Development of Silver Nanoparticles/PEG/Glycerine Composite for Antibacterial Effect using Leaf Extract of Ocimum sanctum and Ocimum basilicum

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Abstract

The main purpose of the experiment is to use green synthesis method for silver nanoparticles (SNP) fabrication using phytochemical and functional groups inherent in aqueous leaf extract of Ocimum sanctum and Ocimum basilicum for formulation of polyethylene glycol (PEG)/Glycerine film. The SNP synthesis reaction is performed under sun condition and change in colour from light brown to dark brown was the initial indication, observed for nanoparticles synthesis. The 95 ml of 0.001 M AgNO₃ is mixed with 5 ml of leaf extract and reaction performed under Sun light at alkaline pH 9 was found efficient to produce stable NP. The synthesized SNP are mixed with [10%, 50%, 100%, 150%, 200% and 250%], polyethylene glycol (PEG)-glycerine (G) in 1:1 ratio to form a film. The UV-spectroscopic analysis confirms absorption at 420-430 nm for synthesized SNP. The FTIR characterization determines alkenes (terminal), 1°, 2° amines, amides, nitriles, alkenyl, alkyl halides functional group from O. sanctum (OS) leaf extract and aldehydes, alkenes (terminal), alkyne, alkene, from O. basilicum (OB) leaf extract responsible for reducing and capping silver nitrate to form nanoparticles. The SEM analysis verify that the O. sanctum based nanoparticles are spherical in shape although O. basilicum based nanoparticles have bright contrast coral reef like morphology. The average zeta potential of silver nanoparticles was found to be 27.74 mV and 23.50 mV that are embedded in polyethylene glycol (PEG)/Glycerine (G) polymer. \[9\] SNP shows antimicrobial activity against E. coli and S. aureus pathogens. The drug release was confirmed by UV spectrophotometry, the drug release from SNP/PEG was found to be 463.2 nm and 43.0 nm. These Sun light mediated SNP shows antimicrobial activity against E. coli and S. aureus pathogens. The SNP/PEG films. Also, the average diameters of SNP in Ocimum sanctum-SNP/PEG and in Ocimum basilicum-SNP/PEG was found to be 463.2 nm and 43.0 nm. These Sun light mediated SNP shows antimicrobial activity against E. coli and S. aureus pathogens.

Key words: Silver Nanoparticles, Ocimum sanctum, Ocimum basilicum, Biopolymer.

1. Introduction

Ocimum is a genus of aromatic annual and perennial herbs, shrubs in the family Lamiaceae, native to the tropical and sub temperate part of the globe, with the large number of species found in Africa and few species of Basil are native to India. The word Ocimum is derived from the Greek word "oio" meaning smell and is called as "king of herbs" due to its immense use in traditional system of medicine, perfumery and pharmaceutical industry [1]. The diversity of Ocimum is geographical distributed in three main regions Tropical and sub tropical regions of Africa, Tropical Asia and Tropical parts of America [Brazil] up to an altitude of about 1800 m from the mean sea level [1]. In India, so far about nine species of Ocimum have been reported including three exotic species namely O. americanum L., O. minimum L., and O. africanum Lour [1]. The Ocimum sanctum also called as Ocimum tenuiflorum (holy basil) is native to the Indian subcontinent and cultivated for religious and traditional medicine purposes of its essential oil. Similarly, Ocimum basilicum (sweet basil) is economic plant grown in India, Iran, and in few warm regions of Africa [2].

The chemical composition of Ocimum species shows clear variation with respect to seasons. The major oil constituents from different Ocimum plant species include 1, 8-cineole, linalool, pinene, eugenol, camphor, methyl chavicol, ocimen, terpinene, and limonene [3]. Ocimum species is widely used in medicine for treating digestive disorders, such as stomach ache, diarrhea, kidney complaints and infections, whooping cough and various types of fever. Moreover, they are used in curing or show antibacterial, ameliorative, antidiabetic, genoprotective, analgesic, antistress, hepatoprotective, memory booster, antipyretic, radioprotective, effect on testicular function, anticancer and anti-cataract effect. It is also effective in Alzheimer's disease, oral sub mucous fibrosis [4], anti-inflammatory and anti-nociceptive [5, 6].

The medicinal plant extracts are used in nanoscience for developments of formulation for varied application in textile, mobile phones, geo-sensing technology, paper, food, fertilizers, pesticides and agrochemical industries. The silver (Ag) is the most compatible with biological system, living organisms and medicine which make ideal candidate to form silver nanoparticles AgNPs (<100 nm) for wound healing, burns and other medical purposes [7]. Recently, the plant mediated silver nanoparticles are complexed with potential, biodegradable polymers to enhance better releasing and effective antimicrobial activity. The one of the important polymer is polyethylene glycol (PEG) which is known as an osmotic-type laxative [8] referred as polyethylene oxide (PEO), that could be cross linked into networks, can hold high water content and forms hydrogels. PEG is a condensation polymer of ethylene oxide and water. Polyethylene glycols do not hydrolyze upon storage, inhibit growth of molds and beneficial for biological, chemical and pharmaceutical applications [9].

Therefore, in present study an attempt have been made to formulate silver nanoparticles using plant extract of O. sanctum and O. basilicum for film/composite development using blend of polyethylene glycols and glycerine asa polymer to give assistance for film formation and to enhance its releasing properties. The characterization and measurements are performed to depict the importance of formulated SNP/PEG/G composite in antibacterial application.

2. Experimental Methods

2.1 General Reagents and Solutions

The 250 ml conical flask, 100 ml measuring cylinder, mortar, pestle, cover slip, various organic solvents and doubled distilled water (Millipore Milli-Q ultrapure water system). The 1 mM sodium hydroxide (NaOH), hydrochloric acid (HCl), silver nitrate (AgNO₃), glycerine (G) polyethylene glycol (PEG) is purchased from Sigma-Aldrich. The plant samples of O. sanctum and O. basilicum have been collected from predefined places mentioned in Table 1 and their leaf extract were used for experimentation.
Table 1: Display the picture, location and possible antibacterial compounds present in Ocimum sanctum and Ocimum basilicum.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Plant species</th>
<th>Location</th>
<th>Possible Antibacterial Compounds</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Ocimum sanctum</td>
<td>N2® 57 E 78° 59’ 27</td>
<td>Monoterpenes, camphor, cineole, estragol, eugenol, 8-elemene, bornyl acetate, α-, β-pinene, sesquiterpenes, camphene, methyl eugenol germacrene, campsterol, caryophyllene, bisabolene, methyl chavicol and eucalyptol [10].</td>
</tr>
<tr>
<td>2</td>
<td>Ocimum basilicum</td>
<td>N2® 57 E 78° 59’ 27</td>
<td>Methyl eugenol, methyl chavicol, terpinolene, eugenol, cubenol, linalool, methyl cinnamate-rich [11], alkaloids, tannins, flavonoids, cholesterol, terpenoids, glycosides, phenols, cardiac glycosides, carbohydrates, and phlobatannins [12].</td>
</tr>
</tbody>
</table>

2.2 Instruments and Bacterial Strains

The UV spectrophotometry (Shimadzu Dual Beam spectrometer, Model UV-1650 PC), Zeiss light microscope of Stemi DV4 (made in Germany), Fourier-transform infrared spectroscopy (FTIR), scanning electron microscopic (SEM), water bath, microwave oven and weighing balance were used in this experiment. Bacterial strains used in present antimicrobial study are Escherichia coli (ATCC no.11105) and Staphylococcus aureus (ATCC no. 6538).

2.3 Leaf Extracts Preparation

The 10 g leaf of O. sanctum and O. basilicum washed with tap water, dried at room temperature and macerated using mortar and pestle in 75 mL autoclaved double distilled water for five min. The extract was filtered using double coated cotton muslin cloth and the filtrate was set for centrifugation at 4000 rpm or 1792 relative centrifugal force (RCF) or G force for 10 min. The 65 mL of supernatant was stored in refrigerator for further fabrication of SNP.

2.4 Silver Nanoparticle Synthesis

The 95 mL of 0.001 M silver nitrate (AgNO₃) is mixed with the 5 mL leaf extract of O. sanctum and O. basilicum independently. The reaction was performed in Sun light (June 2018; temperature 35-45 °C) and after 5 min 0.0001 M NaOH solution was added to mixture solution to maintain alkaline pH 8 (Fig. 1). The reaction mixture of silver nitrate and leaf extract was observed through visual change in colour [7].

2.5 Preparation of SNP/PEG/G Biopolymer

The polyethylene glycol 10%, 50%, 100%, 150%, 200% and 250% mixed with 1 mL of synthesized SNP solution and glycerine was added (1:1) ratio with respect to PEG. The SNP/PEG/G biopolymer complexes solution was then kept in water bath at 70 °C for 30 min. Finally, the SNP/PEG/G biopolymer was set for microwave oven heat burst of 30 sec for five times. The pale yellow colour and homogenous solution of SNP/PEG/G biopolymer complexes are allowed to cool down and poured over cover slip (22 mm size) (Fig. 2).

2.6 Characterization of Silver Nanoparticles

The O. sanctum (OS) and O. basilicum (OB) leaf extract, Ocimum sanctum-nanoparticles (OS-NP) and Ocimum basilicum-nanoparticles (OB-NP) and film (OS-NAV/PEG/G), (OB-NAV/PEG/G) fetched for characterization, measurements and application activities.

2.7 UV-Visible Spectroscopy

The instrument was set for base line correction and auto zero using ddH₂O and 1 µl of extracts, SNP films OS-NAV/PEG/G and OB-NAV/PEG/G were placed on the central top of the cuvette of nanophotometer for measuring UV-Vis spectra at a resolution of 200–800 nm.

2.8 FTIR Spectroscopy

The O. sanctum and O. basilicum leaf extract and their silver nanoparticles (OS-NP) and (OB-NP) were characterised using FTIR in 4000 to 450 cm⁻¹ at a resolution of 4 cm⁻¹.

2.9 Solubility Test

The solubility test of the prepared films (OS-NAV/PEG/G, OB-NAV/PEG/G) has been done by adding distilled water over film in petri dishes. The time to dissolve film, photographs and videos have recorded during the test. Furthermore, the individual solubility of SNP at RT and boiling at 70 °C for 20 min and glycerine at RT was done in various solvents to check solubility potential.

2.10 SEM Analysis

The OS-NP, OB-NP, OS-NAV/PEG/G and OB-NAV/PEG/G films without glycerine were used for scanning electron microscopic analysis using Carl Zeiss SEM-EVO18 with accelerated voltage of 130 kV. The morphology and size of silver nanoparticles and SNP/PEG-films were investigated using SEM specific software. The magnification was set at 4.85 KX for (OS), 9.83 KX (OB), 4.00 KX for OS/NAV/PEG/G and 3.39 KX for OB/NAV/PEG/G.

2.11 DLS Size and Zeta Potential Analysis

The synthesis OS-NAV/PEG and OB-NAV/PEG films were fetched for determination of size analysis and to identify zeta potential surface charge over nanoparticles in films. The nanoparticles are subjected to Brownian motions due to thermal energy that produces Rayleigh scattering. The size of nanoparticles in film are determined by using Particle Sizing Systems Inc. Santa Barbara, Calif., USA at CICR-CIRCOT Mumbai, India and the Number-Weighted Gaussian distribution analysis (solid particle), intensity-wt, volume-wt NICOMP distribution and zeta potential in (mV) were calculated.

2.12 Antimicrobial Test

Antimicrobial properties of O. sanctum and O. basilicum leaf extract, OS-NP and OB-NP, OS-NAV/PEG/G and OB-NAV/PEG/G film against Escherichia coli and Staphylococcus aureus is done as per the protocol followed by [10]. The extract, SNP and their films are placed on culture of E. coli and S. aureus spreader agar plates. The inhibition of bacteria has visualized after 24 h of incubation at 37 °C and the zone of inhibition was measured.

3. Results and Discussion

3.1 Visual Conformation

The change in colour from light brown to dark brown colour was visualised by naked eyes and addition of 1 mM NaOH to the reaction mixture to maintain pH 8 make the solution’s colour more dark (Fig. 1).

3.2 UV-Visible Analysis

The UV-visible analysis has been done for leaf extract of O. sanctum and O. basilicum, their silver nanoparticles (OS-NAV and OB-NAV) and films OS-NAV and OB-NAV were calculated.

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SNP/PEG/G and OB-SNP/PEG/G (Fig. 3). The synthesized SNP exhibited a well-defined absorption peak at 430 nm for O. basilicum and 420 nm for O. sanctum.

Inhibition.

prominent. The other combination of SNP and (PEG+G) shows no inhibition. The Fig. 4 and Table 2 shows the FTIR spectra of leaf extract of O. sanctum and O. basilicum and their SNPs.

3.3 Nanoparticles Concentration

The concentration parameters in nanophotometer was set as wavelength 420 nm, units picomole/µl, pathlength 10 mm and factor 50.0. The concentration parameters of O. sanctum-NP at wavelength 410 nm were found to be 7.0 picomole/µl and absorbance 0.140 Å. Likewise, the concentration parameter of O. basilicum-NP at wavelength 420 nm was found to be 7.500 picomole/µl with absorbance 0.150 Å.

3.4 FTIR Analysis

The FTIR was used to identify the potential functional group, chemical compounds and biomolecules present in the leaf extract of O. sanctum and O. basilicum that are involved in capping and reducing Ag⁺ ions to Ag. The Fig. 4 and Table 2 shows the FTIR spectra of leaf extract of O. sanctum and O. basilicum and their SNPs.

3.5 Light Microscopic Analysis

The best formulated films using O. sanctum and O. basilicum are observed under light microscopy in 10X focus for observation of SNP in PEG/G composite (Fig. 5).

3.6 Solubility Check

The OS-SNP/PEG/G and OB-SNP/PEG/G films were used to check solubility in double distilled water (Fig. 6). From the tested samples of concentration 10%, 50%, 100%, 150%, 200% and 250% glycerine and PEG (1:1) ratio, the sample B and 2 (150%) have optimized for giving best texture, film forming capability and solubility in water. Besides, the solubility of glycerine has performed in different solvents. However, various concentrations of PEG were used to check the solubility potential and dissolve at room temperature (RT), at 70 °C(Table 3).

3.7 Antimicrobial Potential of SNP/PEG/G Film

Antimicrobial potential of OS-SNP/PEG/G and OB-SNP/PEG/G film in (150%PEG+150%) concentration were tested against Escherichia coli and Staphylococcus aureus (Fig. 7). A small inhibition could be seen in O. sanctum/SNP/PEG/G (150%PEG+150%) films however it is not prominent. The other combination of SNP and (PEG+G) shows no inhibition.

Table 2 Functional group detected on the basis of FTIR spectra for O. sanctum (OS) and O. basilicum (OB) leaf extract, OS-SNP and OB-SNP with its absorption range, absorption frequency strength and characteristic bond

<table>
<thead>
<tr>
<th>Peaks</th>
<th>Absorption</th>
<th>Characteristic bond</th>
<th>Functional group</th>
<th>Peaks</th>
<th>Absorption</th>
<th>Characteristic bond</th>
<th>Functional group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ocimum sanctum (OS) leaf extract</td>
<td></td>
<td></td>
<td></td>
<td>Ocimum sanctum-Nanoparticles (OS-SNP)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>3328.169 716.474</td>
<td>–C≡C–H: C–H stretch</td>
<td>alkynes (terminal)</td>
<td>1</td>
<td>3328.169 716.474</td>
<td>–C≡C–H: C–H stretch</td>
<td>alkynes (terminal)</td>
</tr>
<tr>
<td>2</td>
<td>3289.560 –3012.368</td>
<td>–C≡C–H: C–H stretch</td>
<td>alkynes (terminal)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>2270.803 17.519</td>
<td>–C≡C–C–H stretch</td>
<td>alkynes</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>2118.075 17.519</td>
<td>–C≡C–C–H stretch</td>
<td>alkynes</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>1636.350 2845.490</td>
<td>–C≡C–C–H stretch</td>
<td>alkynes</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ocimum basilicum (OB) leaf extract</td>
<td></td>
<td></td>
<td></td>
<td>Ocimumbasilicum-Nanoparticles (OS-SNP)</td>
<td></td>
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<tr>
<td>1</td>
<td>3328.169 716.474</td>
<td>–C≡C–H: C–H stretch</td>
<td>alkynes (terminal)</td>
<td>1</td>
<td>3328.169 716.474</td>
<td>–C≡C–H: C–H stretch</td>
<td>alkynes (terminal)</td>
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<td>–C≡C–H: C–H stretch</td>
<td>alkynes (terminal)</td>
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<td>2118.075 17.519</td>
<td>–C≡C–C–H stretch</td>
<td>alkynes</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>5</td>
<td>1636.350 2845.490</td>
<td>–C≡C–C–H stretch</td>
<td>alkynes</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Fig. 5 Represents the light microscopic structure of SNP/films samples of O. basilicum and O. sanctum synthesized using different concentration of PEG and glycerine (100%PEG+100%G; 150%PEG+150%G)
3.8 SEM Analysis

The SEM analysis of OS-SNP, OB-SNP and the films that are synthesized by leaf extracts of Ocimum species labelled OS/SNP/PEG and OB/SNP/PEG have done. The Fig. 8 shows the SEM analysis of the SNP and film, yet the film or the composite were devoid of glycerine since the dry sample is required for SEM characterization, thus glycerine was not added in film for investigation of SEM characters. The shape of OS-SNP is spherical OB-SNP is like bright contrast coral reef, OS/SNP/PEG composite shape is kind of rosette shape and OB/SNP/PEG is somewhat irregular in shape.

Table 3: Solubility potential of glycerine and PEG at room temperature (RT)

<table>
<thead>
<tr>
<th>SNo</th>
<th>Solvent system</th>
<th>Samples</th>
<th>Cond.</th>
<th>Dissolved time</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Water*</td>
<td>Glycerine</td>
<td>RT</td>
<td>2 sec*</td>
</tr>
<tr>
<td>2</td>
<td>Methanol</td>
<td>Glycerine</td>
<td>RT</td>
<td>4 sec</td>
</tr>
<tr>
<td>3</td>
<td>Ethanol*</td>
<td>Glycerine</td>
<td>RT</td>
<td>1 sec*</td>
</tr>
<tr>
<td>4</td>
<td>Chloroform</td>
<td>Glycerine</td>
<td>RT</td>
<td>Immiscible</td>
</tr>
<tr>
<td>5</td>
<td>Toluene</td>
<td>Glycerine</td>
<td>RT</td>
<td>Immiscible (bubbles at bottom)</td>
</tr>
<tr>
<td>6</td>
<td>Ethyl acetate</td>
<td>Glycerine</td>
<td>RT</td>
<td>Immiscible</td>
</tr>
<tr>
<td>7</td>
<td>Glacial acetic acid</td>
<td>Glycerine</td>
<td>RT</td>
<td>10 sec</td>
</tr>
<tr>
<td>8</td>
<td>Formic acid</td>
<td>Glycerine</td>
<td>RT</td>
<td>2 sec</td>
</tr>
<tr>
<td>9</td>
<td>Butyl alcohol</td>
<td>Glycerine</td>
<td>RT</td>
<td>6 sec</td>
</tr>
<tr>
<td>10</td>
<td>Benzene</td>
<td>Glycerine</td>
<td>RT</td>
<td>Immiscible (bubbles at bottom)</td>
</tr>
<tr>
<td>11</td>
<td>Petroleum ether</td>
<td>Glycerine</td>
<td>RT</td>
<td>Immiscible</td>
</tr>
<tr>
<td>12</td>
<td>Water*</td>
<td>PEG</td>
<td>RT</td>
<td>23 sec</td>
</tr>
<tr>
<td>13</td>
<td>Methanol</td>
<td>PEG</td>
<td>RT</td>
<td>2 min 30 sec</td>
</tr>
<tr>
<td>14</td>
<td>Ethanol</td>
<td>PEG</td>
<td>RT</td>
<td>2 min</td>
</tr>
<tr>
<td>15</td>
<td>Chloroform</td>
<td>PEG</td>
<td>RT</td>
<td>20 sec (white ppt)</td>
</tr>
<tr>
<td>16</td>
<td>Toluene</td>
<td>PEG</td>
<td>RT</td>
<td>Immiscible</td>
</tr>
</tbody>
</table>

Note: *represent quick solubility

3.9 DLS Size and Zeta Potential Analysis

The average zeta potential was found to be 27.74 mV and 23.50 mV for with average mobility is 2.07 M.U. and 1.75 M.U for silver nanoparticles integrated in Ocimum sanctum-SNP/PEG and Ocimum basilicum-SNP/PEG film respectively. The doppler frequency distribution to identify zeta potential and time history for 1 min are represented in Fig. 9. The size and intensities of silver nanoparticles in composite Ocimum sanctum-SNP/PEG and Ocimum basilicum-SNP/PEG is shown in Fig. 10.

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In present study, the silver nanoparticles from the leaf extract of Ocimum sanctum (OS) and Ocimum basilicum (OB) have synthesized successfully and exhibited absorption peak at 420 nm for O. basilicum while 410 nm for O. sanctum. Similarly, the research of Malaparla [13] shows the absorption peak at 438 nm from O. basilicum and 439 nm for O. sanctum. The difference in SNP absorption peak may be due to the involvement of different functional group and the method adopted for leaf extract preparation. The films containing nanoparticles such as OS-SNP, OB-SNP, OB-SNP/PEG have been evaluated for presence of silver nanoparticles using UV spectroscopic analysis in range 500 nm – 800 nm and a sharp peak around 400 – 4500 nm confirms silver nanoparticles incorporated in polymers, besides it was observed that the nanoparticles were stable for 45 days (Fig. 3). The wider range of chemical groups present in aqueous extract of Ocimum sanctum are alyknes (terminal), 1,2 amines, amides, nitriles, alkynes, alkanes and alkyl halides. Similarly, alkyynes (terminal), aldehydes, alyknes and alkyenes are found in Ocimum basilicum are mainly responsible for reduction of Ag⁺ ions to zero valent Ag⁰. The FTIR spectra of the crude aqueous extract and SNPs derived from O. sanctum showed a decrease in stretching frequency from 3333-204 to 3328-169 cm⁻¹ suggesting an alykynes functional group. In the same way, Malaparla [13] suggested the range of 3230-3400 cm⁻¹ for amide region. Further more a shift from 1638.683 to 1636.350 cm⁻¹ is assigned to the N-H bend and alyknes, whereas peaks at 1217.445, 95.397 cm⁻¹ can be attributed to alkyl halides. The other groups that found are 1,2 amines, amides, nitriles, and alkyl halides. In contrast, O. basilicum consist of aldehydes, which shift from 2270.83 to 2119.810cm⁻¹. Overall, the FTIR analysis predicts that SNP have formed using O. sanctum and O. basilicum extracts confirming the involvement of C=Cs:H - C-H stretch and H-C=O: C-H stretch respectively (Table 2).

The nature of Ocimum sanctum-NP and Ocimum basilicum-NP are colloidal and are capped using respective phyto chemical present in extract of Ocimum species. The reference was set to zero using both plant extract as well as water, they have generated the concentration of Ocimum sanctum-NP, Ocimum basilicum-NP using nanophotometer as 0.900 picomole/µL and 3.500 picomole/µL respectively. The film of composite of O. sanctum-NP and O. basilicum-NP was formulated which was used to depict the solubility of in water. In addition, individual PEG and glycerine was also used to check the solubility in different solvents (Table 3). The polymer PEG is dissolved in water by two methods: room temperature and boiling at 70 °C for 20 min. It was observed that the boiling of PEG makes it to dissolve quickly in water when compared with room temperature method. The 10% PEG dissolved in 1 min at room temperature whereas; at 70 °C it was dissolved in 20 sec. Also, 100% PEG in room temperature requires 26 min, however on boiling at 70 °C takes less time of 1 min 40 sec. The boiling process for film formation was found more efficient than room temperature method. There are various papers on silver nanoparticles synthesis using PEG with combination of other polymer such as glucose [14] however none is there with glycerine.

![Image](https://doi.org/10.30799/jnst.161.18040517)


4. Conclusion

In conclusion, this study shows that the aqueous soluble compounds presents in leaf extract of O. sanctum and O. basilicum are efficient for fabrication of silver nanoparticles. In addition, the combination of PEG and glycerine is efficient for metal synthesis in aqueous media. The silver nanoparticles in composite are synthesized in reducing conditions. The FTIR analysis confirms the presence of SNP show that the SNP in same composite have 25% of distribution < 402.8 nm, 50% of distribution < 606.6 nm, 75% of distribution < 972.9 nm, 90% of distribution < 1527.9 nm, 99% of distribution < 3409.4 nm and 80% of distribution < 1099.7 nm. However, mean diameter (nm) in term of intensity weighting and volume weighting in composite Ocimum sanctum/PEG/SNP and OB/SNP/PEG composite found to be 43.0 and 465.4 nm. Additionally, the cumulative result based on number-weighted Gaussian distribution confirms the presence of SNP, 25% of distribution < 122.8 nm, 50% of distribution < 163.9 nm, 75% of distribution < 239.6 nm, 90% of distribution < 354.2 nm, 99% of distribution < 735.0 nm and 80% of distribution < 266.9 nm. Therefore the composite of SNP of average diameter >463.2 nm in Ocimum sanctum/PEG/SNP and 43.0 nm in Ocimum basilicum-SNP/PEG could inhibit pathogens. However we could not observed such activity that may be due to some unknown reasons which is still unclear. Pinzaru [15] used PEG-coated SNPs for in vitro and in vivo studies in mice for a toxicological studies.
extract of *O. sanctum* and alkynes, aldehydes and alkenes in *O. basilicum* as sole stabilizing agents for silver ions. The optimized film *O. sanctum /SNP/PEG/G and O. sanctum /SNP/PEG/G150%PEG150%G* are best in texture, film forming ability, quickly dissolve in water and slow release of SNP. The SEM characterization confirms the spherical shape and bright contrast coral reef for OS-SNP and OB-SNP respectively, although the SNP incorporated film of *O. sanctum* OS/SNP/PEG has rosette shape while *O. basilicum* based OB/SNP/PEG film is irregular in shape. The average zeta potential was found to be 27.74 mV and 23.50 mV for with average mobility is 2.07 MU and 1.75 MU for silver nanoparticles integrated in Ocimum sanctum-SNP/PEG and Ocimum basilicum-SNP/PEG film respectively. The Ocimum basilicum-SNP/PEG have much stable and SNP below 100 nm ie., 43.0 nm as compare to Ocimum sanctum-SNP/PEG which have SNP of average diameters 463.2 nm and could demonstrate study for its antimicrobial potential against *E. coli* and *S. aureus* pathogens which could be possible for development of products helpful in medical application.

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