

A NOVEL ANTI-DANDRUFF FORMULA THROUGH GREEN TECHNOLOGY

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Introduction

Dandruff is an alarming problem among people of all age groups. Dandruff is characterized by scaling of scalp and skin. It is caused by *Malassezia* species such as *Malassezia pachydermatis*, *Malassezia furfur* and *Malassezia sympodialis*. Through antidandruff shampoos, the dandruff can only be eradicated temporarily. Moreover, recurrence and side effects are common problem. However, these problems can be circumvented through nanotechnology.

Nanoparticle synthesis methods can be categorized into three a) chemical methods b) physical methods c) biological methods. During chemical synthesis, the presence of some toxic chemical species adsorbed on the surface of nanoparticles that may have adverse effects in medical applications and is considered to be less biocompatible. Concerning the biological application of nanoparticles, it has been emphasized that methods of synthesis using biological systems viz microbes, algae, diatoms, plants either intra or extracellularly would make the nanoparticles more biocompatible.. But using plants for nanoparticle synthesis can be advantageous over other biological processes because it eliminates the elaborate process of maintaining cell cultures and can also be suitably scaled up for large scale synthesis of nanoparticles. The advantages of using plants for nanoparticle synthesis include use of cheap, non-toxic environmentally benign precursors and simple procedures without time consuming polymerization and problem with treatment of a highly viscous polymeric resin.

We have already reported that the ketoconazole coated gold nanoparticles have synergistic control over the growth of *Malassezia pachydermatis*. We have also observed the same effect with silver nanoparticles, obtained via chemical route. Synthesis of gold nanoparticles is bit expensive. As a next step, we have synthesized silver nanoparticles via biological route, using fenugreek leaves. The particles were tested for antidandruff activity. The activity was compared with that of the silver nanoparticles synthesized via chemical route.

Methodology

Fenugreek, a commonly used spice in India, has wide applications in medicine. In addition, there are also evidences for its non-toxicity. Histopathological studies in Wistar rats show that long term administration of fenugreek does not produce any alteration in the tissue architecture with no side effects. We synthesized nanoparticles by the bio-reduction of silver ions in the aqueous silver nitrate solution using fenugreek aqueous leaf extract as described earlier. Our study had three phases: 1. Green synthesis of silver nanoparticles using fenugreek leaves. 2. Characterisation of the silver nanoparticles. 3. Analysis of the antidandruff activity of the particles, using ketoconazole as a reference drug.

The morphology and the size of the nanoparticles were analysed using Scanning electron microscopy (SEM) as shown in figure 1. The SEM image confirms the nano size range of silver. The particles show polydispersity, and spherical morphology, with size ranging

between 70-90 nm. However if high resolution SEM and TEM images are taken it would better reveal the size of silver nanoparticles.

Dandruff scales were collected from the scalp of volunteers belonging to the age group of 20-30 years. This was done using sterile forceps/comb. The scales were inoculated into petri plate containing Sbouraud Dextrose Agar medium using a sterile cotton swab. The medium was incorporated with streptomycin (50mg/ml) and coconut oil. Sterile filter paper discs of 5mm diameter were impregnated with the following compounds: 1) silver nanoparticles synthesised via biological route 2) silver nanoparticles synthesised via chemical route. 3) ketoconazole 4).DMSO

The petri plates were incubated at 35^oC for two days. After incubation, the plates were observed for inhibition zones. Diameter of the zones was measured in millimetre, using a transparent ruler. Experiments were done in quadruplicate. Results are expressed as average diameter as shown in table 1.

The zone of inhibition produced by biologically synthesized silver nanoparticles is 25.5 mm, which is almost equivalent to the positive control- ketoconazole (26mm). This shows that the silver nanoparticles can be as effective as ketoconazole. The antidandruff activity of biologically synthesized silver nanoparticles is higher than that of chemically synthesized silver nanoparticles. This shows that green synthesis route can be better than chemical route to develop a non-toxic and safer antidandruff formula. Moreover, the side effects will also be reduced by the green synthesis route.

Table 1. Zone of inhibition as a measure of the antidandruff activity of silver nanoparticles and reference compounds. Values are average of experiments done in quadruplicate.

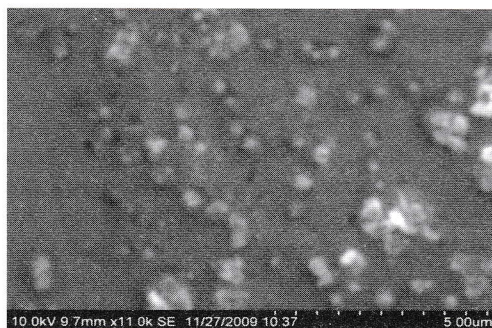
Compound tested	Diameter of the zone of inhibition (mm)
Silver nanoparticles synthesized via biological route (2.5 µg/ml/disc)	25.5
Silver nanoparticles synthesized via chemical route.	23
Ketoconazole - Positive control (2.5 µg/ml/disc)	26
DMSO- Negative control	0

Discussion and conclusion

Our study investigated the antidandruff property of silver nanoparticles synthesised via biological route. The property was compared with that of silver nanoparticles synthesised via chemical route. Ketoconazole and DMSO were used as positive and negative control. Several components of the extracts were capping agents for the synthesis. Silver nitrate was used as a substrate. Ketoconazole is the well known antidandruff agent. In all the cases, the activities were checked using the disk diffusion method, and the diameter of the inhibition zone measured. The anti dandruff activity of biologically synthesized silver nanoparticles is greater than that of chemically synthesized silver nanoparticles, and almost equivalent to that of the reference chemical, ketoconazole. Our results suggest that the silver nanoparticles synthesised using fenugreek leaves can exert a better antidandruff

activity. Hence they can be used to formulate antidandruff shampoos. Further, the proposed side effects of chemically synthesized silver nanoparticles and ketoconazole can also be combated by this formula.

Figure 1. Scanning electron microscopic image of silver nanoparticles synthesized through biological route.



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