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To Study Antibacterial Activity of *Calendula officinalis*

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Monika Bhalchandra Savalkar*[1], Yasmin S Mulani[2], Shital Suhas Gaikwad [3], Tejas Ganesh Kale[4]

Gourishankar Institute of Pharmaceutical Education & Research Limb, Satara.

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ABSTRACT

The present study was carried out to evaluate the *In vitro* Antibacterial activity, ethanolic extracts of *Calendula officinalis* against three important bacteria namely *Staphylococcus aureus*, *E. coli* and *Candida glabrata*. The patterns of inhibition varied with the different plant part extracted as the tested microorganisms. The Antibacterial activity is done with agar well diffusion assay in plates containing nutrient agar media. They showed antimicrobial activity against the test microorganisms. The ethanolic extraction of *Calendela officinalis* shows antibacterial activity and zone of inhibition are 1mm in *Candida glabrata*, 3mm in *Staphylococcus aureus* and increased by 2 mm in *E. coli*.

INTRODUCTION

Aim- "To Study Antibacterial Activity of *Calendula officinalis*"

Calendula officinalis L. (pot marigold) is one of the commonly used medicinal plants in India, China, Europe and US (Muley *et al*., 2009). Calendula was known as "gold's" in old English was associated with Virgin Mary and Queen Mary, hence the name marigold The name of this plant comes from a Latin word 'Calend' meaning the first day of each month, because of the long flowering period of plant. As flowers move in the direction of the sun's radiation, it has become an astronomical sun sign "Leo" (Dinda and Craker, 1998). *Calendula* is an annual herb growing about 80 cm tall, having corymbosely branched stem; a long tap root with numerous secondary roots; hispid, acute, oblanceolate, alternate and sessile leaves; flower head inflorescence (surrounded by two rows of hairy bracts).

The pot marigold extracts possess a wide range of pharmacological effects and are used as antiseptic, stimulant, diaphoretic, antispasmodic and anti-pyretic agents. The flower extracts of the plant have anti-viral effects on HIV. *In-vitro, Calendula officinalis* (CO) plant Published Online on 6 September 2017 770 Nelofer Jan *et al*. extract shows anti-cancerous activity on various tumor cell lines derived from leukemias, fibrosacomas, melanomas, breast, cervix, prostate, pancreas and lung. It has also been internally used for the treatment of gastritis, colitis and bleeding of duodenal ulcers. Due to significant biological activity of *Calendula officinalis* and its constituents it is imperative that the plant be given attention and developed as a medicine.

REVIEW OF LITERATURE

S. Bissa *et al.* (2011)

Studies that different part (roots, leaf, flowers) *Calendula officinalis* were screened for potential antimicrobial activity against some important bacterial strains, namely *Escherichia coli, Salmonella typhi, Klebsiella pneumoniae, Enterobacter aerogenes*. These antimicrobial activity determined in aqueous, alcohol, chloroform and petroleum ether extract using agar disc diffusion method. Although all the plant parts showed significant antimicrobial activity but the highest antimicrobial activity was observed in petroleum ether extract of dried leaves against *Klebsiella pneumoniae*. Phytochemical analysis was also done.

Shrinidhi Maji Shankar *et al*. (2017)

Stated that *Calendula officinalis* is a member of the family Asteraceae which containing flavonoids, and essential oil *etc*. It is known to be effectives against certain gram negatives and gram positives clinical pathogens. Incorporating natural plants extract into periodontal anti-infective therapy is a wise alternative in light of rampant antibiotic resistance amongst periodontal pathogens. To assess the antimicrobial efficacy of *C. officinalis* against five oral microbes, as compared to gold standard chlorhexidine digluconate.

The objective for present work is-

- To determine antimicrobial activity of extract.
- To collect and extract of *Calendula officinalis* by using ethanol and water.
- To determine inhibition zone of extract against bacterial strain.

MATERIALS & METHODS

Sample & Collection

Fresh flowers of *Calendula officinalis* were collected from the Satara nurseries.

MERIGOLD-

Fig. 1. *Calendula officinalis*

Extraction by Maceration

Fresh flowers of *Calendula officinalis* were collected from the nurseries. Petals were washed gently under tap water and left to dry at room temperature for 2 days, the petals of marigold was crushed separately to make powder. 50 gram of the *Calendula officinalis* was mixed separately with 200 ml ethanol in conical flasks. The flasks containing extracts was heated on water bath for 1 h and placed at room temperature for 5 days. The flask was manually shaken daily to obtain maximum extraction. After 5 days, extract added to centrifuge tubes and centrifuged at 4000 rpm for 10 min to separate the supernatant. The supernatant containing extracts of *Calendula officinalis* was transferred into pre-weighed beakers and was left to dry completely on water bath at 60°C to obtain an ethanol free extract residue of *Calendula officinalis.*

Fig. 2. Extraction of *Calendula officinalis*

Cup and Plate Method

This method depends on diffusion of antibiotics from vertical cylinder or a cavity through the solidified agar layer of Petri plate on an extent such that growth of microorganisms is prevented entirely in circular area or inhibitory zone around cylinder or cavity containing solution of antibiotics A previously liquefied medium, appropriate to assay is inoculated with requisite quantity of suspension is added to medium at a temp between 40-50°c and inoculated medium is poured immediately into petriplate or large plate to occupy a depth of 3-4 mm. This prepared plate must be stored such that no significant growth or depth test microorganism occurs before use and the surface of agar layer is dry at time of use.

Solution of known concentration of standard preparation and the test antibiotic are prepared in appropriate buffer solution. These solutions are applied to surface of solid medium sterile

cylinder or cavities prepared in agar the volume added to each cylinder must be uniform and sufficient to fill the holes. The plates are left standing for 1- 4 hours at room temp or at 4℃ to allow diffusion of antibiotics in surrounding media. Plates are then incubated for above 24 hr at 37℃ and the diameter of circle zone of inhibition are measured.

RESULTS AND DISCUSSION

The Ethanolic Extracts of *Calendula officinalis* was tested for their Antibacterial activity. The results of the Antimicrobial activity of Ethanolic Extracts shows that all the concentrations was effective against tested microorganisms with varying zones of inhibition. The zone of inhibition for *C. officinalis* (petals) was observed against *Candida glabrata* 10 mm, S. aureas 16 mm, E. coli 28 mm respectively.

Table no 1: Zone of Inhibition: Antibacterial Activity

Sr. No.	Ethanolic Extract	*Candida glabrata*	*S. aureas*	*E. coli*
1	*C. officinalis*	10 mm	16 mm	28 mm

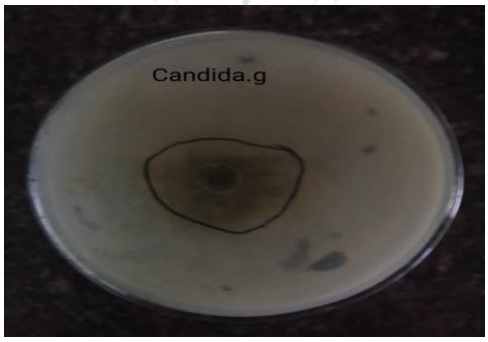

Fig. 3. Zone of inhibition by *Candida glabrata*

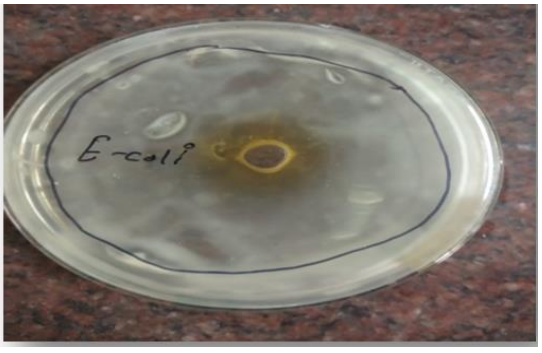

Fig. 4. Zone of inhibition by *Escherichia coli*

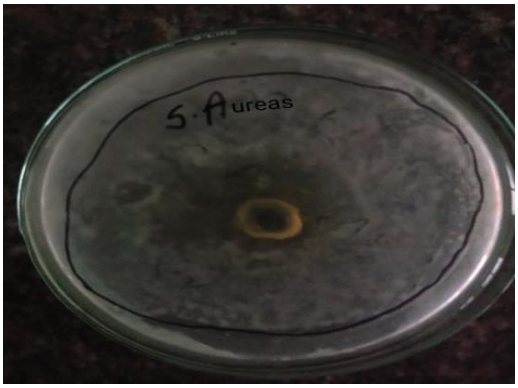

Fig. 5. Zone of inhibition by *Staphylococcus aureus*

CONCLUSION

Finally we can conclude that the following deduction from above experiment & analysis parts:

From the results is conclude that,

This present study suggest that the ethanolic extract of flower *Calendula officinalis* possess antibacterial activity against bacterial strains (*Staphylococcus aureus, candida glaberata, E. coli).*

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