IJARSCT
International Journal of Advanced Research in Science, Communication and Technology
International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal
ISSN: 2581-9429
Volume 6, Issue 2, August 2026
Impact Factor: 8.2

Green Synthesized Silver Nanoparticles for the Treatment of Colon Cancer: Current Advances, Therapeutic Potential, and Future Perspectives

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Abstract: *Colon cancer, also known as colorectal cancer (CRC), remains one of the most frequently diagnosed gastrointestinal malignancies and is a leading cause of cancer-related morbidity and mortality worldwide. Despite significant advances in surgery, chemotherapy, radiotherapy, targeted therapy, and immunotherapy, the clinical management of colon cancer continues to face major challenges, including poor drug selectivity, systemic toxicity, multidrug resistance, and disease recurrence. These limitations have accelerated the development of nanotechnology-based therapeutic strategies aimed at improving treatment efficacy while minimizing adverse effects.*

Among various nanomaterials, silver nanoparticles (AgNPs) have attracted considerable attention because of their unique physicochemical characteristics, high surface-area-to-volume ratio, and diverse biological activities, including antimicrobial, antioxidant, anti-inflammatory, and anticancer properties. Green synthesis has emerged as an eco-friendly and sustainable alternative to conventional physical and chemical methods for the production of AgNPs. In this approach, medicinal plant extracts rich in bioactive phytochemicals act as natural reducing and stabilizing agents, eliminating the need for hazardous chemicals and improving nanoparticle biocompatibility.

Recent studies have demonstrated that green synthesized AgNPs exhibit significant anticancer activity against colon cancer by inducing reactive oxygen species (ROS) generation, oxidative stress, mitochondrial dysfunction, DNA damage, apoptosis, and cell cycle arrest. Furthermore, these nanoparticles serve as efficient carriers for colon-targeted drug delivery, improving drug stability, bioavailability, cellular uptake, and therapeutic selectivity while reducing systemic toxicity. Polyherbal-mediated green synthesis has gained increasing interest because the synergistic action of multiple phytochemicals enhances nanoparticle stability and biological activity.

This review summarizes the current advances in green synthesis of silver nanoparticles, their physicochemical characterization, mechanisms of anticancer action, applications in colon-targeted drug delivery, recent research developments, current limitations, and future prospects. The available evidence suggests that green synthesized silver nanoparticles represent a promising platform for the development of safer and more effective therapeutic strategies for colon cancer management.(1-5)..

Keywords: Colon cancer; Colorectal cancer; Green synthesis; Silver nanoparticles; Nanomedicine; Targeted drug delivery, Polyherbal extrac ,Nanomedicine

I. INTRODUCTION

Cancer is one of the most serious global health challenges and continues to impose a substantial burden on healthcare systems worldwide. It is characterized by uncontrolled cellular proliferation resulting from the accumulation of genetic and epigenetic alterations that disrupt normal mechanisms regulating cell growth, differentiation, and programmed cell

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death. Among different malignancies, colon cancer, commonly referred to as colorectal cancer (CRC), is one of the most prevalent gastrointestinal cancers and remains a major cause of cancer-related mortality. According to recent epidemiological reports, the global incidence of CRC continues to increase because of population ageing, lifestyle modifications, dietary habits, obesity, physical inactivity, smoking, alcohol consumption, chronic inflammatory bowel diseases, and inherited genetic predisposition. Although considerable progress has been achieved in early diagnosis and therapeutic interventions, effective management of advanced-stage colon cancer remains a significant clinical challenge.

Conventional treatment modalities, including surgical resection, chemotherapy, radiotherapy, targeted therapy, and immunotherapy, have substantially improved patient survival over the past decades. Nevertheless, these approaches are frequently associated with limitations such as non-specific drug distribution, severe systemic toxicity, multidrug resistance, poor bioavailability of chemotherapeutic agents, and frequent tumour recurrence. These limitations have encouraged researchers to investigate innovative drug delivery systems capable of selectively targeting tumour tissues while minimizing damage to healthy cells. Nanotechnology has emerged as one of the most promising approaches for addressing these challenges because nanoparticles possess unique physicochemical properties that facilitate controlled drug release, enhanced permeability, improved cellular uptake, and site-specific drug delivery.

Among the various nanomaterials investigated for biomedical applications, silver nanoparticles (AgNPs) have received considerable scientific attention owing to their remarkable biological activities. In addition to their well-established antimicrobial properties, AgNPs exhibit antioxidant, anti-inflammatory, antiviral, and anticancer effects. Their extremely small particle size and high surface-area-to-volume ratio enable efficient interaction with biological membranes and intracellular organelles, thereby improving therapeutic efficacy. Experimental studies have demonstrated that AgNPs inhibit cancer cell proliferation through multiple mechanisms, including excessive generation of reactive oxygen species, mitochondrial dysfunction, DNA damage, induction of apoptosis, and cell cycle arrest. These mechanisms collectively contribute to selective destruction of malignant cells while minimizing toxicity towards normal tissues.

In recent years, green synthesis has emerged as an environmentally friendly and sustainable alternative to conventional chemical and physical methods for nanoparticle production. Green synthesis utilizes biological resources such as medicinal plant extracts, microorganisms, fungi, and algae as natural reducing and stabilizing agents. Plant-mediated synthesis is particularly attractive because medicinal plants contain diverse phytochemicals, including flavonoids, alkaloids, phenolic compounds, terpenoids, proteins, and polysaccharides, which facilitate the reduction of silver ions into metallic nanoparticles while simultaneously stabilizing their surface. Compared with conventional synthesis methods, green synthesis eliminates the use of hazardous chemicals, reduces environmental pollution, improves biocompatibility, and offers a cost-effective approach suitable for biomedical applications.

Polyherbal-mediated synthesis represents a further advancement in green nanotechnology. The combination of extracts from multiple medicinal plants provides a broader spectrum of bioactive phytochemicals, resulting in enhanced nanoparticle stability, improved antioxidant capacity, stronger anticancer activity, and superior pharmaceutical performance. Recent investigations have suggested that these polyherbal green synthesized silver nanoparticles may serve as efficient nanocarriers for colon-targeted drug delivery and could significantly improve therapeutic outcomes in colon cancer treatment.

The present review provides a comprehensive overview of green synthesized silver nanoparticles and their potential applications in colon cancer therapy. The review discusses the pathology of colon cancer, current therapeutic approaches, principles of nanotechnology, green synthesis methods, characterization techniques, molecular mechanisms of anticancer activity, colon-targeted drug delivery systems, recent research developments, existing limitations, and future perspectives. Collectively, these findings highlight the growing importance of green nanotechnology in the development of safer, more effective, and environmentally sustainable strategies for colon cancer management.[1-5]

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II. COLON CANCER

Colon cancer, also known as colorectal cancer (CRC), is a malignant neoplasm originating from the epithelial cells lining the colon or rectum. It is one of the most commonly diagnosed gastrointestinal cancers and remains a major cause of cancer-related morbidity and mortality worldwide. CRC usually develops through a multistep process involving the gradual transformation of normal colonic epithelium into adenomatous polyps and eventually invasive carcinoma following the accumulation of multiple genetic and epigenetic alterations. The adenoma–carcinoma sequence is considered the predominant pathway for colorectal carcinogenesis and often requires several years to progress, providing an opportunity for early detection and prevention through appropriate screening programs.

The global incidence of colon cancer has increased considerably during the past few decades due to changes in lifestyle, dietary habits, increasing life expectancy, urbanization, and environmental factors. According to recent epidemiological observations, colorectal cancer is among the leading causes of cancer-related deaths worldwide. Although the disease predominantly affects individuals over 50 years of age, an increasing number of cases are now being reported in younger adults, indicating the growing influence of modifiable risk factors and changing disease patterns. Early diagnosis and timely therapeutic intervention remain the most effective approaches for improving long-term survival and reducing disease-associated mortality.

Several environmental, genetic, and lifestyle-related factors contribute to the development of colon cancer. Advanced age remains one of the strongest risk factors; however, obesity, physical inactivity, smoking, excessive alcohol consumption, diets rich in processed and red meat, and inadequate intake of fruits, vegetables, and dietary fiber also significantly increase disease susceptibility. Chronic inflammatory bowel diseases, including ulcerative colitis and Crohn's disease, further elevate the risk of malignant transformation due to persistent inflammation of the intestinal mucosa. In addition, hereditary syndromes such as Familial Adenomatous Polyposis (FAP) and Lynch syndrome substantially increase the lifetime risk of colorectal cancer through inherited genetic mutations.

At the molecular level, colorectal carcinogenesis is characterized by sequential alterations in tumor suppressor genes, oncogenes, DNA mismatch repair genes, and signaling pathways regulating cellular proliferation and apoptosis. Mutations affecting the APC gene, KRAS proto-oncogene, TP53 tumor suppressor gene, and DNA mismatch repair mechanisms are among the most frequently reported genetic events associated with CRC progression. These molecular abnormalities promote uncontrolled cell proliferation, impaired apoptosis, angiogenesis, invasion, and metastatic spread to distant organs such as the liver and lungs. A better understanding of these molecular pathways has facilitated the development of targeted therapeutic strategies and personalized treatment approaches for selected patient populations.

The clinical manifestations of colon cancer vary depending on tumor location and disease stage. Patients with early-stage disease may remain asymptomatic, whereas progressive disease commonly presents with persistent abdominal pain, rectal bleeding, altered bowel habits, constipation or diarrhea, unexplained weight loss, fatigue, anemia, abdominal distension, and occult gastrointestinal bleeding. Because these symptoms are often nonspecific, many patients are diagnosed only after the disease has reached an advanced stage, emphasizing the importance of regular screening among high-risk populations.

Accurate diagnosis requires a combination of clinical evaluation, laboratory investigations, endoscopic examination, imaging techniques, and histopathological confirmation. Colonoscopy remains the gold standard for diagnosis because it allows direct visualization of suspicious lesions and facilitates biopsy for microscopic examination. Additional diagnostic tools include fecal occult blood testing (FOBT), fecal immunochemical testing (FIT), computed tomography (CT), magnetic resonance imaging (MRI), positron emission tomography (PET), and serum biomarkers such as carcinoembryonic antigen (CEA), which are useful for disease staging, monitoring therapeutic response, and detecting recurrence.

The prognosis of colorectal cancer depends largely on the stage at diagnosis. Localized tumors confined to the bowel wall generally have favorable survival outcomes following surgical resection, whereas advanced disease with regional lymph node involvement or distant metastasis is associated with significantly poorer prognosis. Therefore, implementation of effective screening programs, increased public awareness, lifestyle modifications, and the

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development of novel therapeutic strategies remain essential for reducing the global burden of colon cancer. Recent advances in nanotechnology and targeted drug delivery systems have created new opportunities for improving therapeutic efficacy while minimizing systemic toxicity, thereby offering promising alternatives to conventional treatment approaches.[1-4]

III. CONVENTIONAL TREATMENT OF COLON CANCER

The treatment of colon cancer depends on several factors, including the stage of the disease, tumor location, molecular characteristics, patient's age, and overall health status. Over the past few decades, considerable advances have been achieved in the management of colorectal cancer through improvements in surgical techniques, chemotherapy, radiotherapy, targeted therapy, and immunotherapy. Although these therapeutic approaches have significantly improved patient survival, complete eradication of the disease remains challenging, particularly in patients with advanced or metastatic colorectal cancer. Treatment failure is frequently associated with drug resistance, tumor recurrence, systemic toxicity, and poor therapeutic selectivity. Therefore, the development of safer and more effective treatment strategies remains an important area of cancer research.

Surgical resection remains the primary treatment option for patients with localized colon cancer. The objective of surgery is complete removal of the primary tumor along with surrounding healthy tissue and regional lymph nodes to minimize the risk of recurrence. Depending on tumor size and location, different surgical procedures including partial colectomy, hemicolectomy, or total colectomy may be performed. Surgical intervention provides excellent clinical outcomes when the disease is detected at an early stage; however, its effectiveness decreases significantly in metastatic disease where complete tumor removal is often impossible.

Chemotherapy is widely employed as adjuvant, neoadjuvant, or palliative treatment depending on disease stage. Commonly used chemotherapeutic agents include 5-fluorouracil, capecitabine, oxaliplatin, and irinotecan, either as monotherapy or in combination regimens such as FOLFOX and FOLFIRI. These agents inhibit rapidly dividing cancer cells by interfering with DNA synthesis and cell division. Despite their clinical effectiveness, conventional chemotherapeutic drugs are associated with several adverse effects including nausea, vomiting, diarrhea, bone marrow suppression, neurotoxicity, mucositis, and alopecia. Furthermore, prolonged chemotherapy frequently results in multidrug resistance, limiting long-term therapeutic success.

Radiotherapy plays a comparatively limited role in colon cancer but remains an important component in the management of rectal cancer. It is commonly administered before surgery to reduce tumor size or after surgery to eliminate residual malignant cells. Advances in radiation planning have improved treatment precision and reduced damage to surrounding normal tissues. However, radiotherapy may still produce adverse effects including gastrointestinal toxicity, fatigue, radiation enteritis, and local tissue injury, particularly during prolonged treatment.

Targeted therapy has emerged as an important advancement in the treatment of metastatic colorectal cancer. Unlike conventional chemotherapy, targeted agents specifically inhibit molecular pathways involved in tumor growth, angiogenesis, and cellular proliferation. Monoclonal antibodies targeting vascular endothelial growth factor (VEGF) and epidermal growth factor receptor (EGFR) have demonstrated improved survival in selected patient populations. More recently, immunotherapy using immune checkpoint inhibitors has shown encouraging results in patients with microsatellite instability-high (MSI-H) or mismatch repair-deficient colorectal cancer. Nevertheless, these therapies are effective only in specific molecular subgroups and are often associated with high treatment costs and immune-related adverse events.

Despite continuous therapeutic advancements, current treatment strategies remain limited by poor tumor selectivity, non-specific drug distribution, severe systemic toxicity, low bioavailability, multidrug resistance, and frequent disease recurrence. These limitations have stimulated extensive research into novel drug delivery systems capable of selectively targeting malignant tissues while reducing exposure of healthy cells to cytotoxic agents. Nanotechnology has emerged as one of the most promising approaches to address these challenges and improve the therapeutic management of colon cancer.[3-5]

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IV. NANOTECHNOLOGY IN COLON CANCER THERAPY

Nanotechnology has revolutionized the field of oncology by enabling the development of nanoscale drug delivery systems with improved therapeutic efficiency and reduced systemic toxicity. Nanoparticles generally range from 1 to 100 nanometers in size and possess unique physicochemical characteristics, including a large surface-area-to-volume ratio, enhanced surface reactivity, tunable physicochemical properties, and the ability to carry therapeutic agents directly to diseased tissues. These features have made nanotechnology an attractive platform for improving the diagnosis, imaging, and treatment of various malignancies, including colon cancer.

One of the major advantages of nanotechnology in cancer therapy is its ability to enhance targeted drug delivery. Nanoparticles preferentially accumulate within tumor tissues through the enhanced permeability and retention (EPR) effect, allowing higher concentrations of anticancer drugs to reach the tumor while minimizing exposure to normal tissues. In addition, nanoparticles can be functionalized with specific ligands, antibodies, peptides, or polymers to achieve active targeting of cancer cells, thereby improving therapeutic selectivity and reducing adverse effects associated with conventional chemotherapy.

Various nanocarriers including liposomes, polymeric nanoparticles, dendrimers, solid lipid nanoparticles, metallic nanoparticles, and mesoporous silica nanoparticles have been investigated for colon cancer therapy. Among these, silver nanoparticles (AgNPs) have attracted considerable attention because of their intrinsic biological activities in addition to their role as drug delivery vehicles. Green synthesized AgNPs possess improved biocompatibility, environmental sustainability, and enhanced biological performance owing to the presence of plant-derived phytochemicals on their surface. These nanoparticles have demonstrated promising anticancer activity through the induction of oxidative stress, mitochondrial dysfunction, DNA damage, apoptosis, and inhibition of tumor cell proliferation.

Furthermore, nanotechnology facilitates controlled and sustained drug release, improves the stability of therapeutic agents, enhances drug solubility, prolongs circulation time, and increases intracellular drug uptake. These advantages collectively contribute to improved therapeutic outcomes and reduced systemic toxicity. Consequently, nanotechnology-based formulations are increasingly recognized as a promising strategy for overcoming the limitations of conventional colon cancer treatment and represent an important area of ongoing pharmaceutical research.[5,21-26]

V. SILVER NANOPARTICLES

Silver nanoparticles (AgNPs) are among the most extensively investigated metallic nanoparticles in nanomedicine because of their unique physicochemical properties and broad spectrum of biological activities. Generally ranging from 1 to 100 nm in diameter, AgNPs possess a high surface-area-to-volume ratio, enhanced surface reactivity, remarkable optical properties, and excellent biological compatibility. These characteristics have enabled their widespread application in pharmaceutical sciences, including antimicrobial therapy, wound healing, biosensing, diagnostic imaging, tissue engineering, drug delivery, and cancer therapeutics. In recent years, increasing attention has been directed towards the potential application of AgNPs in colon cancer treatment owing to their intrinsic anticancer activity and their ability to function as efficient drug delivery carriers.

Unlike conventional chemotherapeutic agents, silver nanoparticles exert cytotoxic effects through multiple intracellular pathways, thereby reducing the likelihood of resistance development. Their small particle size facilitates penetration through biological membranes and promotes efficient uptake by tumor cells through endocytosis. Once internalized, AgNPs interact with various cellular components, including mitochondria, lysosomes, proteins, and nucleic acids, ultimately disrupting cellular homeostasis. Experimental studies have demonstrated that AgNPs inhibit tumor progression by generating reactive oxygen species (ROS), inducing oxidative stress, damaging mitochondrial membranes, causing DNA fragmentation, activating caspase-dependent apoptosis, arresting the cell cycle, and suppressing angiogenesis. These multiple mechanisms contribute to selective inhibition of malignant cells while minimizing toxicity to surrounding normal tissues when appropriately designed.

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The biological activity of AgNPs is strongly influenced by particle size, morphology, surface charge, and synthesis method. Smaller nanoparticles generally exhibit greater cellular uptake and higher biological activity because of their increased surface reactivity. Similarly, surface modification using plant-derived phytochemicals improves nanoparticle stability, dispersibility, and interaction with biological systems. These findings have stimulated considerable interest in the development of green synthesized silver nanoparticles for biomedical applications, particularly in targeted cancer therapy.[6-14]

VI. GREEN SYNTHESIS OF SILVER NANOPARTICLES

The synthesis method plays a crucial role in determining the physicochemical characteristics and biological performance of silver nanoparticles. Conventional chemical and physical synthesis techniques frequently require toxic reducing agents, high energy consumption, elevated temperatures, and expensive instrumentation. Residual chemicals may also compromise nanoparticle biocompatibility and increase environmental hazards. Consequently, there has been growing interest in environmentally sustainable approaches capable of producing biologically safe nanoparticles suitable for pharmaceutical applications.

Green synthesis has emerged as an eco-friendly, cost-effective, and sustainable alternative for the production of silver nanoparticles. This approach utilizes naturally occurring biological resources such as medicinal plant extracts, bacteria, fungi, algae, and biomolecules as reducing and stabilizing agents. Among these biological systems, medicinal plants have become the preferred choice because they are readily available, economical, and rich in phytochemicals capable of simultaneously reducing silver ions (Ag^{+}) into metallic silver (Ag^{0}) while stabilizing the resulting nanoparticles. Unlike conventional methods, green synthesis eliminates the use of hazardous chemicals, minimizes environmental pollution, and significantly improves nanoparticle biocompatibility.

The green synthesis process generally begins with the preparation of a plant extract, followed by its reaction with an aqueous silver nitrate solution under optimized experimental conditions. Phytochemicals such as flavonoids, phenolic acids, tannins, alkaloids, terpenoids, proteins, carbohydrates, and polysaccharides participate in the reduction of silver ions and prevent nanoparticle aggregation by acting as natural capping agents. Successful nanoparticle formation is typically indicated by a visible colour change from colourless to yellowish-brown or dark brown due to surface plasmon resonance. The synthesized nanoparticles are subsequently purified and characterized using appropriate analytical techniques before biological evaluation.

Compared with conventional synthesis methods, green synthesis offers numerous advantages including lower production costs, reduced toxicity, simplified processing, improved pharmaceutical safety, enhanced biological activity, and greater environmental sustainability. These advantages have established green synthesis as one of the most promising approaches for developing silver nanoparticles intended for biomedical and anticancer applications.[6-16]

VII. PLANT-MEDIATED AND POLYHERBAL GREEN SYNTHESIS

Plant-mediated synthesis is currently the most widely investigated green approach for producing silver nanoparticles because medicinal plants contain diverse phytoconstituents capable of acting as both reducing and stabilizing agents. The chemical composition of plant extracts varies according to species, geographical origin, extraction method, and environmental conditions, influencing nanoparticle size, morphology, stability, and biological activity. Numerous medicinal plants have been investigated for AgNP synthesis owing to their rich content of flavonoids, polyphenols, saponins, glycosides, terpenoids, and other secondary metabolites.

Several medicinal plants, including *Azadirachta indica* (Neem), *Curcuma longa* (Turmeric), *Moringa oleifera*, *Ocimum sanctum* (Holy Basil), *Aloe vera*, and *Tinospora cordifolia*, have demonstrated excellent potential for synthesizing biologically active AgNPs. Nanoparticles produced using these plants exhibit enhanced antioxidant, antimicrobial, anti-inflammatory, and anticancer activities because phytochemicals remain adsorbed on the nanoparticle surface, improving their interaction with biological systems. Such surface functionalization also contributes to improved colloidal stability and prolonged biological activity.

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Recently, polyherbal-mediated synthesis has emerged as an advanced strategy that combines extracts from two or more medicinal plants to exploit the synergistic effects of multiple phytochemicals. The combined presence of flavonoids, phenolics, alkaloids, tannins, terpenoids, and polysaccharides enhances nanoparticle formation, stability, antioxidant potential, and anticancer efficacy. Compared with nanoparticles synthesized from a single plant extract, polyherbal AgNPs have shown improved biological performance in several experimental studies. Their enhanced activity is attributed to the complementary actions of different phytoconstituents, which collectively improve reduction efficiency, surface stabilization, and therapeutic activity.

The growing interest in plant-mediated and polyherbal green synthesis reflects the increasing demand for environmentally sustainable nanotechnology capable of producing highly biocompatible nanoparticles with superior therapeutic performance. These nanoparticles not only exhibit intrinsic anticancer activity but also serve as efficient nanocarriers for targeted drug delivery, making them promising candidates for the future management of colon cancer and other malignancies.[8,15,16]

VIII. CHARACTERIZATION OF GREEN SYNTHESIZED SILVER NANOPARTICLES

The successful synthesis of green synthesized silver nanoparticles (AgNPs) must be confirmed through comprehensive physicochemical characterization before their pharmaceutical and biomedical applications can be evaluated. Characterization provides essential information regarding nanoparticle formation, particle size, morphology, crystalline structure, surface chemistry, elemental composition, colloidal stability, and biological functionality. These physicochemical characteristics directly influence the therapeutic efficacy, cellular uptake, biodistribution, drug loading capacity, and safety profile of nanoparticles. Therefore, systematic characterization represents a crucial step in the development of green synthesized AgNPs for colon cancer therapy.

Unlike chemically synthesized nanoparticles, green synthesized AgNPs are naturally coated with plant-derived phytochemicals that function as reducing and stabilizing agents. These biomolecules significantly influence nanoparticle stability, surface charge, biological interactions, and therapeutic activity. Consequently, multiple complementary analytical techniques are required to obtain a complete understanding of nanoparticle characteristics rather than relying on a single analytical method.

8.1 UV–Visible Spectroscopy

Ultraviolet–Visible (UV–Vis) spectroscopy is one of the simplest and most widely employed techniques for the preliminary confirmation of silver nanoparticle synthesis. During the reduction of silver ions (Ag^+) into metallic silver (Ag^0), free electrons present on the nanoparticle surface undergo collective oscillation when exposed to visible light, producing a phenomenon known as **Surface Plasmon Resonance (SPR)**. This characteristic SPR band confirms nanoparticle formation and provides valuable information regarding particle size distribution and aggregation behavior.

Green synthesized AgNPs generally exhibit an SPR absorption peak between **400 and 450 nm**, although slight variations may occur depending on particle size, morphology, concentration, and plant extract composition. A sharp and symmetrical absorption peak usually indicates uniform and well-dispersed nanoparticles, whereas peak broadening or shifting suggests particle aggregation or size heterogeneity. Because UV–Vis spectroscopy is rapid, economical, and non-destructive, it is routinely used for monitoring nanoparticle synthesis during optimization studies.

8.2 Fourier Transform Infrared Spectroscopy (FTIR)

Fourier Transform Infrared Spectroscopy (FTIR) is widely used to identify the functional groups responsible for the reduction and stabilization of green synthesized silver nanoparticles. Medicinal plant extracts contain diverse phytochemicals, including flavonoids, phenolic compounds, alkaloids, proteins, terpenoids, carbohydrates, and polysaccharides, which participate in nanoparticle synthesis. FTIR analysis detects characteristic absorption bands corresponding to these functional groups, thereby confirming their involvement in nanoparticle formation.

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The comparison of FTIR spectra before and after nanoparticle synthesis provides evidence of interactions between phytochemicals and silver ions. Changes in absorption peaks indicate that these biomolecules act as natural capping agents, improving nanoparticle stability, preventing aggregation, and enhancing biological compatibility. Consequently, FTIR plays an essential role in understanding the molecular mechanisms underlying green nanoparticle synthesis.

8.3 Scanning Electron Microscopy (SEM)

Scanning Electron Microscopy (SEM) is employed to examine the external morphology and surface characteristics of silver nanoparticles. SEM images provide valuable information regarding particle shape, size distribution, surface texture, and the degree of nanoparticle aggregation. Green synthesized AgNPs are commonly observed as spherical or nearly spherical particles, although triangular, cubic, rod-shaped, and irregular morphologies may also occur depending on synthesis conditions and plant extract composition.

SEM analysis helps evaluate the uniformity of nanoparticle populations and confirms successful synthesis. Morphological characteristics significantly influence biological activity because particle size and surface architecture affect cellular internalization, drug loading efficiency, and interactions with cancer cells.

8.4 Transmission Electron Microscopy (TEM)

Transmission Electron Microscopy (TEM) is regarded as one of the most accurate techniques for determining nanoparticle size, morphology, and internal structure. Compared with SEM, TEM provides much higher image resolution, enabling direct visualization of individual nanoparticles at the nanometer scale.

TEM analysis allows accurate measurement of particle diameter, shape, dispersion pattern, and crystallinity. It also facilitates the identification of particle aggregation and confirms whether nanoparticles remain uniformly dispersed after synthesis. Since particle size strongly influences cellular uptake, biodistribution, and anticancer activity, TEM is considered indispensable for evaluating the pharmaceutical suitability of green synthesized AgNPs.

8.5 X-ray Diffraction (XRD)

X-ray Diffraction (XRD) analysis is performed to determine the crystalline nature and phase purity of silver nanoparticles. Characteristic diffraction peaks confirm the successful formation of crystalline metallic silver and distinguish AgNPs from unreacted silver salts or other impurities.

XRD also provides information regarding crystal structure, crystallite size, and overall structural stability. The presence of well-defined diffraction peaks indicates high crystallinity, which is an important determinant of nanoparticle stability and biological performance. Therefore, XRD analysis is routinely performed alongside microscopic techniques to establish the structural integrity of synthesized nanoparticles.

8.6 Zeta Potential and Particle Size Analysis

The colloidal stability of silver nanoparticles is commonly evaluated by measuring zeta potential. This parameter reflects the electrical charge present on the nanoparticle surface and predicts the likelihood of particle aggregation during storage. Nanoparticles possessing higher positive or negative zeta potential values generally exhibit greater colloidal stability because electrostatic repulsion prevents aggregation.

Particle size analysis complements zeta potential measurements by determining the average particle diameter and size distribution. Smaller nanoparticles possess larger surface areas, facilitating improved drug loading, enhanced penetration into tumor tissues, efficient cellular uptake, and greater therapeutic efficacy. Consequently, particle size and zeta potential are considered critical quality attributes during the pharmaceutical development of green synthesized AgNPs.

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8.7 Significance of Characterization in Colon Cancer Therapy

Comprehensive characterization ensures that green synthesized silver nanoparticles possess appropriate physicochemical properties for biomedical applications. The combined use of UV–Vis spectroscopy, FTIR, SEM, TEM, XRD, zeta potential, and particle size analysis provides a complete understanding of nanoparticle formation, morphology, stability, crystallinity, and biological functionality. These analytical techniques support the development of reproducible, safe, and effective nanoparticle formulations capable of improving colon-targeted drug delivery and enhancing anticancer efficacy. Accurate characterization therefore represents the foundation for successful translation of green synthesized AgNPs from laboratory research to clinical applications.[13,15-17]

IX. ANTICANCER MECHANISMS OF GREEN SYNTHESIZED SILVER NANOPARTICLES AGAINST COLON CANCER

Green synthesized silver nanoparticles (AgNPs) have emerged as promising nanotherapeutic agents because of their ability to inhibit the growth and progression of colon cancer through multiple molecular and cellular mechanisms. Unlike conventional chemotherapeutic agents that often target a single pathway, AgNPs influence several intracellular processes simultaneously, thereby reducing the possibility of therapeutic resistance. Their small particle size, high surface-area-to-volume ratio, and phytochemical coating facilitate efficient cellular internalization and selective interaction with malignant cells. The synergistic action of silver nanoparticles and plant-derived bioactive compounds further enhances their anticancer potential while improving biocompatibility.

9.1 Cellular Uptake of Silver Nanoparticles

The therapeutic activity of AgNPs begins with their internalization into cancer cells. Due to their nanoscale dimensions, silver nanoparticles are readily taken up by colon cancer cells through endocytosis. Once inside the cell, they accumulate in different intracellular compartments, including the cytoplasm, mitochondria, lysosomes, and occasionally the nucleus. The interaction of AgNPs with these organelles disrupts normal cellular physiology and initiates several cytotoxic events that ultimately result in cancer cell death. Efficient intracellular uptake is therefore considered one of the primary factors responsible for the enhanced therapeutic efficacy of nanoparticle-based drug delivery systems.

9.2 Generation of Reactive Oxygen Species (ROS)

One of the principal mechanisms responsible for the anticancer activity of green synthesized AgNPs is the excessive production of reactive oxygen species (ROS). Following cellular internalization, silver nanoparticles stimulate the formation of superoxide anions, hydroxyl radicals, and hydrogen peroxide, leading to severe oxidative stress within cancer cells. Elevated ROS levels exceed the antioxidant defense capacity of tumor cells, resulting in irreversible oxidative damage.

Oxidative stress promotes lipid peroxidation of cellular membranes, oxidation of intracellular proteins, enzyme inactivation, and damage to nucleic acids. Since cancer cells generally maintain higher basal ROS levels than normal cells, further ROS generation by AgNPs selectively sensitizes malignant cells to oxidative injury, ultimately leading to programmed cell death.

9.3 Mitochondrial Dysfunction

Mitochondria are one of the primary intracellular targets of silver nanoparticles. Increased oxidative stress causes disruption of the mitochondrial membrane potential, resulting in mitochondrial dysfunction and loss of ATP production. Damage to the mitochondrial membrane promotes the release of cytochrome c and other pro-apoptotic proteins into the cytoplasm, activating intrinsic apoptotic pathways.

The impairment of mitochondrial function also enhances ROS production, creating a positive feedback mechanism that amplifies oxidative damage and accelerates cancer cell death. Consequently, mitochondrial dysfunction represents a critical event in AgNP-mediated anticancer activity.

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9.4 DNA Damage and Cell Cycle Arrest

Silver nanoparticles interfere with genomic stability by inducing DNA strand breaks and oxidative modification of nucleic acids. DNA damage activates cellular checkpoint mechanisms that arrest progression through different phases of the cell cycle, preventing replication of damaged genetic material. Persistent DNA injury ultimately triggers apoptosis when repair mechanisms fail to restore genomic integrity.

Cell cycle arrest prevents uncontrolled cellular proliferation, thereby limiting tumor growth and reducing metastatic potential. Experimental studies have consistently demonstrated that green synthesized AgNPs suppress proliferation of colon cancer cells through simultaneous induction of DNA damage and inhibition of cell cycle progression.

9.5 Induction of Apoptosis

Apoptosis is a highly regulated form of programmed cell death that plays an essential role in eliminating damaged or malignant cells. Green synthesized AgNPs promote apoptosis primarily through the intrinsic mitochondrial pathway. Mitochondrial membrane disruption results in activation of caspase enzymes, chromatin condensation, DNA fragmentation, membrane blebbing, and formation of apoptotic bodies.

Unlike necrosis, apoptosis occurs without extensive inflammatory responses, making it an effective mechanism for selective elimination of cancer cells. The phytochemicals associated with green synthesized nanoparticles may further enhance apoptotic signaling, contributing to improved therapeutic efficacy and reduced toxicity toward normal tissues.

9.6 Inhibition of Tumor Growth and Angiogenesis

Tumor progression depends not only on rapid cellular proliferation but also on the formation of new blood vessels that supply oxygen and nutrients to growing tumors. Green synthesized AgNPs have been reported to inhibit angiogenesis by interfering with signaling pathways involved in vascular development. Reduced blood vessel formation limits nutrient availability, suppresses tumor growth, and decreases the likelihood of metastatic spread.

In addition, AgNPs reduce cancer cell migration and invasion by disrupting multiple intracellular signaling pathways associated with tumor progression. These combined effects contribute significantly to the overall anticancer efficacy of nanoparticle-based therapies.

9.7 Overall Therapeutic Significance

The anticancer activity of green synthesized silver nanoparticles results from the coordinated action of multiple molecular mechanisms rather than a single therapeutic target. Efficient cellular uptake, ROS generation, oxidative stress, mitochondrial dysfunction, DNA damage, apoptosis, cell cycle arrest, and inhibition of tumor angiogenesis collectively suppress colon cancer progression. Furthermore, the presence of plant-derived phytochemicals enhances nanoparticle stability, biological compatibility, and therapeutic performance, making green synthesized AgNPs attractive candidates for colon-targeted drug delivery and future nanomedicine applications.

Although encouraging results have been obtained from in vitro and preclinical investigations, further studies are required to optimize nanoparticle formulations, evaluate long-term toxicity, and establish clinical efficacy. Continued advances in green nanotechnology are expected to facilitate the translation of these promising nanoparticles into safe and effective therapeutic agents for the management of colon cancer.[17-19,22]

X. COLON-TARGETED DRUG DELIVERY USING GREEN SYNTHESIZED SILVER NANOPARTICLES

Colon-targeted drug delivery has emerged as an effective strategy for improving the therapeutic management of colorectal cancer by delivering anticancer agents directly to the site of disease while minimizing systemic toxicity. Conventional chemotherapy often results in non-specific drug distribution, exposing healthy tissues to cytotoxic agents and causing adverse effects such as gastrointestinal toxicity, bone marrow suppression, neurotoxicity, and immunosuppression. Nanotechnology-based drug delivery systems have been developed to overcome these limitations by enhancing drug targeting, improving bioavailability, and providing controlled release of therapeutic agents. Green

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synthesized silver nanoparticles (AgNPs) have attracted considerable attention in this field because of their intrinsic biological activity, excellent drug-loading capacity, and favorable biocompatibility resulting from plant-derived surface phytochemicals.

The nanoscale dimensions of AgNPs facilitate efficient transport across biological barriers and promote their preferential accumulation within tumour tissues through the enhanced permeability and retention (EPR) effect. In addition, the surface of green synthesized nanoparticles can be modified with polymers, antibodies, peptides, or other ligands to improve active targeting of cancer cells. Such functionalization enhances selective cellular uptake and reduces exposure of normal tissues to cytotoxic drugs. These properties make AgNPs attractive carriers for colon-targeted drug delivery systems.

Drug molecules can be adsorbed onto or encapsulated within silver nanoparticle-based delivery systems, enabling controlled and sustained release at the tumour site. Controlled drug release maintains therapeutic drug concentrations for prolonged periods, thereby reducing dosing frequency and improving patient compliance. Furthermore, nanoparticle-mediated drug delivery enhances intracellular drug accumulation, improves drug stability, protects labile molecules from premature degradation, and increases therapeutic efficacy. These characteristics are particularly important for anticancer agents with poor aqueous solubility or limited oral bioavailability.

Green synthesized AgNPs also possess intrinsic anticancer properties in addition to serving as drug carriers. Their ability to induce oxidative stress, mitochondrial dysfunction, apoptosis, and inhibition of tumour cell proliferation complements the pharmacological activity of loaded chemotherapeutic agents, producing synergistic therapeutic effects. Consequently, nanoparticle-mediated combination therapy has emerged as a promising strategy for improving colon cancer treatment while reducing the adverse effects associated with high-dose chemotherapy.

Several experimental studies have demonstrated encouraging outcomes following the use of AgNP-based drug delivery systems in colon cancer models. Improved cellular uptake, enhanced tumour selectivity, reduced systemic toxicity, and greater inhibition of tumour growth have been consistently reported in preclinical investigations. Although additional pharmacokinetic studies and clinical trials are required, current evidence indicates that green synthesized silver nanoparticles represent a promising platform for the development of safer and more effective colon-targeted therapeutic systems.[5,21-26,28]

XI. RECENT ADVANCES AND RESEARCH PROGRESS

Over the past decade, substantial progress has been achieved in the application of green synthesized silver nanoparticles for colon cancer therapy. Advances in nanotechnology, medicinal plant research, and pharmaceutical sciences have facilitated the development of highly stable and biologically active nanoparticles with improved therapeutic performance. The incorporation of plant-derived phytochemicals during nanoparticle synthesis has significantly enhanced antioxidant activity, biocompatibility, colloidal stability, and anticancer efficacy compared with conventionally synthesized nanoparticles.

Recent investigations have explored the synthesis of AgNPs using extracts obtained from a wide variety of medicinal plants. Plant-mediated nanoparticles have demonstrated potent cytotoxic activity against colon cancer cell lines through multiple mechanisms, including reactive oxygen species generation, mitochondrial dysfunction, DNA damage, apoptosis, inhibition of cell proliferation, and suppression of tumour angiogenesis. These findings support the growing role of green nanotechnology in the development of environmentally sustainable and biologically effective anticancer therapies.

An important recent advancement is the use of polyherbal extracts for nanoparticle synthesis. Polyherbal formulations combine bioactive phytochemicals from multiple medicinal plants, resulting in synergistic effects that improve nanoparticle formation, stability, antioxidant potential, and therapeutic activity. Several studies have reported that polyherbal-mediated AgNPs exhibit stronger anticancer activity than nanoparticles synthesized using a single plant extract, suggesting that phytochemical diversity contributes significantly to improved biological performance.

IJARSCT
International Journal of Advanced Research in Science, Communication and Technology
International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal
ISSN: 2581-9429 Volume 6, Issue 2, August 2026 Impact Factor: 8.2

Current research has also focused on optimizing synthesis parameters such as pH, temperature, reaction time, silver ion concentration, and plant extract composition to obtain nanoparticles with reproducible physicochemical characteristics. Improvements in characterization techniques, including UV–Visible spectroscopy, FTIR, SEM, TEM, XRD, dynamic light scattering, and zeta potential analysis, have enabled more accurate evaluation of nanoparticle quality and stability. Standardization of these procedures is essential for ensuring reproducibility and facilitating future clinical translation.

Despite encouraging laboratory findings, most available evidence is derived from in vitro experiments and animal studies. Limited clinical data remain one of the major barriers to routine therapeutic application. Future investigations should emphasize comprehensive toxicological evaluation, pharmacokinetic studies, large-scale manufacturing, quality control, and well-designed clinical trials to establish the long-term safety and therapeutic efficacy of green synthesized AgNPs. Integration of nanotechnology with precision medicine, targeted drug delivery, and advanced pharmaceutical formulations is expected to further enhance treatment outcomes and accelerate the clinical application of green synthesized silver nanoparticles in colon cancer management.[15,16,27-30]

XII. ADVANTAGES OF GREEN SYNTHESIZED SILVER NANOPARTICLES

Green synthesized silver nanoparticles (AgNPs) have emerged as promising nanomaterials for biomedical applications because they combine the therapeutic potential of nanotechnology with environmentally sustainable synthesis methods. Unlike chemically synthesized nanoparticles, green synthesized AgNPs utilize plant-derived phytochemicals as natural reducing and stabilizing agents, thereby eliminating the use of hazardous chemicals and improving biological compatibility. This eco-friendly approach has attracted considerable attention for pharmaceutical and anticancer applications.

One of the major advantages of green synthesized AgNPs is their enhanced biocompatibility. The phytochemicals present on the nanoparticle surface reduce toxicity and improve interactions with biological systems. These naturally occurring compounds also contribute antioxidant, anti-inflammatory, and antimicrobial properties that complement the intrinsic biological activity of silver nanoparticles.

Green synthesis is a simple, cost-effective, and environmentally sustainable process. It requires relatively mild reaction conditions, lower energy consumption, and readily available medicinal plant materials. Consequently, the production process is economically attractive and generates minimal environmental pollution compared with conventional physical and chemical synthesis methods.

From a therapeutic perspective, green synthesized AgNPs exhibit significant anticancer activity through multiple molecular mechanisms, including oxidative stress induction, mitochondrial dysfunction, DNA damage, apoptosis, and inhibition of tumour cell proliferation. Their nanoscale dimensions facilitate enhanced cellular uptake and selective accumulation within tumour tissues, improving therapeutic efficacy while reducing systemic toxicity.

In addition to their intrinsic biological activity, AgNPs function as efficient drug delivery carriers. Their high surface-area-to-volume ratio enables efficient drug loading, controlled release, improved bioavailability, prolonged circulation time, and enhanced intracellular drug delivery. These properties make green synthesized AgNPs highly promising candidates for colon-targeted drug delivery and combination cancer therapy.[6-10,21]

XIII. LIMITATIONS AND CHALLENGES

Despite their considerable therapeutic potential, several scientific and technical challenges continue to limit the clinical translation of green synthesized silver nanoparticles. One of the major concerns is the lack of standardized synthesis protocols. Variations in medicinal plant species, geographical origin, extraction procedures, phytochemical composition, reaction conditions, and purification methods frequently produce nanoparticles with inconsistent physicochemical characteristics, affecting reproducibility and therapeutic performance.

Large-scale production also remains challenging. Although laboratory-scale synthesis has demonstrated encouraging results, industrial manufacturing requires highly reproducible protocols capable of producing nanoparticles with

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uniform particle size, morphology, stability, and biological activity. The absence of internationally accepted quality control standards further complicates pharmaceutical development.

Another important limitation is the insufficient availability of long-term toxicological and clinical safety data. Most published investigations have been restricted to in vitro studies or experimental animal models. Comprehensive pharmacokinetic evaluation, biodistribution studies, chronic toxicity assessment, immunogenicity testing, and human clinical trials remain limited. Therefore, additional research is required before routine clinical application can be recommended.

Nanoparticle stability during storage and transportation also requires further optimization. Aggregation, oxidation, and physicochemical instability may influence biological activity and reduce therapeutic effectiveness. The development of standardized storage conditions and advanced formulation strategies will therefore be essential for future commercialization.[25-28]

XIV. FUTURE PERSPECTIVES

Green nanotechnology is expected to play an increasingly important role in the development of next-generation cancer therapeutics. Future investigations should focus on optimizing plant-mediated synthesis methods to achieve reproducible nanoparticle production with well-defined physicochemical characteristics suitable for pharmaceutical applications.

The integration of green synthesized AgNPs with targeted drug delivery systems, ligand-based surface modification, polymeric carriers, and stimulus-responsive nanoplatforms may significantly improve tumour specificity and therapeutic efficacy. Advances in nanomedicine, molecular biology, and pharmaceutical technology are expected to facilitate personalized treatment strategies capable of improving clinical outcomes while minimizing systemic toxicity.

Artificial intelligence and computational modelling may further accelerate nanoparticle design by predicting synthesis conditions, optimizing particle characteristics, and identifying suitable phytochemical combinations for enhanced therapeutic performance. Such interdisciplinary approaches could substantially reduce research costs and improve formulation development.

Future research should also prioritize large-scale manufacturing, quality assurance, regulatory standardization, pharmacokinetic evaluation, and multicentre clinical trials. Establishing internationally accepted guidelines for nanoparticle synthesis and characterization will be essential for successful translation from laboratory research to clinical practice.[26-30]

XV. CONCLUSION

Colon cancer remains one of the leading causes of cancer-related morbidity and mortality worldwide despite significant advances in diagnosis and treatment. The limitations associated with conventional therapeutic approaches, including systemic toxicity, poor drug selectivity, multidrug resistance, and tumour recurrence, have stimulated the search for safer and more effective therapeutic alternatives.

Green synthesized silver nanoparticles represent an innovative and environmentally sustainable nanotechnology-based platform with considerable potential for colon cancer therapy. Their unique physicochemical properties, excellent drug-loading capacity, improved biocompatibility, and intrinsic anticancer activity enable efficient inhibition of tumour progression through multiple molecular mechanisms, including oxidative stress, mitochondrial dysfunction, DNA damage, apoptosis, and suppression of angiogenesis. Furthermore, their application in colon-targeted drug delivery offers significant advantages by improving therapeutic selectivity, enhancing bioavailability, reducing systemic toxicity, and increasing treatment efficacy.

Although encouraging experimental evidence has demonstrated the promise of green synthesized AgNPs, further investigations are required to establish standardized synthesis methods, evaluate long-term safety, optimize pharmaceutical formulations, and validate clinical efficacy through well-designed human studies. Continued advances in green nanotechnology, targeted drug delivery, and translational nanomedicine are expected to facilitate the

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ISSN: 2581-9429 Volume 6, Issue 2, August 2026 Impact Factor: 8.2

development of safer, more effective, and clinically applicable therapeutic strategies for the management of colon cancer.[1-5,22,28-30]

Acknowledgement

The authors express their sincere gratitude to their respective institution for providing academic support, library facilities, and encouragement during the preparation of this review article.

Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this review article.

Funding

The authors received no specific financial support from any funding agency in the public, commercial, or not-for-profit sectors for the preparation of this manuscript.

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International Journal of Advanced Research in Science, Communication and Technology
International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal
ISSN: 2581-9429 Volume 6, Issue 2, August 2026 Impact Factor: 8.2

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