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Original Research Article

EFFECT OF COMBINED EXTRACT FROM DIFFERENT PLANT PART OF *CALENDULA OFFICINALIS* (MARIGOLD) ON ANTIMICROBIAL ACTIVITY

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Abstract: Plants continue to be an important therapeutic aid for alleviating the ailments of humankind. In the present research work different parts of *Calendula officinalis* were screened for potential antibacterial activity against some important bacterial strains, namely Escherichia coli, staphylococcus aureus and fungal strain *Candida glaberata*, Aspergillus flavus .antifungal activity was determined in alcoholic extract using agar disc diffusion method. Although all the plants parts showed significant antimicrobial activity and the higher antifungal activity was observed in alcoholic extract of dried petals against *Candida glaberata*, Aspergillus flavus.

Keywords: *Calendula officinalis*, *Candida glaberata*, Aspergillus flavus .antifungal.

Introduction: Nature has bestowed upon us a very rich botanical wealth and a large number of diverse types of plants grow wild in different parts of our country. In India, the use of different parts of several medical plants to cure specific ailments has been in vogue from ancient times. Calendula officinalis, commonly known as pot marigold, is an annual herb and belongs to Asteraceae family. The flowers are monoecious (individual flowers are either male or female but both sexes can be found on the same plant) and are pollinated by Bees. It is noted for attracting wildlife.[1]

It is one of the best known and versatile herbs in western herbal medicine and is also a popular domestic remedy. The leaves blossoms and buds are used to make a homeopathic remedy. It is used internally in order to speed the healing of wounds. Only the common deep orange flowered variety is considered to be medicinal value. The whole plant, but especially the flowers and the leaves, is antiphlogistic, antiseptic, antispasmodic, astringent, cholagogue, diaphoretic emmenagogue, skin, stimulant and vulnerary. The dried powder of petals and leaves of Calendula sp. has been studied for Antimicrobial activity. The aim of

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the present study was to evaluate antimicrobial activity of various extract of *Calendula officinalis* which is having traditional claims for several diseases.[1, 3, 4]

Material and Methods:

Preparation of plant extract from dried plant parts: - The selected plants were thoroughly washed and then dried under shade about 10 days. The dried plant sample were ground well into a fine powder in a mixer grinder and sieved to give particle size of 50 to 150 mm. The 25gm plant powder weighs. Add 100ml ethanol in plant powder. The macerate was kept for 24 hr. At room temperature to evaporate the solvent. The macerate were squeezed through double layered muslin cloth and filtered through paper. The extract was dissolved in Dimethyl sulfoxide (DMSO) to obtain a concentration of 500mg/ml.

Preparation of inoculum: - Stock culture was maintained at 4°c on slopes of nutrient agar. Active culture for experiment were prepared by transferring a loop full of cells from the stock culture to test tubes of nutrient agar medium and were incubated without agitation for 24hr. The culture were diluted with fresh nutrient agar broth to achieve optical densities corresponding to 2.0-10 colony forming units (CFU/ml) for bacteria.[1, 3, 4, 9]

Antibacterial susceptibility Test:- All the plant extract were screened against pathogenic bacterial strains. The tested organism was Escherichia coli, staphylococcus aureus. The disc diffusion method was used to test the antimicrobial activity of the plant Extract. 20ml of sterilized nutrient agar medium for E.coli, staphylococcus aureus were poured into each sterile Petridis. The plates were allowed to solidify for 5min and 0.1 % inoculum suspension was swabbed uniformly. The entire agar surface of the each plate was inoculated with this swab first in the horizontal direction and then in a vertical direction, which ensure the even distribution of organism over the agar surface. The filter paper disc soaked in 1ml of the plant Extract or loaded with 5mg/disc, of dry extract and were placed on the surface of the bacteria seeded agar plates and the compound was allowed to diffuse for 5min and then the plates were incubated at 37°c for 24hr. At the end of incubation, inhibition zone formed around the disc were measured with transparent ruler in millimetre. These studies were performed in triplicate.[1, 3, 4, 9]

Antifungal susceptibility Test: - All the plant extract were screened against pathogenic fungal strains. The tested organisms were *Aspergillus flavus, Candida glaberata*.The disc diffusion method was used to test the antimicrobial activity of the plant Extract. 20ml of sterilized nutrient agar medium for Aspergillusflavus, *Candida glaberata* were poured into each sterile Petridis. The plates were allowed to solidify for 5min and 0.1 % inoculum suspension was swabbed uniformly. The entire agar surface of the each plate was inoculated with this swab first in the horizontal direction and then in a vertical direction, which ensure the even distribution of organism over the agar surface. The filter paper disc soaked in 1ml of the plant Extract or loaded with 5mg/disc, of dry extract and were placed on the surface of the bacteria seeded agar plates and the compound was allowed to diffuse for 5min and then the plates were incubated at 37 °c for 24hr. At the end of incubation, inhibition zone formed around the disc were measured with transparent ruler in millimetre. These studies were performed in triplicate.[1, 3, 4, 9]

RESULT AND DISCUSSION:-

Observations:-

The essential oil of the flower inhibited the growth of bacteria and fungus. The bacteria are E.Coli, S. Aureus and fungal are Aspergillus flavus and *Candida glaberata*.

In the present study investigation leaves and flowers extract of *Calendula officinalis* were tested for their antimicrobial activity. The dried powder of petals and leaves extract were found to inhibit growth of bacteria are E.Coli, S. Aureus and fungas are Aspergillusflavus , candidaglaberata The extract of leaves proved to be toxic with maximum inhibition zone in

alcoholic extract against fungas are Aspergillus flavus , candidaglaberata again petal extract extract of dried leaves and petals against aspergillus flavus , candidaglaberata.

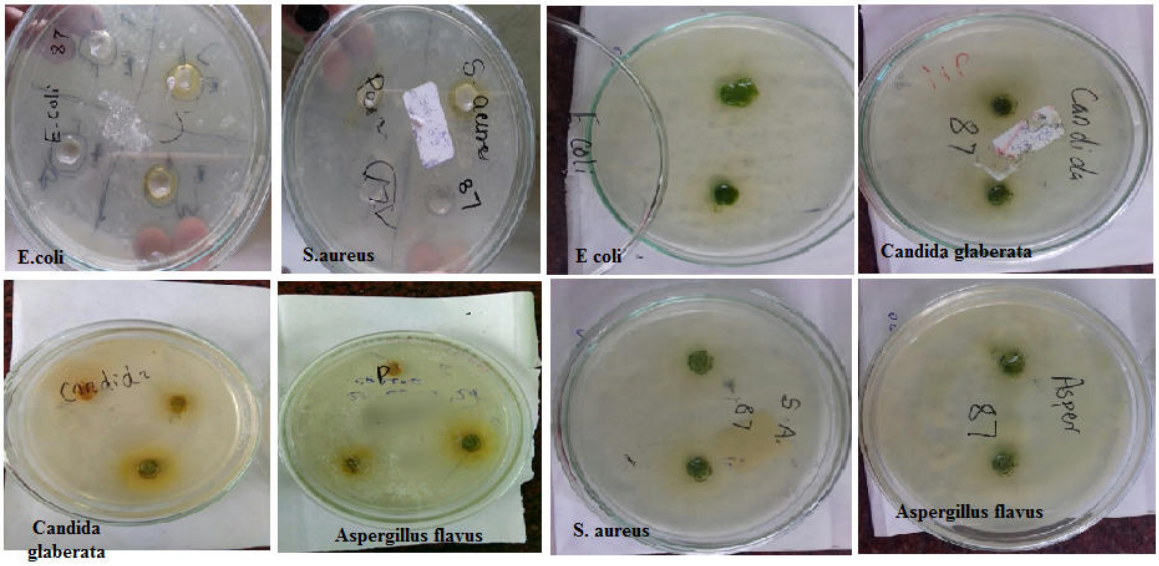

Fig 1: Typical antibacterial and antifungal activity of *Calendula officinalis* leaves and petal extract

Table 1: Zone of inhibitionofCalendula officinalisleaves extract

Sr. no.	E.coli	S. aureus	Candida glabrata	Aspergillus flavus
1.	13mm	16mm	14mm	15mm
2.	12mm	15mm	14mm	16mm
3.	13mm	14mm	15mm	15mm

Table 2: Zone of inhibition of *Calendula officinalis* petals extract

Sr. no	E.coli	S. aureus	Candida glabrata	Aspergillus flavus
1.	12mm	18mm	20mm	25mm
2.	11mm	18mm	20mm	24mm
3.	12mm	17mm	19mm	25mm

The highest antifungal activity was observed in alcoholic extract of dried leaves and petals against *Aspergillus flavus, Candida glaberata*.

Conclusion: - The result of present study indicates that the alcoholic extract of *Calendula officinalis* petals possessed good antimicrobial potential. Both extract showed antifungal & antibacterial activity that is comparable high antifungal activity was showed in the petals.

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