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IN VITRO ANTIBACTERIAL CHARCTERISATION OF NANOGEL: AZIMA TETRACANTHA

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ABSTRACT
The plant A. tetracantha is shown to contain a variety of phytochemicals which are distributed in various parts of the plant. Nutritional composition of leaves of A. tetracantha Potassium and manganese were detected in highest quantity among major and minor elements. Easy to manufactures and chief in cost. Nanogel was first introduced to define cross-linked bifunctional networks a polyion and a non-ionic polymer for delivery of polynucleotides and poly ethylene glycol (PEG). Nanogels are very promising in drug delivery applications due to their high loading capacity.

Keywords: Curcumin, Antibacterial Activity, Gel, Azima tetracantha, Nanogel,

INTRODUCTION

Herbal medicine has become an integral part of standard healthcare based on combination of traditional usage and ongoing scientific research. Burgeoning interest in medicinal herbs has increased scientific scrutiny of their therapeutic potential and safety. Natural products and medicinal plants are believed to be an important source of new chemical substances with potential therapeutic efficacy. WHO has also issued Guidelines for the Assessment of Herbal Medicines (WHO, 1996) [1].

Nutritional composition of leaves of *A. tetracantha* Potassium and manganese were detected in highest quantity among major and minor elements. An appreciable quantity of carbohydrates, proteins, and lipids were detected in the leaf material. The content of Vitamin C was highest when compared to Vitamin E.

Novel drug delivery system is a novel approach to drug delivery that addresses the limitations of the traditional drug delivery systems. Modern medicine cures a particular disease by targeting address for correspondence [2].

If the novel drug delivery technology is applied in herbal medicine, it may help in increasing the efficacy and reducing the side effects of various herbal compounds and herbals.

In a typical polar gel, a natural or synthetic polymer builds a three-dimensional matrix throughout a hydrophilic liquid [3].

The term "nanogel" defined as the nanosized particles formed by physically or chemically crosslinked polymer networks that is sewll in a good solvent. Nanogel was first introduced to define cross-linked bifunctional networks a polyion and a nonionic polymer for delivery of polynucleotides and poly ethylene glycol (PEG). Nanogels are cross-linked nanoscale particles made of flexible hydrophilic polymers [4].

Most topical gels are prepared with organic polymers such as carbomers which impart an aesthetically pleasing, clear sparkling appearance to the product and are easily washed off the skin with water. Gels are two component semisolids systems rich in liquids. In a typical polar gel, a natural or synthetic polymer builds a three-dimensional matrix throughout a hydrophilic liquid [5].

METHODOLOGY

Purchasing of the plants

Azima tetracantha plant purchasing of local market and identified their microscopical characters.

Storage

That powder was packed in locked polythene bags, labelled and stored in the air tight container for study [6].

Extraction Process Of Azima Tetracantha Leaf

Soxhlet Extraction

20 gm *Azima tetracantha* Linn. leaves powdered material was extracted in 200 ml ethanol by soxhlet apparatus at 40-50^{0}C. Filtered extract was kept at room temperature for

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was extracted in 200 ml ethanol by soxhlet apparatus at 40-50^{0}C. Filtered extract was kept at room temperature for elimination of ethanol, 2.29 gm extract was collected. It was diluted in 10 ml ethanol. This extract was stored in a refrigerator for further use and called as the mother solution from which different concentration of extract as 20 mg/ml, 40 mg/ml, 60 mg/ml, 80 mg/ml, and 100 mg/ml was prepared [7].

Formulation Of Azima Tetracantha Nanogel
Emulsion – Solvent Diffusion Method

The nanogel is prepared from modified Emulsion Solvent Diffusion method. It is having 4 steps.

1. A precise amount of *Azima tetracantha* is dissolved in propylene glycol with stirring (organic phase).
2. The second step is the preparation of the aqueous phase by using carbopol -934, sodium alginate, or guar gum dissolved in water and heated continuously in a magnetic stirring for 20 minutes.
3. An emulsion is created by adding organic phase drop by drop into water phase during high speed homogenization for 30 minutes at 6000 rpm. Homogenizer converts emulsion into nanodroplets, resulting in an o/w emulsion.
4. After homogenizing the o/w emulsion for 1 hour at 8000rpm, triethanolamine is added and stirred continuously to form nanogel [8, 9].

***In vitro* Antibacterial effect on formulated nanogel**
Nutrient Agar Medium:

- Adding 200ml of distilled water to the amounts of Beef extract, NaCl, Peptone, and Agar is the appropriate procedure.
- Ensure that the medium is fully dissolved by heating it to boiling.
- After that, they are autoclaved for 15 minutes at 121°C to sterilize them.

A sterilized glass petri plate was transferred into the sterilized medium after autoclaving at 121°C (15 lb/ln2) for 15 minutes. Solidification was allowed to occur at room temperature. A 15l inoculum of bacteria was transferred to each petri plate. Sterile borer was used to bore four wells each 6mm in diameter. A sterile micropipette was used to add different concentrations of drug samples to each cup. In order to allow the solution to diffuse into the medium, the plates were left on sight for 2 hours. After 24 hours, the petri dishes are kept inverted at 37°C x 1°C in an incubator. Each of the wells was measured for the diameter of the zone of inhibition [10, 11].

Preparation of inoculums:
Bacterial inoculums:
The inoculums was prepared by inoculate the Sporres of Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa in Nutrient broth and incubated at 37°C for 24 hours [12].

Agar cup plate method:

- Aseptically transferred into sterilized glass petri plates, the medium was autoclaved at 121°C (15lbs/ln2) for 15 minutes.
- Solidification was allowed to take place at room temperature. It was necessary to transfer 15 liters of inoculums of the bacteria to each petri plate. We drilled four six-millimeter-diameter wells using sterile borer tools.
- Each of the cups was filled with different concentrations of drug samples using a sterile micropipette. To allow the solution to diffuse into the medium, the plates were kept at straight for two hours.
- It was inverted petri dishes that were incubated. In order to screen for antibacterial properties, the samples were incubated for 24 hours at 37°C. Inhibition zone diameter was measured. The results were tabulated [13].

RESULTS
Evaluation of Antibacterial Activity of *Azima tetracantha* nanogel formulation:

Cup plate method was used to test the formulation's antibacterial activity.

Media composition:
Zone of Inhibition:
Table no: 26 **Zone of Inhibition**

Table 1: Nutrient Agar Medium

S.No	Ingredient	Quantity taken
1.	Beef extract	2.0g
2.	NaCl	1.0g
3.	Peptone	2.0g
4.	Distilled water	200.0ml
5.	Agar	4.0g

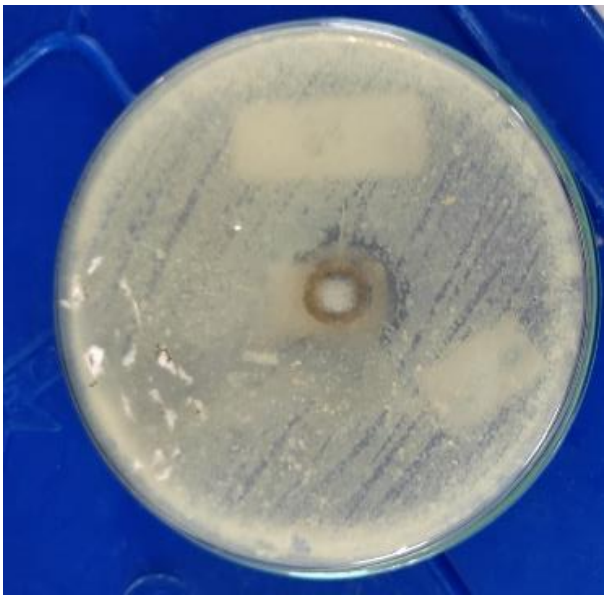
Table 2: Determination of Zone of inhibition

S.No.	Name of organism	Maintenance Media
1.	*Bacillus subtiles*	Nutrient Agar
2.	*Staphylococcus aureus*	Nutrient Agar
3.	*Escherichia coli*	Nutrient Agar

Table 3: 26 Zone of Inhibition

FORMULATION	*Escherichia coli* (mm)
F7	18.4 ± 1.05

Figure 1: Escherichia coli



DISCUSSION

Traditionally, medicinal plants have been used to treat bacterial diseases both externally and internally. Plant products are being investigated all over the world for their ability to inhibit clinically important bacterial strains.

According to estimates by the World Health Organization (WHO), 80% of the world's population uses plant extracts or active ingredients as folk medicine.

Almost all countries have their own indigenous systems of medicine, and many of the formulations have not been scientifically tested for their purported effects and it is imperative that those systems are scientifically proven to be effective in order to gain global acceptance.

As a result of this study, *Azima tetracantha* have antibacterial properties against *Escherichia coli* and are highly potent antibacterial agents when formulated as topical treatments. This may explain why the plant is said to be effective in treating common skin conditions in folk medicine.

Nanogel penetrates the skin better and smoothes, while also inhibiting bacterial growth.

Thus, the noisome gel might have facilitated better drug diffusion than crude extract.

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