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A REVIEW ON GOLD NANOPARTICLES IN NOVEL DRUG DELIVERY SYSTEMS

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Abstract :

Nanoparticles(NPs) are solid,spherical particles of size 100nm,prepared from polymers(natural or synthetic).Hydrophobic and hydrophoilic drugs,vaccines and macromolecules can be delivered by using NPs,which may also allow controlled drug delivery or a targeted administration to a specific cell or organ.

Nanotechnology is an emerging scientific discipline with numerous applications in the field of biomedicine and manufacturing new materials.Plenty and pretty applications of nanoparticles have gained utmost priority now-a-days due to their versatile flexibility in wide range of applications.Some nanoparticles also show bactericidal effects and hence a high surface to volme ratio.

Due to their unique properties, small size and high area to volume ratio ,gold nanoparticles.Show special advantages in this field among nanoparticles ,biosynthesized gold NPs remarkable applications in different and chemicals sensors ,heavy metals ion detection,electrical coatings.During the last two decades ,NPs have been extensively investigated and developed in imaging applications due to the superior narrow range of emission,photo stability,broad excitation wavelength,quantum dots have attracted the attention from scientists and engineers interested in drug targeting,biomarkers and sensors.

Keywords: *Gold nanoparticles, Nanotechnology, drug delivery, biomedical applications.*

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INTRODUCTION :

In recent years, nanotechnology has become one of Physics, Chemistry, Engineering and Biology's most important and exciting front lines. It shows great promise to give us many breakthroughs that in a wide range of applications will change the direction of technological advances. The nanotechnology research area is interdisciplinary, covering a wide range of topics ranging from nanoparticles catalyst chemistry to quantum dot laser physics. As a result, researchers need to go beyond their expertise in any particular area to appreciate the wider implications of nanotechnology and learn how to contribute to this exciting new field. Particles of 1-100 nm size are called nanoparticles, whether dispersed in gaseous, liquid, or solid media. Because NPs are larger than individual atoms and molecules but smaller than bulk solids, materials in the regime of nanometres size show an intermediate behaviour between that of a macroscopic solid and that of an atomic or molecular system.

Nanoparticles great potential is due to the unique properties of elements when their size is reduced to the level of Nano-meter. Overall, particle properties above the Nano-meter size are not significantly different from their bulk counterparts. However, their physical and chemical properties can drastically change if particles are reduced to their Nano-level.Changes in these properties are highly influenced by the shape, size and nature of their surroundings[6].

Because of their increased biocompatibility, stability and oxidation resistance, AuNPs are considered one of the most convenient carrier systems. Colloidal gold is therefore applicable in different fields of medical research, including biosensing and bio detection, catalysis and bioelectronics, drug delivery carriers and macromolecular carriers, bioimaging and photo hyperthermia[7].

Collectively, one can conclude that, during the manufacturing process of Nanosystems with potential applications in controlled and targeted drug therapy, the gold nanoparticle surface charge plays a critical role.Taking into account these unique properties of AuNPs, but also the current challenges to personalized medical care, gold nanoparticles have the attractive potential for engineering novel nanostructured tools for selective tumour targeting and imaging, thus highlighting their tremendous potential in unconventional cancer diagnosis and treatment[8].

AuNPs surface load, estimated in terms of zeta potential, facilitates their physicochemical stability and further implementation in the process and bioaccumulation of cells.As many previous studies have shown, the level of toxicity assigned to AuNPs is heavily dependent on the particle surface load, so the positively charged gold nanoparticles cause cell death at lower concentrations, whereas the neutrally charged particles determine cell death at significantly higher concentrations[9]. Metal nanoparticles, specifically gold nanoparticles, are widely used in biotechnology and biomedicine because they have a large bioconjugation with molecular samples and many optical properties that are mainly concerned with localized plasmon resonance[10].

Nanoparticles were used to enhance the drug delivery system's selectivity and efficiency because they act as drug release mediators.Nanoparticles are extremely small in size and have a high surface area, so their surfaces are available for further modification with hydrophobic, hydrophilic, cationic, anionic or any neutral features in the surrounding environment, so they have a lot of application in biological sciences.Nanoparticular - based drug delivery systems have proven to be a great way to target malignant brain tumours where conventional therapy is less effective. The unique property of nanoparticles to accumulate and interact with tumour cells is enhanced permeability and retention (EPR)[1]

GOLD NANO PARTICLES

Gold nanoparticles provide an outstanding material for study due to the fact that they are one of the most stable, non-toxic, and easy to synthesize nanoparticles and exhibit various fascinating properties like assembly of various types and quantum sizeeffect.[6] The optical behavior of gold nanoparticles is dependent on their surface plasmon resonance (SPR), located in a wide region ranging from visible to the infrared region of the spectrum, which is determined by collective oscillation of conducting electrons. The range of the spectrum depends on various features of gold nanoparticles, including size and shape.[9]Methods have been developed to synthesize these materials reproducibly, which can further be modified using countless chemical functional groups. Many new sensitive and specific assays are based on the gold nanoconjugates.

Gold nanoparticles have emerged as an excellent candidate for the application in delivery of various payloads to the target site.[10,11] These payloads range from small drug molecules including drugs to large biomolecules like DNA, RNA, and

proteins. Some drugs molecules do not require modification of a monolayer of gold nanoparticles for their delivery and can be directly conjugated with gold nanoparticles through physical absorption and ionic or covalent bonding.[12] Whereas for the delivery of other payloads, gold nanoparticles require functionalization like PEGlyation,[13] peptide and amino acid conjugation,[14,15] or functionalization with oligonucleotides.[16] Apart from that, another prerequisite for the efficient delivery of therapeutic agents is their release. Various internal stimuli (glutathione, pH and enzymes)[17–19] and external stimuli (light, etc.)[20] have been investigated for the efficient release of these payloads from gold nanoparticles.Due to the vast amount of information available and the level at which it is being renewed we have chosen the generalized data from the past few years to present this review encompassing the most promising application of gold nanoparticles in drug delivery.

FIGURE 1 | The main morphologies of AuNPs.

Synthesis of AuNPs

For the synthesis of AuNPs, there are two basic strategies that are used, which are "Top-Down" and "Bottom-Up" approaches. The top-down approach involves the synthesis of AuNPs starting from bulk material and cracking it into nanoparticles using different methods. In contrast, the bottom-up approach synthesizes nanoparticles starting from atomic level. Figure 1 shows the basic steps that are involved in the top-down and bottom-up approaches. Synthesis methods that involve the top-down approach include laser ablation,[21] ion sputtering,[22] UV and IR irradiation,[23,24] and aerosol technology,[25] whereas the reduction of Au3+ to Au0 is the bottom-up approach.

The formulation of AuNPs involves two main stages:

In the first stage the gold precursor, which is usually an aqueous gold salt solution, is reduced to gold nanoparticles using a specific reducing agent like citrate.

In the second stage the stabilization of gold nanoparticles is done by a specific capping agent. The capping agents hinder the agglomeration of metallic nanoparticles.

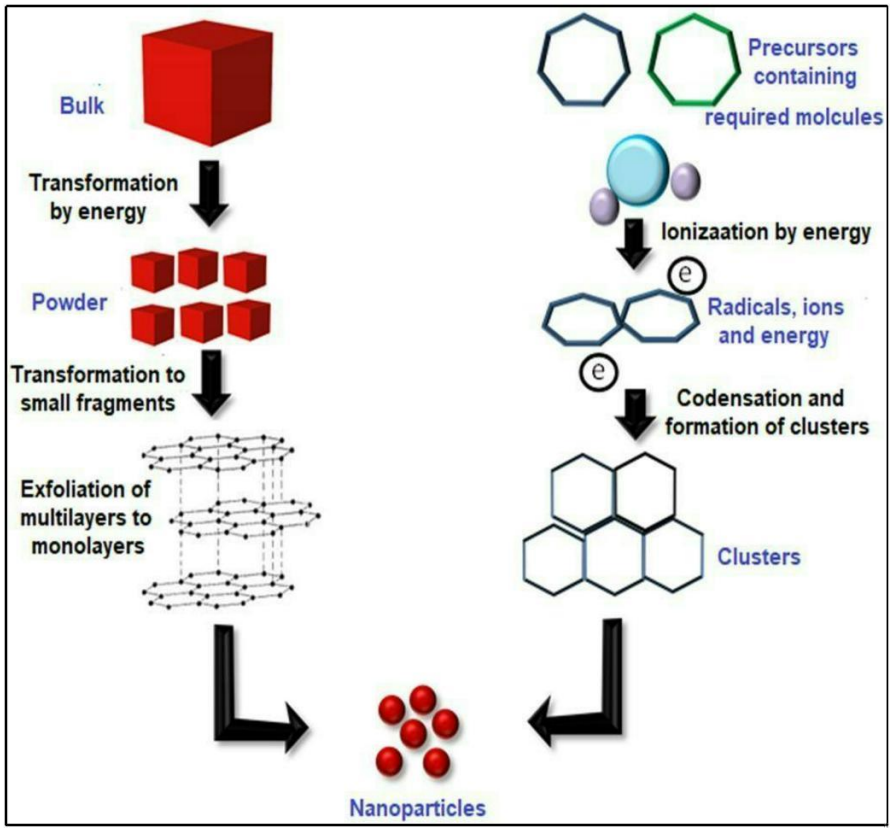

Figure 2 Top-down and bottom-up approaches for the synthesis of NPs. The top-down approach involves the transformation of bulk material by using energy to produce the powder form which is then transformed into smaller fragments with multiple layers and then to the monolayers leading to the formation of nanoparticles. On the other hand, the bottom-up approach uses the precursor molecules which are then ionized by using energy. Radicals, ions, and electrons thus produced are condensed to form clusters which are then transformed to nanoparticles.

Chemical Synthesis

Turkevich Method

This method for the synthesis of AuNPs was first reported in 1951. It is one of the most commonly used techniques for formulation of spherical AuNPs. AuNPs prepared using this method have the size in the range of 1–2 nm.[26] The basic principle of this technique involves the reduction of gold ions (Au3+) to produce gold atoms (Au0) by using some reducing agents like amino acids,[27] ascorbic acid,[28] UV light, or citrate.[29,30] Stabilization of AuNPs is carried out by using different capping/stabilizing agents. At the beginning, the applications of Turkevich method were finite because of the limited range of AuNPs that could be synthesized by this technique.

With the passage of time several advancements in the basic method have enabled researchers to extend the size range of particles synthesized using this method. In 1973, it was established thatby varying the ratio of reducing as well as stabilizing agents, AuNPs of particular size with the range from 16–147 nm can be produced.[31] Figure 3A shows the basic method involved in the Turkevich method.

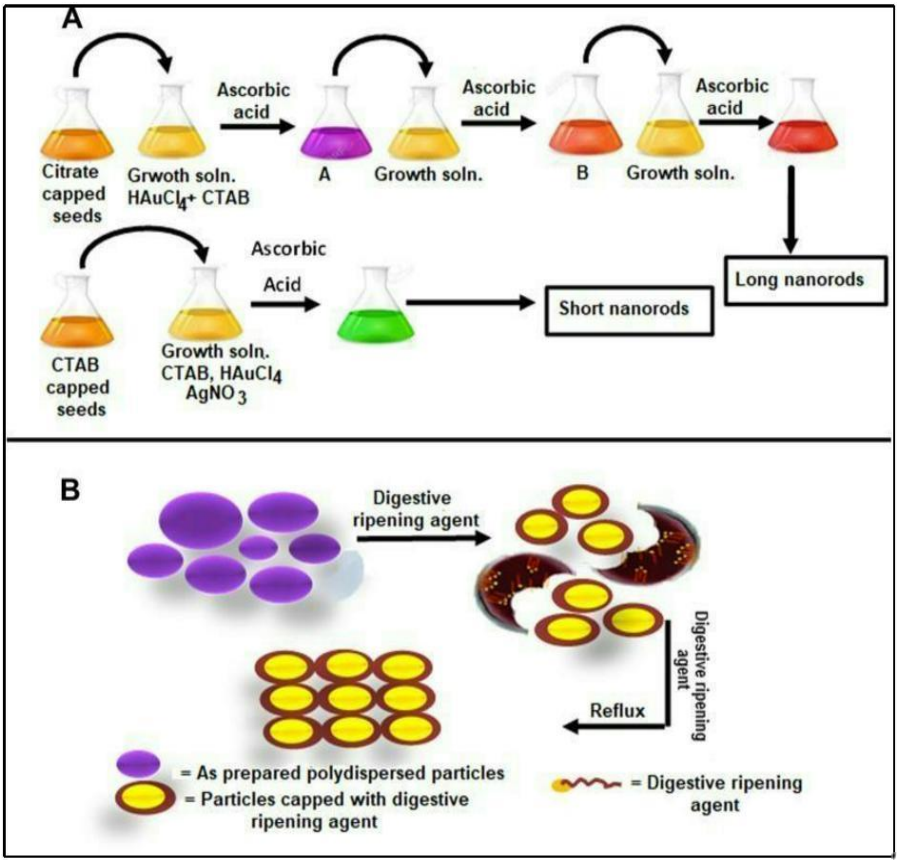

Figure 3 (**A**) Turkevich method for the synthesis of AuNPs. (**B**) Series of steps involved in the Burst method for the synthesis of AuNPs.

The Brust Method

This method was first reported in 1994 and involves a two- phase reaction to synthesize AuNPs with the size range of 1.5–5.2 nm by using organic solvents.The method encompasses the use of a phase transfer such as tetraoctylammonium bromide to carry out transferring of gold salt to organic solvent from its aqueous solution (eg, toluene). The gold is then reduced by the use of a reducing agent such as sodium borohydride along with an alkanethiol. The alkanethiol carries out the stabilization of AuNPs.As a result of this reaction the color changes from orange to brown. Figure 3B shows the schematic illustration of main steps involved in Brust method. **Seed-Mediated**

Growth

The previous two methods can synthesize only spherical AuNPs; however, they can also be formulated in a number of geometries and shape such as rods. The most commonly used technique to synthesize rod shaped AuNPs is seed-mediated growth. This method is based on the fundamental principle which involves first synthesizing seed particles by reducing gold salts. This reaction is done in the presence of reducing agents like NaBH4. The next step involves the transferring of the seed particles to a metal salt and a weak reducing agent like ascorbic acid which prevents further nucleation and speeds up the synthesis of AuNPs of rod shape. Shape and geometry of gold nanoparticles depends on the concentration of reducing agents and seeds. Figure 4A shows schematic illustration of seed-mediated growth of short and long gold nanorods as reported by a study.

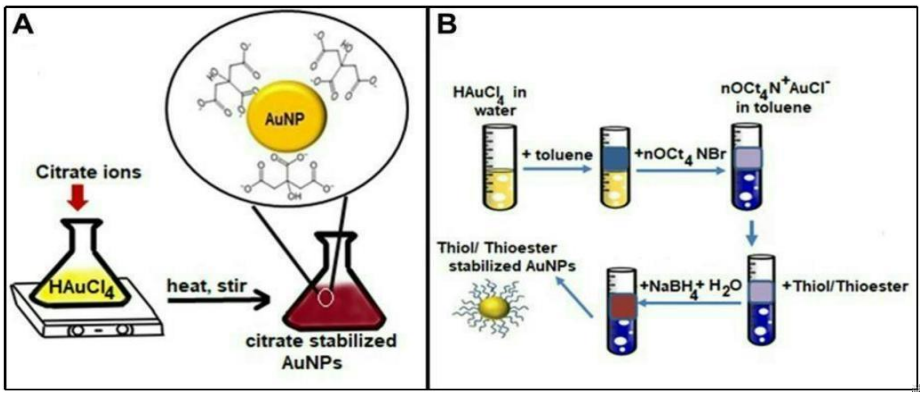

Figure 4 Series of steps involved in the synthesis of AuNPs. (**A**) Seed-mediated method. (**B**) Digestive ripening method.

Digestive Ripening

Digestive ripening is considered to be a convenient method to prepare monodispersed gold nanoparticles in the presence of excessive ligands (digestive ripening agents). The basic process comprises heating a colloidal suspension at high temperatures (~138ºC) for 2 minutes and then heating at 110ºC for 5 hours by using alkanethiol, as shown in Figure 4B. Temperature is the major factor for determining the size distribution of the gold colloids.

In addition to these methods, other methods involve the use of ultrasonic waves for the synthesis of AuNPs.

Advantages and Limitations of the Methods

Turkevich method is a fairly uncomplicated and reproducible procedure for the formulation of spherical particles with the size range 10–30 nm. But, as the size of particles increases above 30 nm, they become less spherical in shape with broader size distribution. Moreover, this reaction gives low yield and involves the use of only water as a solvent. Brust method, on the other hand,

involves an easy strategy for the formulation of thermal and air-stable AuNPs having controlled size and less dispersity. One possible limitation of Brust method is synthesis of AuNPs which are less dispersed and used of organic solvents immiscible with water, therefore, limiting their biological applications. Seed-mediated growth is a reliable method for the synthesis ofrod-shaped AuNPs, but various factors affect the size of rod and so must be carefully controlled.

In a study when higher concentrations of HAuCl4 were used it produced bigger seed rods with smaller aspect ratios. Temperature also plays a significant role in the synthesis of rods and at higher temperatures rods with lower aspect ratio were produced. Also, the number of seeds added to the reaction mixture must be critically considered to stimulate the growth of rods. The digestive ripening method is also an easy and valuable chemical technique to produce monodispersed nanoparticles. Another benefit of this strategy is the high yield of nanoparticles. A possible disadvantage of the digestive ripening method is that controlling the shape of nanoparticles via the digestive ripening process is difficult as it involves very high temperatures.

In addition, chemical methods are inherited with their own limitations which include environmental and biocompatibility concerns. Some of the chemicals that are used in the synthesis of gold nanoparticles during chemical synthesis can affect our environment and are the cause of risks for administering them into the living organisms, thus limiting the biological applications of such NPs.Therefore, various biological methods have been devised for the synthesis of AuNPs to limit these concerns.

Biological Synthesis

Recently, efforts have been made for biological synthesis of AuNPs, which is a clean, dependable, and bio-friendly alternative to harsh chemicals used in chemical synthesis reactions. The biological resources used in synthesis of nanoparticle range from simple bacterial cells to complex eukaryotes. Interestingly, the capability of organisms in synthesis of metal nanoparticles has given rise to a new thrilling approach toward the development of these biological nano-factories. A plethora of organisms have been reported to carry out successful synthesis of AuNPs, ranging from bacteria to plants, algae, and fungi.

Bacteria

Microorganisms have been reported to be an excellent candidate for the synthesis of both intracellular and extracellular AuNPs.The negatively charged cell wall of bacteria can electrostatically interact with positively charged Au(III) ions. During the intracellular synthesis, gold ions are transported inside the cell where enzymes and biomolecules carry out the synthesis of AuNPs. On the other hand, during extracellular synthesis the gold ions are trapped on the cell membrane by membrane enzymes. These enzymes on the membrane or reductase enzymes secreted out by the microbial cell can carry out the synthesis process outside the bacterial cell. Extracellular synthesis, however, is more fascinating as it does not require extra downstream processing steps which are required for the separation of nanoparticles from the intracellular matrix. A study has shown that, during the extracellular synthesis reaction NADPH-dependent enzymes are secreted by bacteria which can reduce Au(III) ions to Au0 such as nitrate reductase secrete by *Pseudomonas denitrificans*. The results showed that the action reductase enzyme diminished once AuNPs had been synthesized. Shah et al reported that both NADH and NADH- dependent enzymes function as a scaffold or nucleating agent for the synthesis reaction. Singh et al reported that *Rhodopseudomonas capsulate* secreted NADH and NADH-dependent enzymes during extracellular synthesis of AuNPs. The transfer of electrons from NADH carried by NADH- dependent enzyme causes the reduction of Au(III) to Au0, resulting in the synthesis of AuNPs.*Thermomonospora* sp. (Order: Actinomycetes) was reported to carry out intracellular enzymes mediated synthesis of AuNPs by achieving the reduction of Au(III) ions at the surface of membrane and mycelia. Similarly, *Shewanella algae* efficiently carried out enzymes mediated bioreduction of $AuCl4^-$ ions to AuNPs which were found to be dispersed in periplasmic membrane of bacterium. Certain materials produced by microbial cells like proteins, enzymes, and organic substances can act as capping agents to stabilize nanoparticles and, hence, prevent their agglomeration. Micro-organisms possess certain reductase enzymes which can reduce metal salts to metal nanoparticles with narrow size distributions and monodispersity. By altering the essential growth parameters, the shape and size of AuNPs can be controlled. Synthesis of AuNPs using bacteria is a tedious reaction and requires additional precautionary measures while handling bacteria, and also takes hours and days as bacterial cultural is a time consuming process. These drawbacks have limited the use of bacteria for the synthesis of AuNPs.

Fungi

Fungi have also been used as a biological source for the synthesis of AuNPs. Fungi secrete a number of biomolecules, including metabolites and extracellular enzymes, such as hemicellulose, acetyl xylem esterase, 3-glucanase, cell wall lytic enzyme β-1, etc., which have been reported to play a role during the synthesis of metallic nanoparticles. Numerous studies have reported the synthesis of gold nanoparticles using unicellular and multicellular fungi. A fungal species *Fusarium oxysporum* was used in a study for the extracellular synthesis of Au- Ag alloy NPs by the reduction action of nitrate-dependent enzyme and shuttle quinone. A fungal species *Verticillium* has also been reported to carry out intracellular synthesis of AuNPs. AuNPs were found to be trapped in the cell membrane and the cell wall of fungi, indicating that $Au3+$ ions were bio-reduced by the reduction action of reductase enzymes in fungi. A study on the biosynthesis of AuNPs from *Phanerochaete chrysosporium* proved that laccase was the enzyme secreted by the fungi for extracellular synthesis of AuNPs and, for intracellular synthesis, ligninase was found to be responsible.

Plants

Phytonanotechnology has gained attention with time as it comprises an eco-friendly, cheap, andrapid process for the synthesis of nanoparticles. A number of studies have reported biosynthesisof AuNPs using different plants or plant extracts involving the use of harmless bio- componentsfrom plants to carry out the reduction and capping of AuNPs, reducing the waste generation andlimiting the requirement for additional purification steps. Numerous bio-components present inplants such as flavonoids, phytosterols, quinones, etc., play a role in the synthesis of AuNPsbecause of the functional groups which speed up the reduction and stabilization of AuNPs.Although nearly every part of plants has been reported to successfully carry out the synthesis ofAuNPs, leaves are most commonly used. The difference in the level of various compounds presentin different plants and even in different parts of a plant affects the synthesis of AuNPs. Forexample, a study has reported the effect of difference in level of phenolic contents present in leavesand fruit of Garcinia mangostana plant on

the synthesis of AuNPs. As the leaves are rich inphenolic content so the rate of synthesis of AuNPs was faster in the presence of leaves than fruit.Moreover, recently the synthesis of gold nanoparticles using medicinal plant *Acorus calamus* and*Cassia auriculate* has been reported.

Reactive compounds; Lignans [(+)-pinoresinol, (+)-medioresinol], alkaloids, flavonoids, steroids(sitosterol- 3-0-glucoside), and terpenoids present in the leaves of *Justicia glauca* have beenreported to complete the synthesis reaction of AuNPs in 1 hour. AuNPs had spherical andhexagonal morphology and were 32 nm in size.71 Leaves of the *Terminalia arjuna* plant alsocarried out the synthesis of AuNPs within 15 minutes. AuNPs synthesized in this study were 20–50 nm in size and had spherical morphology. The author claimed that the reactive compoundsArjunetin, leucoanthoc-yanidins and hydrolysable tannins present in leaves of *Terminalia arjunacontributed to the synthesis of AuNPs. Similarly, the leaves of olive plant and Cassia auriculatawere shown to complete the synthesis reaction of AuNPs in 20 minutes and 10 minutes,respectively. The active metabolites and biomolecules in the leaves of the olive plant are proteins,* oleuropein, apigenin-7-glucoside, and luteolin-7-glucoside, which resulted in the formation of spherical and anisotropic AuNPs with the size range of 50–100 nm. Polysaccharides and flavonoids are the major active substances in the leaves of *Cassia auriculata* and AuNPs synthesized from leaves of this plant were 15–25 nm in size and had spherical and anisotropic morphology.*Mangifera indica* leaves used by Philip synthesized spherical AuNPs within 2 minutes

of reaction time. The size of AuNPs was found to be in the range of 17–20 nm. Terpenoids, flavonoids, and thiamine are the active compounds present in mango fruit, which might have contributed to the synthesis of AuNPs.

Apart from leaves, various other parts of plants, including fruits, roots, stems, etc., have been used for the synthesis of AuNPs. The fruit of *Citrus maxima* was used in one study and synthesized spherical AuNPs with the size range of 15–35 nm within 5 minutes of reaction time. Proteins, terpenes, and ascorbic acid were the major compounds that were claimed to act as reducing agents during reaction. The high phenolic content of *Sambucus nigra* (elderberry) was the major factor in the synthesis of AuNPs. Apart from that, flowers of *Lonicera Japonica*contain amino acids as active compounds and successfully synthesized AuNPs of triangular and tetrahedral morphology with the size range of 8 nm in the reaction time of 1 hour.[7] Similarly flowers of the *Moringa oleifera* plant synthesized AuNPs of size 3–5 nm. This plant was reported to contain a high content of flavonoids, carotenoids, phenols, sterols, and amino acids, which were claimed to be responsible for carrying out the reduction reaction during the synthesis process. Various types of roses have been demonstrated to possess the reducing ability for the synthesis of AuNPs.[7,8] Similarly, banana and mango peels can synthesize AuNPs with the sizes 50 nm and 6.03±2.77 to 18.01±3.67 nm, respectively. Banana peels synthesized spherical shaped AuNPs and mango peel synthesized quasi-spherical shaped AuNPs.

The reaction time for both processes was 20 and 25 minutes, respectively.[18,24] Apart from the above- entioned parts of plants, rhizomes of turmeric,[23]yam beans,[24] ginger,[25] and seeds of cocoa,[26] pulp of green pepper,[27] bark of bay cedar,[28] galls of zebra wood,[29] latex of *Hevea brasiliensis*,[30] nuts of *Areca catechu*,[31] and effluent from palm oil mill92 were found to carry out the synthesis of AuNPs.

Algae

There are a few studies which have demonstrated the synthesis of gold NPs using algae. A few species of both marine and fresh algae were used in these studies. Among the marine red algae, *Gracilaria corticata*, *Acanthophora spicifera*, and *Galaxaura elongata*, and marine brown algae, *Stoechospermum marginatum*, *Ecklonia cava*, *Sargassum wightii*,*Cystoseira baccata*,*Laminaria japonica*, and *Turbinaria conoides* have been previously reported to carry out the synthesis of AuNPs. On the other hand, biomass from freshwater algae including *Prasiola crispa*, *Lemanea fluviatilis*, and *Chlorella* pyrenoidusa can also synthesize AuNPs. Previous studies have shown that hydroxyl and carbonyl groups present in algal biomass can act as reducing agents for carrying out the synthesis of AuNPs. It has also been shown that these group can also act as the capping agent for gold nanoparticles.Table 1 shows the list of various organisms that have been reported to carry out successful synthesis of AuNPs.

Biomolecules

Molecules synthesized by living organisms to speed up their biological processes of the body are known as biomolecules and these include macromolecules such as amino acids, nucleic acids, carbohydrates, and lipids. Previous studies have reported the synthesis of gold nanoparticles using chitosan which does not only act as a reducing agent but also as a stabilizing agent during synthesis reaction. Apart from that, starch is another polysaccharide used for the synthesis of AuNPs. In an alkaline environment starch can be degraded into short chains having carboxyl groups and the – OH group of carboxylic acid can reduce Au3+ ions to gold nanoparticles. Among proteins, consensus sequence tetratricopeptide repeat proteins and corn protein, α-zein can be use to carry out the synthesis reaction of AuNPs. The biological method of synthesis of AuNPs can conveniently overcome the complications of biosafety of the chemicals used for the generation of AuNPs.

Advantages and Limitations of Biological Synthesis

Synthesis of AuNPs using biomass from bacteria is an advantageous process as some species of bacteria are not affected by the presence of heavy metals. Also, the extracellular synthesis approach produces pure nanoparticles as compared to the intracellular synthesis process which requires additional purification steps. Conversely, culturing of bacteria is a slow and tedious process so the synthesis reaction of AuNPs can take a long time comprising hours and even days. On the other hand, fungi produce a large quantity of proteins and reactive compounds. Therefore, the reaction process can be easily scaled up. Moreover, as compared to bacteria it is easier to culture and grow

fungi. But preparing biomass from fungi for the synthesis reaction requires careful steps as it is complicated to separate mycelia from culture filtrates.Manipulation of the genetic makeup. Moreover, changing the concentration of gold salt used for the synthesis reaction, pH, and temperature can also provide control over the size and geometry of AuNPs. Derjaguin Landau VerweyOverbeek theory (DLVO) explains the whole process for stabilization of metallic nanoparticles.The stabilization of NPs done by using various capping agents can be divide into three different categories, including steric, electrostatic, and unification of steric and electrostatic stabilization.

Electrostatic Stabilization

Ionic groups present in the liquid dispersion media can attach to the surface of a colloidal nanoparticle giving rise to a charged layer. As a result, an equal number of oppositely charged ions will border the colloidal nanoparticles giving rise to overall electro-neutral double layers. This stabilization which involves an electric double layer originating from the presence of both repulsive as well as attractive forces between the nanoparticles as a result of the action of some ionic composites is shown in Figure 5A. These ions include polyoxyanions, carboxylates, as well as fluorides. This type of stabilization involving electrostatic repulsions inhibits the agglomeration

of nanoparticles in the solution phase. Electrostatic stabilization is regulated by controlling certain significant variables including pH, concentration, and temperature.

Steric Stabilization

Steric stabilization hinders the free movement of metal nanoparticles during synthesis reactions. Stabilizing agents used in this type of stabilization include various functional groups such as hydroxyl groups, surfactants, and different oligomers/polymers. This results in the generation of a protective layer by the assimilation of the stabilizing agent at the outer surface of nanoparticles which plays an important role in the stability of metallic nanoparticles. The mechanism of steric stabilization is shown in Figure 5B.

Electrosteric Stabilization of AuNPs

The stability of metallic nanoparticles in solution phase can also be maintained by another type of stabilization which involves unification of electrostatic and steric stabilization. A polyelectrolyte employed as a polymeric surfactant gives combined effects of electrostatic and steric stabilization in one molecule. A double electric layer around the nanoparticle is generated by an ionic surfactant possessing extended end chains and polar head group which offers steric repulsion within the nanoparticles, thus preventing the agglomeration and giving rise to a mutual stabilization system.

Figure 5 shows the stabilization of AuNPs by the u nification of steric and electrostatic interactions.

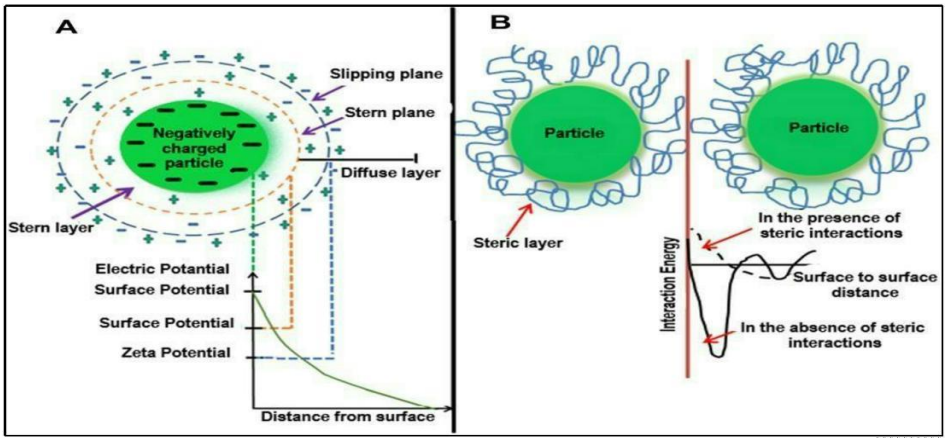

Figure 5 (**A**)Electrostatic stabilization of gold nanoparticles. (**B**) Steric stabilization of gold nanoparticles

Advantages and Disadvantages of Stabilization Methods

Although electrostatic stabilization is easier to maintain in colloidal media, there are certain limitations to it. Firstly, electrostatic stabilization cannot be achieved in electrolyte sensitive media. Additionally, due to strong forces of interactions between oppositely charged ions it is impossible to separate agglomerated particles. Moreover, it cannot be applied to multiple phase systems as different solids establish distinct surface charge and surface potential. As compared to the electrostatic stabilization, which is a kinetic stabilization method, steric stabilization is a thermodynamic stabilization method; therefore, particles can be redispersed. It is also not sensitive to electrolytes and can be applied to multiple phase systems.

Properties of AuNPs

AuNPs exhibit properties which are different from those shown by bulk material. These properties of AuNPs depend on their size and shape. Gold nanoparticles exhibit a wide variety of colors which include brown, purple, blue, orange, and red in the solution form, and the color also depends on the size of the particles. Gold nanoparticles exhibit the SPR band in the range of 500–550 nm, which also depends on the size of nanoparticles. The SPR band arises due to the collective oscillations of conduction electrons caused by the incident photon. Such a band is absent in AuNPs of very small size, particularly those which have a diameter <2 nm as well as in the bulk materials. Apart from the size of the particles, the shape of nanoparticles, ligands, temperature and charge also influence SPR of AuNPs. If the gold nanoparticles exist in the form of aggregates there is a red shift in SPR which results in the broadening of SPB resulting inthe change of color of particles from red to blue.

AuNPs have been widely applied as surface enhanced Raman scattering (SERS) substrate for thedetection of various elements in living cells. The basic mechanism of SERS is caused by two majoramplifications that result in the increase in the cross-section of Raman scattering; first being theelectromagnetic enhancement. The resonance of applied light field along with the collectiveoscillations of electrons of nanostructures cause amplification in the local electric field atnanoparticle surface. Second is the short range chemical enhancement caused by the alteration inpolarizability of molecules due to its charge-transfer interaction with the surface of nanoparticles.Melting point of gold nanoparticles changes with the change in the size of particles. Goldnanoparticles are shown to have a lower melting point as compared to the bulk materials. Thisdecrease in the melting point is because of the fact that the attractive forces of interaction of coreget weaker due to a decrease in the number of neighboring atoms. As a result of this, the interactionbetween inner and surface atoms is reduced and surface atoms get higher surface energy. Thisleads to the decrease in the melting points.Electrical properties of AuNPs are also reported to bedifferent from that of bulk material. As the particle size decreases the surface area is increased whichcauses a decrease in electrical conductivity.However, different materials can be used incombination to enhance their electrical and optical properties. For example, AuNPs can be used

toenhance electrical and optical properties of zinc oxide nanoparticles.

Delivery of Large Biomolecules Using AuNPs

The capability of gold nanoparticles in delivering large biomolecules, such as peptides, nucleicacids, and proteins, has also gained success. Various biomolecules including genes,oligonucleotides, proteins, and peptides are various types of biomolecules which have beendelivered to target cells using AuNPs as delivery vehicles.

Nanoparticle-Based Genetic Therapy

The ideal approach to treat genetically acquired disease is via Gene therapy. Viruses also providea vehicle for highly efficient gene therapy, but they have raised safety concerns which arise due toimmune response and random cytotoxicity.Conversely, at present less efficiency has been reportedvia non-viral gene delivery systems.[28] An effective delivery vehicle should provide efficiententry into the cell, protection of nucleic acid against degradation by nucleases, and release of the nucleic acid in functional form in the nucleus.[29] Nanoparticles, on the other hand, have outstanding therapeutic effects and are capable to deliver all kinds of oligonucleotides such as single stranded DNA (ssDNA), double stranded DNA (dsDNA), plasmids, and single stranded RNA (ssRNA). Gold nanoparticles such as nanorods and nanospheres give protection to nucleic acid and prevent their degradation by nuclease. Oligonucleotide and siRNA-modified AuNPs conjugates are used in gene delivery and gene therapy as intracellular gene regulatory agents which are able to activate immune-related genes.[20]

AuNPs can be conjugated with oligonucleotides using both covalent and non-covalent interactions.Nucleic acid strands can be modified with thiols (–SH) for covalently grafting them onto nanoparticles. In one study, citrate- capped AuNPs were functionalized with antisense oligonucleotides using cyclic disulphides (DTPA) anchoring group and alkyl-thiol anchoring groups to produce tetrathionate particles and mono-thiolated particles. The particles complexes were found to possess high affinity constant for the complementary nucleotide sequence and showed 99% higher cellular internalization without causing any cytotoxicity. When treated with DNAse, AuNPs bound antisense oligonucleotides degraded at a much slower rate than the free antisense oligonucleotide duplexes.[26] The basic mechanism of the study is shown in Figure 6A.

A group has reported the synthesis of polyvalent nucleic acid and AuNPs conjugates by covalently bonding AuNPs with thiol modified nucleic acids. The resultant conjugate was resistant to any degradation by enzymes and showed high cellular internalization. In another study, the same group has applied their “antisense particles” for tumor suppressing. They used mimics of tumor suppressive miRNA-miR-205 for functionalization of AuNPs and sense strand was linked to AuNPs through absorption of alkyl thiol linkage. These conjugates of miR- 205 down-regulated the expression of miRNA target protein and successfully inhibited cancer cell proliferation as compared to non-targeted AuNPs.

Nanocarriers for the Delivery of Protein

Gold nanoparticles can act as nanocarriers of proteins and peptides of interest and reported that cationic tetraalkyl ammonium functionalized gold nanoparticles identify the surface of an anionic protein β- galactosidase via complementary electrostatic interaction and restrain its activity which can be reversed by cellular concentrations of glutathione. The study showed that glutathione-mediated discharge of enzyme β- galactosidase from AuNPs, which depends on the chain length of monolayer, makes it a potential transporter of the protein. In an earlier study, gold nanoparticles functionalized by chitosan have been used to deliver insulin. Chitosan is a non-toxic biopolymer used to synthesize and stabilize the nanoparticles. Chitosan functionalized insulin loaded AuNPs were found to lower the blood glucose level to 30.41% after 2 hours of oral administration. The schematic illustration of this study is shown in Figure 6B. AuNPs conjugated with cell penetrating peptides and lysosomes sorting peptides were tested for their targeted localization into lysosomes. The results showed that these functionalized AuNPs can be efficiently delivered into lysosomes while causing minimum cytotoxicity. Schäffler used gold nanoparticles for conjugation with human serum albumin (alb-AuNP) or apolipoprotein E prior to their intravenous injection.

The outcome of the study demonstrated that protein conjugation extremely reduced the liver retention of AuNPs. This study suggests that the stable conjugation enhances the efficiency and specificity of nanoparticles in the target organ, therefore signifying a potential application in nanopharmacology and nanomedicine.

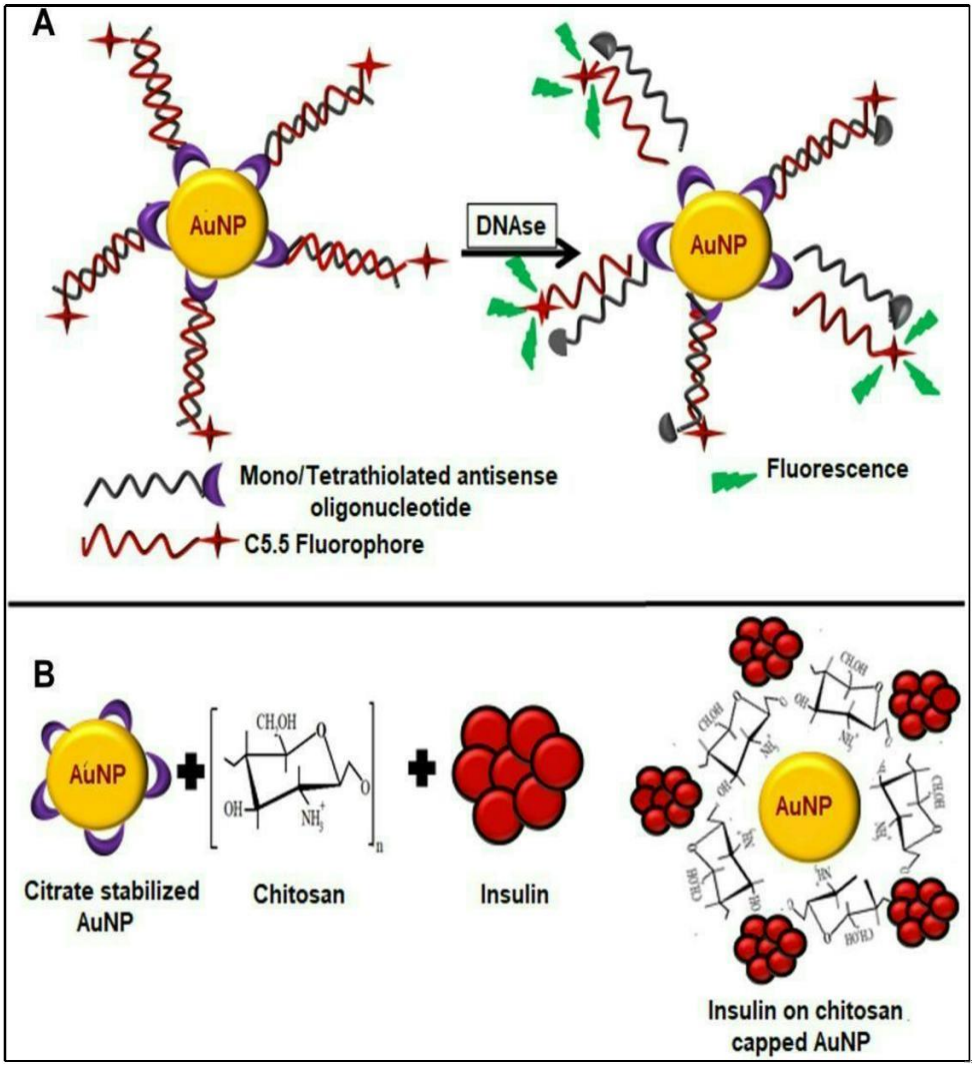

Figure 6 (**A**) capped AuNPs conjugated with mono/tetrathiol modified antisense oligonucleotides treatedwith DNAse. (**B**) Schematic illustration of preparation of insulin loaded AuNPs. Chitosan acts as a reducing andstabilizing agent during the formation of AuNPs. Insulin reacts with chitosan capped AuNPs through hydrogenbonding.

Drug Release from AuNPs

pH-Mediate Drug Release

One of the most appropriate conditions for the release of drug at site of the target over the surrounding tissues is pH.The acidic environment with the pH range from 5.7–7.8 is present in human cancer cells or inside the cell organelles including endosomes and vesicles. These specific pH conditions lead to the cleavage of acid sensitive bond and charge switching due to protonation and morphological alterations of carriers. For example, the acidic conditions (pH 5.0) in lysosomes or endosomes or both can cause the cleavage of the hydrazone bond which is an acid- sensitive bond.This property of hydrazone bond has widely been used in the preparation of pH- responsive supramolecular fabrications for intracellular drug release. A study has reported the AuNPs modified with methyl thioglycolate (MTG) and thiolated methoxy polyethylene glycol (HS-mPEG) having a molar ratio of 1:1. When doxorubicin (DOX) was conjugated with MTG through hydrazine bond the resultant DOX-AuNPs conjugates showed higher pH- sensitive drug release under the pH 5.3 as compared to the normal pH 7. The results showed that after 28 hours of incubation the released DOX drug can be located in the perinuclear region and the nuclei of 4T1 cancer cells.The schematic illustration of this study is presented in Figure 7A. A study has also shown the synthesis of AuNPs functionalized with PEG ligands terminated with DOX having hydrazone bond between PEG and DOX for the release of therapeutics under low pH. These particles were found to enter the cells through endocytosis. Apart from that, DOX-AuNPs having hydrazone bond as compared to the free doxorubicin showed higher drug built-up and retention in MCF-7/ ADR cancer cells which are multidrug resistant cells.

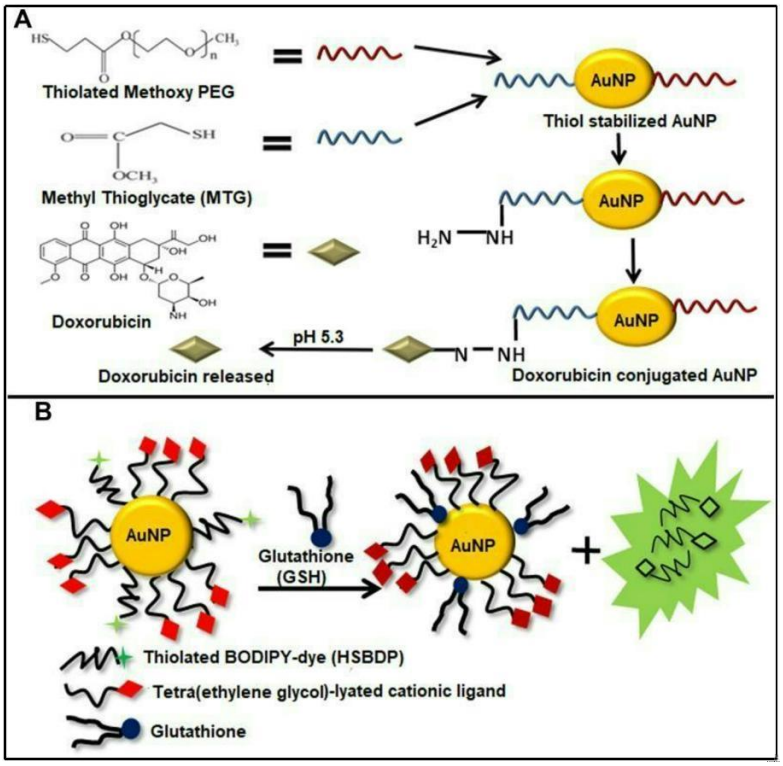

Figure 7 (**A**)Schematic Illustration of pH-mediated release of Doxorubicin from Doxorubicin conjugated AuNPs.First thiol stabilized AuNPs were prepared using Thiolated Methoxy PEG and Methyl Thioglycate (MTG).Doxorubicin was conjugated to AuNPs through hydrazone bond. The acidic conditions (pH 5.4) in the tumor cellscause the breaking of hydrazone bond releasing doxorubicin. (**B**) Glutathione-mediated release of a drug analog(HSBDP) from AuNPs. TTMA and HSBDP functionalized AuNPs when treated with glutathione at 37°C cause therelease of HSBDP which can be detected by the fluorescence it produces in the free form which was quenched when conjugated.

Glutathione (GSH)-Mediated Drug Release

Glutathione-mediated drug release characterizes an alternate non-enzymatic approach for the activation of prodrugs in the intracellular environment. The basic principle of this approach is based on the difference in the concentration of GSH in the intracellular environment (1–10 mM)[as compared to that in the extracellular conditions and the major thiols present in the blood plasma are cysteine (8 μM) and glutathione (2 μM). Previous approaches are based on the disulfide bond between the drugs and drug carriers. Although this approach can be efficacious, modification of the reactivity of the disulfide bond is relatively difficult. Another limitation is that the thiol–disulfide exchange can take place in the presence of cysteines located on the surface of the blood proteins, thus giving rise to a protein–carrier conjugate with different bioaccumulation and pharmacokinetic profiles. In a recent study, hydrophobic dye was used as a model for demonstrating glutathione- mediated hydrophobic drug release using functionalized gold nanoparticles. A monolayer composed of PEGlyated cationic ligands (TTMA) and thiolated bodipy fluorogenic ligands (HSBDP) was presented on the particles. The presence of cationic

ligand enables the passage through the plasma membrane barrier. The release of BODIPY which was not observed when tripeptide was used instead of glutathione indicated that thiol linkage (present in GSH and absent in tripeptide) was required for the release of payload. When AuNPs are conjugated with the dye, BODIPY fluorescence does not occur because the gold core quenches fluorescence through energy and/or electron transfer mechanisms. The fluorescence is produced when AuNPs are triggered with glutathione in cuvette, or cellular thiols present in HepG2 human liver cells. The dye liberation from AuNPs could be controlled by treating embryonic fibroblast cells from mouse with various concentrations of glutathione monoester.[19] Figure 7B shows the schematic illustration of glutathione-mediated drug release from AuNPs.

Enzyme-Mediated Drug Release

Enzymes which are biological catalysts are responsible to sustain life as they catalyze millions of chemical reactions taking place in the living organism. They are substrate specific and not only increase the speed of chemical reactions but also control specificity of metabolic processes.[10] The characteristic degradation by enzyme can be employed for the selective and controlled release of therapeutics from gold nano-conjugates through enhanced permeability and retention effect (EPR). In a study, Hwu et al reported AuNPs and Fe3O4 nanoparticles conjugated with Paclitaxel (PTX) through a phosphodiester bond between thiol-terminated tetraethylene glycol and the C-2′ position of PTX. The phosphodiesterase enzyme present in cancer cells resulted in the cleavage of the phosphodiester linkage. The schematic illustration of this study is presented in Figure 8.

Similarly, another method employed for the designing of prodrugs is esterification and acylation.Enzymes esterases and amidases present in living cells can break the bonds between drugs andnanostructures resulting in liberation of drugs from their prodrugs.19 In order to accurately measure the quantity of drug loading through thermogravimetric analysis efficaciously linked nanocrystals terminated with phenol to a linear derivative of paclitaxel. The mild esterification conditions were used for this reaction and resulted in the high yield of PTX payload.

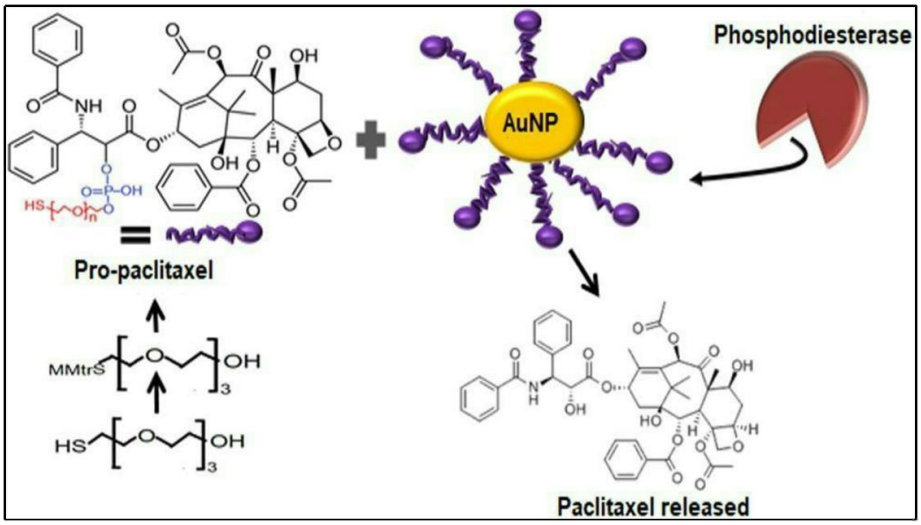

Figure 8 Enzyme-mediated drug release from AuNPs. Pro-paclitaxel was produced by first protecting the thiolgroup of tetraethylene glycol using (mono-4-methoxy) trityl chloride (MMTrCl) and then reacting it with paclitaxel.The thiol terminal of pro-paclitaxel incorporated it onto AuNPs. When treated with enzyme Phosphodiesterase, thePhosphodiester moieties were hydrolyzed causing the liberation of free paclitaxel.

Advantages & Disadvantages of Gold Nanoparticles

Advantages

- Gold nanoparticles is simple for diagnosis.
- It is non-toxic to human beings.
- It is less invasive.
- It provides increased contrast for diagnosis oral cancer.
- It does not photo blinking which is inherent to many other fluorophores.
- As compared to other metallic nanostructures, AuNP provide advantages of their simple and fast preparation and bioconjugation.

Disadvantages

- It leads to acute or chronic toxicity.
- Optical signals of AuNps may not be as strong as quantum dot.
- It exhibits difficulties like biocompatibility, in vivo kinetics, and tumour target efficiency.

CONCLUSION:

In this review article, we have discussed the approaches for the synthesis of gold nanoparticles,stabilization methods, and application in drug and biomolecule delivery. Due to their exclusiveproperties like tunable size and shape, ease of functionalization and fabrication, monodispersity,and low toxicity, AuNPs are considered as exceptionally suitable agents for drug delivery. Greensynthesis of gold nanoparticle is rather an effortless and ecofriendly method which ousts theconcerns associated with biomedical applications of chemically formulated gold nanoparticles.There are wide varieties of biological systems which have been tested for their potential to operateas reducing agents during synthesis reaction. Plants are contemplated to be the most dependableresource for this purpose. Not only do parts of plants (leaves, stems and roots) have reducingabilities, but also the waste produced by plants, such as fruit peels etc. exhibit the same properties.Moreover, the process is quite simple and rapid. There are numerous possibilities for tuning thesurface of AuNPs using different moieties, including PEG, amino acids and peptides,oligonucleotides, and antibodies to facilities the loading of the drug and biomolecules. PEGlyationof AuNPs is considered as the most suitable choice of functionalization for in vivo delivery oftherapeutic agents as it is biocompatible and facilitates nano drug carriers to evade the body'simmune system. Limitations associated with PEGlyation such as loss of ability of AuNPs to bindwith the target receptor can be addressed by

decorating the surface of AuNPs with target specificligands. Delivery of large biomolecules using AuNPs as a delivery vehicle is an innovative andinteresting field and has received a lot of consideration over the past few years, but moreinvestigations are still required to design structures capable of intracellular and intranucleardelivery of conjugates with minimal side effects. Another challenge related to the application ofAuNPs in drug delivery is effective and efficient release of payloads at the target site. Variousstimuli (both external and internal) have been reported to perform this function. Although each ofthese drug release strategies has its own limitations, designing a novel gold nano drug deliverysystem capable of flawlessly carrying and discharging payload at the target site is the subject offuture research. Conclusively, gold nanoparticles offer a promising strategy, but in vivo deliveryefficacy and clinical studies are critically needed.

REFERENCES:

1. Ealias AM, Saravanakumar MP. A review on the classification, characterisation, synthesis ofnanoparticles and their application. *IOP Conf Ser Mater Sci Eng*. 2017;263:3. doi:10.1088/1757-899X/263/3/032019
2. Cai W, Gao T, Hao Hong JS. Applications of gold nanoparticles in cancer nanotechnology. *Nanotechnol Sci Appl*. 2008;1:17–32. doi:10.2147/NSA.S3788
3. Mokhatab S, Fresky MA, Islam MR. Applications of nanotechnology in oil and gas E&P. *J Pet Technol*. 2006;58(04):48–51. doi:10.2118/ 0406-0048-jpt Mu L, Sprando RL. Application of nanotechnology in cosmetics. *Pharm Res*. 2010;27(8):1746– doi:10.1007/s11095-010-0139-1
4. Liu Z, Cai W, He L, et al. In vivo biodistribution and highly efficient tumour targeting of carbon nanotubes in mice. *Nat Nanotechnol*. 2007;2(1):47–52. doi:10.1038/nnano.2006.170
5. Daniel M-C, Astruc D. Gold nanoparticles: assembly, supramolecular chemistry, quantum-size-related properties, and applications toward biology, catalysis, and nanotechnology. *Chem Rev*.2004;104 (1):293–346.
6. Park JW, Benz CC, Martin FJ. Future directions of liposome- and immunoliposome-based cancer therapeutics. *Semin Oncol*. 2004;31 (SUPPL. 13):196– 205.doi:10.1053/j.seminoncol.2004.08.009
7. (7) Jurgons R, Seliger C, Hilpert A, Trahms L, Odenbach S, Alexiou C. Drug loaded magnetic nanoparticles for cancer therapy. *J Phys Condens Matter*. 2006;18(38):38. doi:10.1088/ 0953- 8984/18/38/S24
8. Dykman L, Khlebtsov N. Gold nanoparticles in biomedical applications: recent advances and perspectives. *Chem Soc Rev*. 2012;41(6):2256–2282. doi:10.1039/c1cs15166e
9. Paciotti GF, Kingston DGI, Tamarkin L. Colloidal gold nanoparticles: a novel nanoparticle platform for developing multifunctional tumor-targeted drug delivery vectors. *Drug Dev Res*. 2006;67(1):47–54. doi:10.1002/ddr.20066
10. Paciotti GF, Myer L, Weinreich D, et al. Colloidal gold: a novel nanoparticle vector for tumor directed drug delivery. *Drug Deliv J Deliv Target Ther Agents*. 2004;11(3):169–183. doi:10.1080/ 10717540490433895
11. Chen YH, Tsai CY, Huang PY, et al. Methotrexate conjugated to gold nanoparticles inhibits tumor growth in a syngeneic lung tumor model. *Mol Pharm*. 2007;4(5):713–722. doi:10.1021/ mp060132k
12. Prabaharan M, Grailer JJ, Pilla S, Steeber DA, Gong S. Gold nanoparticles with a monolayer of doxorubicin-conjugated amphiphilic block copolymer for tumor-targeted drug delivery. *Biomaterials*. 2009;30(30):6065–6075. doi:10.1016/j.biomaterials.2009.07.048
13. Zarabi MF, Farhangi A, Mazdeh SK, et al. Synthesis of gold nanoparticles coated with aspartic acid and their conjugation with FVIII protein and FVIII antibody. *Indian J Clin Biochem*. 2014;29(2):154–160. doi:10.1007/s12291-013-0323-2
14. Kalimuthu K, Lubin BC, Bazylevich A, et al. Gold nanoparticles stabilize peptide-drug-conjugates for sustained targeted drug delivery to cancer cells. *J Nanobiotechnology*. 2018;16(1). doi:10.1186/s12951-018-0362-1
15. Storhoff JJ, Mirkin CA. Programmed materials synthesis with DNA. *Chem Rev*. 1999;99(7):1849–1862. doi:10.1021/cr970071p
16. Park C, Youn H, Kim H, et al. Cyclodextrin-covered gold nanoparticles for targeted delivery of an anti-cancer drug. *J Mater Chem*. 2009;19(16):2310–2315. doi:10.1039/b816209c

17. Joshi P, Chakraborti S, Ramirez-Vick JE, et al. The anticancer activity of chloroquine-gold nanoparticles against MCF-7 breast cancer cells. *Colloids Surf B Biointerfaces.* 2012;95:195–200. doi:10.1016/j.colsurfb.2012.02.039

18. Schwert GW, Eisenberg MA. The kinetics of the amidase and esterase activities of trypsin. *JBiol Chem. 1949;179(5):665–72.*

19. Han K, Zhu JY, Wang SB, Li ZH, Cheng SX, Zhang XZ. Tumor targeted gold nanoparticlesfor FRET-based tumor imaging and light responsive on-demand drug release. *J Mater Chem B.*2015;3(41):8065–8069. doi:10.1039/c5tb01659

20. (20) Tangeysh B, Tibbetts KM, Odhner JH, Wayland BB, Levis RJ. Gold nanoparticle synthesis using spatially and temporally shaped femtosecond laser pulses: post-irradiation auto-reduction of aqueous [AuCl4]. *J Phys Chem C.* 2013;117(36):18719–18727. doi:10.1021/jp4056494

21. (21) Birtcher RC, Kirk MA, Furuya K, Lumpkin GR. In situ transmission electron microscopy investigation of radiation effects. *J Mater Res.* 2005;20(7):1654–1683. doi:10.1557/ JMR.2005.0242

22. (22) Sakamoto M, Fujistuka M, Majima T. Light as a construction tool of metal nanoparticles: synthesis and mechanism. *J Photochem Photobiol C.* 2009;10(1):33–56. doi:10.1016/j. jphotochemrev.2008.11.002

23. (23) Zhou Y, Wang CY, Zhu YR, Chen ZY. A novel ultraviolet irradiation technique for shape- controlled synthesis of gold nanoparticles at room temperature. *Chem Mater.* 1999;11(9):2310– doi:10.1021/cm990315h

24. (24) Krinke TJ, Deppert K, Magnusson MH, Schmidt F, Fissan H. Microscopic aspects of the deposition of nanoparticles from the gas phase. *J Aerosol Sci.* 2002;33(10):1341–1359. doi:10.1016/ S0021-8502(02)00074-5

25. (25) Turkevich J, Cooper PHJ. A study of the nucleation and growth process in the synthesis of colloidal gold. *Discuss Faraday Soc.* 1951;55(c):55–75. doi:10.1039/df951110005

26. (26) Wangoo N, Bhasin KK, Mehta SK, Suri CR. Synthesis and capping of water-dispersed gold nanoparticles by an amino acid: bioconjugation and binding studies. *J Colloid Interface Sci.* 2008;323(2):247–254. doi:10.1016/j.jcis.2008.04.043

27. (27) Niidome Y, Nishioka K, Kawasaki H, Yamada S. Rapid synthesis of gold nanorods by the combination of chemical reduction and photoirradiation processes; morphological changes depending on the growing processes. *ChemComm.* 2003;18(18):2376–2377. doi:10.1039/ b307836a

28. (28) Pal A, Esumi K, Pal T. Preparation of nanosized gold particles in a biopolymer using UVphotoactivation. *J Colloid Interface Sci.* 2005;288(2):396–401.doi:10.1016/j.jcis.2005.03.048

29. (29) Kumar S, Gandhi KS, Kumar R. Modeling of formation of gold nanoparticles by citratemethod. *Ind Eng Chem Res.* 2007;46 (10):3128–3136. doi:10.1021/ie060672j

30. (30) Frens G. Controlled nucleation for the regulation of the particle size in monodisperse gold suspensions. *Nat Phys Sci.* 1973;241 (105):20–22. doi:10.1038/physci241020a0