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Original Research

Antifungal Activity of Some Botanicals against Seed-borne Fungi

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Abstract

Synthetic chemicals are extensively used to control plant diseases caused by fungi. Interest in botanicals with antifungal activity increased because of drawbacks associated with the use of synthetic chemicals. The present study was conducted to determine antifungal potential against three seed-borne fungi (*Curvularia* sp., *Alternaria* sp. and *Fusarium* sp.) of aqueous extract from 20 plants belonging to 14 families by poisoned food technique. The inhibition of *Curvularia* sp., *Alternaria* sp. and *Fusarium* sp. by extracts varied between 12.24 to 53.06%, 11.11 to 51.85% and 25.00 to 58.33% respectively. Among fungi, *Fusarium* sp. was inhibited to higher extent by majority of extracts. Highest inhibitory activity against *Curvularia* sp. was displayed by *Harpullia arborea*. Extract of *Solanum virginianum* exhibited stronger inhibitory activity against *Alternaria* sp. while extract of *H. arborea* and *Azima tetracantha* inhibited *Fusarium* sp. to higher extent. These botanical extracts can be exploited in the control of seed-borne fungi and other phytopathogenic fungi.

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INTRODUCTION

Plant diseases are one among the several important factors which influence directly the global agricultural productivity. Fungi are considered as the leading pathogens of plants as they cause numerous diseases in plants leading to huge economic losses in severe cases. The management of plant diseases caused by fungi is often followed with the extensive use of chemical fungicides, but this strategy for control of phytopathogens is associated with negative environmental impacts, potential damage to humans and other non-target organisms and deposition of residues on the agricultural produces. Besides, many pathogens are likely to develop resistance against synthetic fungicides. Hence there is a great demand for safer, alternative and effective agent for controlling phytopathogenic fungi. Nowadays the search for natural products with antifungal activity is triggered immensely. Aromatic and other plants are promising and have been shown to control many phytopathogenic fungi which cause dreadful diseases in several crops (Khan and Nasreen, 2010; Al-Reza *et al*., 2010; Prince and Prabakaran, 2011; Bahraminejad *et al*., 2013; Rodino *et al*., 2014; Neela *et al*., 2014; Baize *et al*., 2014; Sundaramoorthy *et al*., 2014; Kekuda *et al*., 2016; Sales *et al*., 2016). The present study investigated antifungal potential of aqueous extract of 20 plants (14 families) collected from different places of Shivamogga district, Karnataka, India.

MATERIALS AND METHODS

Collection and Identification of Plants

A total of 20 plants belonging to 14 families were collected from different places of Shivamogga district, Karnataka during January-February 2017. The plants were identified by referring standard flora (Manjunatha *et al*., 2004; Bhat, 2014) along with the help of taxonomists. Details on the family, place of collection and parts of the plants used is presented in Table 1.

Extraction

The selected parts from the plants were washed under clean water to remove adhering matter and dried under shade. The shade dried plant materials were ground into fine powder in a blender. Extraction was carried out by transferring 10g of powdered material into 100ml of distilled water and boiling for 15 minutes. The content was then filtered through 4-fold muslin cloth followed by Whatman filter paper No. 1 (Al-Manhel and Niamah, 2015). The filtrate was used to assess antifungal activity.

Antifungal Activity of Aqueous Extract of Selected Plants

The antifungal effect of aqueous extracts was screened by Poisoned food technique employed by Kekuda *et al*. (2016). The sterile control (without extract) and poisoned (10%) Potato dextrose agar (PDA) plates were inoculated aseptically at the centre with the well sporulated cultures of test fungi namely *Alternaria* sp.,

Curvularia sp. and *Fusarium* sp. (isolates recovered previously from sorghum seeds; maintained on PDA slants in refrigerator) the plates were incubated in upright position at room temperature for 96 hours. The diameter of fungal colonies in mutual perpendicular directions was measured using a ruler and the antifungal effect of extracts in terms of inhibition of mycelial growth (%) was determined using the formula:

Mycelial growth inhibition (%) = (A – B / A) x 100

where 'A' and 'B' denotes the diameter of fungal colony on control and poisoned plates respectively

Table 1: Plants selected for this study

No.	Name of the Plant	Family	Part Used	Place of Collection
1	*Kigelia africana* (Lam.) Benth.	Bignoniaceae	Leaf and flower	Shikaripura
2	*Clerodendrum philippinum* Schauer	Verbenaceae	Leaf and flower	Shikaripura
3	*Harpullia arborea* (Blanco) Radlk.	Sapindaceae	Leaf	Shiralakoppa
4	*Hydnocarpus pentandrus* (Buch.-Ham.) Oken	Achariaceae	Leaf	Shiralakoppa
5	*Flacourtia indica* (Burm. f.) Merr.	Salicaceae	Leaf	Sagara
6	*Gardenia gummifera* L.f.	Rubiaceae	Leaf and fruit	Sagara
7	*Salix tetrasperma* Roxb.	Salicaceae	Leaf	Siddarahalli
8	*Azima tetracantha* Lam.	Salvadoraceae	Leaf	Matturu
9	*Bixa orellana* L.	Bixaceae	Leaf and fruit	Malalakoppa
10	*Kirganelia reticulata* (Poir.) Baill.	Euphorbiaceae	Leaf	Malalakoppa
11	*Couroupita guianensis* Aubl.	Lecythidaceae	Leaf and flower	Shikaripura
12	*Solanum virginianum* L.	Solanaceae	Aerial parts	Indiranagara
13	*Nicotiana plumbaginifolia* Viv.	Solanaceae	Aerial parts	Matturu
14	*Capparis zeylanica* L.	Capparaceae	Leaf	Shikaripura
15	*Ixora brachiata* Roxb.	Rubiaceae	Aerial parts	Shiralakoppa
16	*Canthium dicoccum* (Gaertn.) Merr.	Rubiaceae	Leaf	Sagara
17	*Capparis sepiaria* L.	Capparaceae	Leaf	Shikaripura
18.	*Antigonon leptopus* Hook. & Arn.	Polygonaceae	Leaf	Matturu
19.	*Ipomea* sp.	Convolvulaceae	Whole plant	Shiralakoppa
20.	*Solanum torvum* Sw.	Solanaceae	Whole plant	Shikaripura

RESULTS AND DISCUSSION

Interest in plants with antifungal activity against phytopathogenic fungi has increased due to severe effects associated with the indiscriminate use of synthetic fungicides. Most of these chemical agents are residual in nature and are not easily degraded leading to pollution problems. Besides, these chemicals are costly and are not easily afforded by poor individuals. Botanicals offer cheaper, biodegradable and safest alternate for disease control (Thembo *et al*., 2010; Masih *et al*., 2014; Ngadze, 2014; Daniel *et al*., 2015; Kekuda *et al*., 2016). In this study, we evaluated the potential of aqueous extracts prepared from selected plants to inhibit the mycelial growth of three fungi by Poisoned food technique. Poisoned food technique has been extensively used by several researchers to investigate antifungal potential of plant extracts against a range of phytopathogenic fungi (Khan and Nasreen, 2010; Farooq *et al*., 2010; Ngadze, 2014; Daniel *et al*., 2015; Omidpanah *et al*., 2015; Kekuda *et al*., 2016). The result of inhibitory potential of aqueous extracts of selected plants is shown in Table 2.

Poisoning PDA medium with the aqueous extracts resulted in inhibition of mycelial growth of test fungi. The extent of inhibition of *Curvularia* sp., *Alternaria* sp. and *Fusarium* sp. by extracts varied between 12.24 to 53.06%, 11.11 to 51.85% and 25.00 to 58.33 % respectively. Among fungi, *Fusarium* sp. was inhibited to higher extent by majority of extracts (22 extracts out of 25 extracts). Highest inhibitory activity against *Curvularia* sp. was displayed by *H. arborea* while least inhibition of *Curvularia* sp. was shown by *Ipomea* sp. and flower of *K. africana*. Extract of *S. virginianum* exhibited stronger inhibitory activity against *Alternaria* sp. while least inhibition of *Alternaria* sp. was displayed by *I. brachiata* and *C. zeylanica*. Extract of *H. arborea* and *A. tetracantha* inhibited *Fusarium* sp. to higher extent while *H. pentandrus* displayed least inhibitory activity against *Fusarium* sp. In case of *C. guianensis*, flower extract exhibited stronger inhibition of test fungi when compared to leaf extract. Among leaf and fruit extract of *G. gummifera*, marked antifungal activity was shown by leaf extract. Similarly, leaf extract of *H. arborea* exhibited high antifungal activity when compared to fruit extract. Leaf and flower extract of *C. philippinum* exhibited similar inhibitory activity against *Curvularia* sp. and *Fusarium* sp. while flower extract caused high inhibition of *Alternaria* sp. when compared to leaf extract. Similarly, in case of *K. africana*, leaf extract caused high inhibition of *Curvularia* sp. and *Fusarium* sp. while flower extract caused high inhibition of *Alternaria* sp.

Table 2: Colony diameter of test fungi on control and poisoned plates

No.	Extract / Control	Colony diameter in cm (% inhibition of fungi)		
		***Curvularia* sp.**	***Alternaria* sp.**	***Fusarium* sp.**
1	*Control*	4.9	5.4	4.8
2	*S. virginianum*	3.3 (32.65)	2.6 (51.85)	2.8 (41.66)
3	*C. dicoccum*	4.0 (18.36)	4.0 (25.92)	2.5 (47.91)
4	*C. guianensis* leaf	4.0 (18.36)	4.0 (25.92)	2.6 (45.83)
5	*C. guianensis* flower	3.6 (26.53)	3.0 (44.44)	2.3 (52.08)
6	*G. gummifera* leaf	3.6 (26.53)	3.4 (37.03)	2.4 (50.00)
7	*G. gummifera* fruit	4.0 (18.36)	3.8 (29.62)	2.6 (45.83)
8	*S. tetrasperma*	3.0 (38.77)	3.6 (33.33)	2.4 (50.00)
9	*A. tetracantha*	4.1 (16.32)	4.6 (14.81)	2.0 (58.33)
10	*I. brachiata*	4.0 (18.36)	4.8 (11.11)	3.0 (37.50)
11	*B. orellana*	4.0 (18.36)	4.0 (25.92)	2.8 (41.66)
12	*N. plumbaginifolia*	3.8 (22.44)	3.6 (33.33)	2.6 (45.83)
13	*Ipomea* sp.	4.3 (12.24)	4.0 (25.92)	3.4 (29.16)
14	*H. arborea* leaf	2.3 (53.06)	3.5 (35.18)	2.0 (58.33)
15	*H. arborea* fruit	2.5 (48.97)	3.6 (33.33)	2.6 (45.83)
16	*K. africana* leaf	4.0 (18.36)	4.0 (25.92)	2.9 (39.58)
17	*K. africana* flower	4.3 (12.24)	3.8 (29.62)	3.2 (33.33)
18	*H. pentandrus*	3.8 (22.44)	4.2 (22.22)	3.6 (25.00)
19	*S. torvum*	3.0 (38.77)	3.1 (42.59)	2.8 (41.66)
20	*C. zeylanica*	3.5 (28.57)	4.8 (11.11)	3.2 (33.33)
21	*C. sepiaria*	3.6 (26.53)	4.4 (18.51)	3.3 (31.25)
22	*C. philippinum* leaf	4.0 (18.36)	4.4 (18.51)	3.2 (33.33)
23	*C. philippinum* flower	4.0 (18.36)	4.0 (25.92)	3.2 (33.33)
24	*F. indica*	3.4 (30.61)	3.6 (33.33)	2.8 (41.66)
25	*K. reticulate*	3.8 (22.44)	4.0 (25.92)	3.3 (31.25)
26	*A. leptopus*	3.6 (26.53)	3.7 (31.48)	3.2 (33.33)

CONCLUSIONS

It was found that most of the plants used in this study displayed marked inhibitory activity against test fungi. Formulations containing these plants can be exploited as fungicidal agents effective against seed-borne fungi and other phytopathogenic fungi. The observed bioactivity of could be attributed to the presence of secondary metabolites having antifungal activity.

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Conflict of Interest

None Declared

REFERENCES

Al-Manhel, A.J. and Niamah, A.K. (2015). Effect of aqueous and alcoholic plant extracts on inhibition of some types of microbes and causing spoilage of food. *Pakistan Journal of Food Science* 25(3): 104-109.

Al-Reza, S.M., Rahman, A., Ahmed, Y. and Kang, S.C. (2010). Inhibition of plant pathogens *in vitro* and *in vivo* with essential oil and organic extracts of *Cestrum nocturnum* L. *Pesticide Biochemistry and Physiology* 96: 86–92.

Bahraminejad, S., Amiri, R., Ghasemi, S. and Fathi, N. (2013). Inhibitory effect of some Iranian plant species against three plant pathogenic fungi. *International Journal of Agriculture and Crop Sciences* 5(9): 1002-1008.

Baize, S., Ayalew, A. and Woldetsadik, K. (2014). Antifungal activity of some plant extracts against (*Colletotrichum musae*) the cause of postharvest banana anthracnose. *Journal of Plant Pathology and Microbiology* 5: 226. doi:10.4172/2157-7471.1000226.

Bhat, G.K. (2014). Flora of South Kanara. Akriti Prints, Mangalore, India.

Daniel, C.K., Lennox, C.L. and Vries, F.A. (2015). In-vitro effects of garlic extracts on pathogenic fungi *Botrytis cinerea*, *Penicillium expansum* and *Neofabraea alba*. *South African Journal of Science* 111 (7/8), 8 pages. http://dx.doi.org/10.17159/sajs.2015/20140240.

Farooq, M.A., Iqbal, U., Iqbal, S.M., Afzal, R. and Rasool, A. (2010). In-vitro evaluation of different plant extracts on mycelial growth of *sclerotium rolfsii* the cause of root rot of sugar beet. *Mycopath* 8(2): 81-84.

Kekuda, T. R. P., Raghavendra, H. L., Solomon, T. and Duressa, D. (2016). Antifungal and antiradical potential of *Moringa stenopetala* (Baker f.) Cufod (Moringaceae). *Journal of Bioscience and Agriculture Research* 11(01): 923-929.

Khan, Z.S. and Nasreen, S. (2010). Phytochemical analysis, antifungal activity and mode of action of methanol extracts from plants against pathogens. *Journal of Agricultural Technology* 6(4): 793-805.

Manjunatha, B.K., Krishna, V. and Pullaiah, T. (2004). Flora of Davanagere district, Karnataka, India. Regency Publications, New Delhi, India.

Masih, H., Peter, J.K. and Tripathi, P. (2014). A comparative evaluation of antifungal activity of medicinal plant extracts and chemical fungicides against four plant pathogens. *International Journal of Current Microbiology and Applied Sciences* 3(5): 97-109.

Neela, F.A., Sonia, I.A. and Shamsi, S. (2014). Antifungal activity of selected medicinal plant extract on *Fusarium oxysporum* Schlechtthe causal agent of Fusarium wilt disease in tomato. *American Journal of Plant Sciences* 5: 2665-2671.

Ngadze, E. 2014. In vitro and greenhouse evaluation of botanical extracts for antifungal activity against *Phythopthora infestans*. *Journal of Biopesticides* 7(2):198-203.

Omidpanah, S., Sadeghi, H., Sarcheshmeh, M.M. and Manayi, A. (2015). Evaluation of antifungal activity of aqueous extracts of some medicinal plants against *Aspergillus flavus*, pistachio aflatoxin producing fungus in vitro. *Drug Development and Therapeutics* 6: 66-9.

Prince, L. and Prabakaran, P. (2011). Antifungal activity of medicinal plants against plant pathogenic fungus *Colletotrichum falcatum*. *Asian Journal of Plant Science and Research* 1(1): 84-87.

Rodino, S., Butu, M., Petrache, P., Butu, A. and Cornea, C.P. (2014). Antifungal activity of four plants against *Alternaria alternata*. *Scientific Bulletin. Series F. Biotechnologies* 18: 60-65.

Sales, M.D.C., Costa, H.B., Fernandes, P.M.B., Ventura, J.A. and Meira, D.D. (2016). Antifungal activity of plant extracts with potential to control plant pathogens in pineapple. *Asian Pacific Journal of Tropical Biomedicine* 6(1): 26-31.

Sundaramoorthy, S., Usharani, S. and George, A.P. (2014). Antifungal activity of plant products for the management of fruit rot infection in chillies. *Plant Pathology Journal* 13(2): 87-99.

Thembo, K.M., Vismer, H.F., Nyazema, N.Z., Gelderblom, W.C.A. and Katerere, D.R. (2010). Antifungal activity of four weedy plant extracts against selected mycotoxigenic fungi. *Journal of Applied Microbiology* 109: 1479–1486.