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## ***In vitro* evaluation of plant extracts and cow urine against anthracnose of *Cymopsis tetragonoloba* (Cluster bean)**

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**KAMINI DUBEY<sup>\*1</sup>, REETI SINGH<sup>2</sup> AND SWETA PRAKASH<sup>3</sup>**

<sup>1</sup>Govt. PG College, Narsinghgarh, Rajgarh, MP, India. 46566;

<sup>2</sup>Plant Pathology Dept. College of Agriculture, Gwalior, MP, India;

<sup>3</sup>School of Study in Botany, Jiwaji University, Gwalior, MP, India.

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In the present work, anthracnose of *Cymopsis tetragonoloba* (Cluster bean) was studied in the Plant Pathology laboratory, Agriculture college, Gwalior. Due to the poisonous impact of chemical fungicides, it is easier to use the plant extracts (botanicals) for a safer solution to combat the pathogen. Anthracnose is a severe disease in Guar growing areas and due to this farmers suffer a huge loss. The cow urine (at three concentrations) and six plant extracts were examined. The growth of pathogen was found to be prevented substantially by *Calotropis procera*, cow urine 30ml/l (conc2), cow urine 45ml/l (conc3), *Azadirachta indica* and cow urine 15ml/l (conc1).

**Key words:** Botanicals, *Colletotrichum capsici*, guar, legume crop, pathogen, poisoned food technique

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### **INTRODUCTION**

Cluster bean is an important legume crop in Madhya Pradesh's northern region. It is cultivated for nutritive fodder as well as cereals. It can be used as edible crop and green manure. A valuable gum is extracted from the seeds of cluster bean which is used in industries such as pharmaceuticals, paper and textile. Moreover, it plays an important role as binding agent in medicines and used for thickening of the lotions.

Anthracnose disease occurs in kharif seasons and its symptoms appear on leaves, pod and stem (cotyledon) in shape of black spots. In country like India the cluster bean is cultivated over 3 million hectare in different states of north-west region. In view of this, every year this disease causes a heavy agricultural loss.

### **MATERIALS AND METHODS**

#### ***Preparation of plant extract***

The young, healthy part of the plant ( Table 1 ) was collected from the field. The healthy leaves of the

plants under study were systematically cleaned in running tap water. Then these leaves were dried for two days in oven at 60° C. The powders of different leaves were obtained by drying, grinding and finally sieving through a sieve of mesh size 52. These powders were packed in different air tight containers. The plant powders were mixed in water and botanical solutions of 10 % concentration of each were prepared. The potato sucrose agar (PSA) was prepared and melted. The 10 ml botanical solution was mixed with 90 ml melted PSA.

#### ***Poisoned food technique***

The poisoned food technique of *in-vitro* plant products (Nene, 1971) is an effective method for testing their toxicity of fungi. The studies were performed using potato sucrose agar media. In twenty ml of distilled water, 10 grams of powder from plant extract was dissolved. The plant extract was sieved after 24 hours. This extract was combined with PSA in an autoclave and sterilized. The poisoned medium was poured in each sterilized petriplate and was inoculated with a 10-day-old culture disc (5 mm). Three replications have been recorded. These plates were incubated at 25±2°C. Then diameter of fungus colony was measured after three, five and seven days.

## RESULTS AND DISCUSSION

It can be observed from Table 2 that, under *in vitro* conditions, all plant extracts significantly inhibited the growth of anthracnose pathogen. Among the plant extracts and cow urine examined, *C. procera* (52.5 mm), cow urine 30 ml/l (53.3 mm), cow urine 45 ml/l (54 mm), *A. indica* (57.8 mm) and cow urine 15 ml/l (59 mm) were found to be substantially superior to all other treatments, with the exception of *L. camara* (68.6 mm) in fungal growth reduction (Fig. 1 and Fig. 2). Less radial growth was found

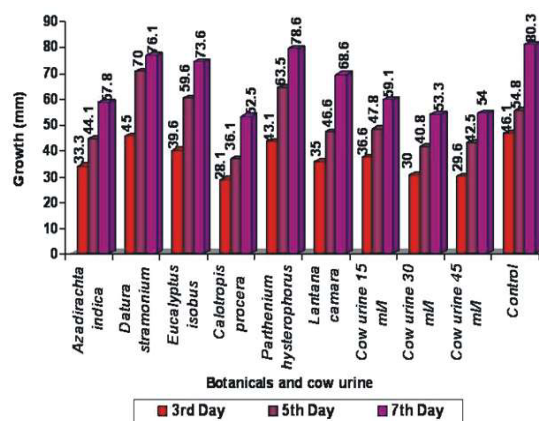
among the other tested botanicals in *D. stramonium* (76 mm) and *P. hysterophorus* (78.6 mm). The maximum growth of mycelium was observed in the control (80.3 mm). Sporulation was abundant (65 conidia/ microscopic field) in control and medium in *E. isobus* and *C. procera* (20-25 conidia /microscopic field). Very poor sporulation was observed in *D. stramonium*, *P. hysterophorus*, *L. camara*, cow urine (conc1), cow urine (conc2) and cow urine (conc3) (2-5 conidia/ microscopic field). The fungus did not sporulate in *A. indica* treatment.

**Table 1:** List of plant extracts (botanicals) used in the experiments

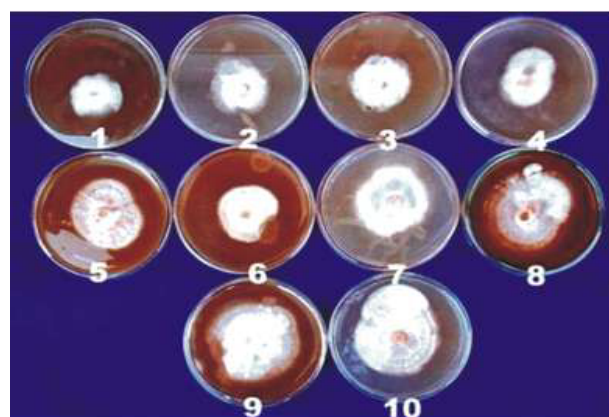
Plant extracts	%/ml/ltr.	Local name	Family
<i>Azadirachta indica</i> ( <i>A. indica</i> )	10	Margosa/Neem tree	Meliaceae
<i>Datura stramonium</i> ( <i>D. stramonium</i> )	10	Ghaneri/ Papardani	Verbenaceae
<i>Eucalyptus isobus</i> ( <i>E. isobus</i> )	10	Dhatura	Solanaceae
<i>Calotropis procera</i> ( <i>C. procera</i> )	10	Lemon scented eucalypt	Myrtaceae
<i>Parthenium hysterophorus</i> ( <i>P. hysterophorus</i> )	10	Madar, Safedak,	Asclepiadaceae
<i>Lantana camara</i> ( <i>L. camara</i> )	10	Gajar Ghas	Compositae
Cow urine (conc1)	15		
Cow urine (conc2)	30		
Cow urine (conc3)	45		
Control			

**Table 2:** Impact of various plant extracts and cow urine on colony (Mycelial) growth and sporulation of anthracnose pathogen

Plant extracts	Concentrations (Percent)	Sporulation	Mycelial growth (mm/day)		
			3	5	7
<i>Azadirachta indica</i> ( <i>A. indica</i> )	10	No sporulation	33.3	44.1	57.8
<i>Datura stramonium</i> ( <i>D. stramonium</i> )	10	Very poor (2 - 5)	45	70	76.1
<i>Eucalyptus isobus</i> ( <i>E. isobus</i> )	10	Medium (20 -25)	39.6	59.6	73.6
<i>Calotropis procera</i> ( <i>C. procera</i> )	10	Medium (20 -25)	28.1	36.1	52.5
<i>Parthenium hysterophorus</i> ( <i>P. hysterophorus</i> )	10	Very poor (2 - 5)	43.1	63.5	78.6
<i>Lantana camara</i> ( <i>L. camara</i> )	10	Very poor (2 - 5)	35	46.6	68.6
Cow urine (conc1)	15 ml/l	Very poor (2 - 5)	36.6	47.8	59.1
Cow urine (conc2)	30 ml/l	Very poor (2 - 5)	30	40.8	53.3
Cow urine (conc3)	45 ml/l	Very poor (2 - 5)	29.6	42.5	54.0
Control		Abundant (65)	46.1	54.8	80.3
SE(m)±			1.843	2.140	1.939
CD(at 5%)			5.517	6.408	5.806



**Fig. 1 :** *In vitro* evaluation of plant extracts and cow urine against Anthracnose pathogen



**Fig. 2 :** *In vitro* evaluation of plant extract and cow urine against anthracnose pathogen (1) *Calotropis procera* (2) Cow urine (conc2)(3) Cow urine (conc3)( 4) *Azadirachta indica* (5) Cow urine (conc1)( 6) *Lantana camara* (7) *Eucalyptus isobus* (8) *Datura stramonium* (9) *Parthenium hysterophorus* (10) Control

In the plant extracts, various researchers confirmed the existence of an antifungal compound (Rahman *et al.* 2005; Ushakiran *et al.* 2006). The fungitoxic properties of *A. indica*, *C. procera* against *Colletotrichum capsici*, were also reported (Verma *et al.* 2018). The growth of *C. capsici* was inhibited by Neem extract (Pardhi and Raut 2011) the same result was also reported by Hegde *et al.* (2002). *D.*

*stramonium* efficiently prevented the colony growth of anthracnose pathogen (*C. capsici* and *G. piperatum*) (Gomathi and Kannabiran 2000). The cow urine was found as effective inhibitor of *C. capsici* in *In vitro* conditions (Salam *et al.* 2018; Kamar *et al.* 2013). Bagri *et al.* (2004) evaluated the antimicrobial property of *Datura* against *C. capsici*. In another research, *A. indica*, *D. stramonium* and *O. sanctum* effectively inhibited the growth of *C. capsici* (Sinha *et al.* 2004).

In this research work among the applied plant extracts (*C. procera*, *A. indica*, *L. camara*, *D. stramonium*, *E. isobus*, *P. hysterophorus*) and three concentrations of cow urine, *A. indica*, *C. procera* and cow urine at 15 ml/l (conc1), 30 ml/l (conc2) and 45 ml/l (conc3) showed significant efficacy in reducing the intensity of anthracnose fungus. The effect of *Azadirachta indica* (neem) against the fungus (*C. capsici*) under green-house conditions was also stated by Hedge *et al.* (2002)

An analysis on the utility of plants in crop disease control was published by many researchers and concluded that the use of plant extract pesticides is of immense value in crop disease control and will hold a privileged role in the coming future (Kumaran *et al.* 2003; Ajith *et al.* 2012). Rajput *et al.* (2014) also reported that, *Ocimum sanctum* followed by *Adathoda vesica* prevented the growth of *C. capsici*. Rewale *et al.* (2018) also reported that, significantly highest average inhibition was found with *A. indica* followed by *Z. officinale*. *A. cepa*, *P. hystrophorus* and *P. pinnata*.

## CONCLUSION

The six plant extract and three concentrations of cow urine were observed to be appreciably prevented the fungal growth of pathogen, but *Calotropis procera* and cow urine (conc2 and conc3) significantly reduce the growth of pathogen.

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