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Research Article

ANTIFUNGAL ACTIVITY OF SELECTED MEDICINAL PLANT EXTRACTS AGAINST PLANT PATHOGENIC FUNGI; *Rhizoctonia solani, Colletotrichum musea andFusarium oxysporum*

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ABSTRACT

The aim of this work was to find an alternative to chemical fungicides currently used in the control plant pathogenic fungi *Rhizoctonia solani* ,*Colletotrichum musae* and*Fusarium oxysporum*,. The antifungal activity of the methanol extracts of six medicinal plants used in native medicine in Sri Lanka is reported.All plant extracts were screened for their fungistatic, fungicidal activities and minimum inhibitory dilution (MID) against above fungi. The media amended with methanol and recommended fungicide for respective fungal strain were consider as negative and positive control respectively.Results showed that radial growth in all the three tested organisms was significantly impaired (p<0.05) by the addition of the extracts in the culture medium used. The test fungi differed in their reaction to the different extracts but on the whole, growth inhibition increased with the concentration of each extract. The most active extracts, shows a marked effect of the 20% methanol extracts from sweet flag with inhibition values of 91%, 86% and 84 % for *F. oxysporum,R. solani and C.mucea* whereas those from wild basil inhibited the growth of the same pathogens by 89%, 84% and 74%.The results showed minimal inhibitory concentrations (MIC) were 5 % (v/v) for sweet flag and wild basil and 20% (v/v) for all other plant crude extracts. Out of six plants extract screened, wild basil and sweet flag showed more than 80% fungal inhibition after 6 hour immersion and other extracts could not exceed 60% inhibition after any exposure time. The study revealed that methanol crude extract of sweet flag and wild basil exhibit strong fungistatic and fungicidal activities against tested fungi. These results support the potential use of these plant extracts in the management of diseases caused by tested plant pathogenic fungi.

Keywords: Antifungal activity, *Fusarium oxysporum*,*Rhizoctonia solani* and *Colletotrichum musae,* plant extracts, sweet flag and wild basil

INTRODUCTION

Rhizoctonia solani, *Fusarium oxysporum* and *Colletotrichum musae* are plant pathogenic fungi with a wide host range and worldwide distribution [1].At present quick and effective management for most of plant pathogenic fungi is generally achieved by the use of synthetic fungicides. The massive use of synthetic fungicides in crop defense from plant pathogenic fungi had severe environmental impact.[2] The inappropriate use of agrochemicals especially fungicides were found to possess adverse effects on ecosystems and a possible carcinogenic risk than insecticides and herbicides together.[3] Moreover, resistance by pathogens to fungicides has rendered certain fungicides ineffective.[4,5,6] In recent years, a large number of synthetic pesticides have been banned in the western world because of their undesirable attributes such as high and acute toxicity, long degradation period, accumulation in food chain and an extension of their power to destroy both useful organism and harmful pests[7]. Due to the aforementioned considerations, necessitate the search for alternative control measures to reduce the dependence on the synthetic fungicides.

Medicinal plants represent a rich source of antimicrobial agents [8]. Many of the plant materials used in traditional medicine are readily available in rural areas at relatively cheaper price [9]. Medicinal plants extracts are promising as alternative or complementary control means because of their anti-microbial activity, nonphytotoxicity, systemicity as well as biodegradability.[10,11]Plants produce a great deal of secondary metabolites, many of them with antifungal activity. Well-known examples of these compounds include flavonoids, phenols and phenolic glycosides, unsaturated lactones, sulphur compounds, saponins, cyanogenic glycosides and glucosinolates.[12,13]Although hundreds of medicinal plants are used medicinally in different countries as a source of many potent and powerful drugs and the vast majority of them have not been adequately explored against plant pathogenic fungi[14] . Plants are the sources of natural pesticides that make excellent leads for new biopesticide development. [15,16,17]

The use of plants for their antifungal properties which could be used against the pathogen in the organic farming system becomes an area of interest for the ecofriendly mode of disease management. Considering the vast potentiality of plant as sources for antimicrobial drugs with reference to antimicrobial agents, a systematic investigation was undertaken to screen the antifungal activity of medicinal plant species ,*Oxalis corniculata* (creeping woodsorrel red and green), *Ocimum gratissimum* (wild basil) and bulbs of *Acorus calamus* (sweet flag), *Zingiber officinale* (ginger),wild sunflower (*Tithonia diversifolia)* and siam weed (*Chromolaena odorata*) against fungal strains *F. oxysporum*,*R.solani*,*and C. musea* .

MATERIALS AND METHODS

Preparation of plant extracts:

Fresh leaves of *Oxalis corniculata* (creeping woodsorrel red and green), *Ocimum gratissimum* (wild basil), wild sunflower (*Tithonia diversifolia)* and siam weed (*Chromolaena odorata*) and bulbs of *Acorus calamus* (sweet flag) and *Zingiber officinale* (ginger) were collected from the surrounding areas of Belihuloya,

Sri Lanka . The leaves and bulbs were washed in clean water and dried in room temperature. The dried plant materials were milled to a fine power using grinder and stored in the dark at room temperature inairtight containers. The air dried plant materials were ground to a fine power and 1 g of finely ground plant material was used for the extraction in 20 ml of methanol. The solution was kept overnight at room temperature and filtered using filter papers (Whatman filter paper No. 1) and stored at 4^0C temperature.

Effect of Different Concentrations of Extracts on Radial Growth of test Organisms:

Plant extracts were tested for their efficiency against the pathogen by using an agar dilution technique.[18] Different concentrations of the extracts; 20%, 15%, 10%, and 5% were obtained by amending PDA. The amended medium was dispensed into sterile Petri plates and allowed to solidify with streptomycin (100 µg/ml). Each plate was inoculated with *F. oxysporum,R.solani*, and *C. musea*. A 4-mm diameter mycelia disc of each of the test organisms was inoculated on each amended agar plate. Inoculated plates were incubated at 25±2°C and growth measured along the perpendicular lines. Daily radial growth of each test organism in any of the test extracts was recorded for 7 days. Each treatment was replicated thrice with appropriate untreated controls.

In here three replications were prepared for each treatment. Then all the culture plates were incubated at 25±2°C in dark condition. The mycelia growth of fungus was measured after 24, 48, 72 and 96 hours. Calculate the percent inhibition of the mycelia growth over control by using the following formula [19].

$$\% \text{ of inhibition} = \frac{\text{Diameter of control colony} - \text{Diameter of treated colony}}{\text{Diameter of control colony}} \times 100$$

Minimum Inhibition Concentration (MIC):

The minimal inhibition concentration was taken from the results of the fungistatic activity. The lowest bio extracts and chemical concentrations with highest inhibition percentage were taken as MIC.

Fungicidal activity:

Further study about fungicidal activity of each plant extract and Carbendazim were done for the minimal inhibition concentrations (MIC). Fungicidal activities of each treatment against *F. oxysporum* , *R.solaniand C.musea* were done by immersing the fungal block in minimal inhibition concentration of each solution separately for 5, 10, 30, 60 and 120 minutes and 24 hours. PDA media plates were prepared and treated fungal blocks were inoculated aseptically in the center of the plate. The agar blocks were washed prior to inoculate on PDA plates to remove the crude extracts. Fungal blocks which were dipped in methanol for the above time periods were used as controller. Three replications were prepared for each treatment. All culture plates were incubated at room temperature in dark condition. The mycelia growth was measured by taking the colony diameter after 36, 72 and 96 hour.

The comparison of the fungicidal activity of minimal inhibition concentrations of plant extracts and Carbendazim were done by calculating the present inhibition of the mycelia growth over control by using the

formula [19].

Data analysis:

The experiment was conducted using a completely randomized design. Standard errors of means of three replicates were computed using computer software Microsoft Excel. All the data were subjected to analysis of variance followed by mean separation through Duncan's Multiple Range Test using computer software.

RESULTS AND DISCUSSION

Antifungal activity of six botanical extracts was assayed and the effect of plant extracts on the growth of fungal strain *F. oxysporum*,*R.solani*,*and C.musea* was observed. The data revealed that significant reduction in growth of fungal strain *F. oxysporum*,*R.solani*,*and C. musea* against six medicinal plants. The antifungal effects of the studied plant extracts creeping woodsorrel, wild basil, wild sunflower, siam weed, sweet flagand ginger on the tested fungi strain *F. oxysporum*,*R.solani*,*and C.musea* were compared with the control .The results showed that the growth inhibition of the tested fungi produced by plants extracted at concentration ranging from 5 - 20% were significantly different from control values.

All the plant extracts exhibited different degrees of antifungal activity against *F.oxysporum.* The growth of *F. oxysporum* was highly inhibited by all the tested concentrations (5 - 20%) of methanol extracts of sweet flagand wild basilcompared with control, the correspond inhibition ranging from 91% - 75%. The plant extract of creeping woodsorrel, wild sunflower,siam weed showed comparatively very low activity against *F.oxysporum* . However, *Ginger* showed moderate level of antifungal activity (75%-42%)followed by siam weed (44%-8%) creeping ws (red) (42%-8%), creeping ws (green) (13%-2%), wild sunflower (flower) (15%-13%) and wild sunflower (leaves) (8%-1%) (Figure 01).

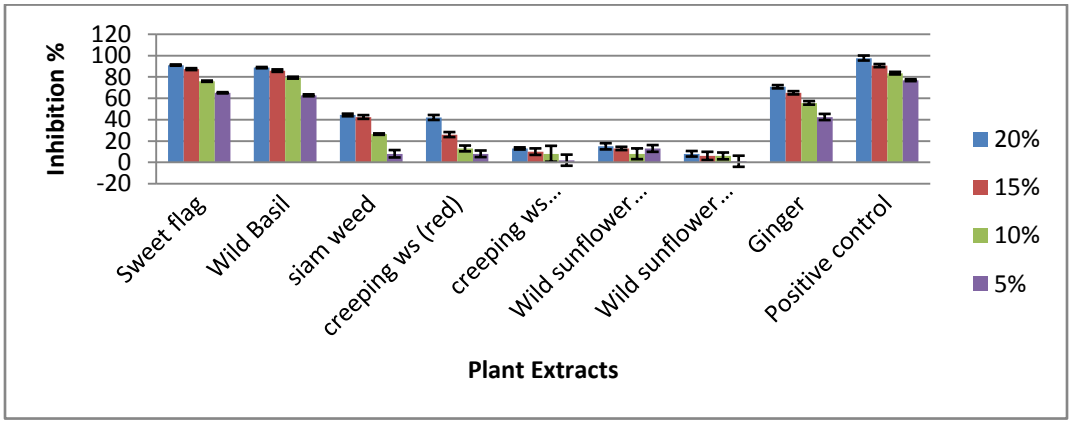

Figure 1: Mycelium growth inhibition effect at various concentrations of*Oxalis corniculata* (creeping woodsorrel red and green), *Ocimum gratissimum* (wild basil), wild sunflower (*Tithonia diversifolia)* and Siam Weed (*Chromolaena odorata*) *Acorus calamus* (sweet flag)and *Zingiber officinale* (ginger) against *Fusarium oxysporum*

All the plant extracts exhibited different degrees of antifungal activity against *R. solani.* The growth of *R. solani* was highly inhibited by all the tested concentrations (5 - 20%) of methanol extracts of sweet flagand wild basilcompared with control, the corresponding inhibition ranging from 91% - 75%. The plant extract of ginger, creeping woodsorrel (red and green), wild sunflower and siam weed showed comparatively very low activity against *R. solani.*Ginger showed inhibitory effect ranging from 41%-10%followed by siam weed (24%-2%) , creeping ws (red)(18%-2%), creeping ws (green) (23%-14%), wild sunflower (flower)(15%-3%) and wild sunflower (leaves) (12%-5%)(Figure 02). However, there was no significant difference between them.

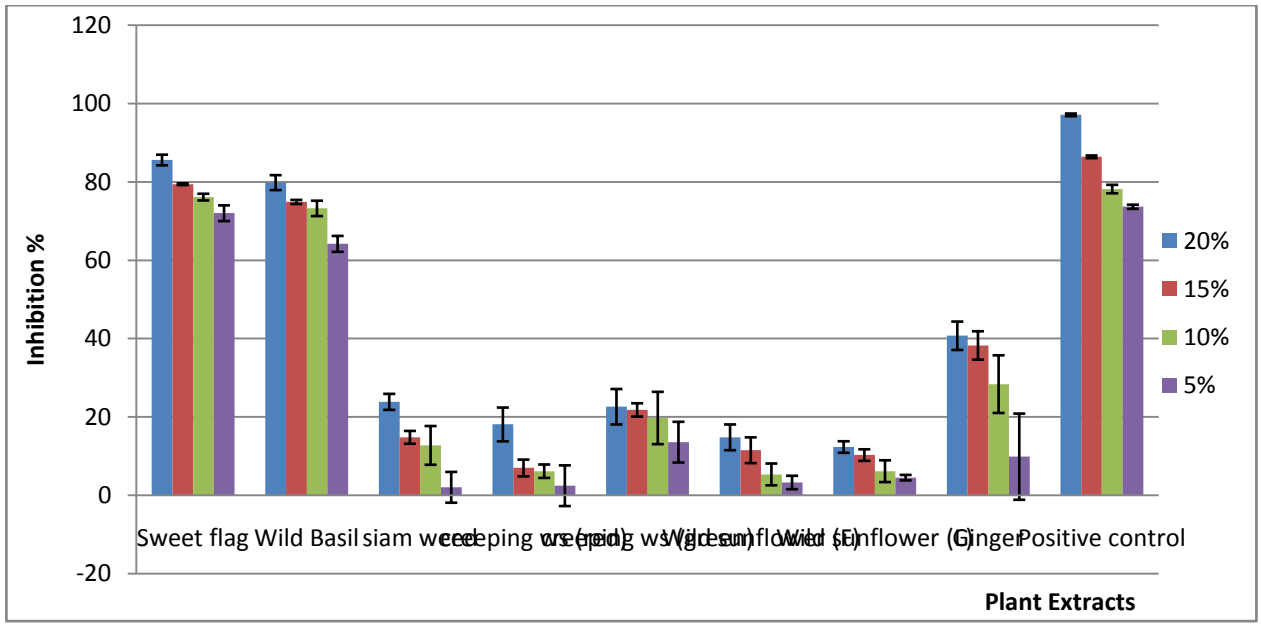

Figure 2: Mycelium growth inhibition effect at various concentrations of*Oxalis corniculata* (creeping woodsorrel red and green), *Ocimum gratissimum* (wild basil), Wild sunflower (*Tithonia diversifolia)* and Siam Weed (*Chromolaena odorata*) *Acorus calamus* (sweet flag) and *Zingiber officinale* (ginger) against *Rhizoctonia solani*

All the plant extracts exhibited different degrees of antifungal activity against *C. musea.* The growth of *C. musea* was highly inhibited by all the tested concentrations (5 - 20%) of methanol extracts of sweet flagand wild basil compared with control, the corresponding inhibition ranging from 91% - 75%. The plant extract of creeping woodsorrel (red and green), wild sunflower,siam weed showed comparatively very low activity against *F.oxysporum.* However, gingershowed 61%-34%followed by Siam weed (23%-3%) , creeping ws (red)(7%-1%), creeping ws (green) (18%-5%), wild sunflower (flower)(20%-1%) and wild sunflower (leaves) (23%-5%)(Figure 03). However, there was no significant difference between them.

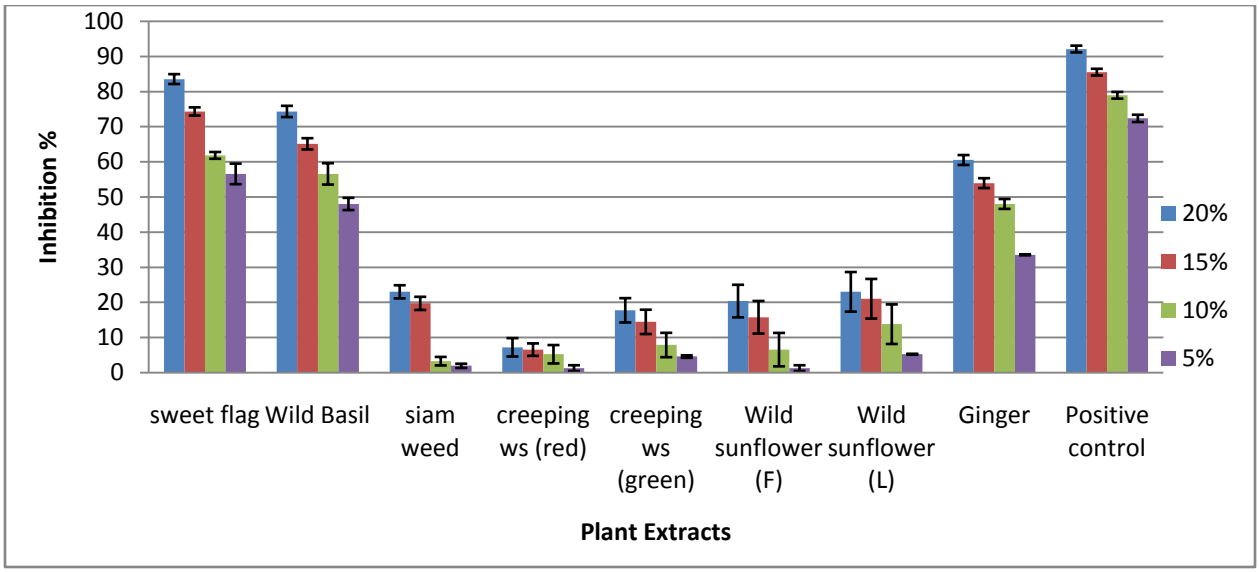

Figure 3: Mycelium growth inhibition effect at various concentrations of*Oxalis corniculata* (creeping woodsorrel red and green), *Ocimum gratissimum* (wild basil), Wild sunflower (*Tithonia diversifolia)* and Siam Weed (*Chromolaena odorata*) *Acorus calamus* (sweet flag) and *Zingiber officinale* (ginger) against *Colletotrichum musea*

Methanol extracts of wild basil and sweet flagwere found highly effective in suppressing the growth of *F. oxysporum , R. solaniand C.musea*evenat 5%concentration (Figure 04). However creeping woodsorrel red and green, ginger, siam weed and wild sun flower cause very low growth inhibition against F. *oxysporum, R. solani* and *C.musea* even at 20% of concentration.Minimal inhibition concentration (MIC) of sweet flag and wild basil was 5%, whereas MIC for ginger was recorded as 20 %(figure 04).

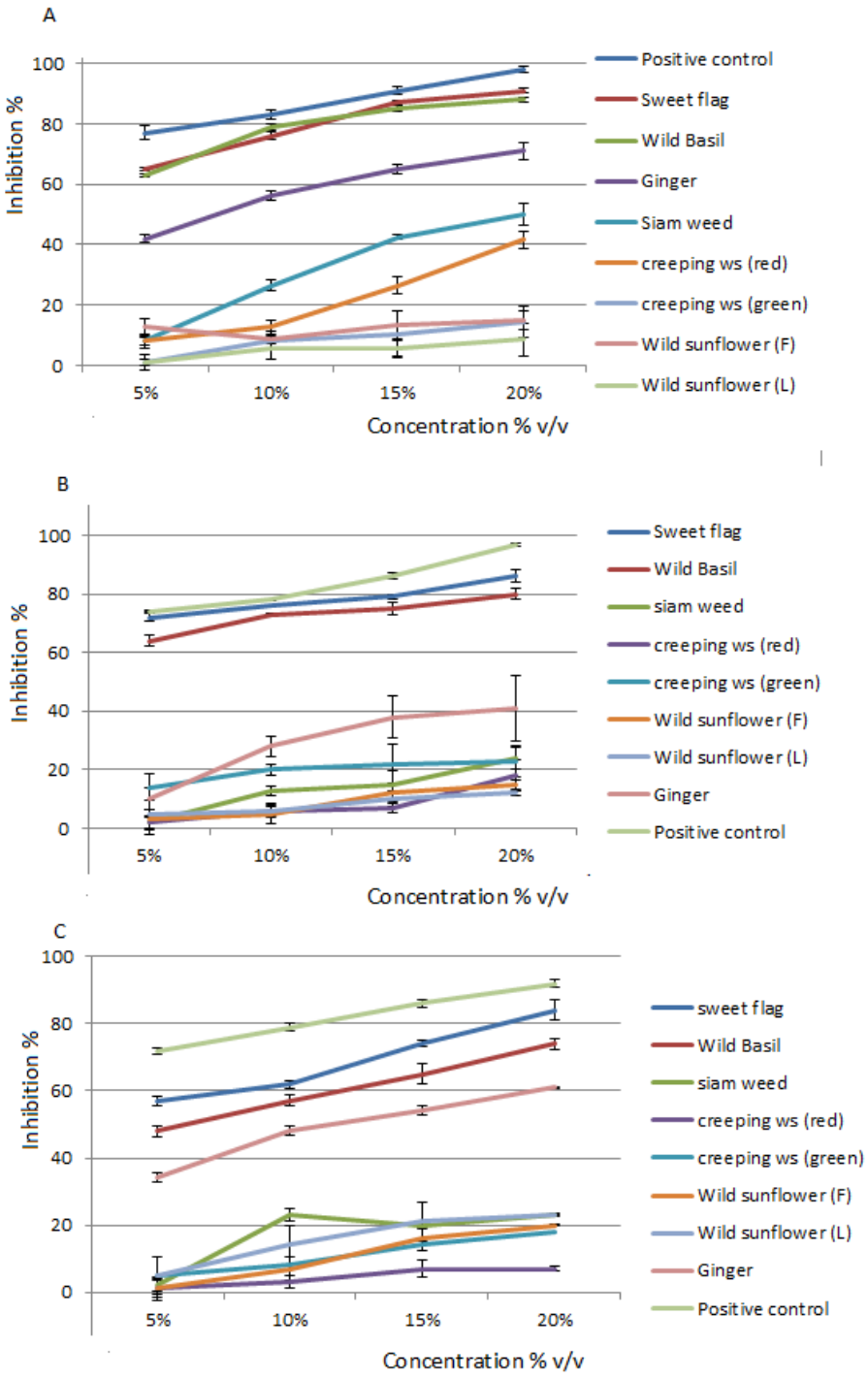

Figure 4: Effect of *Oxalis corniculata* (creeping woodsorrel red and green), *Ocimum gratissimum* (wild basil), wild sunflower (*Tithonia diversifolia)* and siam Weed (*Chromolaena odorata*) *Acorus calamus* (sweet flag)and *Zingiber officinale* (ginger) treatment at different concentration against *F. oxysporum*(A), *R. solani* (B) *and C.musea* (C)

The fungicidal activities were tested to understand the ability of different plant extract to kill the vegetative forms of *F. oxysporum, , R. solaniand C.musea* after immersed in solutions for different time periods. According to results, only the wild basil and sweet flagwas shown the significant inhibition (more than 85%) of the vegetative growth of the fungus after 6 hours immersion time for sweet flagand 6 hours of immersion time for wild basil, respectively (Table 02). In addition, the fungicidal activity was varied with exposure time. Although all the other plant extracts showed inhibition of the fungal growth compared to the control, after 24 hours of immersing, they didn't show any significant fungicidal effect on *F. oxysporum, , R. solaniand C.musea.*

Time	Inhibition ratio of fungal growth																							
	Methanol extracts of																							
	sweet flag			Ginger			Wild basil			Wild sun flower (L)			Wild sun flower (L)			Creeping Wood (red)			Creeping Wood (red)			Siam Weed		
	F	*R*	*C*	*F*	*R*	*C*	*F*	*R*	*C*	*F*	*R*	*C*	*F*	*R*	*C*	*F*	*R*	*C*	*F*	*R*	*C*	*F*	*R*	*C*
1 hr	53 .3^{y}	60 .4^{y}	58 .8^{y}	44 .6^{x}	33 .5^{x}	37 .5^{x}	55 .5^{y}	56 .5^{y}	50 .3 y	18 .5^{x}	10 .2^{x}	11 .2^{x}	16 .3^{x}	14 .5^{x}	20 .2^{x}	12 .3^{x}	22 .2^{x}	17 .4^{x}	12 .5^{x}	16 .4^{x}	15 .3 2^{x}	28 .3^{x}	18 .2^{x}	20 .2^{x}
3 hr	73 .3^{z}	70 .6^{z}	70 .4^{z}	48 .3^{x}	39 .6^{x}	46 .2^{x}	65 .5	60 .4^{y}	65 .3^{y}	20 .3^{x}	15 .3^{x}	12 .2^{x}	19 .0^{x}	20 .2^{x}	22 .2^{x}	19 .0^{x}	25 .2^{x}	20 .2^{x}	16 .8^{x}	21 .2^{x}	20 .6 2^{x}	29 .0^{x}	20 .2^{x}	22 .2^{x}
6 hr	95 .8^{z}	87 .7^{z}	92 .0^{z}	56 .4^{y}	46 .5^{x}	53 .8^{x}	85 .5	87 .3^{z}	86 .5^{z}	23 .0^{x}	18 .2^{x}	16 .3^{x}	20 .0^{x}	22 .2^{x}	26 .3^{x}	24 .0^{x}	28 .2^{x}	24 .3^{x}	20 .5^{x}	23 .5^{x}	24 .5^{x}	30 .0	24 .2^{x}	26 .3^{x}
12hr	98 .8^{z}	93 .3^{z}	95 .3^{z}	59 .8^{y}	51 .2^{x}	58 .5^{y}	95 .3	90 .3^{z}	94 .3^{z}	22 .4^{x}	23 .2^{x}	19 .3	22 .4^{x}	25 .2^{x}	29 .3	26 .4^{x}	30 .2^{x}	28 .3	23 .4^{x}	25 .2^{x}	28 .3	30 .4^{x}	30 .2^{x}	29 .3
24hr	10 0^{z}	10 0^{z}	10 0^{z}	65 .2^{y}	55 .2^{y}	59 .3^{y}	10 0	10 0^{z}	10 0^{z}	29 .6^{x}	25 .3. 2^{x}	25 .2^{x}	24 .8^{x}	33 .2^{x}	31 .2^{x}	29 .8^{x}	34 .2^{x}	31 .2^{x}	26 .5^{x}	29 .8^{x}	30 .2^{x}	39 .8	33 .2^{x}	35 .2^{x}

Table 2: Fungicidal activity of various concentration of crude extracts against the growth of *Fusarium oxysporum*, *Rhizoctonia solani and Colletotrichum musae*

F: *Fusarium oxysporum* *R: Rhizoctonia solani* *C: Colletotrichum musae*

Numbers followed by the same letters in each row were not significantly different according to Duncan's Multiple Range test at (P = .05).

Minimal inhibition concentration (MIC) of sweet flag and wild basil was 5%whereas, MIC for ginger was determine as 20%.

Biological control had attained importance in modern agriculture to curtail the hazards of intensive use of chemicals for pest and disease management.[20] Accordingly, the efficacy of different plant extracts against *F. oxysporum,R. solani* and *c.musea* the causal agents of wilt diseases of tomato, damping off and anthracnose disease of banana were studied *in vitro.*

Methanol extract of creeping woodsorrel (red and green), wild sunflower, ginger and siam weed at all concentrations, showed a slightly inhibition for the three selected pathogens. Thus those extracts were

excluded from further studies whereas sweet flag and wild basil were found to be highly effective in controlling the growth of the three tested pathogens. All types of extracts from sweet flag and wild basil showed different levels of antimicrobial activity and the relative differences were found to vary within the tested extracts and with increasing concentration of the extracts, a gradual increase in the inhibition potential of the tested fungi was recorded.

In this study, four concentrations of the sweet flag and wild basil extracts effectively suppressed mycelial growth of the three pathogens, the 20% concentration completely suppressed the growth of one of them, *F. oxysporum* (100%). These results are in agreement with [21] who found that the growth of four pathogens (*F. oxysporum*, *R. solani*, *Sclerotium rolfsii* and *Sclerotinia sclerotiorum*) which incite wilt and rot in *Cicer arietinum*, was inhibited in liquid medium by extracts of leaf, trunk park and oil from the aromatic ginger and wild basil.

Comparing leaf extracts of with methanol, the inhibition percentage in each increased gradually with the extract concentration, both completely suppressed the growth of *F. oxysporum*. Results of the present study are in agreement with[22] who noticed that methanol extracts of wild basil showed fungitoxic properties against 5 pathogenic fungi ; *Alternaria brassicola*, *Colletotrichum capsici*, *F. oxysporum*, *R. solani* and *Sclerotinia sclerotiorum*) when tested under laboratory conditions at 500 and 1000 µg/ml. Several plant extracts including wild basil have been tested for antifungal activities and used to control of plant fungal diseases caused by different formae *speciales* of *F.oxyporum* , *R. solani* and *C.musae.*[23, 24, 25, 26 ,27 ,28]However according to our knowledge this is the first report regarding antifungal activities of sweet flag against *F.oxyporum* , *R. solani* and *C.musae*.

The fungitoxic effects of the phyto-extracts indicate the potentials of selected plant species as a source of natural fungicidal material. These extracts exhibit significant fungicidal properties that support their traditional use as antiseptics. In the case of fungal infection, these mechanisms include synthesis of bioactive organic compounds [29] and antifungal proteins [30]and peptides. [31]

CONCLUSION

Antifungal activity was confirmed by all of the selected plant species and the results revealed wild basil and sweet flag are the most effective inhibitor for the mycelia growth of three tested pathogens. The finding of the present investigation could be an important step towards the possibilities of using natural plant products as biopesticides in the control of plant diseases caused by *F. oxysporum*,*R. solani* and *C.musea*. Further studies are needed to determine the chemical identity of the bioactive compounds responsible for the observed antifungal activity.

REFERENCES

1. Agrios GN., Plant pathology, sixth edition. Academic Press, New York. 2005.
2. Osman KA and Al-Rehiayam S. Risk assessment of pesticide to human and the environment. Saudi J. Biol. Sci. 2003: 10:81-106.
3. Stranger RN and Scott PR. Plant Diseases: a threat to global food security; Annual Review of phytopathology.2005:43:83-116
4. Zhonghua MA and Michailides TJ. Advances in understanding molecular mechanisms of fungicide resistance and molecular detection of resistant genotypes in phytopathogenic fungi. Crop Prot. 2005: 24:853-863.
5. Cohen ML. Epidemiology of drug resistance implications for a post antimicrobial era. Science. 1992: 257:1050-1055.
6. Nascimento G., Juliana Locatelli, G.F., Paulo C. Freitas, G. Silva, L. 2000.Antibacterial activity of plant extracts and phytochemicals on antibiotic-resistant bacteria**.***Braz.J. Microbiol.* 31: 247 – 256.
7. Gatto MA, Ippolito A, Linsalata V, Cascarano NA, NigroF, Vanadia S. and Di Venere, D. Activity of extracts from wild edible herbs against postharvest fungal diseases of fruit and vegetables. Postharvest Biol Technol: 2011: 61:72–82.
8. Mahesh B and Satish S .Antimicrobial activity of some important medicinal plant against plant and human pathogens. World J. Agri. Sci. 2008: 4: 839-843.
9. Mann A, Banso A and Clifford LC .An antifungal property of crude plant extracts from *Anogeissus leiocarpus* and *Terminalia avicennioides*. Tanzania J.Health Res. 2008: 10:1 34-38.
10. Talibi I, Askarne L, Boubaker H, Boudyach EH, Msanda F, Saadi B. and Ait Ben Aoumar A. Antifungal activity of Moroccan medicinal plants against citrus sour rot agent Geotrichum candidum. Lett Appl Microbiol: 2012:55:155–161.
11. Gómez Garibay R, Reyes Chilpa, Quijano L , Calderón Pardo J.S, Ríos Castillo T.*Methoxifurans auranols* with fungostatic activity from *Lonchocarpus castilloi* Phytochemistry. 1990:29:459–463
12. Bennett RN and Wallsgrove RM. Secondary metabolites in plant defence mechanisms New Phytology. 1994: 127 :617–633
13. Osbourne A.E .Preformed antimicrobial compounds and plant defence against fungal attack. The Plant Cell. :1996:8:1821–1831
14. Gangadevi V, Yogeswari S, Kamalraj S, Rani G and Muthumary J .The antibacterial activity of *Acalypha indica* L. Indian J. Sci. Technol. 2008:1 :6:1-5.
15. Satish S, Raghavendra MP, Mohana DC and Raveesha KA .Antifungal activity of a known medicinal plant *Mimusops elengi* L. against grain molds. J. Agri. Technol. 2008: 4:1: 151-165.
16. Shanmugavalli N, Umashankar V and Raheem .Anitmicrobial activity of *Vanilla planifolia*. Indian J.

Sci.Technol. 2009: 2 :3: 37-40.

17. Srivastava AK, Bihari, B and Lal B. Studies on biofungicidal properties of leaf extract of some plants. Indian Phytopathol. 1997. 50:3: 408-411.
18. Jorgensen JH, Turnidge JD and Washington JA. Antibacterial susceptibility tests: dilution and disk diffusion methods. In: Murray PR, Pfaller MA, Tenover FC,Baron EJ, Yolken RH, ed. Manual of clinical microbiology, 7th ed.. Washington, DC: ASM Press; 1999:1526-1543
19. Vincent JM. Distoration of fungal hyphae in the presence of certain inhibitors. Nature, 1947: 159:850
20. Tuber MJ and Baker R. Every other alternative biological control. Bio Science. 1988: 38: 660
21. Singh UP, Singh HB and Singh RB. The fungicidal effect of neem (*A. indica*) extracts on some soil borne pathogens of gram (*Cicer arietinum*). Mycologia, 1990: 72: 1077-1093
22. Shivpuri A, Sharma OP and Jhamaria SL. Fungitoxic properties of plant extracts against pathogenic fungi. J. Mycol. and Pl. Pathol. 1997: 27:1:29-31.
23. Amadi JE, Salami SO and Eze CS .Antifungal properties and phytochemical screening of extracts of African Basil (*Ocimum gratissimum* L.) Agriculture and biology journal of North America. 2010:12: 163-166
24. Ketut Suada I ,Wayan Suniti NI. Suppression ability of crude extract derived from marine biota against *Fusarium oxysporum* f.sp. *vanilla*. Journal biology 2010: xiv :1 : 7 - 10
25. Ogechi N. Agbenin, P.S. Marley. In-vitro assay of some plant extracts against *Fusarium oxysporum* f. sp. *Lycopersici* causal agent of tomato wilt. Journal of plant protection research. 2006: 46:3:215-219
26. Jaya Gujar and Dhananjay Talwankar. Antifungal potential of crude plant extract on some pathogenic fungi. World Journal of Science and Technology .2012: 2:6:58-62
27. Mangang HC and Chhetry GKN. Antifungal Properties of certain Plant Extracts against *Rhizoctonia Solani* Causing Root Rot of French bean In Organic Soil of Manipur. International Journal of Scientific and Research Publications. 2012: 2: 5:1-5
28. Stangarlin JR, Kuhn O J , Assi and Schwan-Estrada L K R F. Control of plant diseases using extracts from medicinal plants and fungi. Science against microbial pathogens: communicating current research and technological advancesA. Méndez-Vilas (Ed.) 2011;1033-1042
29. Domenico R, Filomena M and Elisabetta S.Inhibitory Effect of Plant Extracts on Conidial Germination of the phytopathogenic Fungus *Fusarium oxysporum.* American Journal of Plant Sciences. 2012: 3:1693-1698.
30. Morrisey JP and Osbourn A. Fungal resistance to plant antibiotics as a mechanism of pathogenesis. Microbiology and Molecular Biology Reviews. 1999: 63:708-724.
31. Selitrennikoff CP. Antifungal proteins. Applied and Environmental Microbiology. 2001; 67:2883-2894.