ABSTRACT

The present invention relates to Nanosolve mouth-dispersing granules of silver nanoparticles of lemon fruit extract, which do not require water for oral administration and process for preparation thereof. The mouth-dispersing granules of the present invention used as antimicrobial and antibacterial therapy.

E 101/11434/2014.

We claim,

1. A mouth dispersing granular pharmaceutical formulation which comprises;

a. silver nanoparticles of lemon fruit extract

b super disintegrant

c. Sweetener

d. organic acid and

e. At least one other pharmaceutically acceptable excipient.

2. A pharmaceutical formulation according to claim 1 wherein said formulation is in granule form.

3. A pharmaceutical formulation according to claim 1 wherein said formulation is in sachet/packet form

4. A pharmaceutical formulation according to claim 1 wherein said formulation do not require water for oral dosing.

5. A pharmaceutical formulation according to claim 1 to 4 wherein said formulation is prepared as a single composition comprising silver nanoparticle of lemon fruit extract.

6. A pharmaceutical formulation according to claim 1 to 4 wherein silver nanoparticle is of lemon fruit extract

7. A pharmaceutical formulation according to claim 6 wherein silver nano particle of lemon fruit extract is used 5-50% of total formulation.

8. A pharmaceutical formulation according to claim 1 wherein disintergrant is selected from a group comprising crospovidone, croscarmellose sodium and sodium starch glycollate.

9. A pharmaceutical formulation according to claim 1 wherein sweetener is selected from a group of natural and artificial sweeteners comprising sucrose, fructose, dextrose and glucose, and sweet polyols like glycerin, sorbitol ,mannitol aspartame, sucralose, acesulfame potassium, lactilol, cyclamates and saccharin,

10. A formulation according to claim 1 wherein said formulation is used for treatment of antimicrobial and antibacterial therapy

Dated this the day of March 2014

Name of Applicants:

Name	Sign
Waghulde, Sandeep Onkar	
Juvatkar, Pritam Vijay	
Gorde, Nilesh Keshavrao	
Naik, Pravin Pandurang	
Jain, Vishal Ramesh	
Shirodkar, Prabhakar Yashwant	
Kale, Mohan	

To,

The Controller of Patents

The Patent Office,

at Mumbai.

21 MAR 2014

E-01/11433/2014

Technical field of the invention

The present invention relates to Nanosolve mouth-dispersing granules of silver nanoparticles of lemon fruit extract, which do not require water for oral administration and process for preparation thereof. The mouth-dispersing granules of the present invention used as antimicrobial and antibacterial therapy.

Background and prior art of the invention

The synthesis of nanoparticles has become the matter of great interest in recent times due to its various advantageous properties and applications in various fields. Though physical and chemical methods are more popular for nanoparticle synthesis, the biogenic production is a better option due to eco-friendliness. This invention relates to the potential of plants i.e. "green chemistry" to synthesize nanoparticles not only in the laboratory scale but also in their natural environment.

U. S. Patent 8,314,078 by Vascular Vision Pharma. Co. discloses a silver nanocomposite, a formation method for forming the silver nanocomposite, and an application method utilizing the silver nanocomposite. The silver nanocomposite includes a silver nanoparticle conjugated to a glycosaminoglycan (GAG) or glucose. The formation method includes chemically reacting silver nitrate with a reducing agent to form a silver nanoparticle conjugated to the reducing agent of a GAG or glucose. The application method may include topically applying the silver nanocomposite to a wound or burn as an anti-microbial with respect to an antibiotic-resistant genotype in the wound or burn, wherein the silver nanocomposite topically applied includes the silver nanoparticle conjugated to the GAG of 2,6-diaminopyridinyl heparin (DAPHP) or hyaluronan (HA). The application method may include applying the silver nanocomposite as a coating to plastic, a catheter, or a surgical tool, wherein the silver nanocomposite applied as the coating includes the silver nanoparticle conjugated to the GAG of DAPHP.

Nusatara Bioscience, Vol. 3, No. 2, Pp. 59-6, by Shital Bonde, discloses a biogenic approach for green synthesis of silver nanoparticles using extract of *Foeniculum vulgare* and its activity against *Staphylococcus aureus*and *Escherichia coli.*

International Journal of Bioassays (IJB), Volume 01, Number 07, July 2012; by Ravendra PS Chauhan, Charu Guptaand Dhan Prakash, discloses the methodological advancements in green nanotechnology and their applications in biological synthesis of Herbal Nanoparticles.

Nanoparticles of noble metals such as silver, calcium, zinc, copper, etc. have many pharmacological properties like anticancer, antibacterial, antimicrobial, etc.; metallic nanoparticles being the most useful, from this point of view. The present invention further highlights a cost effective and environment friendly technique for green synthesis of silver nanoparticles. Biosynthesis of nanoparticles by lemon fruit extracts is currently under exploitation. The silver nanoparticles synthesized by reaction of biomass of aqueous extracts from lemon fruit with aqueous solutions of silver nitrate (AgNO3). To the best of our knowledge, biological approach using juice of lemon has been used for the first time as a reducing material as well as surface stabilizing agent for the synthesis of spherical-shaped Ag-NPs.

Plant extracts is not expensive and ecofriendly and thus can be an economic and efficient alternative for the large-scale synthesis of nanoparticles. In the present study, synthesis of silver nanoparticles is achieved through simple eco-friendly nontoxic, inexpensive, abundantly available route. The silver nanoparticles show efficient antimicrobial activity compared to other salts due to their extremely large surface area, which provides better contact with microorganisms. Therefore, silver is ideally suited for effective control of germs, molds and fungus. The rapid breakdown of silver nanoparticles releases ionic silver that inactivates vital bacterial enzymes by interacting with essential thiol groups. Silver ions can inhibit bacterial DNA replication, damage bacterial cytoplasm membranes, depleting levels of intracellular adenosine triphosphate (ATP) and finally

cause cell death. The synthesized nanoparticles were confirmed by XRD and Scanning Electron Microscope (SEM). This novel green approach is a rapid, facile and used for large scale production of metallic Nanoparticle.

None of the aforementioned prior art discloses Nanosolve mouth-dispersing granules of silver nanoparticles of lemon fruit extract and process for preparation thereof. Another invention relates to rapid and green synthesis of silver nanoparticles using leaf extracts of lemon fruits.

The present invention provides mouth-dispersing granules of comprising silver nanoparticles of lemon fruit extract as active ingredient, superdisintegrant(s), sweetener(s), organic acid(s) and other pharmaceutically acceptable excipients; wherein the mouth-dispersing granules do not require water for oral administration.

Summary of invention

The present invention relates to Nanosolve mouth-dispersing granules of silver nanoparticles of lemon fruit extract, which do not require water for oral administration. The invention further relates to Nanosolve technology helps in faster dissolution of mouth-dispersing granules of silver nanoparticles. Another invention relates to synthesis of silver nanoparticles. The mouth-dispersing granules of the present invention can be used as antimicrobial and antibacterial therapy.

Detailed description of the drawings

Figure 1 shows the general biosynthesis of metal nanoparticles from biological sources.

Detailed description of the invention

The invention will now be described in detail so that various aspects thereof may be more fully understood and appreciated.

The thrust to develop environmental friendly procedures for production of Nanoparticles arises from the very fact that current nanotechnology research uses a lot of chemicals,

which are potential threat to both environment and public health. The present invention of safe eco-friendly methods for biogenetic production is now of more interest due to simplicity of the procedures and versatility.

Due to their amenability to biological functionalization, the biological nanoparticles are finding important applications in the field of medicine. Various processes for the synthesis of nano and micro- length scaled inorganic materials which have contributed to the development of relatively new and largely unexplored area of research based on the biosynthesis of nanomaterials. Synthesis using bio-organisms is compatible with the green chemistry principles. "Green synthesis" of nanoparticles makes use of environmental friendly, non-toxic and safe reagents. In several studies, the biological routes to the synthesis of nanoparticles have been proposed by exploiting microorganisms and vascular plants. However, the utilization of herbal and medicinal plant extracts for the synthesis of nanoparticles is a relatively recent activity. In a number of studies, gold and silver have been used mostly for the synthesis of stable dispersions of nanoparticles using the extracts of herbal and medicinal plants.

Bio-Nanotechnology combines biological principles with physical and chemical approaches to produce nano-sized particles with specific functions. It also represents an economic substitute for chemical and physical methods of nanoparticles formation. Metallic nanoparticles have a high definite surface area and a high fraction of surface atom; Certain nano crystals are attractive probes of biological markers because of: small size (1-100nm) large surface to volume ratio, chemically alterable physical properties, change in the chemical and physical properties with respect to size and shape, strong affinity to target particularly proteins, structural sturdiness in spite of atomic granularity, enhanced or delayed particles aggregation depending on the type of the surface modification, enhanced photoemission, high electrical and heat conductivity and improved surface catalytic activity. e.g. Silver, Aluminum, Gold, Zinc, Carbon, Titanium, Palladium, Iron, Fullerenes, Copper etc have been rottenly used for the synthesis of nanoparticles. Syntheses of silver nanoparticles (Ag NPs) have gained considerable interest due to their unique properties such as excellent electrical and thermal

conductivity, chemical stability, catalytically activity, nonlinear optical behavior, anti-microbial and anti-bacterial effects. They also show a very peculiar nature of collective oscillation and absorption of light by surface electrons termed as surface plasmon resonance (SPR) in the UV-Vis region, which makes them potential biomaterials. As both size and shape of Ag NPs affect the optical and electronic properties, a number of methods have been reported for the preparation of Ag NPs with varying sizes and shapes. Most of the methods generally involve chemical reduction of metal salt with different external reducing agents.

Synthesis using bio-organisms is compatible with the green chemistry principles. "Green synthesis" of nanoparticles makes use of environmental friendly, non-toxicand safe reagents. The plant kingdom and its use for the synthesis of nanoparticles is relatively less explored. Some work has been carried out for the production of Ag NPs using either the whole plant or plant parts extracts. Usage of plants or plant products give a distinctive edge over the microorganism routes since one need not undergo the cumbersome process of maintaining cultures. In this invention the report an effective room temperature synthetic procedure for the reduction of Ag1 to Ag0 using lemon juice, commonly known as lemon.

Locally available lemons, was used for preparation of juice and distilled water with are sensitivity was used as medium to dissolve the silver salt and for making the different conc. of juices. 0.5 mL of juice were pipette out(vitamin C content measured in juice before used) and transferred into a 50 ml volumetric flask and was filled up to the mark with water. The contents of the flask were thoroughly agitated for 1 hour in a shaking water bath at 30 °C and then allowed to stay overnight. The reddish brown color solution was decanted and then centrifuged at 500 rpm to remove any un-dissolved sediments. The aliquot was recovered by filtering the solution through a filter paper and used for further experiments. A dilute solution of silver (0.5 M) was prepared by dissolving suitable amount of $AgNO_3$ in water. To carry out the $Ag+ \rightarrow Ag0$ reaction, 5ml each of 0.5 M $AgNO_3$ solutions were pipetted out into four identical transparent plastic vials marked as experiments 1, 2, 3 and 4, to which selective volumes of 1, 2, 3 and 5 ml of tea

aliquot was added respectively. The vials were capped to prevent external contamination and evaporation of solvent. The solutions were left to react for three hours. The above-obtained emulsions were characterized using UV-Vis spectrometer.

The present invention relates to preparation of lemon fruit extract involves the part of plant will be finely cut into small pieces and the aqueous extract was prepared by mixing 10 g of dried part of plant powder with 500 mL of water (boiled and cooled distilled water) with constant stirring on a magnetic stirrer.

The method for preparation of silver nanoparticles of lemon fruit extract comprising the fresh fruit of lemon solution is prepared by taking 500g of thoroughly washed and in a 300 mL Erlenmeyer flask along with 100 mL of sterilized double distilled water and then boiling the mixture of 60° C for 15 min before finally decanting it. After boiling, the solution was cooled, decanted, and 12 mL of this solution was added to 88 mL of 1mM aqueous AgNO3 solution and the resulting solution having some color. This extract was filtered through nylon mesh (spectrum) followed by Millipore hydrophilic filter (0.22μm) and used for further experiment extract and color intensity of the extracts was measured at 420 nm for different intervals (15, 30, 60, 120, 240 and 300 min respectively).

Another method for the synthesis of silver nanoparticles, 1mM silver nitrate (AgNO3) solution was prepared and stored for further use. 5ml of fruit extract (5%) was taken into conical flask and to this 50ml of 1mM AgNO3 solution was added drop wise with constant stirring at 50-60°C and observed the color change (17-22). The color change of the solution was checked periodically. The conical flask was incubated at room temperature for 48 hours. The change in color of the solution to the dark brown indicates the synthesis of silver nanoparticles from the fruits extract. This content was centrifuged at 10,000 rpm for 15min. The supernatant was used for the characteristics of the silver nanoparticles through characterization techniques.

In summarized the bio-reduction of aqueous Ag^+ ions by the *lemon fruit* extract has been demonstrated. In this present invention found that lemon fruit extract used as good source

for synthesis of silver nanoparticles. This green chemistry approach towards the synthesis of silver nanoparticles has many advantages such as, ease with which the process can be scaled up, economic viability, etc. The "green" route for nanoparticle synthesis is of great interest due to eco-friendliness, economic prospects, feasibility and wide range of applications in nano medicine, catalysis medicine, nano-optoelectronics, etc.

Conventional solid oral dosage forms such as tablets and capsules, are widely used for administration of various active drugs. Many patients such as children, old-aged persons and patients who are busy or traveling with little or no access to water, find it very difficult to swallow such tablets and capsules. Hence, conventional solid oral dosage forms of silver nanoparticles of lemon fruit extract for antimicrobial and antibacterial used as poor patient-compliant dosage forms.

The present inventors have found that mouth-dispersing granules of silver nanoparticles of lemon fruit extract, improve patient compliance and convenience as well as bioavailability, while overcoming the difficulty of swallowing conventional solid oral dosage forms such as tablets and capsules.

The mouth-dispersing granules of silver nanocarriers of lemon fruit extract of the present invention, comprise lemon fruit extract as active ingredient, superdisintegrant(s), sweetener(s), organic acid(s) and other pharmaceutically acceptable excipients.

When mouth-dispersing granules of are placed on the tongue, they rapidly dissolve or disintegrate in the saliva within few seconds, to release active drugs.

The mouth-dispersing granules of silver nanoparticles of lemon fruit extract of present invention disperse in patient's mouth within few seconds without the need of water or chewing, while overcoming the difficulty of swallowing conventional oral dosage forms such as tablets and capsules. The mouth-dispersing granules of silver nanoparticles of lemon fruit extract get dissolved/disintegrated in the saliva within few seconds, and turn into smooth suspension/solution for easy swallowing, thus proving to be beneficial to

children, old-aged patients, bed-ridden patients, busy patients and traveling patients, who find it difficult to swallow the silver nanoparticles of lemon fruit extract tablets/capsules in antimicrobial therapy.

The superdisintegrant used in the present invention leads to explosion of silver nanoparticles of lemon fruit extract granules, and hence aids in rapid disintegration of the granules, which offers smaller particle size and higher surface area for dissolution of silver nanoparticles of lemon fruit extract granules in the mouth. The superdisintegrant(s) used in the present invention are selected from but not limited to crospovidone, croscarmellose sodium or sodium starch glycollate, and used in an amount of 0.5 to 20% w/w of the total formulation.

The sweeteners used in the present invention aid to provide a palatable taste. It also helps in faster dissolution. The sweeteners used in the present invention are in a combination of artificial sweetener and natural sweetener. The natural sweetener(s) are selected from but not limited to sugars such as sucrose, fructose, dextrose and glucose, and sweet polyols like glycerin, sorbitol and mannitol, and used in an amount of 2 to 50% w/w of the total formulation. The artificial sweetener(s) are selected from but not limited to aspartame, sucralose, acesulfame potassium, lactilol, cyclamates and saccharin, and used in an amount of 0.02 to 10% w/w of the total formulation

The organic acid used in the present invention increases salivation, and hence offers high volume of saliva for dissolution of silver nanoparticles of lemon fruit extract granules. The pharmaceutically acceptable organic acid alone or in combination with alkaline salts, used in the present invention is selected from but not limited to citric acid, tartaric acid, maleic acid and their salts either alone or in combination, in an amount of 0.5 to 20% of the total formulation.

The other pharmaceutically acceptable excipients used in the present invention include but are not limited to binder(s), glidant(s), antioxidant(s), stabilizer(s), flavoring agent(s) and colorant(s).

The mouth-dispersing silver nanoparticles of lemon fruit extract granules of the present invention are available within sachets/packets with proper dosing amounts.

The mouth-dispersing granules of silver nanoparticles of lemon fruit extract of the present invention can be used as an antimicrobial and antibacterial therapy.

The present invention is exemplified by the following examples which are provided for illustration only and should not be construed to limit the scope of the invention.

Examples:

Silver nanoparticles of lemon fruit extract floating mouth-dispersing granules

Example 1:

Sr No.	Ingredients	Quantity (mg/2gm)
1	Silver nanoparticles of Lemon fruit Extract	1000
2	Sodium alginate	200
3	Sucralose	2
4	Mannitol	607
5	Sodium Chloride	20
6	Crospovidone	40
7	Citric acid	100
8	Quinoline yellow	2
9	Trusil lemon	25
10	Colloidal silicon dioxide	4

Preparations & dissolution profiles are remaining

In vitro dissolution profile for Silver nanoparticles of lemon fruit extract of mouth dispersible granules:

Time (hrs)	Drug dissolution (%)
0.15	88
0.20	95
0.30	98

Example 2:

Sr	Ingredients	Quantity

No.		(mg/2gm)
1	Silver nanocarrier of Lemon fruit extract	1000
2	Sodium alginate	200
3	Sucralose	2
4	Mannitol	607
5	Sodium Chloride	20
6	Crospovidone	40
7	Citric acid	100
8	Sunset yellow Supra	2
9	Orange flavor	25
10	Colloidal silicon dioxide	4

Table 1: Moisture contain of Granules:

Examples		**1**	**3**
Duration		3 months	3 months
Moisture uptake Studies 40 °C/75% RH in Open Petri dish	% LOD	4.1%	4.4%

E 101/11434/2014.

We claim,

1. A mouth dispersing granular pharmaceutical formulation which comprises;

a. silver nanoparticles of lemon fruit extract

b super disintegrant

c. Sweetener

d. organic acid and

e. At least one other pharmaceutically acceptable excipient.

2. A pharmaceutical formulation according to claim 1 wherein said formulation is in granule form.

3. A pharmaceutical formulation according to claim 1 wherein said formulation is in sachet/packet form

4. A pharmaceutical formulation according to claim 1 wherein said formulation do not require water for oral dosing.

5. A pharmaceutical formulation according to claim 1 to 4 wherein said formulation is prepared as a single composition comprising silver nanoparticle of lemon fruit extract.

6. A pharmaceutical formulation according to claim 1 to 4 wherein silver nanoparticle is of lemon fruit extract

7. A pharmaceutical formulation according to claim 6 wherein silver nano particle of lemon fruit extract is used 5-50% of total formulation.

8. A pharmaceutical formulation according to claim 1 wherein disintergrant is selected from a group comprising crospovidone, croscarmellose sodium and sodium starch glycollate.

9. A pharmaceutical formulation according to claim 1 wherein sweetener is selected from a group of natural and artificial sweeteners comprising sucrose, fructose, dextrose and glucose, and sweet polyols like glycerin, sorbitol ,mannitol aspartame, sucralose, acesulfame potassium, lactilol, cyclamates and saccharin,

10. A formulation according to claim 1 wherein said formulation is used for treatment of antimicrobial and antibacterial therapy

Dated this the day of March 2014

Name of Applicants:

Name	Sign
Waghulde, Sandeep Onkar	
Juvatkar, Pritam Vijay	
Gorde, Nilesh Keshavrao	
Naik, Pravin Pandurang	
Jain, Vishal Ramesh	
Shirodkar, Prabhakar Yashwant	
Kale, Mohan	

To,

The Controller of Patents

The Patent Office,

at Mumbai.

21 MAR 2014