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## Research Paper

### Screening Of Anti-Microbial And Anthelmintic Activity In The Fruit Extract Of Medicinal Plant *Cucumis Callosus*

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#### **Abstract**

In the current research investigation on the plant of Cucurbitaceae family , the Anti-microbial activity and Anthelmintic activity of the plant *Cucumis callosus* was investigated. The Ethanolic extract taken of the fruit pulp of the plant showed antibacterial activity only against *Staphylococcus aureus* and *Klebsiella pneumonia*. Data also revealed that decrease in concentration of extract either lowers activity or eliminate the activity as shown mostly in case of concentration 1.25 mg in most of the cases. In the Anthelmintic activity investigation of of plant *Cucumis callosus* it was found from the results that the aqueous extract exhibited more potent anthelmintic activity (paralysis time 2-3 min and death time 3-5 min) even more than standard drug while the ethanolic extract has taken long time for death of worms

**Key Words :** *Cucumis callosus* , anti- microbial , Anthelmintic , Fruit pulp , anti-bacterial

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## INTRODUCTION

Herbal medicines are being used by about 80% of the world population primarily in the developing countries for primary health care. They have stood the test of time for their safety, efficacy, cultural acceptability and lesser side effects. The phytoconstituents present in them are a part of the physiological functions of living flora and hence they are believed to have better compatibility with the human body.

*Cucumis callosus* (Rottl.) Cogn (Cucurbitaceae) commonly known as “Kachri” in Rajasthan has been claimed in traditional literature to be valuable against a wide variety of diseases. The herb is distributed throughout India in arid zones. The herb is much branched very common prostrate, perennial herb, Leaves are cordate, suborbicular, deeply palmately 5-7 lobed. Flowers are yellow. Fruits are smooth, obovoid. ellipsoid, green variegated stripes. Fruiting in August-November. Fruit is traditionally used to prevent insanity to strong memory and remove vertigo. The seeds are cooling and astringent and useful in bilious disorder. Seeds are powdered and given in twice a day traditionally in diabetics in Sri Lanka, exert cooling effect, improve appetite, easy bowel syndrome, relieves stomach pain, vomiting and constipation (Figure 1 And 2).



**Fig 1:** *Cucumis callosus* plant



**Fig 2 :** *Cucumis callosus* Fruit & Seeds

## MATERIALS AND METHODS

### Plant material

Plant materials were procured from western arid zone of Rajasthan . Plant was identified and authenticated at Botany Department of the Apex University, Jaipur.

### **Preparation of extracts**

About 20 grams of fruit pulp powder were extracted using ethanol, water, and ethyl acetate individually for duration of 24 hours. After filtering, the extracts underwent evaporation to concentrate on water bath and dried.

#### **Method:** Disc Diffusion Method

- **Standard:** Streptomycin
- **MIC** by using Nutrient Broth as medium

The ethanol and aqueous extracts of fruits of *Cucumis callosus* were screened against a total of 6 bacterial strains and one fungal strain. The test strains were *Bacillus subtilis*, *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and one fungal strain *Aspergillus niger*.

### **Preparation of inoculum**

Cultures were maintained at 4°C on slopes of nutrient agar. Active cultures were prepared by transferring a loopful of cells from the stock cultures to test of Nutrient Agar Broth for bacteria and Sabouraud dextrose broth (SDB) for vet incubated without agitation for 24 hrs at 37°C and 25°C respectively. The cultures were and with fresh Nutrient Broth and Sabouraud dextrose broth to achieve optical densities armopcoding to  $2.0 \times 10^8$  colony forming units (CFU/ml) for bacteria and  $2.0 \times 10^6$  sporo/ml is hagal strains.

### **Antimicrobial susceptibility test**

The disc diffusion method (Bauer, 1966; Elizabeth, 2001) was used to screen the microbial activity. In vitro antimicrobial activity was screened by using Nutrient Broth rained from Himedia (Mumbai). The plates were prepared by pouring 15 ml of molten into sterile petri plates. The plates were allowed to solidify for 5 minutes and 0.1% inculan suspension was swabbed uniformly and the inoculum was allowed to dry for 5 The different concentrations of extracts (1.25, 2.5 and 5 mg/disc) were loaded on 6 sterile discs . of Streptomycin (10µg) and ketoconazole (10µg) were used as standard and shints as negative control. The loaded discs were placed on the surface of medium and the compound was allowed to diffuse for 5 minutes and the plates were kept for incubation at 370 for 24 hrs. At the end of incubation, inhibition zones formed around the dise were moistened with transparent ruler in millimeter. These studies were performed in triplicate.

## ANTHELMINTIC ACTIVITY

Indian adult earthworms (*Pheretima posthuma*) collected from moist soil and washed normal saline to remove all matters were used for all study. The earthworms of 3-5 cm length and 0.1-0.2 cm in width were used for all the experimental protocol due to its anatomical and physiological resemblance with the intestinal roundworm parasites of human beings.

The extracts were suspended in normal saline. All the drugs and extracts were prepared freshly before starting the experiment.

Six groups of six earthworms each were released into 10 ml of desired formulation as normal saline, Albendazole (40 mg/ml), ethanol and aqueous extracts (20 mg/ml and 40mg/ml, each) of fruits of *Cucumis callosus*.

Observation was made for the time taken to paralysis and death of individual worms 4 hrs of the test period.

Paralysis was said to occur when the worms did not revive even in normal saline.

## RESULT and Discussion

### ANTIMICROBIAL ACTIVITY

The inhibition zone assay revealed primarily two types of observations which were discs without any surrounded clear or inhibition zones which could be attributed to the absence of any inhibitory activity and clear inhibition zone representing the antimicrobial action of the tested plant extract. The results of the antimicrobial screening of the crude extracts of *Cucumis callosus* are shown in Table 1.

**Table 1: Antimicrobial screening of the extracts of *Cucumis callosus* .**

Extract	Conc.	Bs	Sa	Pa	Kp	Ec	An
		Zone of Inhibition in mm					
Streptomycin	10 $\mu$ g	28	13	27	18	17	-
Ketoconazole	10 $\mu$ g	-	-	-	-	-	25
AECA	1.25mg	-	-	-	-	2	-
	2.5 mg	-	-	4.5	3	4.5	-
	5 mg	-	-	10	9.5	11.5	-
EECA	1.25mg	-	-	-	2.5	-	-
	2.5 mg	-	1.5	-	6	-	-
	5 mg	-	4	-	14	-	-

The results of the microbial assay indicate that both the extracts of *Cucumis callosus* showed variable activity with different organisms. Activity was not shown against fungal strain by both the extracts indicating absence of antifungal activity. Both the extracts also displayed concentration dependent antibacterial activities. Aqueous extract (2.5 mg and 5 mg) showed antibacterial activity against *Pseudomonas aeruginosa* and *Klebsiella pneumonia* but no measurable effect for aqueous extract (1.25 mg). Aqueous extract do not show activity against *Bacillus subtilis* and *Staphylococcus aureus* at any concentration.

Ethanol extract showed antibacterial activity only against *Staphylococcus aureus* and *Klebsiella pneumonia*. Data also revealed that decrease in concentration of extract either lowers activity or eliminate the activity as shown mostly in case of concentration 1.25 mg in most of the cases.

Amongst all the extracts tested the ethanolic extract proved to be the most effective against *Klebsiella pneumonia*. It showed maximum zone of inhibition (14 mm).

### ANTHELMINTIC ACTIVITY

The anthelmintic activity of ethanol and aqueous extract of *Cucumis callosus* fruit was studied against the adult earthworms, *Pheretima posthuma*. Time for paralysis and death of worms were noted to assess anthelmintic activity (Table 2).

**Table 2: Anthelmintic Activity of Various Extracts of *Cucumis callosus***

Treatment	Concentration mg/ml	Time Taken for Paralysis	Time Taken for Death
Control	-	-	-
Albendazole	20	3.43 ± 0.130	5.45 ± 0.008
	40	1.38 ± 0.056**	4.18 ± 0.008**
Aqueous Extract of <i>Cucumis callosus</i>	20	2.18 ± 0.009**	4.34 ± 0.015**
	40	1.46 ± 0.062**	3.45 ± 0.014**
Ethanolic Extract of <i>Cucumis callosus</i>	20	14.26 ± 0.006*	18.14 ± 0.014*
	40	9.24 ± 0.004*	12.45 ± 0.33*

P\* < 0.05, p\*\* < 0.01

From the results shown in the table, aqueous extract of *Cucumis callosus* exhibited anthelmintic activity in dose dependent manner giving shortest time of paralysis and deaths compared to Albendazole. From the results it is observed that aqueous extract exhibit potent anthelmintic activity (paralysis time 2-3 min and death time 3-5 min) even more than

standard drug while the ethanolic extract has taken long time for death of worms. It was taken 10-15 min to bring paralysis and 15-20 min. to bring death of worms.

## CONCLUSION

In conclusion, the pharmacognostic parameters reported in this study will be useful in the development of pharmacopeial standards for the future studies. The physicochemical Aqueous extract of *Cucumis callosus* has showed varied extent of antimicrobial activity against the chosen test microorganisms. Ethanol extract showed antimicrobial activity but to a very less extent. Thus in search of novel broad spectrum antimicrobial agent, the formulation comprising different proportions of these extracts may be proven good. This study has not only shown the scientific basis for some of the therapeutic uses of traditional plant, but also confirmed the ethnomedicinal claims for the selected plant constants are important parameters for detecting adulteration or improper handling of drugs.

Extraction of plant part was done to performs screening of these extracts for activities like , antimicrobial, and anthelmintic activity. Aqueous extract of *Cucumis callosus* fruit exhibited potent anthelmintic activity . Results obtained will be useful for establishment of scientific standards of these plant to utilise for treatment of vaious ailments.

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