INTRODUCTION

Microorganisms cause a number of diseases in crops than any other pathogens and results in major crop losses. The harvest losses of crops are much higher in developing countries. Pathogens such as species of Fusarium, Alternaria, Pythium, Sclerotium, Phytophthora, Curvularia, Botrytis, Ralstonia, Xanthomonas etc., cause severe damages to agricultural crops before and after harvesting. The plant diseases caused by microorganisms are usually controlled by the use of chemicals. However, the use of synthetic compounds to control phytopathogens suffers from two main drawbacks viz., potential development of resistance in pathogens and the risk of toxicity. Due to this, research focused on compounds derived from natural sources such as plant extracts and their possible application in agriculture is being intensified. Many natural products, including plant extracts, have been shown to possess marked inhibitory activity against a variety of pathogens (Ojala et al., 2000; Benkeblia, 2004; Bhai et al., 2005; Bajpai et al., 2008; Paret et al., 2010; Ranaware et al., 2010; Zhao et al., 2011; Tiwari and Das, 2011; Bhardwaj et al., 2011; De Britto et al., 2011).

From the ancient period in India, cow urine has been used for several medicinal purposes and the description on its use has been in several classical Ayurveda texts like Charaka samhita and Shushruta samhit. Cow is believed to be a sacred animal in India its urine is known to cure several diseases. In Veda, cow urine is compared with the nectar (Krishnamurthi et al., 2004; Gururaja et al., 2011). Cow urine has got applications in agriculture. It has been found that cow urine has potential to control Meloidogyne incognita in Lycopersicon esculentus (Abubakar et al., 2004) and aphids and pickleworms in watermelon cultivation (Burubai and Eribo, 2012).


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f.sp. cucubita (Basak et al., 2002b), Bipolaris sorokiniana (Akhter et al., 2006) and Xanthomonas oryzae pv. oryzae (Murugan et al., 2012). It has been shown that cow urine extract of certain plants as well as cow urine in combination with certain plant extracts are found to possess marked inhibitory effect on human pathogens as well as plant pathogens (Akhter et al., 2006; Yadav et al., 2008; Rajapandiy et al., 2011; Tiwari & Das, 2011).

Ginger (Zingiber officinale Rosc., Zingiberaceae) is an important commercial crop grown for its aromatic rhizomes being used as spice and medicine. India is the largest producer of ginger and accounts for about 1/3rd of total world output. Ginger is grown in Kerala, Karnataka, West Bengal, Andhra Pradesh, Orissa, Arunachal Pradesh, Sikkim and other parts of India (Kumar et al., 2008; Sharma et al., 2010). The production of ginger is influenced largely by a number of diseases caused by bacteria, fungi, viruses, mycoplasma and nematodes. Main diseases of ginger are bacterial wilt caused by Ralstonia solanacearum, rhizome rot caused by Pythium species, Fusarium species, Sclerotium species, Pseudomonas species and others (Dake and Edison, 1989; Senapati and Ghose, 2005; Paret et al., 2010; Sharma et al., 2010; Kavyashree, 2009). Soft rot is a serious disease and has drastic effects on crop and eventually leads to rhizome loss. It is manifested initially by foliar yellowing and later water soaked lesions appears on the collar of the pseudostem which extend to rhizomes and leaves resulting in rotting of the entire plant. The disease is both seed and soil-borne (Bhai et al., 2008). In the present study, we have determined the inhibitory activity of cow urine extracts of selected plants against the pathogens viz., Fusarium oxysporum f.sp. zingiberi, Pythium aphanidermatum and Ralstonia solanacearum causing rhizome rot of ginger.

MATERIALS AND METHODS

Collection of Cow Urine

Urine was collected in a sterile container from a local cow variety called Malnad gidda at early morning 6:30am. The urine was filtered through Whatman No. 1 and stored in airtight container.

Preparation of Cow Urine Extract of Selected Plants

Table 1 represents the plants used in the present study. The plants were shade dried, powdered mechanically and used for preparation of extract. A known quantity (10g) of powdered plant material was added to 100ml of cow urine and left for 15 days. Later, the contents were filtered through muslin cloth followed by Whatman no. 1 and the filtrates were stored in refrigerator until use.

### Table 1: Plants used in the study.

<table>
<thead>
<tr>
<th>Name of the plant</th>
<th>Family</th>
<th>Part used</th>
</tr>
</thead>
<tbody>
<tr>
<td>Artocarpus lakoocha Roxb.</td>
<td>Moraceae</td>
<td>Leaf</td>
</tr>
<tr>
<td>Maesa indica (Roxb.) Wallich</td>
<td>Myrsinaceae</td>
<td>Leaf</td>
</tr>
<tr>
<td>Polyalthia longifolia Thw.</td>
<td>Annonaceae</td>
<td>Leaf</td>
</tr>
<tr>
<td>Hemedesmus indicus R. Br</td>
<td>Asclepiadaceae</td>
<td>Leaf</td>
</tr>
<tr>
<td>Swertia chirata (Roxb. ex Fleming) H. Karst.</td>
<td>Gentianaceae</td>
<td>Whole plant</td>
</tr>
<tr>
<td>Croton roxburgii Balak.</td>
<td>Euphorbiaceae</td>
<td>Leaf</td>
</tr>
<tr>
<td>Elaeagnus kologa Schlecht</td>
<td>Elaeagnaceae</td>
<td>Leaf</td>
</tr>
<tr>
<td>Gnidia glauca (Fresen.) Gilg</td>
<td>Thymelaceae</td>
<td>Leaf</td>
</tr>
<tr>
<td>Fahrenheitia zeylanica (Thw.)</td>
<td>Euphorbiaceae</td>
<td>Leaf</td>
</tr>
</tbody>
</table>

Antifungal Activity

Poisoned food technique was employed to screen the antifungal efficacy of cow urine extracts of selected plants (Dileep et al., 2013). In brief, Potato dextrose agar (HiMedia, Mumbai) media amended with cow urine extracts (10%) were autoclaved and poured into sterile petriplates. Fungal discs of 5mm diameter were cut with the help of sterile cork borer from the periphery of 5 days old culture of F. oxysporum f. sp. zingiberi and P. aphanidermatum and the discs were transferred aseptically on PDA plates poisoned with cow urine extracts and incubated for 5 days at 28°C. Colony diameters in mutual perpendicular directions were measured on the 5th day with the help of a ruler. The experiment was repeated two times and average colony diameter was noted. Antifungal activity of cow urine extracts was recorded in terms of inhibition of mycelial growth (%) and was calculated using the formula:

\[
\text{Mycelial growth inhibition} \% = \frac{\text{C} - \text{T}}{\text{C}} \times 100
\]

where ‘C’ is average colony diameter in control plates and ‘T’ is average colony diameter in poisoned plates.

Antibacterial Activity

In order to assess antibacterial activity of cow urine extracts against R. solanacearum, we have employed Agar well diffusion method (Kekuda et al., 2012). The bacterium was inoculated into sterile Nutrient broth (HiMedia, Mumbai) tubes and incubated for 24 hours at 37°C. The broth culture was swabbed on sterile Nutrient agar (HiMedia, Mumbai) plates using sterile cotton swabs. With the help of a sterile cork borer, wells of 0.6cm diameter were punched in the inoculated plates and cow urine extracts and standard (Streptomycin, 1mg/ml) were transferred into respectively labeled wells. The plates were incubated at 37°C for 24 hours and the zone of inhibition formed around the wells was measured. The experiment was repeated twice and the average value was recorded.
RESULTS
The result of inhibitory effect of cow urine extracts of selected plants against *F. oxysporum* and *P. aphanidermatum* is presented in Table 2 and Figure 1. The growth of test fungi, in terms of diameter of the fungal colony in poisoned plates was measured and compared with the control plates. The colony diameter of test fungi was lesser in poisoned plates in comparison with that of colony diameter in control plates indicating antifungal potential of cow urine extract of plants. The test fungi were found to be sensitive to all the extracts. Among fungi, high susceptibility was recorded in case of *F. oxysporum* with growth inhibition of >50% produced by all extracts. Only 3 extracts caused >50% inhibition of *P. aphanidermatum*. Cow urine extract of *E. kologa* & *P. longifolia* caused high and least inhibition of *P. aphanidermatum* respectively. In case of *F. oxysporum*, higher inhibition was produced by *A. lakoocha*, *H. indicus*, *C. roxburghii* and *M. indica*.

### Table 2: Antifungal activity of Cow urine extracts of selected plants.

<table>
<thead>
<tr>
<th>Cow Urine Extract</th>
<th>Colony diameter in cm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><em>P. aphanidermatum</em></td>
</tr>
<tr>
<td>Control</td>
<td>2.8</td>
</tr>
<tr>
<td><em>A. lakoocha</em></td>
<td>1.1</td>
</tr>
<tr>
<td><em>H. indicus</em></td>
<td>1.6</td>
</tr>
<tr>
<td><em>E. kologa</em></td>
<td>1.0</td>
</tr>
<tr>
<td><em>G. glauca</em></td>
<td>1.8</td>
</tr>
<tr>
<td><em>P. longifolia</em></td>
<td>1.9</td>
</tr>
<tr>
<td><em>C. roxburghii</em></td>
<td>1.5</td>
</tr>
<tr>
<td><em>F. zeylanica</em></td>
<td>1.1</td>
</tr>
<tr>
<td><em>S. chirata</em></td>
<td>1.8</td>
</tr>
<tr>
<td><em>M. indica</em></td>
<td>1.7</td>
</tr>
</tbody>
</table>

DISCUSSION
The term rhizome rot of ginger is accepted generally for soft rot and yellow disease complex as soft rot and yellows are generally found together affecting the plants and symptoms often mixed up. Soft rot is a serious disease leading to drastic effects on crop (Bhai et al., 2005; Senapati and Ghose, 2005). The rhizome rot disease management involves cultural, biological and chemical approaches for suppression of the pathogens. However, the control of the disease by the use of chemical agents is not so beneficial due to high cost, breakdown of resistance, residual problem and deleterious effect on non-target organisms including humans. This has necessitated search for alternatives for controlling the rhizome rot of ginger (Bhai et al., 2005; Pandey et al., 2010). Plants have been shown to possess inhibitory effect against fungi causing rhizome rot of ginger. Sagar et al. (2007) showed the fungitoxic efficacy of some plant extracts against *P. aphanidermatum* & *F. solani* isolated from rhizome rot specimen of ginger. It was found that *Azadirachta indica* and *Ferula asafoetida* showed maximum inhibition of mycelial growth of *P. aphanidermatum* and *F. solani* respectively. In an earlier study, we have shown the potential of ripe and unripe pericarp extract of *Polyalthia longifolia* against *P. aphanidermatum* and *F. solani* isolated from ginger rhizome rot (Dileep et al., 2013).
CONCLUSION
A marked inhibition of rhizome rot pathogens by cow urine extracts of selected plants was observed in this study. The extracts may find a possible use in agriculture as potent agents against pathogens. Further studies involving field trials is needed to justify the results of the present study.

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REFERENCES
Rakesh et al.,


