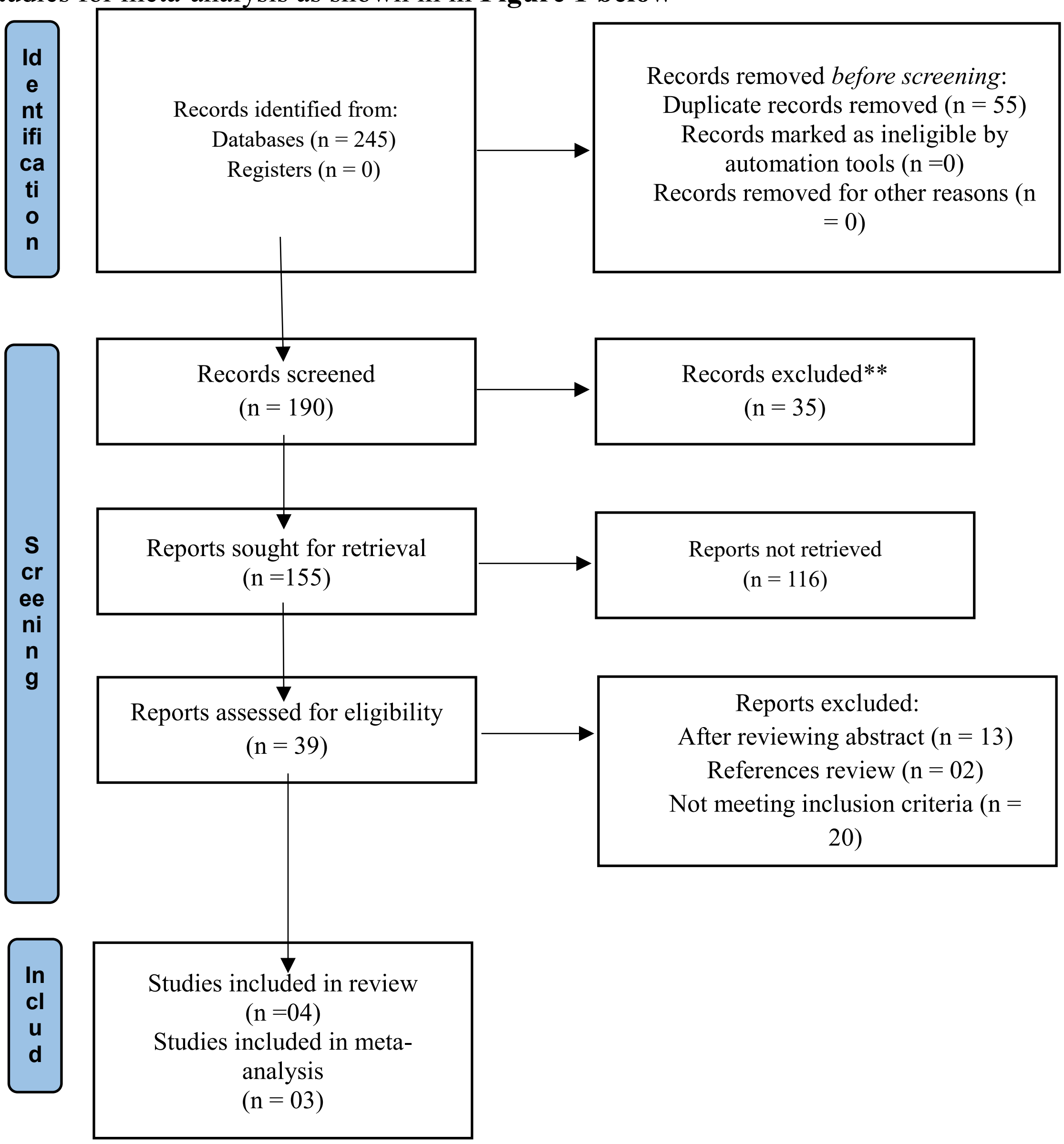

Figure 1. PRISMA 2020 Flow Diagram

Study Characteristics

As shown in **Table 1,** data was evaluated from four studies[21-24] which assessed and evaluated the antibacterial efficacy of silver nanoparticles and chitosan coating on titanium (Ti) surfacesagainst gram-positive and gram -negative bacteria in terms of colony forming units (CFU) for *Staph. Aureus* and cell viability on Ti surfaces. All the included studies had in vitro study design. Among the included studies, two studies[21,24] were conducted in China and one study each in Italy[22] and India[23]. The antibacterial efficacy was assessed in terms of CFU, minimal inhibitory concentration (MIC) and zone of inhibition (ZOI) of *Staph. Aureus,P.aeruginosa*, *P. gingivalis* and *E. coli,* cell viability on Ti surfaces, alkaline phosphatase (ALP) and lactate dehydrogenase(LDH) activity. From the results of the studies, it was found that antibacterial coating is highly effective in preventing implant associated infections and these coatings showed good anti-bactericidal property by inhibiting surface adhesion of gram positive and gram- negative bacteria with minimal cytotoxicity.

Author, years of study	**Country**	**Study design**	**Outcome evaluated**	**Parameters assessed**	**Estimation method**	**Conclusion**
Zhong et al., 2016[21]	China	In vitro	To establish Ag-Np coating on Ti implant surface and assess its antibacterial efficacy against planktonic and adherent bacteria	Cell viability, CFU, ALP and LDH activity of *S. aureus*	CFU –beef extract peptone (BEP) Cell viability - Fluorescence staining	Long term antibacterial coating is highly effective in preventing implant associated infections
Cometa et al., 2017[22]	Italy	In vitro	To prevent microbial colonization on implant surface by surface characterization through silver -chitosan coating	CFU of *S. aureus* and *P.aeruginosa*(24 & 48 hrs), cell viability	Anti-microbial activity was measured using optical density. Coatings were incubated at 37^0c at 24 & 48 hrs	Effective coatings prevent PII occuring from pathogens in immunocompromise d patients
Divakar et al., 2017[23]	India	In vitro	To evaluate antimicrobial efficacy of Ag+CH Np coating on implant surface	MIC and ZOI of *S. mutans* and *P. gingivalis*	ZOI - Kirby-Bauer disc diffusion method performed using Mueller- Hinton agar plates (MHA) MIC	Ag- Np coatings provided corrosion resistance of Ti dental implants
Cheng et al., 2019[24]	China	In vitro	To assess antibacterial efficacy of AgNps/CACS coating on Ti implants surface on surface inhibition of bacterial cells	ZOI and cell viability of *S. aureus&E. coli*	ZOI –agar plate inoculation	AgNps/CACS coatings showed good anti-bactericidal property by inhibiting surface adhesion of gram positive and gram-negative bacteria with minimal cytotoxicity.

ALP: alkaline phosphatase; Ag: silver nanoparticles; CS: chitosan; CFU: colony forming units; LDH: lactate dehydrogenase; MIC: minimal inhibitory concentration; Np: nanoparticles; PAA: polyacrylic acid; PII: prosthetic implant infection; ZOI: zone of inhibition

Table 1: showing descriptive study details of included studies

Evaluation of methodological quality

The greatest risk of bias (ROB) was observed in random sequence generation followed by allocation concealment and blinding of participants and personnel. However, all the studies included in the analysis reported moderate to the lowest levels of ROB overall. Domains such as blinding of outcome assessment, incomplete outcome data, selective reporting and other biases were assigned the lowest levels of ROB. Detailed assessments of ROB across various domains and individual studies are visually represented in **Figures 2 and 3**.

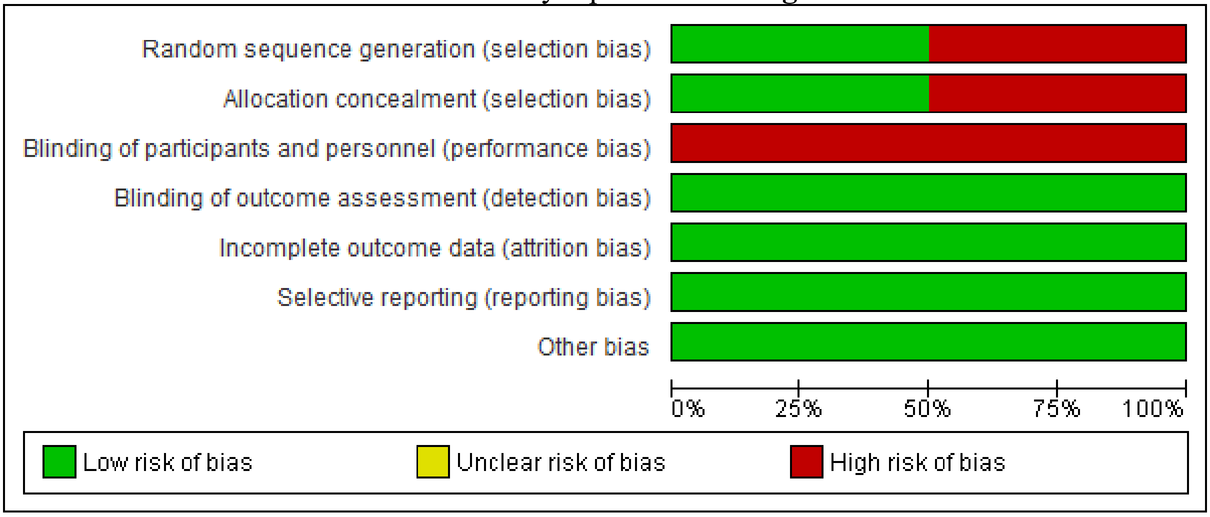

Figure 2: showing ROB graph: presented as percentages across all included studies.

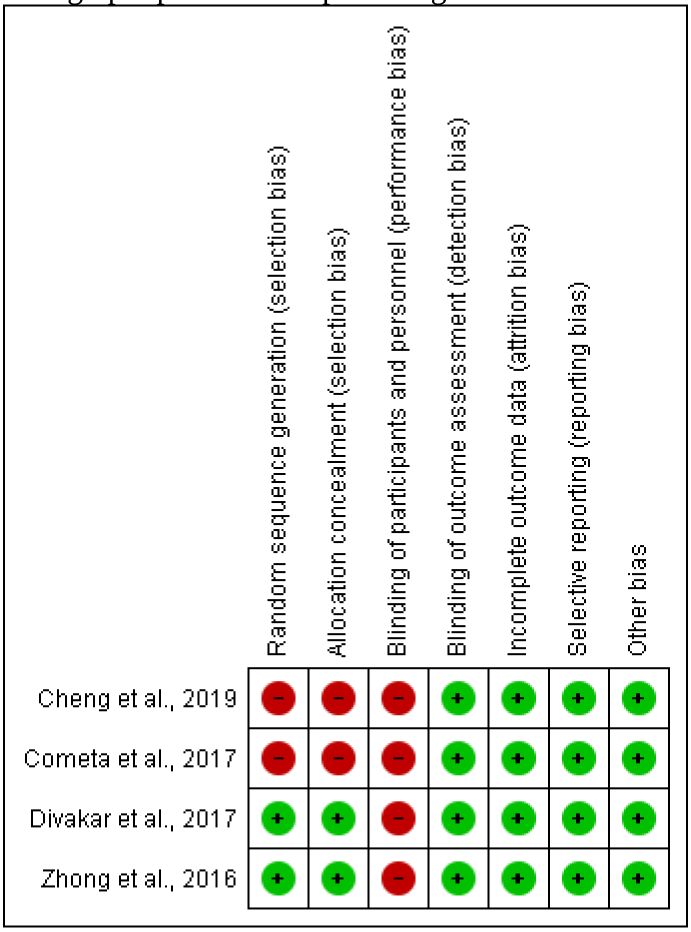

Figure 3:showing ROB summary: for each included study

Synthesis of results

The meta-analysis was performed to assess the better antimicrobial efficacy of silver nanoparticles and chitosan coating in terms of colony forming units (CFU) for *Staph. Aureus* and cell viability**as shown in figure 4 – 7.**

1. **Colony Forming Units (CFU)**

Two studies[21,22] containing data on 16 specimens, of which (*n*=8) specimens were evaluated by silver nanoparticles with chitosan coating in Ti coated disc and (*n*=8) specimens by uncoated Ti disc for the evaluation of better antimicrobial efficacy in terms of greater reduction in rate of colony forming units for *Staph. aureus*.As shown in **Figure 4.** the SMD is -12.66 (-19.54 – 5.78) and the pooled estimates signifies that reduction in rate of colony forming units of*Staph. aureus*on an average is 12.66 times more by silver nanoparticles and chitosan coated group(p<0.05).

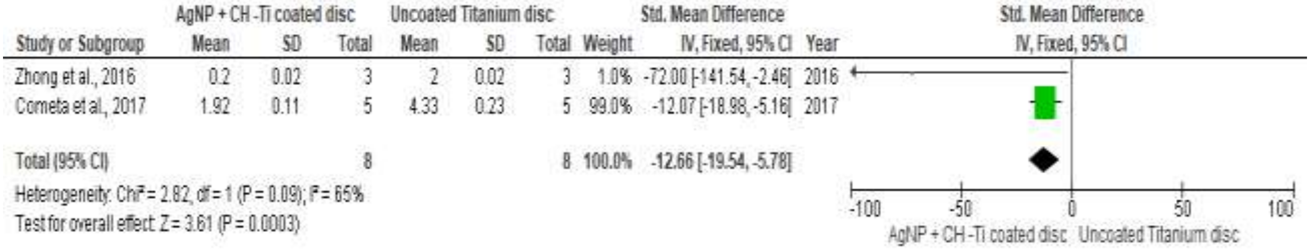

Figure 4: CFU between silver nanoparticles with chitosan coated and uncoated Ti disc

The funnel plot did not show significant asymmetry, indicating absence of publication bias as shown in **Figure 5.**

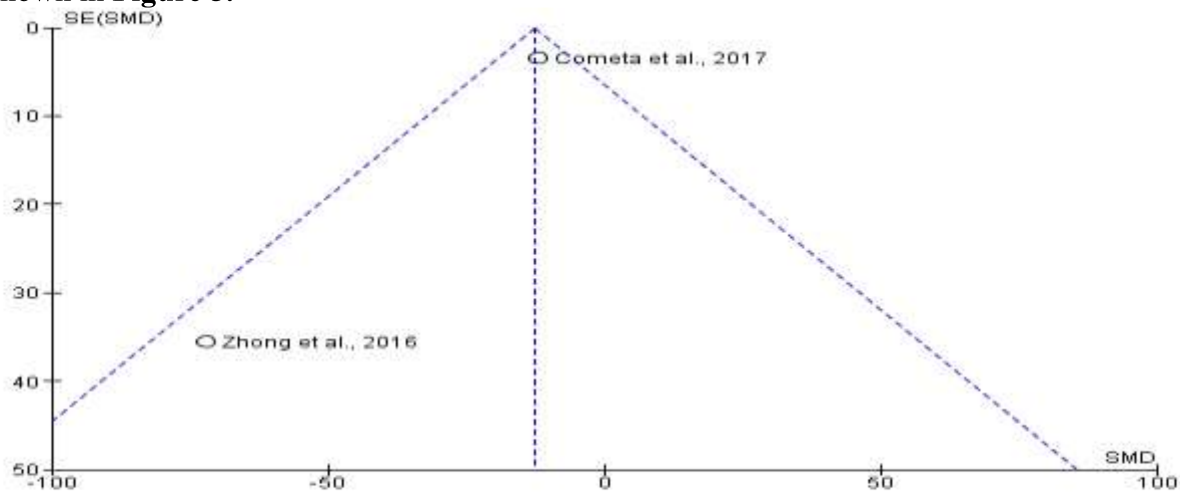

Figure 5: showingBegg's Funnel plot with 95% confidence intervals demonstrating an absence of publication bias.

2. **Cell Viability**

Three studies[21,24]containing data on 12 specimens, of which (*n*=6) specimens were evaluated by silver nanoparticles with chitosan coatingon Ti coated disc and (*n*=6) specimens by uncoated Ti disc for the evaluation of better antimicrobial efficacy in terms of greater cell viability.As shown in **Figure 6.** the SMD is 0.41 (-1.10 – 1.91) and the pooled estimates signifies that greater cell viabilityon an average is 0.41 times more by silver nanoparticles and chitosan coated group(p>0.05).

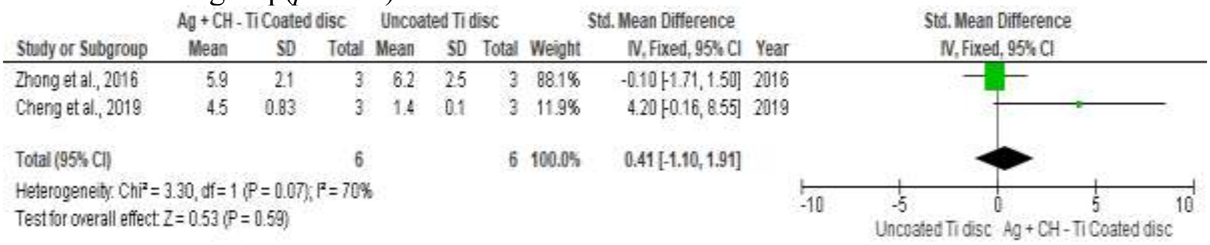

Figure 6: Cell viability between silver nanoparticles with chitosan coated and uncoated Ti disc

The funnel plot did not show significant asymmetry, indicating absence of publication bias as shown in **Figure 7.**

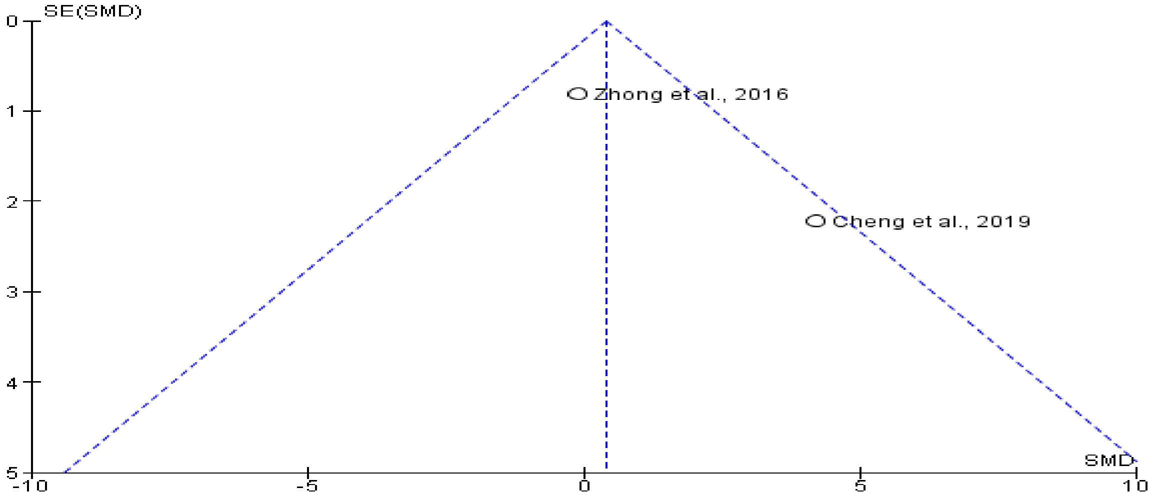

Figure 7: showingBegg's Funnel plot with 95% confidence intervals demonstrating an absence of publication bias.

Discussion

Antimicrobials, have been integrated with the Ti-based materials as the active bactericidal coatings.[12] Among them, silver-based antibacterial agents have long been known and used due to their superior and long-lasting antimicrobial properties against a wide range of microbes.[13]

Sun et al., 2021[25]conducted systematic review of existing literature to evaluate the in -vivo bactericidal outcomes of nanostructures on the Ti implant surface. Databases were searched which yielded five preclinical studies (in vivo) which were included in final review. From the results of the studies, it was found that in vivo bactericidal effect of nanoparticles on Ti implant surface was considerably poor. It was concluded that to overcome these shortcomings and drawbacks, surface coatings with metallic or antibiotic elements are best practical and clinical approaches.

de Oliveira Rigotti et al., 2023[26]conducted systematic review to assess the influence the effect of various antibacterial surface treatment on the cell viability of dental implants. Databases were searched for the studies which reported the results in terms of the antibacterial activity and cytotoxicity on osteoblastic cells of titanium and their alloy when treated superficially. Databases were searched till 2022 which yielded 12 studies included in final review. From the results of the studies, it was found that adding various nano-materials reduced/decreased the chances of bacterial resistance by controlling their adhesion by electrical forces.

Ubaldi et al., 2024[27] conducted a systematic review and meta-analysis to assess and evaluate the antimicrobial efficacy of various photocatalysts applied by various different coating methods on different surfaces. Databases were searched from 2006 till 2023 which yielded 45 articles been included in systematic review. From the pooled data analysis, it was found that TiO_2 (titanium dioxide) coating showed a substantial decrease in bacterial strain (gram positive as well as gram negative) (99.4%). It was concluded that photocatalytic coating provided an alternative approach in decreasing/ reducing the spread of micro-organisms on surfaces.

Tardelli et al., 2024[28] conducted a systematic review to evaluate the chitosan coatings on titanium-based-implants. The relationship between the physical and chemical properties pf polymeric chitosan coatings on titanium implants on antibacterial activity and osteoblast viability was assessed. The surface characterization effects of chitosan coating was assessed according to technique of fabrication, adhesion, roughness, wettability, antibacterial activity and osteoblast viability. From the results of the studies, it was found thatchitosan coatings overall showed good anti-bacterial activity and cytocompatibility.

There have been few systematic reviews and meta-analysis[25-28] published in past but due to presence of data heterogeneity, none of them actually could provide a comprehensive qualitative and quantitative analysis on providing a comparative analysis on the antibacterial efficacy of silver nanoparticles and chitosan coating on titanium surfaces against gram-positive and gram -negative bacteria. According to our knowledge, this is the first systematic review and meta-analysis which assessed and evaluated the the antibacterial efficacy of silver nanoparticles and chitosan coating on titanium surfaces in terms of colony forming units (CFU) for *Staph. Aureus* and cell viability**.**

Databases were searched from January 2000 to September 2024 for in-vitro and comparative analytical studies which evaluated the antibacterial efficacy of silver nanoparticles and chitosan coating on titanium surfaces in terms of colony forming units (CFU) for *Staph. Aureus* and cell viability. Four studies[21-24] were included in for qualitative synthesis and three studies[21,22,24] for meta-analysis. Included studies reported presence of low risk of bias overall. Meta-analysis showed that reduction in CFU of *Staph. aureus*was (SMD-12.66 (-19.54 – 5.78) more by silver nanoparticles and chitosan coated group($p<0.05$) while greater cell viability on Ti surface was (SMD - 0.41 (-1.10 – 1.91) more by silver nanoparticles and chitosan coated group ($p>0.05$).Funnel plot did not show any presence of possible publication bias in meta-analysis.

The adherence to PRISMA guidelines, thorough literature search, and rigorous methodology, including Cochrane risk of bias assessment, underscored the credibility of these systematic reviews. With high overall study quality and minimal bias across the included studies, the evidence base supporting therapeutic recommendations for optimizing the use of silver nanoparticles and chitosan coating on titanium surfaces against infective pathogens is robust and actionable.

However, there were also some limitations. A review of the evidence shows that the literature on antibacterial efficacy of silver nanoparticles and chitosan coating on titanium surfaces is sparse. Even after an unlimited search and eligibility criteria, there were very few studies with qualitative synthesis and quantitative synthesis. Only four studies were included in the final assessment. More in vitro studies or comparative analytical studieswith larger sample size should be conducted to validate the findings of this study.

A systematic review is a transparent and repeatable procedure for identifying, selecting and critically assessing published or unpublished data to address a well-defined research question. Meta-analyses, a statistical analysis that incorporates numerical data from related studies, are frequently paired with systematic reviews. The best evidence is generally regarded as systematic reviews and meta-analyses. However, the calibre of the included studies has an impact on how strong the evidence is. In the present review, sufficient studies with a brief observation period and a known risk of bias were included. As a result, the presently available evidence is sufficient to make therapeutic recommendations in response to the current systematic review's focus question.

Conclusion

From the results of the studies, it was found that antibacterial coating is highly effective in preventing implant associated infections and these coatings showed good anti-bactericidal property by inhibiting surface adhesion of gram positive and gram- negative bacteria with minimal cytotoxicity.

References

1. Shah FA, Trobos M, Thomsen P, Palmquist A. Commercially pure titanium (cp-Ti) versus titanium alloy (Ti6Al4V) materials as bone anchored implants—Is one truly better than the other?. Materials Science and Engineering: C. 2016;62:960-6.
2. Takasaki AA, Aoki A, Mizutani K, Schwarz F, Sculean A, Wang CY, Koshy G, Romanos G, Ishikawa I, Izumi Y. Application of antimicrobial photodynamic therapy in periodontal and peri-implant diseases. Periodontology 2000. 2009;51(1):109-40.
3. Wainwright M. Photodynamic antimicrobial chemotherapy (PACT). The Journal of antimicrobial chemotherapy. 1998;42(1):13-28.

4. Javadpour MM, Juban MM, Lo WC, Bishop SM, Alberty JB, Cowell SM, Becker CL, McLaughlin ML. De novo antimicrobial peptides with low mammalian cell toxicity. Journal of medicinal chemistry. 1996;39(16):3107-13.
5. Zhang Q, Neoh KG, Xu L, Lu S, Kang ET, Mahendran R, Chiong E. Functionalized mesoporous silica nanoparticles with mucoadhesive and sustained drug release properties for potential bladder cancer therapy. Langmuir. 2014;30(21):6151-61.
6. Rai M, Yadav A, Gade A. Silver nanoparticles as a new generation of antimicrobials. Biotechnology advances. 2009;27(1):76-83.
7. Rinaudo M. Chitin and chitosan: properties and applications. Progress in polymer science. 2006;31(7):603-32.
8. Prashanth KH, Tharanathan RN. Chitin/chitosan: modifications and their unlimited application potential—an overview. Trends in food science & technology. 2007; 18(3):117-31.
9. John J, Saranathan R, Adigopula LN, Thamodharan V, Singh SP, Lakshmi TP, CharanTej MA, Rao RS, Krishna R, Rao HS, Prashanth K. The quorum sensing molecule N-acyl homoserine lactone produced by Acinetobacter baumannii displays antibacterial and anticancer properties. Biofouling. 2016;32(9):1029-47.
10. Reck M, Tomasch J, Wagner-Döbler I. The alternative sigma factor SigX controls bacteriocin synthesis and competence, the two-quorum sensing regulated traits in Streptococcus mutans. PLoS genetics. 2015;11(7):20-33.
11. Socransky SS, Haffajee AD. Dental biofilms: difficult therapeutic targets. Periodontology 2000. 2002;28(1):12-55.
12. Tonetti MS, Schmid J. Pathogenesis of implant failures. Periodontology 2000. 1994;4(1):127-38.
13. Größner-Schreiber B, Griepentrog M, Haustein I, Müller WD, Briedigkeit H, Göbel UB, Lange KP. Plaque formation on surface modified dental implants. Clinical oral implants research. 2001;12(6):543-51.
14. Durgesh BH, Basavarajappa S, Ramakrishnaiah R, Al Kheraif AA, Divakar DD. A review on microbiological cause of periodontal disease: disease and treatment. Reviews in Medical Microbiology. 2015;26(2):53-8.
15. Khoushab F, Yamabhai M. Chitin research revisited. Marine drugs. 2010;8(7):1988-2012.
16. Moher D, Liberati A, Tetzlaff J, Altman DG, PRISMA Group. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. Annals of internal medicine. 2009;151(4):264-9.
17. Corbett MS, Higgins JP, Woolacott NF. Assessing baseline imbalance in randomised trials: implications for the Cochrane risk of bias tool. Research Synthesis Methods. 2014;5(1):79-85.
18. DerSimonian R, Laird N. Meta-analysis in clinical trials revisited. Contemporary clinical trials. 2015;45(2):139-45.
19. Higgins JP, Thompson SG. Quantifying heterogeneity in a meta-analysis. Statistics in medicine. 2002;21(11):1539-58.
20. Sterne JA, Becker BJ, Egger M. The funnel Plot. Publication bias in meta-analysis: Prevention, assessment and adjustments. 2005;10(3):75-98.
21. Zhong X, Song Y, Yang P, Wang Y, Jiang S, Zhang X, Li C. Titanium surface priming with phase-transited lysozyme to establish a silver nanoparticle-loaded chitosan/hyaluronic acid antibacterial multilayer via layer-by-layer self-assembly. PLoS One. 2016;11(1):46-57.
22. Cometa S, Bonifacio MA, Baruzzi F, de Candia S, Giangregorio MM, Giannossa LC, Dicarlo M, Mattioli-Belmonte M, Sabbatini L, De Giglio E. Silver-loaded chitosan coating as an integrated approach to face titanium implant-associated infections:

Analytical characterization and biological activity. Analytical and bioanalytical chemistry. 2017;409(12):7211-21.

23. Divakar DD, Jastaniyah NT, Altamimi HG, Alnakhli YO, Alkheraif AA, Haleem S. Enhanced antimicrobial activity of naturally derived bioactive molecule chitosan conjugated silver nanoparticle against dental implant pathogens. International journal of biological macromolecules. 2018;108(17):790-7.
24. Cheng YF, Zhang JY, Wang YB, Li CM, Lu ZS, Hu XF, Xu LQ. Deposition of catechol-functionalized chitosan and silver nanoparticles on biomedical titanium surfaces for antibacterial application. Materials Science and Engineering: C. 2019;98(10):649-56.
25. Sun Y, Yang Y, Jiang W, Bai H, Liu H, Wang J. In vivo antibacterial efficacy of nanopatterns on titanium implant surface: a systematic review of the literature. Antibiotics. 2021;10(12):1524.
26. de Oliveira Rigotti RL, Tardelli JD, Dos Reis AC. Influence of antibacterial surface treatment on dental implants on cell viability: A systematic review. Heliyon. 2023;9(3):12-23.
27. Ubaldi F, Valeriani F, Volpini V, Lofrano G, Romano Spica V. Antimicrobial Activity of Photocatalytic Coatings on Surfaces: A Systematic Review and Meta-Analysis. Coatings. 2024;14(1):92-102.
28. Tardelli JD, Schiavon MA, Dos Reis AC. Chitosan coatings on titanium-based implants-From development to characterization and behaviour: A systematic review. Carbohydrate Polymers. 2024;15(8):12-24.