RESEARCH ARTICLE

Launaea procumbens as a Natural Fungitoxicant against *Aspergillus niger*

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ABSTRACT

Launaea procumbens (Asteraceae) is an aromatic plant has potential of antifungal activity. Methanol extract of leaves of *Launaea procumbens* has shown a striking zone of inhibition of 14.33 mm against *Aspergillus niger* in well diffusion method. The least minimum inhibitory concentration (MIC) was found out of plant extract *Launaea procumbens* with optical density 1.533 in 0.4ml dilution. This study thus confirmed the fungitoxicant potential of *Launaea procumbens*.

Keywords: *Launaea procumbens, A. niger*, Antifungal activity, Fungitoxicant.

INTRODUCTION

Fungal contamination of stored commodities is a very serious problem in tropical warm regions of the world. Contamination by storage fungi and their mycotoxins is of great concern in herbal drug and food industry. Fungi, especially the species of *Aspergillus* and *Penicillium* are among the major reported genera having the ability to produce mycotoxins during storage (Gautam & Bhadauria, 2009). These fungi producing related mycotoxins reduce the quality of food products and the medicinal potential of herbal drugs. *Aspergillus niger* (commonly known as black *Aspergilli),* was recorded as a most dominating fungal species to be associated with herbal drugs during storage (Bugno, Almodovara, Pereira, Pinto, & Sabino, 2006). *A. niger* is a saprophytic and filamentous fungus found in soil, forage, organic debris and food product, causing black mould of onion, shallot; stem rot of Draceana; root stalk rot of Sanseveiria; and boll rot of Cotton; spoilage of cashew kernels, dates, figs, vanilla pods and dried prune (Bobbarala, Katikala., Naidu, & Penumajji, 2009). This fungi commonly cause food poisoning by aflatoxin formation and can also cause aspergillosis, a severe opportunistic infection, in humans (Zhirong, 1999). Medicinal plants are natural resources, yielding valuable herbal products which are often used in the treatment of various ailments (M. Chandrasekaran *et al., 2002). Launaea procumbens* is a naturally occuring population which belongs to family Asteraceae. Considering the vast potentiality of plant as sources for antimicrobial drugs with reference to antifungal agent, a systematic investigation was undertaken to screen the local flora for antifungal activity from *Launaea procumbens*.

MATERIAL AND METHODES

Preparation of Plant Extract

Launaea procumbens was collected from garden of Botany Department, Campus, RTM Nagpur University. Matured leaves and young flowering shoots were thoroughly washed in running water and kept in shade dry. Dry materials were then ground finely by a blender and stored in zip lock polythene bags. Methanol was used as a solvent for extraction. 5g of powdered plant material was extracted by maceration with 20ml methanol and shaken on an orbital shaker for 48 hours.

Antifungal Activity

1. Well Diffusion Method:

The potato tubers were peeled off and weighed for about 200g tubers were chopped into small pieces into the sterile conical flask containing 1000ml distilled water. After boiling, the supernatant were collected and dextrose (20g) with agar (20g) to dissolve the ingredients. Finally, the medium was transferred into measuring cylinder of 1 litre capacity and made the volume to 1 litre by adding more distilled water into it. The medium was poured into two or more Erlenmeyer flasks, cotton plug were put and the plugs were covered with aluminium foil and were autoclaved at 121˚C for 20 minutes. The flasks were taken out when temperature cools down and were used for antifungal activity. Antifungal activity was screened by Agar well diffusion method (Perez *et al.,1990).* The methanol and seven different plant extracts were tested against fungus *Aspergillus niger*. We used the method suggested by Ambikapathy *et al., 2011* with slight modification. After Potato Dextrose Agar (PDA) media was prepared, we added fungal culture in it and pour it in the sterilized petri plates and allowed to solidify. Next day, wells (6 mm) were made in the medium using sterile cork borer. 200µl of each extracts were transferred into the separate wells. The plates were incubated at 27˚C for 24 hrs. After the incubation, the plates were observed for formation of clear inhibition zone around the well indicated the presence of antifungal activity. The zone of inhibition was recorded (Ambikapathy *et al., 2011). Griseofulvin* was used as a standard.

2. Minimum Inhibitory Concentration (MIC)

50 ml nutrient broth was taken in a sterilized test tube to make the fungal broth by inoculating the fresh fungal strain in it and kept at room temperature for 24 hrs for incubation. Five sterilized test tubes of each plant extract were taken. The solution was prepared by adding 4 ml nutrient broth in each test tube. First test tube filled with 4 ml plant extract and shaken well. Then 4 ml solution transferred from first test tube to the second one and subsequently transferred in rest of the tubes for concentrations of 0.4, 0.2, 0.1, 0.05, 0.025 mg/ml. Each tube was then inoculated with 50 µl of fungal strain and incubated at 37˚C for 48 hours. The tubes were examined for visual turbidity. The turbidity was measured in terms of optical density (OD) at 610 nm by spectrophotometer. The MIC values were taken as the lowest concentration that inhibited the visual growth of the *Launaea procumbens*.

RESULT AND DISCUSSION

The methanol extract of *Launaea procumbens* exhibited maximum antifungal activity (14, 13 and 12mm) than the *Griseofulvin* (12, 12 and 12mm) zone of inhibition. Hence, the leaf extract of *Launaea procumbens* showed significant antifungal activity than the standard used, against *Aspergillus niger* (Table 1). MIC value is used to evaluate antimicrobial nature of plant extracts and minimum quantity of antimicrobial compound required to kill or arrest multiplication of all microorganisms present in the medium or body fluid. This result may open important perspectives in alternative antifungal therapies. Percentage growth inhibition was, calculated only with the finding of 4 ml of each concentration, which indicated that the values of optical density were further decreased by reducing

Table 1: Effect of Antifungal Activity of leaf extract of *Launaea procumbens* against *Aspergillus niger*

Sample Name	Zone of Inhibition(mm)			Mean
	Z_1	Z_2	Z_3	
Griseofulvin	12	12	12	12
Launaea procumbens	14	13	12	14.33

Table 2: Minimum inhibitory concentration (MIC) of methanolic leaf extract of Launaea procumbens against *Aspergillus niger*

Plants	Absorbance in different Concentrations(mg/ml)				
	0.4	0.2	0.1	0.05	0.025
Griseofulvin	0.847	0.247	0.154	0.618	1.713
Launaea procumbens	1.533	0.659	0.566	1.208	0.986

visual growth or turbidity of fungal strain, shown in Table 2 and inhibition increases with decrease in concentration of plant extract. The least minimum inhibitory concentration (MIC) was found out of plant extract *Launaea procumbens* with optical density 1.533 in 0.4ml dilution. The plant which was selected for the present study have aromatic compounds and hence they show the antifungal potential. In the present study, the antifungal activity of a new combination was studied by microdilution assay against *A. niger*.

CONCLUSION

The present study revealed that the leaf extract of *Launaea procumbens* from Asteraceae family possess noteworthy antifungal activity and it leads to discover novel antifungal drugs. It is interesting to note that the methanol extract of this species, could be used mainly against the fungi *Aspergillus niger* to control the infectious diseases aspergillosis in an effective manner. In conclusion, the findings of this experiment confirmed that plant extracts can be used as natural fungitoxicant to control the growth of pathogenic fungi (*A. niger*) against infected plants and thus reduce the dependence on the synthetic fungicides.

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