



Review

Silver Nanoparticles – A Review

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Abstract

Demand of silver nanoparticle (AgNPs) is increasing rapidly in many of the streams like in medical, pharmaceutical companies, healthcare, food, consumer, cosmetics etc. Due to its uses it has been used for its several applications like antibacterial properties, household, medical devices, and food industry, wound dressing, in diagnostic, orthopaedics and an anticancer agent. These nano-sized particles are found to be in unique in nature and are also able to change their physical, chemical and biological property that's why they can be exploited in for various purposes. Methods for preparation for preparation of AgNPs are physical, chemical and biological. . Out of all three methods biological method is found to be simple, environmental, commercial and single step method and doesn't need elevated temperature, pressure, force and deadly chemicals. Before application of nanoparticles in any of the purpose like medicine, human welfare, or in healthcare industry, it is very important to characterize the prepared nanoparticles so as to check the safety issue of any of the prepared nanoparticle. Analytical techniques that are used for the analysis of this AgNPs are UV-Vis spectroscopy, XRD, FTIR, DLS, XPS, SEM, TEM, AFM etc. AgNPs have applications like anticancer, antifungal, anti-bacterial, anti-cancer etc.

Keywords: Biological, cancer, diagnosis, nanoparticles, silver nanoparticles

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Silver has been used extensively from last 5000 years for its antibacterial nature. Ag is preferred as nanoparticle for the reason that it has antibacterial property and non-toxic to human beings. Either killing or reducing the growth of bacteria without affecting surrounding cells is known as antibacterial activity. Various methods are used for preparation of silver nanoparticles like physical, chemical and biological. Demand of silver nanoparticle is increasing rapidly in many of the streams like in medical, pharmaceutical companies, healthcare, food, consumer, cosmetics etc. Due to its uses it has been used for its several applications like antibacterial properties, household, medical devices, and food industry, wound dressing, in diagnostic, orthopaedics

and an anticancer agent.^[1] These nano-sized particles are found to be in unique in nature and are also able to change their physical, chemical and biological property that's why they can be exploited in for various purposes. Various methods are used for the synthesis of these AgNP to fulfil the requirements of AgNPs.^[2]

Normally, physical and chemical methods are found to be costly and dangerous. But the nanoparticles that are prepared from biological method they show high yield, high solubility as well as high stability. Out of all three methods biological method is found to be simple, environmental, commercial and single step method and doesn't need elevated temperature, pressure, force and deadly chemicals.^[3] Differ-

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ent materials like leaf extract, bark, root, stem, leaf, fungi etc are used for the synthesis of nanoparticles.

So, after the synthesis of these nanoparticles their particle characterization is important, as the biological properties could be highly impact by the physicochemical properties of particle. Before application of nanoparticles in any of the purpose like medicine, human welfare, or in health-care industry, it is very important to characterize the prepared nanoparticles so as to check the safety issue of any of the prepared nanoparticle[3]. Before finding the toxicity or biocompatibility, it is important to check the characteristic features of any of the nano material like particle size, particle size distribution, shape, surface area, aggregation, solubility etc.

Analytical techniques that are used for the analysis of this AgNPs are UV-Vis spectroscopy, XRD, FTIR, DLS, XPS, SEM, TEM, AFM etc.^[4]

Development of AgNPs with controlled structure i.e. Uniformity in size, morphology and functionality are important for different bio-medical applications. The bioavailability of any of the drug after systemic as well as localized administration of AgNP can be enhanced with the physicochemical properties of the drug.^[5]

As we all know about cancer that cancer is a complex disease in which there is an uncontrolled growth of the cells which may be due to multiple reasons like combination of genetic, internal or may be external factors. It is well known that treatment of cancer includes chemotherapy, surgery, radiation therapy; targeted therapy is very expensive and painful also.^[6] So it is required to find effective, cheap and sensitive molecules for the treatment of cancer. It has been found that AgNPs are found to have application in cancer as anticancer agent as well as in diagnosis. Apart from this AgNPs have other applications like antifungal, anti-bacterial, anti-cancer etc.^[7]

Synthesis of Silver Nanoparticles

Using Physical and Chemical Methods

As I have discussed AgNPs can be prepared by three methods; Physical, Chemical and biological methods. Using evaporation-condensation method having tube furnace at environmental temperature these nanoparticles are prepared in physical method. Before some conventional methods were used to prepare the nanoparticles these are pyrolysis and spark discharging. As no toxic chemicals are used in this method so it is found to be safe.^[8] Another advantage is speed and radiation i.e. used as reducing agent. Apart from this some of the disadvantages are like low yield, contamination in solvent, consumption of high amount of energy and sometime lack of uniform distribution.^[8]

In chemical methods, the AgNPs can be prepared by using either water or some other organic solvents. In this chemical process basically 3 components are there; metal precursors, reducing agent and capping agents. Basically there are two steps in reduction of metal and i.e. Nucleation and growth of subsequent.^[9] By "top-down" method and "bottom-up" method AgNPs can be prepared. In "top-down" method, with the use of colloidal protecting agent there is using of mechanical grinding of large size metals with consequent stabilization. In second method i.e. "bottom-up" methods chemical reduction, sono-decomposition and electrochemical methods are included. In this case the main advantage of chemical method is high yield but the disadvantage is that the methods are very expensive in mature. Secondly, the materials that are used for the synthesis of AgNPs using this chemical method are very toxic and hazardous in nature.^[9]

Green Synthesis Approach (Biological Method)

Green synthesis of silver nanoparticles is preferred because this method is environmental, commercial and single step method and doesn't need elevated temperature, pressure, force and deadly chemicals. Different materials like leaf extract, bark, root, stem, leaf, fungi etc are used for the synthesis of nanoparticles. Apart from this small molecules like vitamins and amino acids are also used as an alternative of chemical methods for the preparation of not only AgNP but some other nano-particles also.^[10] Many of the studies have report that these AgNPs are prepared without use of any toxic and hazardous chemicals and also cost-effective. In the preparation of AgNPs different type of bacteria are used these are; Lactobacillus strains, Brevibacterium casei, Pseudomonas stutzeri, Escherichia coli and fungi that are used are Fusarium oxysporum et.^[11,12] Biological synthesis includes main 3 components these are solvent, reducing agent and non-toxic material. In the biological method we are able to synthesize nano-particle of controlled size and shape, which is one of the most important requirements for preparation of nanoparticle.^[13] Availability of amino acids and proteins is the main advantage of biological method as in this eco-friendly material is used which is less toxic towards both environment and humans. Apart from all this other advantages of AgNPs are availability of vast resources, less time involvement, stability etc. As we know size and shape are most important characteristics of AgNPs, this is because the biological activity of AgNPs depend upon this only.^[14]

If we see in literature small size nanoparticles are found to be more effectual as well as they have better properties.

Characterization

For safety and efficacy of every nanoparticle it is very essential to monitor the physicochemical properties. So, to

check or evaluate the functional properties and properties of synthesized nanoparticles, depiction is very important. As I have discussed earlier there are many techniques that are used for the characterization of these nanoparticles can be done by various techniques like XRD, FTIR, DLS, UV-Vis spectroscopy, TEM, SEM, XPS, so with the use of these technique we are able to determine various characteristics of synthesized nanoparticles.^[15] Techniques for characterization are given in Figure 1.

UV-Vis Spectroscopy

For primary characterization of prepared nanoparticles this UV-Vis technique is one of the easiest and reliable method, apart from this the synthesis as well as the stability of nanoparticles can also be checked using this technique. In this technique, prepared AgNPs are able to interact with the specific wavelength of the light. In case of colloidal suspension calibration of sample or that suspension is not requires.^[16] That's why this technique is found to be easy, reliable, sensitive, simple and effective and selective in many of the nanoparticles. In AgNP it has been found in the studies that valence band and conduction band are found to be very close to each one which results in the freely movement of electrons and then these free electrons give rise to Surface Plasmon Resonance (SPR) band, which is generally due to combined fluctuation of AgNPs electrons in resonance with light wave. The assimilation of AgNP from sample is mainly depend upon three things, these are particle size, dielectric medium and chemical surroundings. In case of SPR observation of the peak is well assigned for the particles that are ranging from 2-100 nm.^[16]

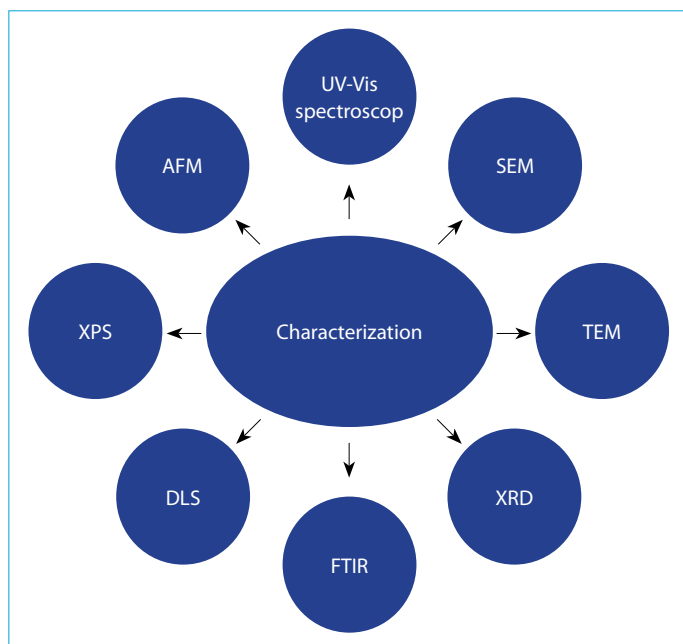


Figure 1. Characterization techniques of AgNP.

X-Ray Diffraction Spectroscopy (XRD)

Here, the study of both molecular as well crystalline structure, determination of quantitative movement in chemical species, degree of crystallinity, particle size etc. can be done by using this technique i.e. XRD. Structural features of wide range of compounds like glasses, superconductors, inorganic catalysts, polymers can be done with the use of this XRD technique.^[17] When light falls on the crystal it results into formation of many patterns of diffraction and then those patterns are able to reflect the physiochemical properties of the structure of crystal. In case if specimen use is powder than the beams that are diffracted they typically come from sample and then that beam will be able to reflect physiochemical structure of the product.^[17]

Basically XRD is the primary or main technique to identify the crystalline nature of the product. Apart from this, this method is used for phase identification measurements, conduction of qualitative analysis, and also to determine imperfection in the structure in many of the streams like pharmaceutical, environmental, geological, and sometimes forensic science also.^[17]

Disadvantage of this XRD technique is that sometimes there is difficulty in growing the crystals. This is the only disadvantage of XRD.^[17]

Dynamic Light Scattering (DLS)

For study of biological activities by using radiation scattering technique, physiochemical parameters or evaluation of synthesized nanoparticle is an important parameter. Particle size ranging from submicron to one nanometre can be determined using DLS. Particles ranging from 2-500 nm can be determined with this DLS, which means smaller particles can be easily determined by this.^[18] This method is basically depends upon the interaction between light and particles. It is the most commonly used technique in measurement of particle size and particle size distribution. DLS measures that light which is scattered from a laser which will be able to pass through a colloid, and will mainly rely on Rayleigh that is scattering from that of nanoparticles are on the edge. Then, the modulation of the scattered light force that is acting as a purpose of time is analyzed, and the hydrodynamic size of particles can be resolute. It has been observed that size that is obtaining from DLS is better as compared to TEM, this is possibly will be due to Brownian Motion. This method is basically used for determination of particle size in aqueous solution.^[18]

Fourier Transform Infrared Spectroscopy (FTIR)

With the use of FTIR it becomes possible to determine the small changes in absorbance i.e. up to 10⁻³, which will eventually help in performance of difference spectroscopy,

which will be then helpful for determination of little combination bands of functionally dynamic residues that left from the large absorption bands of proteins. This method provides accuracy as well as reproducibility and also with method we are able to determine that whether biomolecules are involved in the synthesis of nanoparticles. Apart from FTIR is non-invasive technique. FTIR is also used in some other aspects like authentication of functional molecules that are graft onto silver, gold, silver and graphene nanoparticles, carbon nanotubes etc. FTIR is able to provide strong data, rapid collection of data, large signal-to-noise ratio and also a smaller amount of sample heat up. Overall if we see this FTIR is simple, accurate, valuable, cost-effective, non-invasive technique that is used to confirm the function of biomolecules.^[19]

X-Ray Photoelectron Spectroscopy (XPS)

To estimate empirical formulae this XPS technique is used. The other name of XPS is Electron Spectroscopy for Chemical Analysis (ESCA). This technique is generally used under high vacuum conditions. With this XPS technique we are able to find, identify and characterize the specific groups of macromolecules aromatic rings. It also helps in giving the access of the information regarding qualitative, quantitative and speciation that concerns about the surface of sensor.^[20]

Scanning Electron Microscopy (SEM)

In order to learn much more about nanotechnology and nanosciences many of the techniques are used which includes many techniques of electron microscopy are used. Among which one is SEM. SEM is a technique which is basically used to determine the morphology of the particles. AS SEM is surface imagining method so with the use of SEM we are able to determine the particle size, particle size distribution, surface morphology and nanomaterial shapes. With the use of SEM we are able to find the morphology of particles and then we are able to either draw histogram or we can also count the number of particles either manually or by using some other software. To determine morphology of silver powder and to conduct the analysis of chemical composition SEM is used along with EDX.^[21]

Disadvantage of SEM is that with this technique we are not able to identify the internal structure of the particle but the advantage is that purity and degree of particle aggregation can be determined using SEM.^[22]

Transmission Electron Microscopy (TEM)

TEM is one of the most frequently and important technique i.e. used for the characterization of particles. With this TEM technique we can find quantitative dimension of particle size, particle size distribution and particles morphology

characters¹⁵. The proportion of distance between objective lens and specimen and the distance between that objective lens and image plane will help to determine the magnification of TEM. TEM is able to provide better resolution and better analysis than SEM.^[23]

Limitation of SEM is that it requires high rate of vacuum, sample section should be very thin and sometimes time consuming also.^[23]

Atomic Force Microscopy (AFM)

To find dispersion and aggregation of nanomaterials AFM is used generally. Three more different scanning modes are available apart from their size, shape, and structure and these three modes are known to be as contact mode, non-contact mode and intermittent sample contact mode. The communication of nanomaterials with their supported with lipid bilayer can be characterized using AFM which cannot be done by other electron microscopy techniques.^[24]

Limitation of AFM is that due to the size of canilever there is overestimation of lateral dimensions of the sample, so to remove error it should require much more attention.^[25]

Applications

Silver has been used extensively from last 5000 years for its antibacterial nature. Ag is preferred as nanoparticle for the reason that it has antibacterial property and non-toxic to human beings. Either killing or reducing the growth of bacteria without affecting surrounding cells is known as antibacterial activity. Apart from its antibacterial activity, AgNPs are used extensively for other purpose also like for their antiviral activity, antifungal, anti-angiogenic, cosmetics, water treatment, health care, anti-oxidative, biosensing, anti-inflammatory, drug carrier, imagining purpose and textiles etc. Applications of silver nanoparticles are listed as in Figure 2.

Antibacterial Activity

Either killing or reducing the growth of bacteria without affecting surrounding cells is known as antibacterial activity. Ag is preferred as nanoparticle for the reason that it has antibacterial property and non-toxic to human beings. AgNPs are able to overcome the resistance that has been due to antibiotics. It has been observed that due to presence of their large surface-to-volume ratio as well as crystallographic structure of surface AgNPs are found to be potential as anti-bacterial agent.^[21] AgNPs are able to destroy multiple drug resistant strains that indicates that these AgNPs have the potential to act as a antibacterial agent.^[5]

It has been observed that Gram-negative bacteria show more antibacterial effect as compared to Gram-positive

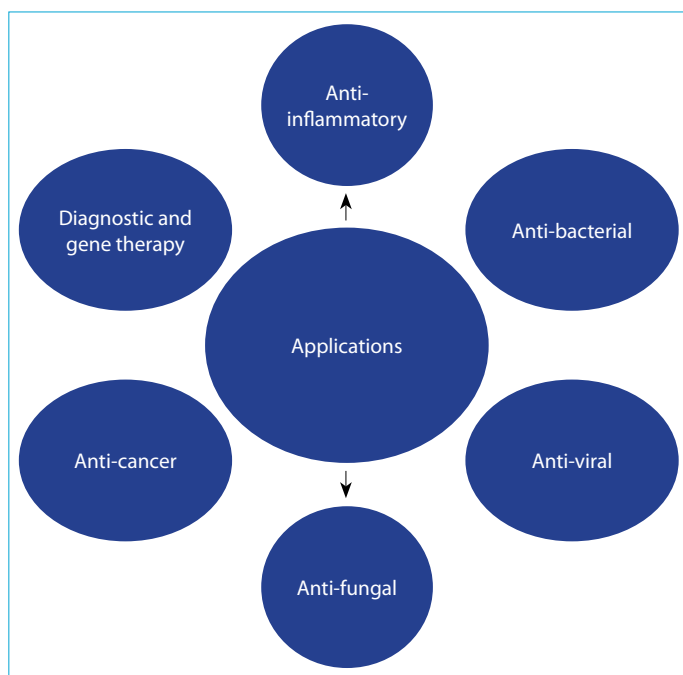


Figure 2. Applications of AgNPs.

bacteria. It is due to presence of cell wall i.e. peptidoglycan layer which is found to be thick in gram-positive bacteria i.e. of 30nm, in case of gram-negative bacteria it is 3-4 nm which is very thin as compared to gram-positive bacteria.^[14] Another reason is that cellular membrane is of negatively charged that is may be due to presence of carboxyl, phosphate and amino group whereas positive charge in bacteria cause attraction between AgNPs and cellular membrane i.e. negatively charged which will result in the attachment of AgNP with cellular membrane which will alter the antibacterial activity of AgNP^[14] Since one-pot synthesis favours the formation of small AgNPs attached to the polymer, which can be distributed in media with pH 6.3, the antimicrobial activity of the chitosan–Ag–nanoparticle composite was found to be higher than that of its components at their respective concentrations.

Antiviral Activity

In the whole world, viral infections and disease are found to be very common, so it's very important to make antiviral agents that results in showing prominent results. AgNPs are found to be prominent in showing such results this is due to their very small size and their shape also. It has been observed that silver is found to be relatively non-toxic towards humans as well as animals and found to be effective against viruses.^[23]

In case of HIV AgNPs found to give permissible result. AgNP acts as anti-viral in this case at an early stage of viral replication, mainly acts as virucidal or may be inhibit the entry of virus. AgNP will bind to gp120 and will prevent CD-4 de-

pendent virion binding and infectivity which will result in acting as an effective virucidal agent against cell free virus. Apart from this AgNPs they inhibit post-entry stages of HIV-1 life cycle.^[23]

Antifungal Activity

Persons having less immunity are more prone to fungal infections. To overcome the fungus related diseases it is found that this process is found to be very tedious in nature. There are very limited number of antiviral drugs that are available in the market.^[24] Anti-viral drugs should be biocompatible, non-toxic as well as environmental friendly. AgNPs are found to be prominent against many diseases that are caused due to fungi^[7] By inhibiting conidial germination, biologically synthesized AgNPs showed good antifungal activity against *Bipolaris sorokiniana*. Indoor fungal species such as *Penicillium brevicompactum*, *Aspergillus fumigatus*, *Cladosporium cladosporoides*, *Chaetomium globosum*, *Stachybotrys chartarum*, and *Mortierella alpine* cultured on agar media are also inhibited by AgNPs.

Anti-inflammatory Activity

Inflammation is the state in which some part of the body becomes swollen, red, hot and sometime painful also and this may occur due to certain injury or sometime infection also. Inflammation is also found to give an immunological response that is against some foreign particles. As AgNPs are known to be their antibacterial and anti-microbial activities but their response to act as anti-inflammatory reagent are limited but they also play important role in this anti-inflammatory field.

Anticancer Activity

Cancer is basically an uncontrolled growth of cells in specific area in a body. Every 1 in 3 person in the world is suffering from one or another type of cancer. There are many treatments like chemotherapy, radiation therapy etc are there for patients who are suffering from cancer but the side effects of these treatments are very harsh and the process is also very painful. It is well known that treatment of cancer includes chemotherapy, surgery, radiation therapy; targeted therapy is very expensive and painful also.^[7] So it is required to find effective, cheap and sensitive molecules for the treatment of cancer. Several studies have been done to know the promising result of AgNPs.^[6] It is found to be most suitable as well as an alternative for other cancer treatments. They have ability to target specific cells or tumour at that site only by encapsulation of therapeutic agent in nanoparticle and then used as drug delivery system.

After 6 hours of exposure to *Albizia adianthifolia* leaf extract synthesized AgNPs (AA-AgNPs), A549 cells showed

21% and 73% cell viability, respectively, and normal peripheral lymphocytes showed 117 and 109 percent viability. This means that AgNPs aren't harmful to normal PLs cells. At 43 g/mL of AA-AgNPs, 50 percent cell inhibition of A549 cells was achieved, and cell death was induced by the generation of ROS, resulting in apoptosis. After 48 hours of Hoechst staining, MCF-7 cells treated with *Sesbania grandiflora* mediated AgNPs (20 g/mL) display nuclear condensation, cell shrinkage, and fragmentation. These changes mean that DNA repair has been enabled as a result of the cleavage.^[7]

Challenges for Cancer therapy Using AgNPs

Although AgNPs are used as an alternative treatment in cancer therapy because of their site specific, reduction in toxicity, better efficacy and some other advantages but there are certain limitations of AgNPs for cancer therapy. These limitations are of physiological barriers, enhanced permeation and retention effect (EPR), limited carrying capacity and mainly manufacturing issues also.^[7]

Antimicrobial Activity

The AgNPs made from the inflorescence of the *Cocos nucifera* inhibited the growth of *V. alginolyticus*, *K. pneumoniae*, *P. aeruginosa*, *B. subtilis*, and *P. shigelloides*. The ability of AgNPs to bind to the bacterial cell wall has been demonstrated microscopically. AgNPs synthesized with lemon peel extract had the highest antidermatophytic activity and the largest inhibition region. *T. mentagrophytes* and *C. albicans* showed 3SD and 5SD activity, respectively, but no activity against *T. Rubrum*. The leaf extracts of *Caesalpinia coriaria* developed triangular, hexagonal, and spherical AgNPs with sizes ranging from 78 to 98 nm that had better antibacterial activity against *Escherichia coli* (12 mm) and *Pseudomonas aeruginosa* (18 mm). The generation of reactive oxygen species and decreased development of hydroxyl radicals initiated by the phytoconstituents capped on the synthesized AgNPs can cause apoptosis in *C. albicans* and *S. Cerevisiae*.^[26]

Various Silver Nanoparticles Prepared Till Date

List of silver nanoparticles prepared till date are shown in Table 1.

Conclusion

Silver nanoparticles are now used extensively because of their numerous uses in many of the sectors. It has been extremely used as an anti-bacterial, apart from this AgNPs have their uses as anti-viral, anti-inflammatory, gene therapy, anti-fungal, in diagnostic and imaging purpose and many more. Now, AgNPs is an area of explore in case of cancer therapy as an alternative to other conventional

Table 1. Various silver nanoparticles prepared till date

Drug	Part taken	Reference
<i>Pedaliu murex</i>	Leaf extract	27
White sugar		28
<i>Carpesium cernuum</i>	Extract	29
<i>Coriander sativum</i>	Leaf extract	30
<i>Salicornia brachiata</i>	Aqueous extract	31
<i>Amaranthus gangeticus</i>	Leaf extract	32
<i>Ocimum sanctum</i> (Tulsi)	Leaf extract	33
<i>Parthenium</i>	Slurry	34
Gum acacia	Gum	35
Red apple	Fruit extract	36
<i>Coffea Arabica</i>	Seed extract	37
<i>Syzygium aromaticum</i> (Clove)	Bud extract	38
<i>Putranjiva, drypetes roxburghii</i> (Wall)	Fruit extract	39
<i>Lippia nodiflora</i>	Aerial extract	40
<i>Origanum vulgare</i>		41
<i>Soymida febrifuga</i>	Aqueous fruit extract	42

therapies like chemotherapy, radiation therapy etc. As these AgNPs are used as an anti-angiogenic so it is found to be one of the interesting approach for cancer therapy. These AgNPs can be prepared by three methods these are physical, chemical and biological method. Out of all three biological methods is mainly used as it is environmental, non-toxic and eco-friendly. It is found to be safe and site specific in case of cancer treatment. So, it can be say that these AgNPs are safe, simple, reliable treatment for many of the diseases.

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