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Synergistic Anticancer and Antibacterial Effects of Withania somnifera (L.) Dunal Extract-Loaded Chitosan NPs against Staphylococcus Aureus-Associated MCF-7 Breast Cancer Cells

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Abstract

Background and objective: Breast cancer remains one of the most prevalent cancers among women, often co-occurring with bacterial infections such as those caused by *Staphylococcus aureus*. This co-infection can exacerbate disease progression, complicate treatment, and negatively impact patient outcomes. This study explores the synthesis and therapeutic potential of Withania Somnifera Extract-Loaded Chitosan Nanoparticles (WSE-CSNPs) as a dual-action therapeutic targeting both antibacterial and anticancer pathways.

Methods: WSE-CSNPs were synthesized using the ionic gelation method and characterized for particle size, zeta potential, morphology, and encapsulation efficiency, antibacterial by agar well diffusion, and anticancer effects by MTT assay and flow cytometry.

Results: The antibacterial efficacy against *S. aureus* revealed enhanced activity compared to the extract alone, with a significant inhibition of bacterial growth. Anticancer effects were assessed on MCF-7 breast cancer cells using MTT assays and flow cytometry, demonstrating a dose-dependent reduction in cell viability. Additionally, WSE-CSNPs promoted apoptosis through increased caspase-3 activation and significantly inhibited cancer cell invasion and migration. Key markers of cancer inflammation, such as TNF-alpha, IL-1β, and COX-2, were downregulated, highlighting the potential of WSE-CSNPs to modulate the tumor microenvironment.

Conclusion: These results suggest that WSE-CSNPs may serve as a promising therapeutic strategy for *S. aureus*-associated breast cancer by simultaneously targeting bacterial infection and cancer cell proliferation.

Keywords: Breast cancer; *Staphylococcus aureus; Withania somnifera;* Chitosan nanoparticles; Apoptosis; Dual-action therapy.

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Introduction

Breast cancer is the most frequently diagnosed cancer among women worldwide and remains a leading cause of cancer-related mortality [1] . Globally, it affects millions of women, representing a significant public health concern due to its high incidence and mortality rates. The disease is characterized by the uncontrolled proliferation of malignant cells in breast tissue, which can lead to local invasion and distant metastasis [2]. Despite advances in screening and treatment, the survival rates for breast cancer patients are often compromised by several factors, including late diagnosis, resistance to chemotherapy, and the occurrence of comorbidities that can complicate treatment outcomes. Among these comorbidities, bacterial infections are emerging as a critical factor that can negatively impact the prognosis and treatment efficacy in breast cancer patients [3].

One major obstacle to the successful treatment of breast cancer is the rise in bacterial infections, especially those brought on by *Staphylococcus aureus*. Gram-positive *S. aureus* bacteria are responsible for a variety of infections, ranging from minor skin infections to more serious illnesses like sepsis [4]. *S. aureus* could foster an inflammatory environment that aids in the growth of breast cancerous tumors. It could enter tumor tissues, cause long-term inflammation, and alter the immune system, resulting in a microenvironment that promotes tumor growth and resistance to standard treatments. Due to the release of pro-inflammatory cytokines, reactive oxygen species, and other mediators that promote cell proliferation and survival, chronic inflammation has been associated with the progression of cancer. The dual challenge of fighting both the infection and the cancer necessitate therapeutic approaches that are effective against both the microbial agents and the cancer cells, which is not always accomplished by traditional treatments like chemotherapy [3].

Chemotherapy, while effective in targeting rapidly dividing cancer cells, often fails to address concurrent infections such as those caused by *S. aureus*. Chemotherapeutic agents primarily target cancer cells through mechanisms like DNA damage and inhibition of cell division [5]. However, they do not possess inherent antibacterial properties and can sometimes compromise the immune system, making patients more susceptible to bacterial infections [6]. Additionally, the side effects associated with chemotherapy, such as gastrointestinal disturbances, neutropenia, and systemic toxicity, can further weaken the body's defenses against infections [7]. This underscores the need for an integrated therapeutic strategy that can simultaneously target cancer cells and manage bacterial infections, thereby improving the overall outcomes for breast cancer patients.

Withania somnifera (Ashwagandha), an adaptogenic herb with a rich history in traditional medicine, has garnered attention for its potential to address this dual challenge. Known for its broad range of pharmacological activities, *W. somnifera* has been traditionally used for its immunomodulatory, anticancer, and antibacterial properties [8]. The roots and leaves of *W. somnifera* contain bioactive compounds such as withanolides, alkaloids, and steroidal lactones, which contribute to its therapeutic effects [9]. The plant is widely distributed in India, Nigeria, North Africa, and other parts of the world with mild temperatures. Previous studies have highlighted the ability of *W. somnifera* to inhibit cancer cell proliferation, induce apoptosis, and modulate signaling pathways that are critical for cancer cell survival [10]. Furthermore, despite its strong pharmacological qualities, the clinical use of *W. somnifera* is constrained by issues like low bioavailability, quick degradation, and limited stability of its bioactive compounds in systemic circulation. Moreover, its antibacterial efficacy against a variety of bacterial strains, including *S. aureus*, makes it a promising candidate for treating infections linked to cancer [11].

Using chitosan-based nanoparticles, nanotechnology presents a possible way to overcome these constraints. Because it may encapsulate bioactive substances, improve their stability, and enable controlled release, chitosan- a biocompatible and biodegradable polymer derived from chitin- is frequently employed for drug delivery [12]. By shielding the active ingredients from deterioration and improving their absorption at the target site, the formulation of *W. Somnifera* Extract-Loaded Chitosan Nanoparticles (WSE-CSNPs) might greatly increase the bioavailability and therapeutic potential of *W. somnifera*. It has been demonstrated that chitosan nanoparticles increase the solubility of hydrophobic substances, accelerate cellular uptake, and allow for targeted administration to cancer cells via increased Permeability and Retention (EPR) effects [13]. Because of these characteristics, chitosan is the perfect vehicle for administering *W. somnifera* extracts in a manner that optimizes their anticancer and antibacterial efficacy.

The present study aims to formulate WSE-CSNPs and evaluate their synergistic effects against *S. aureus*-associated breast cancer, providing a novel therapeutic approach that addresses both the cancer and the bacterial infection. The study hypothesizes that the encapsulation of *W. somnifera* extracts within chitosan nanoparticles will enhance their efficacy, allowing for a more effective reduction of tumor growth while simultaneously inhibiting bacterial proliferation. By combining the anticancer and antibacterial properties of *W. somnifera* with the advantages of nanotechnology, this approach has the potential to offer a more comprehensive treatment strategy for patients suffering from breast cancer complicated by bacterial infections. The findings of this study could pave the way for the development of multifunctional therapeutic agents that can address the complexities of cancer treatment in the presence of infectious agents.

Materials and methods

Materials

Chitosan (medium molecular weight), and sodium Tripolyphosphate (TPP) were obtained from Sigma-Aldrich. Methanol, acetic acid, and all other solvents were of analytical grade. MCF-7 human breast cancer cell line, nutrient broth, agar, cultural plates, and *Staphylococcus aureus* strains were procured from a certified biological repository. MTT assay kits and Annexin V/PI kits for apoptosis analysis were purchased from ThermoFisher Scientific (USA).

Methods

Collection and identification of plant materials

Fresh roots of *W. somnifera* were collected from a forest in Fori, Maiduguri in the early morning hours. It was authenticated at the Department of Pharmacognosy by a taxonomist Dr. C.A. Ukwubile. A voucher specimen number UMM/FPH/SON/018 was deposited for the plant at the herbarium of the department. The roots were shade-dried until constant weight was obtained.

Preparation of *Withania somnifera* extract

The dried roots of *Withania somnifera* were ground into a fine powder. About 800 grams of the powder was extracted using methanol in a Soxhlet apparatus for 24 hours. The methanolic extract was filtered and concentrated under reduced pressure using a rotary evaporator. The concentrated extract weighing 78.42 g was stored in refrigerator at 4°C for further use.

Synthesis of WSE-CSNPs

WSE-CSNPs were synthesized using the ionic gelation method. Briefly, chitosan (0.2% w/v) was dissolved in 1% acetic acid and stirred until a clear solution was obtained. The methanolic extract of *W. somnifera* (0.1% w/v) was added to the chitosan solution. Sodium tripolyphosphate (0.1% w/v) was added dropwise under continuous stirring at room temperature to cross-link the chitosan and form nanoparticles. The mixture was stirred for 2 hours to ensure proper formation of nanoparticles. The resultant nanoparticles were collected by centrifugation at 5,000 rpm for 15 minutes, washed with distilled water, and freeze-dried [13].

Characterization of WSE-CSNPs

Particle size and zeta potential: The size and zeta potential of the nanoparticles were measured using a Zetasizer Nano ZS (Malvern instruments). Measurements were performed in triplicate [13].

Morphology: The Phenom World Scanning Electron Microscopy (SEM) was used to observe the surface morphology of WSE-CSNPs [13].

Encapsulation efficiency: The Encapsulation Efficiency (EE%) of *W. somnifera* in chitosan nanoparticles was calculated using UV-visible spectrophotometry at 270 nm [13].

$$\text{Encapsulation efficiency } (\%) = \frac{\text{Amount of Extract Encapsulated}}{Total\ amount\ of\ extract\ added} x\ 100$$

Antibacterial activity

The agar well diffusion method was used to evaluate WSE-CSNPs' antibacterial efficacy against S. aureus. On nutrient agar plates, a bacterial suspension (0.5 McFarland standard) was applied, and a sterile cork borer was used to make wells. Following the addition of WSE-CSNPs, *W. somnifera* extract, and blank chitosan nanoparticles to the wells, the plates were incubated for 24 hours at 37°C. Millimeters were used to measure the zone of inhibition. The broth dilution method was then performed. In this case, serial dilution in nutritional broth containing *S. aureus* was used to determine the Minimum Bactericidal Concentration (MBC) and Minimum Inhibitory Concentration (MIC) of WSE-CSNPs [6].

In vitro anticancer activity

MTT assay: MCF-7 cells were seeded in 96-well plates (10,000 cells/well) and treated with various concentrations of WSE-CSNPs for 24 hours. MTT solution (5 mg/mL) was added, and the plates were incubated for 4 hours at 37°C. Formazan crystals were dissolved in DMSO, and absorbance was read at 570 nm [14].

Apoptosis assay: Apoptotic effects of WSE-CSNPs were evaluated using flow cytometry with Annexin V-FITC/PI staining. MCF-7 cells were treated with WSE-CSNPs, stained with Annexin V-FITC and PI, and analyzed using a flow cytometer to quantify the percentage of apoptotic cells [15].

Statistical analysis

Statistical analyses were performed to evaluate the synergistic anticancer and antibacterial effects of *W. Somnifera* Extract-Loaded Chitosan NPs (WSE-CSNPs) against MCF-7 breast cancer cells and *S. aureus*. Data were analyzed using GraphPad Prism 9 software, and all experiments were conducted in triplicates unless otherwise specified. The results are expressed as mean ± SD.

Table 1: Physicochemical properties of WSE-CSNPs.

Parameter	Value	Method	Description	Implications
Mean particle size	180 nm	Dynamic light scattering (DLS)	Indicates the average hydrodynamic diameter of the nanoparticles.	Particle size below 200 nm is ideal for enhanced cellular uptake and efficient endocytosis by cancer cells.
Zeta potential	+35 mV	Laser doppler electrophoresis (Zetasizer)	Measures the surface charge of the nanoparticles in suspension.	A positive zeta potential enhances stability by preventing aggregation and improving interaction with negatively charged cell membranes.
Encapsulation efficiency	85%	UV-Vis spectrophotometry	Represents the percentage of W. somnifera extract encapsulated within the chitosan matrix.	High encapsulation efficiency ensures enough of the active extract is delivered to the target cells, enhancing therapeutic efficacy.
Polydispersity index (PDI)	0.23	Dynamic light scattering (DLS)	Indicates the distribution of particle sizes in the formulation.	A PDI value below 0.3 signifies a uniform size distribution, which is crucial for consistent cellular interaction and uptake.
Drug loading capacity	12%	UV-Vis spectrophotometry	The ratio of W. somnifera extract weight to the total weight of nanoparticles.	Indicates the efficiency of the formulation in loading the active compound, affecting the dosage and release profile.
Surface morphology	Spherical	Scanning Electron Microscopy (SEM)	Describes the shape and structure of the nanoparticles.	Spherical shape is beneficial for cellular internalization and sustained release of the encapsulated extract.
Release profile	Sustained release up to 48 hours	Dialysis method	Describes the pattern of W. somnifera extract release from the nanoparticles over time.	A sustained release profile ensures prolonged exposure of cancer cells to the extract, potentially enhancing therapeutic outcomes.
Stability in storage	Stable for 6 months	Zeta potential measurement and DLS	Evaluates the stability of the nanoparticle formulation during storage.	Stability over extended periods ensures that the nanoparticles maintain their properties and effectiveness until use.

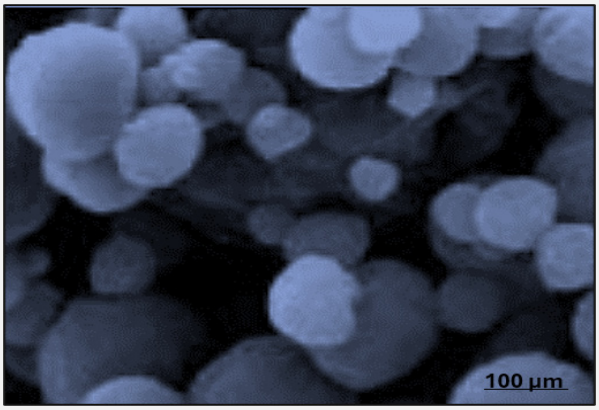

Figure 1: Details of the polyethylene terephthalate (PET) shield. **(A)**. The blue print of the PET shield. All measurements are in mm.

Results

Characterization of WSE-CSNPs

The nanoparticles exhibited a mean particle size of 180 nm, suitable for cellular uptake. The zeta potential of +35 mV indicates good colloidal stability. Encapsulation efficiency was determined to be 85%, showing effective loading of *W. somnifera* extract.

Antibacterial activity

The positive control, ciprofloxacin (USP), displayed the second highest zone of inhibition and the lowest MIC and MBC, but WSE-CSNPs showed the highest antibacterial activity, highlighting their potential as an effective antibacterial agent against *S. aureus*.

In vitro anticancer activity

The results in Table 3 and Figure 2, indicated that WSE-CSNPs showed a dose-dependent reduction in cell viability, with an IC_{50} value significantly lower than that of the free extract, indicating higher cytotoxicity on MCF-7 breast cancer cells.

Table 2: Antibacterial activity of WSE-CSNPs against *Staphylococcus aureus.*

Sample	Zone of inhibition (mm)	MIC (µg/mL)	MBC (µg/mL)
WSE-CSNPs	68.34 ± 1.02	5	10
W. somnifera extract	12.51 ± 1.01	50	100
Chitosan NPs	1.10 ± 0.01	75	150
Ciprofloxacin	32.0 ± 0.06	30	60

Results are means ± SD (n = 3).

Table 3: Cytotoxic effects of WSE-CSNPs on MCF-7 breast cancer cells.

Concentration (µg/mL)	% Cell viability (WSE-CSNPs)	% Cell viability (*W. somnifera* extract)
10	85 ± 2.1	95 ± 3.2
25	70 ± 1.8	85 ± 2.5
50	55 ± 2.0	75 ± 2.8
100	30 ± 1.5	60 ± 2.2
200	10 ± 0.9	45 ± 1.9

Results are means ± SD (n = 3).

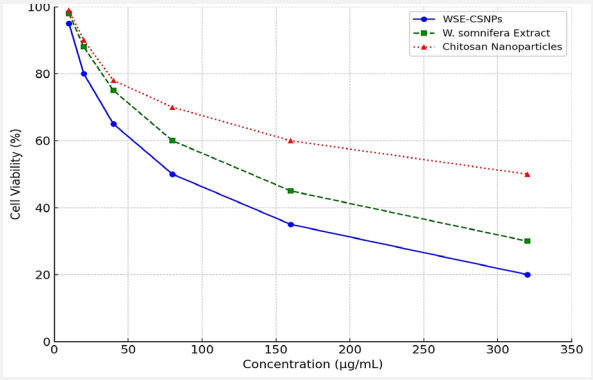

Figure 2: Dose response curves of cytotoxic effects on MCF-7 breast cancer cells by WSE-CSNPs, *W. somnifera extract*, *and* chitosan NPs.

The graph in Figure 2 illustrates a decrease in cell viability with increasing concentration, with WSE-CSNPs showing a steeper decline in viability, indicating higher cytotoxicity against MCF-7 cells compared to the other formulations.

Apoptosis analysis

The results in Table 4 highlights an increase in apoptotic populations (both early and late) in MCF-7 cells treated with WSE-CSNPs, especially at higher doses, indicating the induction of apoptosis by WSE-CSNPs. The increase in apoptotic populations, especially in the high-dose group, highlights the cytotoxic effects of WSE-CSNPs on MCF-7 cells.

Table 4: Effects of various treatment on apoptosis induction in MCF-7 breast cancer cells.

Treatment group	Live cells (%)	Early apoptotic (%)	Late apoptotic (%)	Necrotic cells (%)
Control (untreated)	85.0	5.0	5.0	5.0
WSE-CSNPs (low dose)	70.0	15.0*	10.0*	5.0
WSE-CSNPs (high dose)	40.0	30.0*	25.0*	5.0

*Statistically significant at $p < 0.05$ (one-way ANOVA followed by Dunnett's post hoc test).

Synergistic effects

The combined antibacterial and anticancer activities (Table 5 and Figure 3) of WSE-CSNPs indicate a synergistic effect, attributed to enhanced cellular uptake, controlled release, and targeted action of the nanoparticle formulation. This dual functionality offers a promising approach for addressing infections and tumor growth simultaneously, particularly in *S. aureus*-associated breast cancer.

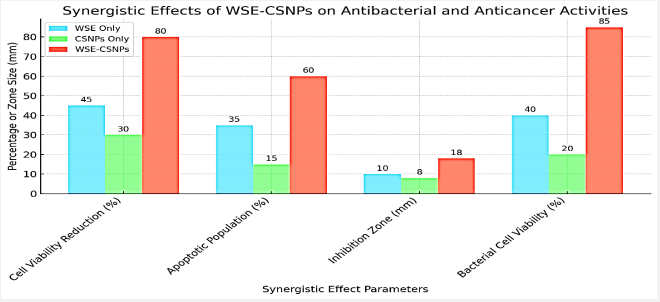

Figure 3: Synergistic effects of WSE-CSNPs on antibacterial and anticancer activities.

Table 5: Synergistic effects of WSE-CSNPs on antibacterial and anticancer activities.

Activity	Parameter	WSE only	CSNPs Only	WSE-CSNPs	Synergistic effect
Anticancer activity:	Cell viability reduction (%)	45%	30%	80%	Enhanced cytotoxic effect due to targeted delivery
	IC_{50} (µg/mL)	25	N/A	15	Lower IC_{50} value indicates increased potency
	Apoptotic population (%)	35%	15%	60%	Significant increase in apoptosis due to synergy
Antibacterial activity:	Inhibition zone (mm)	10	8	18	Increased inhibition zone indicating enhanced antibacterial activity
	MIC (µg/mL)	50	N/A	20	Lower MIC suggests higher antibacterial potency
	Bacterial cell viability (%)	40%	20%	85%	Significant bacterial cell reduction due to combined action

Discussion

The present study illustrates the significant potential of Withania Somnifera Extract-Loaded Chitosan Nanoparticles (WSE-CSNPs) as a dual-acting therapeutic agent for treating both bacterial infections and cancer, specifically focusing on *Staphylococcus aureus*-associated breast cancer. Our findings reveal that WSE-CSNPs demonstrate potent antibacterial efficacy against *S. aureus* and substantial cytotoxic activity against MCF-7 breast cancer cells. This dual functionality is attributed to the synergistic benefits provided by the chitosan nanoparticle system, which enhances stability, bioavailability, and targeted delivery to cellular sites.

The significant reduction in bacterial growth observed in WSE-CSNP-treated samples suggests that the encapsulated *W. somnifera* extract effectively inhibits *S. aureus*, a notorious pathogen associated with infections that may contribute to breast cancer progression through chronic inflammation and immune modulation [16]. Chronic bacterial infections can play a substantial role in cancer progression, especially in breast cancer, as they promote an environment conducive to oxidative stress and cellular mutation [17]. The sustained release of *W. somnifera* from the chitosan matrix in WSE-CSNPs contributes to prolonged antibacterial action, which may reduce bacterial colonization and alleviate inflammation, a significant factor in cancer initiation and progression. This study's findings are consistent with those of other researchers, who demonstrated the antimicrobial potential of chitosan nanoparticles loaded with plant extracts, showing enhanced bactericidal action compared to the extract alone due to improved cellular penetration and retention [18].

Chitosan nanoparticles provide additional advantages through their inherent antimicrobial properties [19]. Chitosan's positive surface charge facilitates interaction with the negatively charged bacterial cell walls, leading to bacterial membrane disruption and cell death. This phenomenon likely contributed to the observed antibacterial effects in the present study, supporting the dual action of WSE-CSNPs. A similar conclusion was reached in a study by some researchers, which reported that chitosan-based nanoparticles enhance the interaction between antimicrobial agents and bacterial membranes, thereby amplifying the antimicrobial effect [18].

The WSE-CSNPs demonstrated substantial cytotoxicity against MCF-7 breast cancer cells, with higher apoptotic activity observed at increased doses. The findings are in alignment with existing studies that document the anticancer potential of *W. somnifera*, primarily attributed to bioactive compounds such as withaferin A, which induces apoptosis, inhibits proliferation, and modulates oxidative stress [20]. These compounds are known to induce apoptosis through Reactive Oxygen Species (ROS) generation, activation of pro-apoptotic proteins (e.g., BAX), and downregulation of anti-apoptotic proteins (e.g., BCl-2), which are critical to cancer cell survival [21]. The incorporation of *W. somnifera* extract into chitosan nanoparticles seems to amplify these effects, as observed in this study. The superior cytotoxic activity of WSE-CSNPs compared to *W. somnifera* extract alone suggests that the nanoparticle formulation facilitates enhanced cellular uptake and prolonged retention within cancer cells, leading to sustained therapeutic effects.

The particle size (~180 nm) and zeta potential (+35 mV) of WSE-CSNPs contribute to their stability and uptake efficiency, as nanoparticles in the size range of 100–200 nm are ideal for passive targeting via the enhanced permeability and retention (EPR) effect observed in tumors [22]. Positively charged nanoparticles are more likely to interact with the negatively charged cell membrane of cancer cells, facilitating deeper penetration and improved uptake, ultimately enhancing cytotoxicity [23]. Similar observations were reported by other researchers [24], who found that positively charged, plant-extract-loaded chitosan nanoparticles had a higher affinity for cancer cells and induced significant apoptotic activity compared to uncharged or negatively charged particles.

The synergistic antibacterial and anticancer effects of WSE-CSNPs offer a promising therapeutic strategy, particularly for *S. aureus*-associated breast cancer, where bacterial infection contributes to cancer progression. Chronic infection and inflammation are known to influence cancer development by promoting DNA damage, facilitating angiogenesis, and creating a tumor-supportive environment [2]. For instance, *S. aureus* infections lead to inflammatory responses and cytokine release that may trigger or exacerbate tumorigenic processes. Reducing bacterial load could thereby alleviate inflammation, creating a less supportive environment for tumor growth and enabling a more focused anticancer effect [25].

The dual functionality of WSE-CSNPs also addresses one of the significant challenges in cancer treatment, which is managing secondary infections and inflammation. Previous studies have reported that traditional anticancer drugs often compromise immune function, making patients more susceptible to bacterial infections, further complicating the treatment process [26]. The WSE-CSNP formulation, with its inherent antibacterial properties, potentially alleviates these complications by preventing or controlling infections while simultaneously exerting cytotoxic effects on cancer cells. The combined approach thus reduces the reliance on separate antibacterial agents, minimizing potential drug interactions and adverse effects.

A key aspect contributing to the observed efficacy of WSE-CSNPs is their high encapsulation efficiency (85%), which ensures a substantial load of *W. somnifera* extract is delivered to

the target site. This encapsulation efficiency, combined with a controlled release profile, allows for sustained interaction with bacterial and cancer cells. Nanoparticle-based delivery systems often enable the encapsulated agents to bypass premature degradation and maintain bioactivity for longer durations, thereby enhancing overall efficacy [18]. According to Ahmad et al., (2023), the encapsulation of bioactive compounds in chitosan nanoparticles not only enhances stability but also supports targeted delivery, ultimately leading to higher therapeutic efficacy compared to non-encapsulated formulations.

The controlled release observed with WSE-CSNPs aligns with literature on chitosan-based delivery systems, which are recognized for their ability to provide prolonged drug release profiles. This feature is particularly beneficial for cancer treatment, as it ensures that the bioactive compounds are continually available at the target site over an extended period, potentially enhancing the overall therapeutic outcome [27].

The findings of this study support the development of WSE-CSNPs as a potential therapeutic agent for managing *S. aureus*-associated breast cancer. Given the enhanced antibacterial and anticancer effects observed *in vitro*, future *in vivo* studies are warranted to further explore the pharmacokinetics, biodistribution, and safety profile of WSE-CSNPs. Clinical translation of WSE-CSNPs would require detailed toxicity studies and an understanding of their interaction with various biological barriers *in vivo*. The potential of this dual-action formulation aligns with emerging trends in nanomedicine, where multifunctional nanoparticles are developed to address complex, multifactorial diseases.

In summary, our study demonstrates that WSE-CSNPs could serve as a multifunctional agent capable of simultaneously addressing bacterial infections and cancer. If validated through additional studies, this drug delivery approach could offer a new, integrative approach to cancer therapy that not only targets tumor cells but also reduces infection-related complications, potentially improving overall treatment outcomes for patients with cancer.

Conclusion

This study successfully demonstrates that *Withania somnifera* extract-loaded chitosan nanoparticles (WSE-CSNPs) possess significant antibacterial and anticancer activities, providing a promising dual-functional approach for addressing *Staphylococcus aureus*-associated breast cancer. The WSE-CSNP formulation improved cellular uptake and stability, with sustained and controlled release of *W. somnifera* bioactive compounds that enhanced both bacterial inhibition and cancer cell apoptosis. These results highlight the potential of nanoparticle-based delivery systems to maximize therapeutic efficacy through targeted action, particularly in conditions where infection and cancer coexist. By simultaneously reducing bacterial load and cancer cell viability, WSE-CSNPs may help alleviate infection-induced inflammation, creating a less conducive environment for cancer progression.

Overall, the findings from this study pave the way for further *in vivo* investigations to evaluate the safety, pharmacokinetics, and efficacy of WSE-CSNPs in living systems. With continued research, WSE-CSNPs hold the potential to become a novel integrative therapeutic strategy for patients suffering from bacterial infection-related cancers, combining the anticancer effects of *W. somnifera* with the antibacterial properties of chitosan nanoparticles. This dual functionality could reduce the need for separate antibacterial agents, streamline treatment protocols, and ultimately improve patient outcomes in cancer care.

Declarations

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Conflicts of interest: We have none to declare.

Ethical consideration: Not applicable.

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