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FORMULATION OF *CLEOME GYNANDRA* MICROSPHERES AND EVALUATION OF ANTIMICROBIAL ACTIVITY

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ABSTRACT

Objective: The main aim of study was to compare antimicrobial activity of extracts of Cleome gynandra and it's Egg Albumin Loaded microsphere against pathogenic strains. **Methods:** Collection of plant. Preparation of leaf extract from Cleome gynandra using water. Preparation of Egg Albumin loaded microsphere of extract of Cleome gynandra by solvent evaporation technique. Comparison of antimicrobial activity of leaf extracts of Cleome gynandra and it's Egg Albumin Loaded microsphere by paper disc diffusion method. **Results:** The leaf extract of Cleome gynandra and its egg albumin loaded microsphere showed good antimicrobial activity against all the tested microorganisms. **Conclusion:** On comparing to fresh leave extract, microsphere had shown good antimicrobial activity against bacteria and fungi. This is may be due to the egg albumin loaded with extract showed greater release characteristics.

Keywords: Antifungal, Antibacterial, solvent evaporation, Paper disc diffusion

INTRODUCTION

Novel technologies have been introduced recently for delivery of medicines at the target specific site. Modified release of herbal drugs has been achieved by Novel drug delivery technology which in terms increases therapeutic value and reduces toxicity **[1]**. In recent times the use of liposomes, ethosomes, phytosomes, emulsion, microspheres, solid lipid nano

particles has improved the therapeutic effects for the herbal drugs **[2]**.

Microspheres may be a superior choice of drug release system as it has the improved target specificity, better patient compliance. Herbal drugs were broadly used all over the world for the enhanced therapeutic value and fewer adverse effects as compared with these days modern medicines **[3]**. The drug obtained from natural origin can be made use in a better form with at most efficacy by designing as microsphere drug delivery system for herbal constituents. Microspheres are solids, approximately spherical particle ranging in size from 1-300µm **[4]**. Microspheres contains drugs dispersed in each matrix that are made of polymeric substance The absorption rate and bioavailability are increased due to the microsphere properties of having high surface area and low particle size .Microsphere can able to offer controlled release and site specific drug delivery **[5]**.

Cleome gynandra is a plant species of cleome which grows in open grass lands. In India they were not cultivated as it grows instinctively everywhere, mostly found in road sides and can be seen as weed in paddy fields. They are erect, upright, branched plant mostly between 25cm and 60cm height. Their sparse leaves are made up of 3-5 ovalage **[6]**. In ayurvedic treatment these are used as an anthelmentic, pruritis, in ear disease and some other disease like GIT disorder and also have antimicrobial action **[7]**. The purpose of this study is to examine antimicrobial activity of extracts of *Cleome gynandra* and it's Egg Albumin Loaded microsphere against pathogenic strains.

MATERIALS AND METHODS

Plant collection

The plant *Cleome gynandra* (CG) was collected from in and around Erode. This was taxonomically authenticated by Dr. C. Murugan, scientist 'D', Botanical survey of India in Coimbatore with a voucher specimen numbered BSI/ SRC/ 5/ 23/ 2017/ Tech./-1021.

Preparation of plant extract

25g of fresh leaves of CG were taken and washed thoroughly with distilled water. This was transferred to 500ml round bottom flask and 100ml distilled water was added. The contents were boiled for 15min. it was then filtered to obtain the extract. The extract was evaporated to get residue **[8]**.

Preparation of microsphere

1g egg albumin was dissolved in 15ml of chloroform to get clear solution. Then 1g of extract was dispersed into the polymeric solution and stirred for 30mins to ensure uniform dispersion of the extract into the polymeric solution **[9, 10]**. This polymer – extract dispersion was poured into 125ml liquid paraffin containing 1.5% tween 80 and stirred continuously for 4 hours at 480-

500rpm at room temperature using mechanical stirrer. After complete evaporation of solvent, microsphere were formed and separated by filtered through whatmann filter paper no.44. Then it was washed 3 to 4 times with sufficient quantity of diethyl ether at room temperature and dried overnight. The microsphere obtained by this method was spherical, free of flow and with no aggregation in the size range of 50-100µm. The size of microspheric particle was determined by means of optical microscope **[11, 12, 13]**.

Antimicrobial activity

Paper Disc Diffusion Method

Sterilenutrient agar / Saborauds dextrose agar medium were prepared and it was poured in to sterile petridishes aseptically **[14, 15, 16]**. After solidification, 0.1ml of inoculum from standardized culture of test organisms were spreaded on agar plate uniformly using spreader **[17, 18, 19]**. Sterile filter paper disc of 6mm having extract and microsphere at the concentration of 1000 g/ml and the standard disc (streptomycin for bacteria and Amphotericin for fungi) were placed on respective medium. All the plates were incubated at 37^0C for 24hrs for bacteria and 27^0C for 48hrs for fungi **[20, 21, 22]**.

RESULTS AND DISCUSSION

Leaf extract and microsphere were evaluated for antimicrobial activity by paper disc diffusion method.

Zone of inhibition for fresh leaves extract of CG and its egg albumin loaded microsphere was measured and shown below in **Table 1 and 2**.

From the result, the leaf extract of CG and its egg albumin loaded microsphere showed good antimicrobial activity against all tested microorganism. Moreover, the extract and microsphere had shown better antibacterial activity against gram positive organism than gram negative bacteria. On comparing to fresh leave extract, microsphere had shown good antimicrobial activity against bacteria and fungi. These were represented in **Figure 1**.

The inhibition percentage was calculated for leaf extract and egg albumin loaded microsphere and tabulated below in **Table 3**.

Based on the results it is seen that on *Lactobacillus acidophilus* microsphere shows maximum of 95% inhibition followed by *Escherichia coli* -84.21%, *Staphylococcus aureus* -77.27%, *Salmonella typhi* -72.22% and *Candida albicans* -68.18%. In leaf extract the maximum inhibition percentage obtained to be 70% on *Lactobacillus acidophilus* followed by *Escherichia coli* -63.15%, *Salmonella typhi* -61.11%, *Staphylococcus aureus* -59.09% and *Candida albicans* -59.09%.

Table 1: Antimicrobial activity of leaves extract of CG

Micro-organism	Zone of Inhibition (in mm) for *Cleome gynandra*	Zone of Inhibition (in mm) for Standard
Staphylococcus aureus	13	22
Lactobacillus acidophilus	14	20
Escherichia coli	12	19
Salmonella typhi	11	18
Candida albicans	13	22

Table 2: Antimicrobial activity of leaf extract – egg albumin loaded microsphere

Micro-organism	Zone of Inhibition(mm) for Microsphere	Zone of Inhibition(mm) for Standard
Staphylococcus aureus	17	22
Lactobacillus acidophilus	19	20
Escherichia coli	16	19
Salmonella typhi	13	18
Candida albicans	15	22

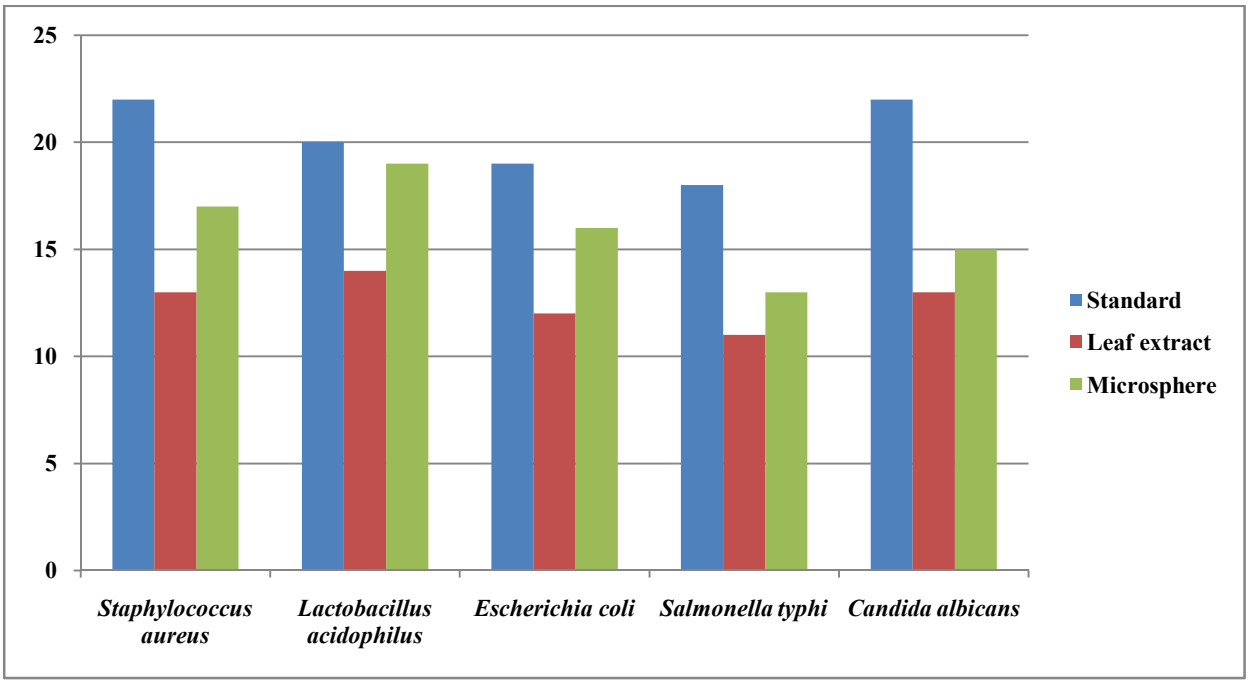

Figure 1: Antimicrobial activity of leaf extract and egg albumin loaded microsphere (Zone of inhibition in mm)

Table 3: Percentage activity of leaf extract and egg albumin loaded microsphere

Micro-organism	Percentage of inhibition for Leaf extract	Percentage of Inhibition for Microsphere
Staphylococcus aureus	59.09%	77.27%
Lactobacillus acidophilus	70%	95%
Escherichia coli	63.15%	84.21%
Salmonella typhi	61.11%	72.22%
Candida albicans	59.09%	68.18%

CONCLUSION

The leaf extract and the microsphere were showed good antimicrobial activity against gram positive microorganism than the gram negative. Comparatively, egg album loaded with leaf extract microsphere had shown better antimicrobial activity than leaf extract which may be the effect of longer release characteristics. Hence, we concluded that the egg albumin loaded leaf extract microsphere had better action against bacteria and fungi.

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