Synthesis and evaluate silver nanoparticles containing *Momordica charantia* Linn.
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Abstract: The physicochemical and optoelectronic properties of metallic nanoparticles are strongly dependent on the size and size distribution of the nanoparticles. In this study, the silver nanoparticles were synthesized from the leaf extract of *Momordica charantia* at room temperature as well as stirred at 60°C. The effects of different leaf extract concentrations, metal ions concentration, reaction times and reaction temperatures on the synthesis of silver nanoparticles were evaluated. The nanoparticles were characterized by UV-Visible, XRD, SEM, and FTIR. The UV-Vis spectra showed that the Surface Plasmon Resonance peak of silver colloids synthesized from *Momordica charantia* leaf extract was observed at 426 nm for stirred at 60°C and room temperature condition. X-ray diffraction (XRD) analysis confirmed that the nanoparticles were crystalline in nature with Face Centered Cubic structure. Scanning Electron Microscopy (SEM) analysis showed that silver nanoparticles were spherical in shape. The FTIR measurement was carried out to identify the possible functional groups responsible for the efficient stabilization of silver nanoparticles.

Key words: *Momordica charantia*; size and size-distribution; XRD; Silver Nanoparticles

Introduction
Nanotechnology is a broad-based science involving manipulation of atoms, electrons, protons and neutrons in a variety of ways to generate new understanding of how materials can be developed to solve many problems in medicine, engineering, agriculture, surface science, marine science, and geology. It involves in the dimensions at nanoscale size ranging up to 100 nm. Nanoparticles has potential applications in various fields such as healthcare, food and feed, cosmetics, environmental health, biomedical science, chemical industries, drug and gene therapy, electronics, mechanics, and space industries. It also has been achieved extensively in the drug delivery system for the treatments of cancer, diabetes, allergy, infection and inflammation.

In recent years Green synthesis provides an advancement over chemical and physical method as it is cost effective, environment friendly, easily scaled up for large scale synthesis. This technique eliminates the use of energy, high pressure, temperature and toxic chemicals. As plant medicated nanoparticles preparation is easy to handle, safe and economical. It finds more advantages over chemical and physical method. In biological method plants have been used for the synthesis of nanoparticles were coated by the plant extract which has medical benefits and can be used as drug and cosmetic application.

Figure: Nanoparticles
*Momordica charantia* or Bitter Melon is a Tropical vegetable, is a common food in Indian cuisine and is used extensively in folk medicine as a remedy for diabetes. Bitter melon has been used in various Asian traditional medicines for a long time. The
major parts used are leaves, fruits and flowers. Leaves are simple; usually palmate 5-7 lobed, tendrils unbranched or 2 branched. The herbaceous, tendril bearing vine grows to 5 m. It bears simple, alternate leaves 4-12 cm across, with 3-7 deeply separated lobes. The main constituents of M. Charentia are Alkaloids, Charentia, Momerdica, ascorbic acid, phenol and protein. In the present study, the biosynthesis of silver nanoparticles using the leaf extract of *Momordica charantia* has been reported which belongs to the family Cucurbitaceae. Synthesized nanoparticles were characterized with UV-Visible spectroscopy, XRD, FTIR, and SEM and the antidiabetic activity.

**Materials and Methods**

*Momordica charantia* Purchased from Sunpure Extract Private Limited, Silver Nitrate Purchased from Myochem Mumbai.

**Preformulation studies:**

**Physical Characteristics:**
By visual examination, the drug was identified for physical characters like colour, texture, smell, taste.

**Solubility studies:**
Solubility of *Momordica charantia* was studied in distilled water, organic solvents and phosphate buffer.

**Compatibility study**

FTIR was used to find whether any kind of interaction between drug and Excipients. IR spectra for nanoparticles were taken separately to know the interactions. using Perkin Elmer Spectrum Version 21. The scanning range used was from 4000-400 cm$^{-1}$ at a scan period of 3 min.

**Determination of λmax by UV Spectroscopy:**

UV-visible spectrophotometer was used to determine the absorbance maximum (λ-max) of *Momordica charantia* using a digital double - beam recording spectrophotometer with scanning range of 200-400 nm. Solution of Nanoparticles was prepared using double distilled water. Shimadzu UV-visible spectrophotometer UV-1800 with spectral bandwidth of 1 nm, ± 0.3 nm wavelength accuracy and 10 mm pair of quartz cells were used to record the spectral and absorbance readings.

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**Figure 1:** Different Method of Nanoparticles

**Green synthesis of silver nanoparticle**

**Synthesis of *Momordica charantia* Linn Silver Nanoparticles**

**Preparation of *Momordica charantia* Linn. Extract:**
Dried powdered *Momordica charantia* 5 gm was mixed 100 ml distilled water then the solution was kept for continuous heating at 80°C for 1 hr at room temp. with frequent shaking. After that extract were filter by using whatman no 1 filter paper. The extract was collected and stored at 40°C for further use.

**Synthesis of Silver Nanoparticles from *Momordica charantia* Linn.:** 10ml of aqueous extract of *Momordica charantia* Was added into 90ml of aqueous solution of 1ml silver nitrate the mixture was exposed to a range of controlled temp for 24 hr. appearance of brown colour in solution intended the formation of agnp. The solution was then kept in dark for further analysis collected and stored for 4°C for further use.

**Separation of Silver Nanoparticles**
The synthesized *Momordica charantia* Linn silver nanoparticles were separated by centrifugation using REMI centrifuge at 10000 rpm for 15 minutes the supernatant liquid was discarded and the pellets were collected & store.

**Table 1: Formulation Table**

<table>
<thead>
<tr>
<th>Formulation Code</th>
<th>Extract of <em>Momordica charantia</em> (ml)</th>
<th>1mm Silver Nitrate solution (ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>80</td>
<td>20</td>
</tr>
<tr>
<td>F2</td>
<td>85</td>
<td>15</td>
</tr>
<tr>
<td>F3</td>
<td>90</td>
<td>10</td>
</tr>
</tbody>
</table>

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Results and Discussion

Figure 2: Green synthesis of Silver Nanoparticles

Results and Discussion

Physical Appearance:

Table 2: Physical Appearance

<table>
<thead>
<tr>
<th>Sr.No.</th>
<th>Properties</th>
<th>Observation</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Description</td>
<td>powder</td>
</tr>
<tr>
<td>2</td>
<td>Colour</td>
<td>Dark green</td>
</tr>
<tr>
<td>3</td>
<td>Odour</td>
<td>Characteristic</td>
</tr>
<tr>
<td>4</td>
<td>Taste</td>
<td>Bitter</td>
</tr>
</tbody>
</table>

Solubility study:

Table 3: Solubility Study

<table>
<thead>
<tr>
<th>Solubility</th>
<th>Solvent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Freely Soluble</td>
<td>Methanol</td>
</tr>
<tr>
<td>Slightly Soluble</td>
<td>Ethanol, Acetic Acid</td>
</tr>
<tr>
<td>Practically Soluble</td>
<td>Petroleum Ether</td>
</tr>
</tbody>
</table>

Compatibility study:

This mixture was then scanned over a wave number range of 4000 to 400 cm⁻¹. The FTIR of pure drug and physical mixture of formulation ingredients of optimized batch were measured using Fourier Transform Infrared Spectrophotometer (Model FTIR8400S, Shimadzu). The amount of each formulation ingredient in the physical mixture was same as that in the optimized batch. The pure drug and physical mixture were then separately mixed with IR grade.

Calibration curve:

10mg of drug in 10ml Phosphate Buffer ph 6.8 to give a concentration of 1 mg/ml. It appropriately diluted with Phosphate buffer ph 6.8 into get a concentration 100μg/ml. The standard solution was scanned in the wavelength region of 200-400 nm & observed at 279 nm.

Table 4: Concentration Verses Absorbance

<table>
<thead>
<tr>
<th>Concentration</th>
<th>Absorbance</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>4</td>
<td>0.070</td>
</tr>
<tr>
<td>8</td>
<td>0.150</td>
</tr>
<tr>
<td>12</td>
<td>0.244</td>
</tr>
<tr>
<td>16</td>
<td>0.350</td>
</tr>
<tr>
<td>20</td>
<td>0.430</td>
</tr>
</tbody>
</table>

In the Standard Curve Concentration Verses Absorbance Were plotted and given in the figure

Figure 4: Calibration Curve

Evaluation of Nanoparticles

Scanning Electron Microscopy
Transmission Electron Microscopy
PX-ray Diffraction
Fourier Transmission infrared Spectroscopy
UV-Visible Spectroscopy
Entrapment Efficiency

Scanning Electron Microscopy
SEM analysis of the synthesized Momordica charantia Linn silver Nanoparticles was performed to evaluate the surface morphology of nanoparticles. Silver nanoparticles were prepared and dried well to remove the moisture content and images were taken by using Hitachi X650, Tokyo, Japan. The SEM images were taken in different magnification such as 1000 X, 3000X and shown in the figures
Transmission electron microscopy  
TEM analysis of the synthesized *Momordica charantia* Linn silver Nanoparticles was performed to evaluate the nanoparticles. In the observation size determined by 34.13 nm, 17.69 nm, and 35.37 nm. Silver nanoparticles were prepared and dried well to remove the moisture content and images were taken by using Hitachi X650, Tokyo, Japan.

**XRD analysis**
XRD spectrum showed distinct diffraction peaks around 27.86°, 32.28°, 46.25° in the 2θ range 10-90° which are indexed by the cubic face-centered silver. Thus, XRD pattern clearly showed that the silver nanoparticles formed in this synthesis were crystalline in nature. These sharp Bragg peaks might have resulted due to capping agent stabilizing the nanoparticles. Intense Bragg reflections suggest that strong X-ray scattering centers in the crystalline phase and could be due to capping agents. Independent crystallization of the capping agents was ruled out due to the process of centrifugation and redispersion of the pellet in Millipore water after nanoparticles formation as a part of purification process. Therefore, XRD results also suggested that the crystallization of the organic phase occurs on the surface of the silver nanoparticles or vice versa. Generally, the broadening of peaks in the XRD patterns of solids is attributed to particle size effects. Broader peaks signify smaller particle size and reflect the effects due to experimental conditions on the nucleation and growth of the crystal nuclei.

**Table 4: P-XRD Range of *Momordica charantia* Silver Nanoparticles.**

<table>
<thead>
<tr>
<th>Angle 2-Theta °</th>
<th>D value Angstrom</th>
<th>Intensity count</th>
<th>Intensity %</th>
</tr>
</thead>
<tbody>
<tr>
<td>27.86</td>
<td>3.19</td>
<td>391</td>
<td>59.2</td>
</tr>
<tr>
<td>32.28</td>
<td>2.77</td>
<td>660</td>
<td>100</td>
</tr>
<tr>
<td>46.25</td>
<td>1.96</td>
<td>226</td>
<td>34.2</td>
</tr>
</tbody>
</table>

**FTIR spectroscopy of *Momordica charantia* Linn. Silver nanoparticles**
FTIR measurements were carried out to identify the biomolecules responsible for capping and stabilization of metal nanoparticles synthesized. The IR spectrum of *Momordica charantia* Linn. Silver nanoparticles have following interpretations.

**Table 5: interpretations of Silver nanoparticles.**

<table>
<thead>
<tr>
<th>Functional Group</th>
<th>Standard Peak</th>
<th>Observed Peak</th>
</tr>
</thead>
<tbody>
<tr>
<td>C-H Bending</td>
<td>700-850</td>
<td>844.77</td>
</tr>
<tr>
<td>C-N Vibration</td>
<td>1000-1400</td>
<td>1014.63</td>
</tr>
</tbody>
</table>

**Figure 5:** Sem Analysis of *Momordica charantia* Linn Silver Nanoparticles at 3000x Magnification.

**Figure 6:** Tem Analysis of *Momordica charantia* Linn Silver Nanoparticle.

**Figure 7: XRD Analysis of *Momordica charantia* Linn Silver Nanoparticle.

**Figure 8: FTIR Spectrum of *Momordica charantia* Linn Silver Nanoparticles.**

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UV visible spectral analysis
UV visible spectral analysis characterizes the formation and completion of silver nanoparticles. The reduction of silver ions was monitored by measuring Uv- Vis spectrum of reaction medium from the wavelength of 200 – 800 nm by using distilled water as blank.

The reduction of Ag+ to Ag0 via the active bio molecules present in the *Momordica charantia* Linn. was indicated by a colour change from Pale yellow to brown colour. Silver nanoparticles exhibit Plasma absorption band in the visible region. The metal nanoparticles have free electrons which gives the surface Plasma resonance absorption band due to the combined vibrations of electrons of metal nanoparticles in resonance with light wave.

![Figure 9: Uv-visible Spectra of *Momordica charantia* Linn Silver Nanoparticle.](image)

Silver nanoparticles are known to exhibit UV – Visible absorption in the range of 400-500nm. The sharp absorption bands of *Momordica charantia* Linn silver nanoparticles were observed at around 426 nm.

Drug entrapment efficiency
Drug entrapment can be determined from the supernatant liquid of *Momordica charantia* Linn silver nanoparticles after centrifugation by uv Spectrophotometry at 426 nm. A calibration curve was plotted between concentrations vs. absorbance. From this standard curve amount of drug present in supernatant liquid was determined by Centrifugation Method.

\[
\text{% Entrapment efficiency} = \left( \frac{\text{Total drug content} - \text{drug content in supernatant}}{\text{Total drug content}} \right) \times 100
\]

The % entrapment of drug or drug content of *Momordica charantia* Linn. Silver nanoparticles were found to be 92.88%.

Conclusion
Synthesis of silver nanoparticles is the novel approach to enhance the bioavailability of Antidiabetic herbal drugs like *Momordica charantia*.

References


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