

Short Communication

Inhibitory Effect of Cow Urine against *Colletotrichum capsici* Isolated from Anthracnose of Chilli (*Capsicum annuum* L.)

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Abstract

Cow urine has got several applications in agriculture. It is shown to possess inhibitory activity against many phytopathogenic fungi and bacteria. Among various diseases of Chilli, anthracnose is the most important disease which results in drastic reduction in yield. The present study was conducted with an aim to determine antifungal efficacy of cow urine against *Colletotrichum capsici* isolated from anthracnose of chilli (*Capsicum annuum* L.). Poisoned food technique was employed to determine antifungal activity of different concentrations of cow urine (5, 10 and 15%). Cow urine was found to display concentration dependent inhibitory activity against fungal growth. An inhibition of >50% was observed at 5% concentration. In conclusion, the use of cow urine can be the cost-effective and eco-friendly approach for controlling anthracnose in chilli.

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INTRODUCTION

Chilli, belonging to the genus *Capsicum* (Solanaceae) is an herbaceous, annual, dicotyledonous flowering plant and is an important crop grown worldwide in tropical and subtropical regions. It is grown extensively for its consumption, nutritional and economy purposes. It is used as spice (ripe and dried form) and vegetable (green fruit). India is known to be the largest producer of chilli in terms of international trade. Chilli is known to contain a number of chemicals viz., steam-volatile oils, fatty oils, capsaicinoids, carotenoids, vitamins, protein, fibre and mineral elements. The production of chilli is influenced by several factors. The chilli cultivars are susceptible to various diseases caused by fungi, bacteria and viruses which account for marked reduction in productivity. Among various diseases, anthracnose (both pre-harvest and post-harvest) is the most important disease which results in drastic reduction in yield, deterioration of the quality of fruit. The typical anthracnose symptoms on chilli fruit include sunken necrotic tissues with concentric rings of acervuli. Anthracnose disease of chilli is reported to be caused by a number of *Colletotrichum* species such as *C. capsici*, *C. acutatum*, *C. gloeosporioides*, *C. coccodes* and *C. dematium*. The anthracnose may result in yield loss up to 50%. Among the species of *Colletotrichum*, *C. capsici* is the most important pathogen implicated in

causing anthracnose (Ushakiran *et al.*, 2006; Anand *et al.*, 2007; Ratanacherdchai *et al.*, 2007; Than *et al.*, 2008; Kim *et al.*, 2010; Narasimhan and Shivakumar, 2012; Susheela, 2012; Masoodi *et al.*, 2013).

Cow is believed to be a sacred animal in India. The urine of cow is used for several medicinal purposes since ancient time. The description on the use of cow urine has been mentioned in classical Ayurveda texts such as Charakasamhita and Shushrutasamhita. The urine of cow is compared with the nectar in Veda. Cow urine is known to cause weight loss, and reverse certain cardiac and kidney problems, indigestion, stomach ache and edema. It is suggested that cow urine is also useful in treating renal colic, jaundice, anemia, diarrhoea, gastric infections, piles and skin diseases. It is also considered as an appetizer and is known to reverse inflammation, or a diuretic as well as a nephroprotective agent. Experiments have shown that cow urine has several biological activities such as antimicrobial, antidiabetic, antioxidant, antitumor, molluscicidal and others (Krishnamurthi *et al.*, 2004; Gururaja *et al.*, 2011; Rakesh *et al.*, 2013a). Cow urine has got several applications in agriculture. Cow urine is shown to control root knot nematode in tomato and melon aphids and pickle worms in watermelon cultivation

(Abubakar *et al.*, 2004; Burubai and Eribo, 2012). Cow urine, cow urine extracts of plants and cow urine in combination with plants were found to exhibit inhibitory activity against phytopathogenic fungi and bacteria (Basak *et al.*, 2002a; Basak *et al.*, 2002b; Akhter *et al.*, 2006; Murugan *et al.*, 2012; Rakesh *et al.*, 2013a; Rakesh *et al.*, 2013b). In the present study, we have determined the effect of different concentrations of cow urine against *C. capsici* isolated from anthracnose of Chilli (*Capsicum annum* L.).

MATERIALS AND METHODS

Isolation of *Colletotrichum capsici*

The test fungus *C. capsici* was isolated from anthracnose of chilli by cutting and placing the affected fruit part on potato dextrose agar (PDA). The fungus was identified based on morphology and microscopic features (Barnett and Hunter, 1998; Shenoy *et al.*, 2007). The fungus produced dense whitish to dark grey aerial mycelium, reverse dark, conidial mass pale buff to salmon. Sclerotia absent, setae were abundant. Conidia are falcate, fusiform, with acute apices. Appressoria are abundant, medium brown and clavate to circular. The isolate was maintained in refrigerator on PDA slants.

Cow Urine

Urine of an indigenous cow variety called Malnad gidda was collected at early morning in a sterile container. The urine was filtered through Whatman No. 1, stored in airtight container and used for antifungal study (Rakesh *et al.*, 2013b).

Antifungal Activity of Cow Urine against *C. capsici*

In order to evaluate inhibitory potential of cow urine, Poisoned food technique was employed. Here, petriplates containing PDA medium amended with different concentrations of cow urine *viz.*, 5%, 10% and 15% were inoculated at the centre with the 5mm fungal discs (cut from the periphery of 5 days old test fungus). The plates were incubated at 28°C for 5 days. Colony diameters were measured in mutual perpendicular directions on 5th day. Antifungal effect of cow urine was recorded in terms of inhibition of mycelial growth (%) and was calculated using the formula:

$$\text{Inhibition of mycelia growth (\%)} = (C - T / C) \times 100$$

where C is average diameter of fungal colony in control plates and T is the average diameter of fungal colony in poisoned plates (Rakesh *et al.*, 2013a).

RESULTS AND DISCUSSION

The result of antifungal effect of cow urine against *C. capsici* is presented in Table 1 and Figure 1. Cow urine was found to exhibit concentration dependent inhibition of test fungus. The colony diameter of test fungus drastically reduced in PDA plates poisoned with cow urine. An inhibition of >50% was observed at 5% concentration and >75% inhibition was observed at cow urine concentration 10 and 15%.

One of the widely used strategies for controlling anthracnose causing fungi is the use of chemical approach. However, the use of chemicals has some ill-effects such as toxicity, development of resistant fungal strains and environmental pollution. Hence, various

approaches have been tried to find alternatives for the chemical agents. Inoculation of chilli plants with bacteria such as *Pseudomonas fluorescens* (Anand *et al.*, 2007) and *Burkholderia* sp. strain TNAU-1 (Madhavan *et al.*, 2011) resulted in induction of systemic resistance. It has been found that the plant extracts (Ushakiran *et al.*, 2006; Tiwari *et al.*, 2008; Aderonke *et al.*, 2011) and biocontrol agents such as *Trichoderma harzianum*, *Trichoderma viridae*, *Pseudomonas fluorescens*, *Bacillus* species (Ekbote, 2005; Ushakiran *et al.*, 2006; Tiwari *et al.*, 2008; Narasimhan and Shivakumar, 2012) have shown to be effective against the *Colletotrichum* species isolated from anthracnose of chilli.

Table 1: Radial growth and percentage inhibition of *C. capsici* in plates poisoned with different concentrations of cow urine.

Concentration (%)	Colony diameter (cm)	Inhibition (%)
0 (control)	3.45±0.05	0.0
5	1.50±0.00	56.5
10	0.85±0.05	75.4
15	0.70±0.00	79.7

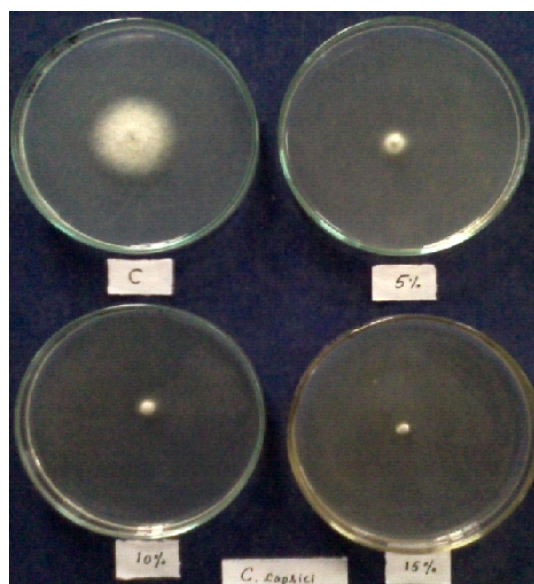


Figure 1: Radial growth of *C. capsici* in plates poisoned with different concentrations of cow urine.

Cow urine has shown to be promising against phytopathogenic fungi and bacteria. Studies on Cow urine alone/combination of cow urine with plants showed inhibition of *Sclerotinia sclerotiorum* causing *Sclerotinia* rot in cucumber (Basak *et al.*, 2002a), *Fusarium solani* f.sp. *cucurbitae* causing root rot disease of cucumber (Basak *et al.*, 2002b), *Bipolaris sorokiniana* causing leaf blight of wheat (Akhter *et al.*, 2006), *Xanthomonas oryzae* pv. *oryzae* causing leaf blight of paddy (Murugan *et al.*, 2012) and *Fusarium oxysporum* f.sp. *zingiberi*, *Ralstonia solanacearum* and *Pythium aphanidermatum* causing rhizome rot of ginger (Rakesh *et al.*, 2013a; Rakesh *et al.*, 2013b). In our study, cow urine has shown to exhibit marked inhibition of *C. capsici* isolated from anthracnose lesions on chilli. The inhibitory activity was concentration dependent *i.e.*, with increase in the concentration, inhibition was also increased. In an earlier study, Rakesh *et al.* (2013b) showed similar concentration dependent

inhibitory activity of cow urine against *Fusarium oxysporum* f. sp. *zingiberi* isolated from rhizome rot specimen of ginger.

CONCLUSION

Cow urine has shown marked concentration dependent inhibition of *C. capsici* in this study. The use of cow urine can be the cost-effective and eco-friendly approach for controlling anthracnose in chilli.

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