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Novel Synthesis of Antimicrobial Cotton Fibers Embedded with Enriched Zinc Chloride Nanoparticles

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ABSTRACT
An attempt is made to synthesisze the cotton fibers embedded with enriched zinc chloride nanoparticles. The Zinc chloride solution was treated with green reducing agent d-glucose in autoclave. Autoclave treatment time was optimized based on single type of particle presence which is followed by microwave radiation cycles for selected time intervals. Synthesized nanoparticles were analysed using UV spectroscopy, IR spectroscopy and Scanning electron microscopy SEM. Purified particles were analyzed for presence of antibacterial and antifungal activity. Cotton fibers were treated in presence of obtained nanoparticles and starch as capping agent. Treated fibers also retained antibacterial activity.

Keywords: zinc chloride nanoparticle, autoclave, microwave treatment, infrared spectroscopy,scanning electron microscopy, antibacterial cotton fibers.

INTRODUCTION

It is wel known fact that the Nanotechnology deals with small structure and small size material of dimension in the range of few nanometers to less than 100 nm (1, 2, 3). Properties like particle size,shape and interparticle interaction of synthesized metal nanoparticles are determined by change in absorbance**.** In synthesis and assembly strategies of nanoparticles precursors from liquids, solids or gas phase are used applying physical and chemical deposition approach (4,5,6). Over the last few decades, the applications of nanotechnology in

medicine have been extensively explored as antibacterial and antifungal agents. The green synthesis of zinc chloride nanoparticles involves three main steps selection of solvent medium, selection of environmentally benign reducing agent, and selection of nontoxic substances (7, 8, 9). Among the many possible natural products, Polysaccharides and biologically active plant products having hydroxyl groups, a hemiacetal reducing end can play important roles in reduction and the stabilisation of metallic nanoparticles (Y. Park Et al Polysaccharides and phytochemicals: a natural reservoir for the green synthesis of gold and silver nanoparticles, IET Nanobiotechnol., 2011). Microwave based synthesis was recent approach for green synthesis (10, 11). In present work d-glucose is used as the reducing agent and starch as the capping agent for zinc chloride nanoparticle synthesis. Cotton fibers are used as the source for embedment. Medicinal application of this present work is tested against the most commonly isolated serotypes of bacteria and fungi.

METHODOLOGY

The synthesis method and optimization of autoclave treatment time for ZnCl nanoparticles:
50 ml 2% starch (Merck Pvt.ltd) solution was prepared. 1mM D-glucose (Merck Pvt.ltd) and 1 mM zinc chloride (Merck Pvt.ltd.) was added in it. This solution was autoclaved at 15 lb/inch^2 pressure for 10, 15 and 20 minutes and analyzed by UV spectroscope (Elico ltd.) followed by addition of 1mM NaOH (Merck Pvt.ltd) to make pH alkaline.

The optimization of Microwave treatment cycles:
Microwave radiation treatment for autoclaved solution was given at 2.45 GHz frequency in domestic microwave oven (LG make). Each cycle consist of 15 second exposure to microwave irradiation followed by cooling time interval of 15 second. Maximum 12 cycles had given. Resulting solution was centrifuged at 14000 rpm for 20 minute followed by washing with 70% ethanol.

Characterization of zinc chloride nanoparticles:
Harvested particles were made into fine powder. This powder was dissolved in double distilled water and analyzed on UV spectrophotometer (Elico make) from 190 nm to 600 nm wavelength ranges followed by FTIR spectroscopy. Powder was also analyzed by using scanning electron microscopy (SEM).

The specific antibacterial and antifungal activity of zinc chloride nanoparticles:
Zinc chloride nanoparticle powder was further analyzed for antibacterial and antifungal activity by disc diffusion test against *Salmonella typhi*, *Klebsiella pneumoniae*, *Staphylococcus aureus* and *Pseudomonas aeruginosa* bacteria and *Rhizopus*, *Candida albicans* and *Aspergillus niger* fungi. Sterile 6mm filter paper disc were aseptically placed on Luria Bertani (LB) agar surface for antibacterial activity and potato dextrose agar for antifungal activity. Sterile disc was soaked in zinc nanoparticle solution for 30 min. After 30 min ,disc were placed on medium .The plates were left at ambient temperature for 15 min to allow excess prediffusion of solution prior to incubation at 37^0 C for 24 hrs for antibacterial activity and 30^{o}C for 48 hrs for antifungal activity. After incubation Inhibition zones were measured.

The preparation of Zinc chloride nanoparticle (ZnClNP) embedded cotton fibers:
The Cotton fibers were cut in small pieces and deeped in solution containing 10mM zinc chloride 1mM glucose, 2 % starch. This solution was autoclaved at 15 lbs/ inch^2 for 15 minutes. After autoclaving solution was cooled to normal temperature. pH of solution was made alkaline followed by 9 cycles of Microwave treatment. Antibacterial activity of cotton fibers was analyzed by incubating the bacteria in presence of treated cotton fibers.

The antibacterial and antifungal activity of ZnClNP embedded cotton fibers:
In this, Liquid culture medium dilution method was used to measure the minimum inhibitory concentration (MIC) for each bacterial and fungal strain using Luria Bertani medium and potato dextrose agar respectively. 900 µL of medium was placed in a sterilized test tube. 1 gm nanoparticle embedded cotton fibers and 10 µl of the cultured bacterial solution (final bacterial count of 1 × 10^6 cfu/ml) were added. The tube was incubated for 48 hrs at 37^{0}C for bacterial growth and 30^{0}C for fungal

growth. No bacterial and fungal growth were observed confirming antibacterial and antifungal action of nanoparticle embedded cotton fibers.

RESULTS AND DISCUSSION

Optimization of autoclave treatment time for zinc chloride nanoparticle synthesis: Present investigation utilizes autoclave assisted microwave mediated synthesis approach with d-glucose as reducing agent. In alkaline condition, Glucose oxidizes itself by reducing zinc chloride solution in water. Comparative zinc chloride nanoparticle synthesis was analyzed by observing the absorbance at 350 nm. Increase in autoclave treatment time showed increase in absorbance at 350 nm. This reaction was performed in autoclave at 15 lb/inch^2 pressure for 15 minute at 121^{0}C.

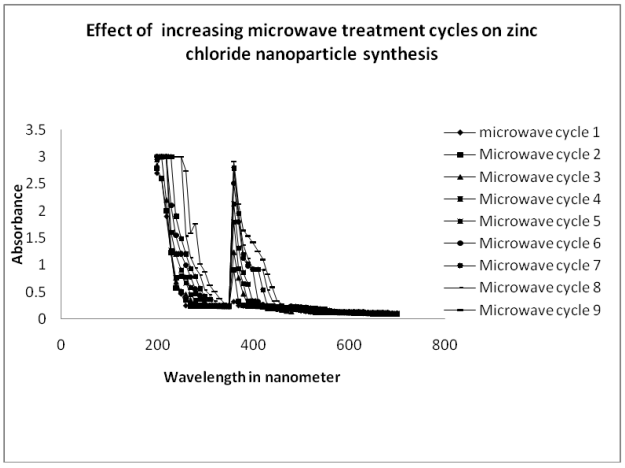

Fig.1

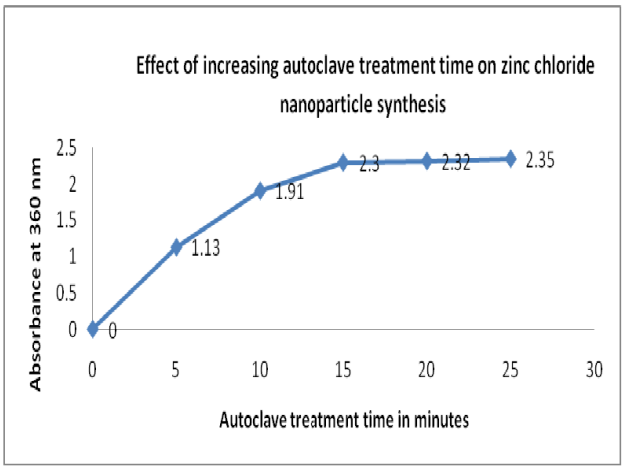

Fig. 2

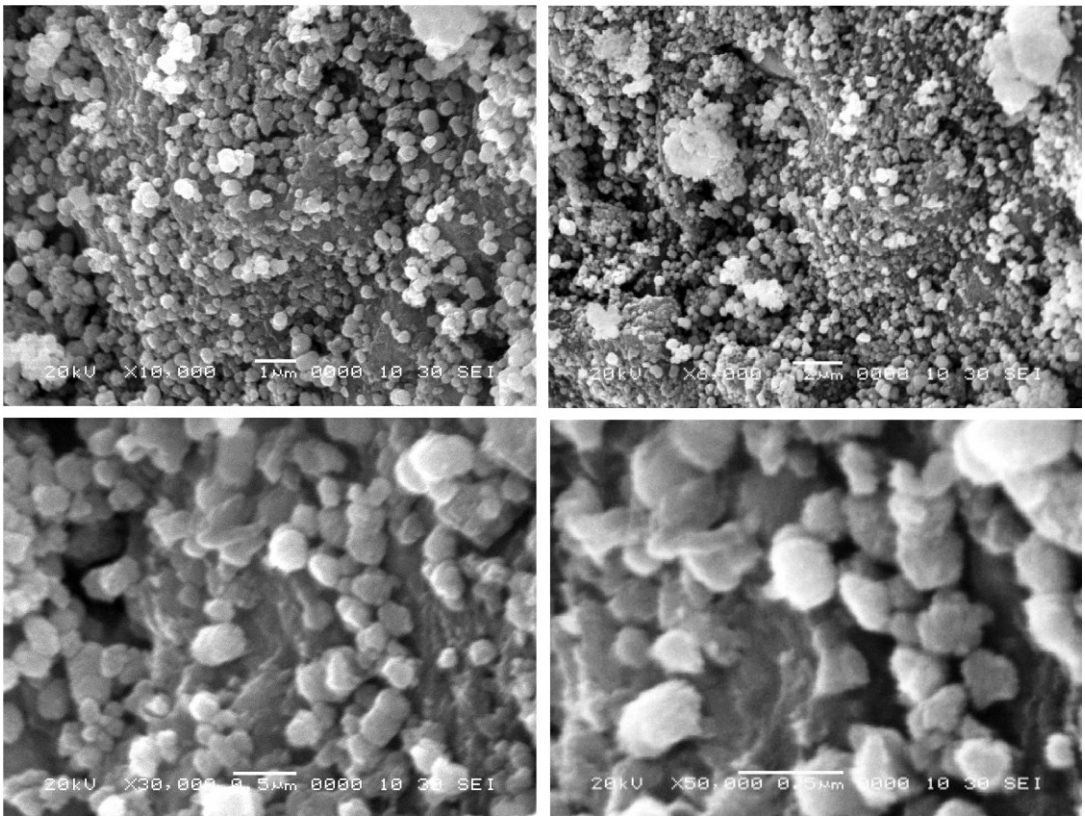

Fig.2 SEM image of synthesized zinc chloride nanoparticles

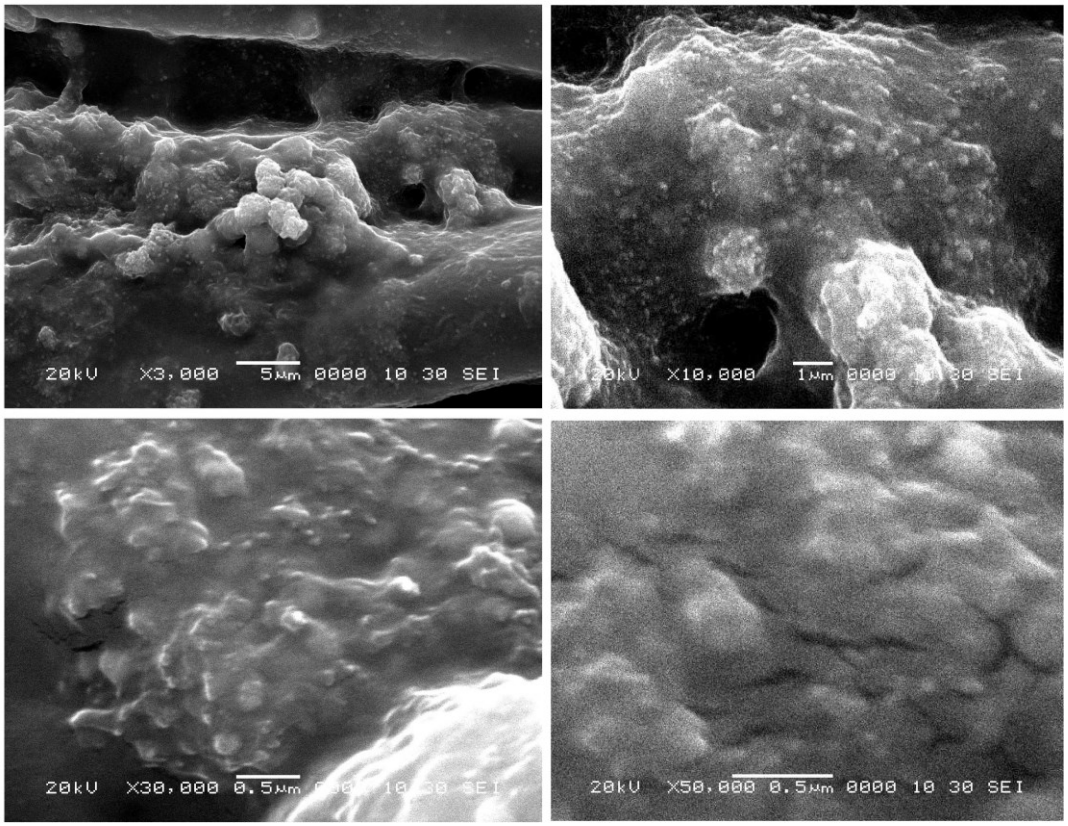

Fig. 3 : SEM image of treated fiber

	5ppm*	10 ppm	15ppm
Name of bacteria	Zone of inhibition (diameter in mm)		
Salmonela typhi	11mm	15mm	22mm
Staphyloccus aureus	03mm	04mm	07mm
Psudomonas aurogenosa	12mm	23mm	28mm
Klebsiella	4mm	9mm	16mm
Name of fungus			
Rhizopus	12mm	19mm	24mm
Candida albicans	14mm	17mm	25mm
Aspergillus niger	7mm	9mm	16mm

*1ppm zinc chloride nanoparticle solution was prepared by dissolving 1miligram of zinc chloride nanoparticle powder in 1000ml of double distilled deionized water.

Optimization of Microwave treatment cycles for zinc chloride nanoparticle synthesis:

Microwave treatment cycle was optimized at 9 cycles each comprising of 15 second duration. Treated samples showed maximum absorbance at 350 nm Each Microwave treatment cycle showed significant absorbance of autoclaved zinc chloride solution at 350 nm. Figure 1 shows the zinc chloride nanoparticle synthesis as increase inmicrowave treatment cycle confirming presence of zinc chloride nanoparticles.

FTIR Characterization

The spectrum in the range 300-4000 cm^{-1} was showing IR absorption due to the various vibrations involved. The FT-IR report of the synthesized zinc nanoparticles showed the fingerprint region peaks at 441.71, 497.86, 515.01 cm^{-1} was attributed to the ZnCl stretching mode frequency, 705.97, 731.05, 859.42 cm^{-1} due to 3, 6-anhydro - β- galactose skeletal bending in starch, 1014.59, 1134.18 cm^{-1} corresponds to the ester-sulfate link vibration, 1228.7 cm^{-1} due to C-O stretching,

1319.35 cm^{-1} due to C-H stretching, 1425.44 cm^{-1} due to C=O ring stretching and significant peaks at 1626.05 cm^{-1} corresponds to O-H bending of absorbed water, 2430.39, 2895.25 cm^{-1} due to carboxylic acid O-H stretch, 3255.95 and 3540.14 cm^{-1} corresponds to alcohol phenol O-H stretch associated with starch. All the obtained peaks indicate that the ZnCl nano particles are embedded in the starch matrix.

SEM analysis

Scanning Electron Microscopy (SEM) is used for the morphological studies of the ZnCl nano particles and starch embedded ZnCl nano particles . This characterization was done at the medium energy electrons in the range 5-50Kv in a fine beam scanning the specimen. Both X-rays and secondary electrons are emitted by the sample. SEM image of synthesized zinc chloride nanoparticles is shown in figure 2. SEM images confirmed synthesis of nanoparticles with size ranging from 50 nm to 100 nm.Figure 3 show the SEM images of ZnCl – starch nanocomposite at different magnifications. The SEM observation clearly illustrates that the Zinc chloride nanoparticles are formed in agar matrix varying in size from 50 nm – 100 nm. The low-magnification images demonstrate that the ZnCl nanoparticles are dispersed in the starch matrix (Figure 3)

The antimicrobial activity of zinc chloride nanoparticles

Antimicrobial activity of zinc chloride nanoparticles was observed as zone of inhibition. Table 1showed antibacterial and antifungal activity of Zinc chloride nanoparticles at varying concentration. ZnClNP inhibited growth of all selected bacteria and fungi. In bacteria, *Psudomonas aurogenosa* showed maximum zone of inhibition and *Staphyloccus aureus* showed minimum zone of inhibition while in fungi, *Candida albicans* showed maximum zone of inhibition and *Aspergillus niger* showed minimum zone of inhibition. Stability in inhibition activity further remained for more than 6 months.

Antimicrobial activity of zinc chloride nanoparticle embedded cotton fibre

SEM image of treated fiber (Fig. 4) further confirmed embedment of synthesized zinc chloride nanoparticles Treated cotton fibers showed significant inhibition activity against all selected bacterial and fungal pathogens.

Conflicts of interest: The authors stated that no conflicts of interest.

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