Review Article

Effectiveness of Biosynthesized Silver Nano particles on Oral pathogens – A Systematic Review

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ABSTRACT

Background: Silver nanoparticles (AgNPs) synthesized via biosynthesis have physicochemical properties and broad-spectrum biological activities and possess remarkable antimicrobial, anti-inflammatory, and wound-healing properties, making them useful in healthcare, pharmaceuticals, and dentistry. This systematic review aims to evaluate the effectiveness of biosynthesized silver AgNPs in promoting oral health by assessing their antimicrobial, antibiofilm, and biocompatibility properties in in vitro studies. **Materials and Methods:** A comprehensive literature search was conducted using PubMed and Science Direct databases up to May 2025. In vitro studies using biosynthesized AgNPs targeting oral pathogens were included. Only studies reporting measurable outcomes such as minimum inhibitory concentration (MIC), zone of inhibition (ZOI), biofilm reduction, or cytotoxicity were considered. The quality of the included studies was assessed using the QUIN (Quality Assessment Tool for In Vitro Studies). **Results:** Out of 1731 identified articles, 14 in vitro studies met the inclusion criteria. The biosynthesized AgNPs, showed significant antimicrobial effects against key oral pathogens. Reported MIC values ranged from 10.6 µg/ml to 50 µg/ml, and ZOI measurements varied between 9 mm and 29 mm. Few studies reported favorable cytotoxicity outcomes. However, quality assessment revealed that 13 out of 14 studies exhibited a high risk of bias due to methodological limitations. **Conclusion:** Biosynthesized AgNPs demonstrate promising antimicrobial potential and could serve as eco-friendly alternatives to conventional chemical-based oral antimicrobials.

Key words: Silver nanoparticles, biofilm, biocompatibility, nanoparticles.

Biosynthesis has emerged as a sustainable alternative to chemical and physical synthesis methods for producing nanoparticles, where microorganisms, plants, and other biological entities act as reducing and stabilizing agents, enabling the synthesis of nanoparticles under mild and non-toxic conditions, minimizing the use of hazardous chemicals, ensuring environmental safety and biocompatibility [1, 2, 3]. Silver nanoparticles (AgNPs) synthesized via this method have garnered significant attention due to their unique physicochemical properties and broad-spectrum biological activities [4]. They are highly versatile and possess remarkable antimicrobial, anti-inflammatory, and wound-healing properties, making them useful in healthcare, pharmaceuticals, and dentistry. In dentistry, AgNPs have been extensively studied for their potential to combat oral pathogens, promote tissue regeneration, and enhance the efficacy of dental materials [5, 6].

When introduced into the oral tissues or dental materials, AgNPs release silver ions, which interact with bacterial cell membranes, leading to structural damage and the disruption of vital cellular functions [7]. Their interaction with DNA and proteins inhibits bacterial replication and metabolism [8]. It also modulates inflammatory responses, enhancing tissue healing while minimizing microbial colonization [9]. Their small size and large surface area facilitate better tissue absorbance and targeted action, making them highly effective in oral health challenges such as dental caries, periodontitis, and periimplantitis [10].

Unlike conventional chemical-based mouth rinses, which are often limited by cytotoxicity, short action time, and growing antimicrobial resistance, biosynthesized AgNPs are a superior alternative due to their green synthesis, enhanced biocompatibility, and broad-spectrum antimicrobial activity [11].

This review specifically focuses on the role of biosynthesized AgNPs in combating oral pathogens, preventing biofilm formation, ensuring biocompatibility, safety, and therapeutic potential in dental applications by critically appraising current in vitro studies, laying the foundation for future translational and clinical research. This targeted approach is expected to guide innovation in

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nanodentistry and promote more effective, natural, and patient-friendly alternatives in oral healthcare.

METHODS

The preferred reporting system will follow the PRISMA-P (Preferred Reporting Items for Systematic Reviews and Meta-Analysis) guidelines [12]. This protocol was registered with the International Prospective Register of Systematic Reviews (PROSPERO): CRD42024599265.

The studies published in English and available as full-text articles in peer-reviewed journals published between 2018 and 2023 were included which were in vitro experimental studies evaluating biosynthesized/green-synthesized (e.g., plant extracts, microbial agents, natural reducing agents) AgNPs. Those studies that focus on oral health, including antimicrobial or antibiofilm activity on oral pathogens, or incorporation into dental materials (e.g., mouthwashes, denture liners, composites) were selected. The studies reporting quantitative or measurable outcomes such as Zone of inhibition (ZOI), Minimum inhibitory concentration (MIC), Minimum bactericidal concentration (MBC), Cytotoxicity or biocompatibility were only included. These inclusion criteria ensured that the review is built upon robust, experimentally validated findings relevant to the potential clinical application of biosynthesized silver nanoparticles in dentistry, as an alternative to conventional chemical mouth rinses and antimicrobial agents.

The studies in which chemically or physically synthesized AgNPs without a biological component were excluded. The Non-oral focus studies that are not relevant to oral pathogens or dental applications, type-Review articles, editorials, case reports, conference abstracts, letters without original experimental data, or incomplete data, and those studies that did not report outcome measures or lacked sufficient methodological detail were also excluded.

Literature Search and Search Strategy

A comprehensive literature search was conducted using electronic databases, including PubMed and Science Direct up to May 2025. The search strategy focused on retrieving studies evaluating the effectiveness of biosynthesized AgNPs in oral health applications. Keywords employed in the search included: [biosynthesized silver nanoparticles], [green synthesis], [silver nanoparticles], [oral pathogens], [AgNPs in dentistry], and [biogenic nanoparticles]. Boolean operators [AND] and [OR] were used to combine keywords, and search terms were modified appropriately for each database.

Types of participants/samples

In vitro research was included; preparation of samples of materials was reported.

Study Screening and Selection

The articles collected were organized using Mendeley Reference Manager. Following the initial compilation, duplicate records were identified and removed. Two independent reviewers screened the remaining titles and abstracts based on the predefined inclusion and exclusion criteria. Each manuscript was analyzed for methodological quality according to a prepared checklist based on the Checklist for Reporting In-vitro Studies (CRIS) Guidelines [13]. Any disagreements during the screening process were resolved through discussion or consultation with a third reviewer when necessary. Full-text articles were assessed to determine final eligibility, and studies not meeting the criteria were excluded for documented reasons. The screening and selection process adhered to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 guidelines to ensure transparency and reproducibility (Figure 1).

Data Extraction

All the studies included were thoroughly reviewed, and key data were systematically extracted and documented in a Microsoft Excel spreadsheet. For each study, the following information was recorded: author(s), publication year, country of origin, plant or biological source of AgNPs, characterization techniques, particle size and shape, test organisms, methods used, control substances, and outcomes related to antimicrobial and cytotoxic effects. This structured synthesis enabled cross-comparison and evaluation of trends in biosynthesis methods, efficacy outcomes, and potential translational applications in oral care.

Risk of Bias Assessment

The risk of bias was determined for each included publication using the Quality Assessment Tool for In Vitro Studies (QUIN), which consists of 12 criteria [14]. Each criterion is scored from 0 to 2 (0: not specified; 1: inadequately specified; 2: adequately specified; NA: not applicable). The final score is determined by the following formula: final score = (total score × 100)/ (2 × number of criteria applicable). According to the final score, the article is graded as having low, medium, or high risk of bias (>70%, low risk of bias; 50–70%, medium risk of bias.

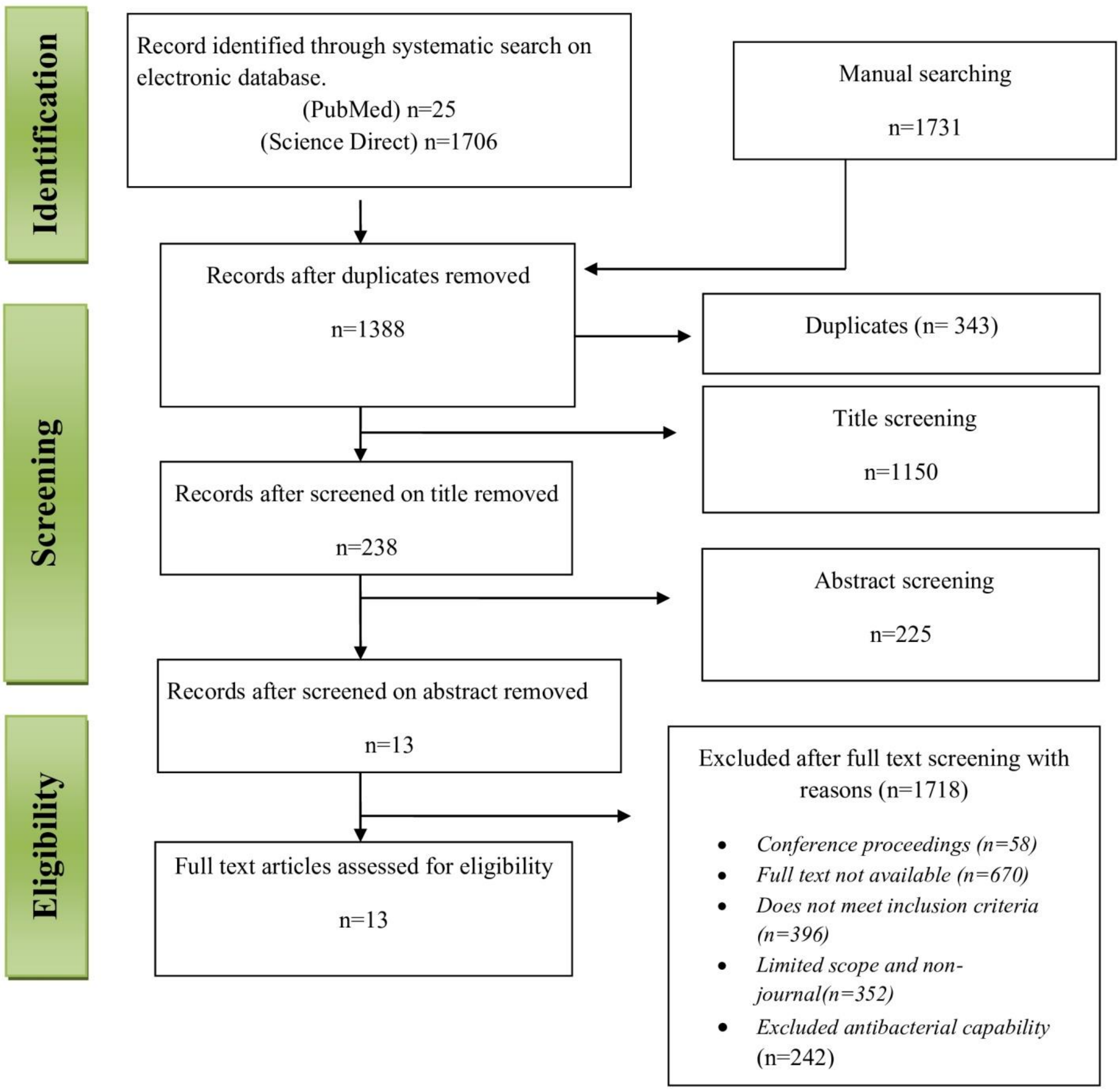

Figure No.1: PRISMA flow diagram of the screening and selection process.

Table No.1: Details of the included publications with their methodology in the included articles.

Sl no.	Author, Year, Place [citation]	Plant	Component	TEM	XRD	SEM	FTIR	Wavelength	Crystallinity	Shape	Size	Method Used	Organisms	Control	Cont	Outc	PF	LOE
1	Hanaa Ghabban, 2022, Saudi Arabia [15]	*Astragalus spinosus*	Fruit	Yes	Yes	Yes	Yes	Not specified	Yes	Spherical	5–60 nm	MIC, MBC	S. mutans, A. viscosus	CHX	10.6 µg/ml	13.3 µg/ml	25.5 %	5c
2	Enas Tawfik Enan,2021, Saudi Arabia [16]	*Cupressus macrocarpa*	Leaf	Yes	Yes	-	Yes	Not mentioned	Yes	Not mentioned	-	ZOI, Biofilm	S. mutans, S. aureus	CHX	29 mm	15 mm	92.2 %	5c
3	Kiran R.	*Fusarium*	Fungi	-	-	Yes	-	-	Yes	-	-	MIC,	P. gingivalis, B. pumilus, E.	CH	20	30 µg/m	Similar to	5c

	Halkai, 2018, India [17]	*semitectum*										Biofilm	faecalis	X	µg/ml	l	CHX	
4	Muhammad Ramzan, 2022, Pakistan [18]	*Zingiber zerumbet*	Rhizome	Yes	Yes	Yes	Yes	400–500 nm	Yes	Spherical	-	MIC, ZOI	S. aureus, S. mutans, E. faecalis	CHX	50 µg/ml	ZOI present	-	5c
5	Erika Jardón-Romero, 2022, Mexico [19]	*Syzygium aromaticum* [Clove]	Flower bud	Yes	Yes	Yes	-	400–500 nm	Yes	Quasi-spherical	4–16 nm	MIC, MEC	General oral microbes	CHX	2–4 mm	Effective	-	5c
6	Wagner Bernardo, 2022, Brazil [20]	*Syzygium cumini*	Leaf	Yes	Yes	Yes	Yes	-	Yes	Various	31.2–250	MIC, MBC, Biofilm	Multiple oral pathogens	CHX	125–8000 µg/ml	Effective	-	5c
7	Shanmugam Rajeshkumar, 2021, India [21]	*Cissus arnottiana*	Leaf, Stem, and Roots	Yes	Yes	Yes	Yes	-	Yes	Spherical	-	ZOI	S. aureus, C. albicans, E. faecalis	CHX	High Gram+	Effective	-	5c
8	Shatha S. ALHarthi,2021, Saudi Arabia [22]	*Myrrh*	Resin	-	-	Yes	Yes	-	Yes	-	-	ZOI	P. gingivalis	CHX	16–18 mm	Effective	-	5c
9	Wagner Boriollo, 2021, Brazil [23]	*Syzygium cumini*	Flower and Seed	Yes	Yes	Yes	Yes	-				ZOE,MMC,MIC	*A. naeslundii, C. albicans,F. nucleatum, S. aureus, S. epidermidis, S. mutans, S. oralis and V. dispar*					5c
10	Widadh, Ismai, 2023, South Africa [24]	*B. lanuginose, H. cymosum, and S. crenata*	Flowers	Yes	Yes	-	Yes	-	Yes	-	-	MIC ZOI, Cytotoxicity	*Candida albicans*	CHX	<0.015 mg/ml	Effective	-	5c
11	Omnia Ahmed, 2022, South Africa [25]	*Gum Arabic*	Gum	Yes	Yes	Yes	Yes	-	Yes	Not specified	-	MIC, ZOI	Oral pathogens	CHX	Not mentioned	Effective	-	5c
12	Indumathy Pandiyan, 2022, India [26]	*O. tenuiflorum + S. rebaudiana*	Leaf	-	-	Yes	Yes	-	Yes	-	-	ZOI, Cytotoxicity	Oral pathogens	CHX	Not specified	Effecrtive	-	5c
13	M. Rangeela, 2021, India [27]	*Ficus benghalensis*	Leaf	-	-	-	Yes	-	Yes	-	-	ZOI	S. aureus, S. mutans, E. faecalis	CHX	9–21 mm	Effective	-	5c

*****Transmission Electron Microscopy (TEM), Scanning Electron Microscopy (SEM), Fourier Transform Infrared Spectroscopy (FTIR), X-ray Diffraction (XRD), and chlorhexidine (CHX).

Table No.2: JBI Levels of evidence [28]

Level 1: Experimental designs	
Level 1a	Systematic review of Randomized Controlled Trials (RCTs)
Level 1b	Systematic review of RCTs and other study designs Level
Level 1c	RCT Level
Level 1d	Pseudo-RCTs
Level 5: Expert Opinion and Bench Research Level	
Level 5a	Systematic review of expert opinion Level
Level 5b	Expert consensus Level
Level 5c	Bench research/ single expert opinion

Table No.3: Quality Assessment of In Vitro Studies using the QUIN Tool

0–2 (0: not specified; 1: inadequately specified; 2: adequately specified; NA: not applicable). Final score = (total score × 100)/ (2 × number of criteria applicable). According to the final score, the article is graded low, medium, or high risk of bias (>70%, low risk of bias; 50–70%, medium risk of bias.

Not specified (red)
Inadequately specified (yellow)
Adequately specified (green)

First Author	Aims/Objectives	Sample Size Calculation	Explanation of Sampling Technique	Details of Control Group	Explanation of Methodology	Operator Details	Randomization	Method of Measurement of Outcome	Outcome Assessor Details	Blinding	Statistical Analysis	Presentation of Results	score	Risk of Bias
Hanaa Ghabban [15]	2	0	0	1	2	0	0	2	0	0	2	2	45.83 %	High
Enas Tawfik Enan [16]	2	1	1	2	2	0	0	2	0	0	2	2	58.33 %	Medium
Kiran R [17]	2	0	0	1	2	0	0	2	0	0	2	2	45.83 %	High
Muhammad Ramzan [18]	2	0	0	1	2	0	0	2	0	0	0	2	37.5 %	High
Erika Alejandra [19]	2	0	0	1	2	0	0	2	0	0	0	2	37.5 %	High
Wagner Luis [20]	2	0	0	1	2	0	0	2	0	0	2	2	45.83 %	High
Shanmugam Rajeshkumar [21]	2	0	0	1	2	0	0	2	0	0	0	2	37.5 %	High
Shatha Subhi ALHarthi [22]	2	0	0	0	2	0	0	2	0	0	0	2	37.5 %	High
Wagner Luis [23]	2	0	0	0	2	0	0	2	0	0	0	2	37.5 %	High
Widadh Klein [24]	2	0	0	0	2	0	0	2	0	0	2	2	41.67 %	High
Omnia Ahmed [25]	2	0	0	0	2	0	0	2	0	0	2	2	41.67 %	High
M. Rangeela A [26]	2	0	0	0	2	0	0	2	0	0	0	2	33.33 %	High
Indumathy Pandiyan [27]	2	0	0	0	2	0	0	2	0	0	0	2	33.33 %	High

RESULTS

A total of 1731 articles were initially identified through the comprehensive database search conducted in PubMed, Science Direct, and manual hand-searching. After excluding 343 duplicate records, 1150 and 225 articles were screened by their titles and abstracts. Following the application of the predefined inclusion and exclusion criteria, 1718 studies were excluded for reasons such as: use of chemically synthesized silver nanoparticles (n=396), focus on non-oral applications (n=352), review articles, conference abstracts, or editorials without original data (n=58), and studies lacking measurable outcomes relevant to antimicrobial or biocompatibility assessment (n=242). Consequently, 1542 full-text articles were assessed in detail. Out of these, 670 articles were excluded primarily due to insufficient methodological details, absence of oral relevance, or incomplete outcome reporting. Finally, 13 in vitro studies met the eligibility criteria and were included in this systematic review. The detailed screening and selection process is illustrated in Figure 1**.**

The selected studies focused on evaluating the antimicrobial, antibiofilm, and biocompatibility effects of biosynthesized AgNPs against oral pathogens. The included studies demonstrated considerable diversity in the biological sources used for nanoparticle synthesis, ranging from plants such as Astragalus spinosus, Cupressus macrocarpa, and Syzygium aromaticum to microbial or gum-based sources. Most studies employed characterization techniques such as TEM, SEM, FTIR, and XRD to confirm nanoparticle formation, size, shape, and crystallinity.

The antimicrobial outcomes, detailed in Table 1, revealed that the biosynthesized AgNPs exhibited promising antimicrobial activity against key oral pathogens, including Streptococcus mutans, Porphyromonas gingivalis, Staphylococcus aureus, *and* Enterococcus faecalis. The reported ZOI ranged from 9 mm to 29 mm, while the MIC values were as low as 10.6 µg /ml in certain studies. Several studies also assessed biofilm inhibition, with biosynthesized AgNPs showing comparable or superior results to conventional CHX in vitro. The particle size of the AgNPs ranged broadly between 4 nm and 60 nm, with spherical or quasi-spherical shapes most reported, factors that may contribute to their enhanced bioactivity. A small number of studies evaluated cytotoxicity, indicating favorable biocompatibility at effective antimicrobial concentrations.

The quality of the included studies was assessed using the QUIN (Quality Assessment Tool for In Vitro Studies). The outcomes of this evaluation are summarized in Table 2. Most studies (13 out of 14) were classified as having a high risk of bias, with scores ranging from 33.33% to 45.83%, primarily due to the absence of sample size calculations, inadequate reporting of randomization, blinding, and insufficient statistical analysis. Only one study [16] achieved a medium risk of bias with a score of 58.33%. None of the included studies met the threshold for low risk of bias (>70%). This finding highlights methodological weaknesses across the current body of in vitro research on biosynthesized AgNPs.

The inference drawn from the QUIN-based risk of bias assessment shows the need for improved study designs in future research. Standardizing experimental protocols, ensuring methodological transparency, and including proper statistical validation are essential for generating more robust and clinically translatable evidence. Nevertheless, despite the identified limitations, the reviewed studies collectively support the antimicrobial efficacy of biosynthesized silver nanoparticles and suggest their potential role as a natural, eco-friendly alternative in oral health management.

DISCUSSION

The present systematic review supports the antimicrobial potential of biosynthesized AgNPs in oral healthcare. Across the 13 included studies, AgNPs demonstrated significant activity against a broad spectrum of oral pathogens, including Streptococcus mutans, Porphyromonas gingivalis, Staphylococcus aureus, and Enterococcus faecalis. The observed antimicrobial effects were often comparable to or, sometimes, superior to conventional antimicrobial agents such as CHX.

The diversity in the biological sources used for nanoparticle synthesis. including plant leaves, fruits, gums, and microbial agents, underscores the versatility of green synthesis techniques. This eco-friendly approach not only reduces the need for hazardous chemicals but also imparts bioactive properties through phytoconstituents that may synergistically enhance antimicrobial effects.

Despite these promising outcomes, this review also identified substantial methodological limitations in the current research landscape. The quality assessment using the QUIN tool revealed that most studies had a high risk of bias due to poor reporting of sample size calculations, randomization, blinding, and statistical rigor. Additionally, while antimicrobial effects were the primary focus, fewer studies evaluated the cytotoxicity or long-term biocompatibility of biosynthesized AgNPs, which is essential for safe clinical translation.

The findings from this review are consistent with previous research demonstrating that biosynthesized AgNPs possess potent antimicrobial properties. Khorrami et al. emphasized the role of biological synthesis in producing nanoparticles with enhanced bioactivity and reduced toxicity compared to chemically synthesized counterparts [29]. Similarly, Wypij et al. reported that plant-mediated AgNPs exhibit superior antimicrobial efficacy due to the presence of bioactive phytochemicals that act synergistically with silver ions [30]. These observations align with the present review's findings, where plant-based sources such as Astragalus spinosus, Cupressus macrocarpa, and Syzygium aromaticum yielded AgNPs with strong antimicrobial activity against oral pathogens.

The antimicrobial mechanism of AgNPs has been extensively studied. Dakal et al. described how silver ions disrupt bacterial cell membranes, interfere with DNA replication, and inhibit protein synthesis, leading to bacterial death [31]. Le Ouay and Stellacci demonstrated that the shape and size of nanoparticles significantly influence their antibacterial efficacy, with smaller, spherical nanoparticles showing greater activity [32]. The current review corroborates these findings, as the included studies predominantly reported spherical AgNPs in the 4–60 nm range, which exhibited effective antimicrobial outcomes.

In the context of oral health, several studies have highlighted the potential of AgNPs in combating dental caries and periodontal diseases. Hernández-Sierra et al. demonstrated that AgNPs effectively inhibit the growth of Streptococcus mutans, the primary causative agent of dental caries [33]. Besinis et al. reported that AgNPs incorporated into dental materials significantly reduced bacterial colonization and biofilm formation on tooth surfaces [34]. Furthermore, Hernández-Gómora et al. observed that green-synthesized AgNPs showed enhanced antimicrobial activity against oral pathogens with minimal cytotoxic effects on human cells [35].

About the biocompatibility of biosynthesized AgNPs, Vazquez-Muñoz et al. noted that while AgNPs possess excellent antimicrobial properties, their toxicity profile must be carefully evaluated to ensure safe use in medical applications [36]. A limited number of studies in this review assessed cytotoxicity, with results indicating favorable biocompatibility at effective antimicrobial concentrations. However, Rónavári et al. cautioned that long-term exposure and high concentrations of AgNPs could pose potential health risks, emphasizing the need for comprehensive safety assessments [37].

Siddiqi et al highlighted the importance of establishing uniform guidelines for nanoparticle characterization and antimicrobial testing [38]. This review also identified a lack of standardized synthesis protocols and inconsistent outcome measures across studies, making it difficult to draw definitive conclusions about the optimal formulation, concentration, or delivery method for AgNPs in dental practice. Future research should prioritize well-designed, standardized in vitro and in vivo studies with adequate controls, transparent reporting, and comprehensive safety assessments. Exploring synergistic effects with natural bioactives, developing controlled-release systems, and evaluating patient-centered outcomes should be the critical steps toward clinical implementation.

CONCLUSION

This systematic review showed that biosynthesized AgNPs exhibit potent antimicrobial properties against oral pathogens, positioning them as promising eco-friendly alternatives to conventional chemical-based oral antimicrobials. However, the high risk of bias in most included studies necessitates cautious interpretation and the need for rigorous, standardized research and clinical trials to validate their safety and effectiveness in real-world dental applications

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