

## ANTIFUNGAL ACTIVITIES OF SOME EXTRACTS OF ALOE VERA

### INTRODUCTION

Plants belonging to the genus *Aloe* have been known for their medicinal value (Ali et al., 1999). Juice of *Aloe vera* is useful in treating wounds from thermal burns and radiation injury. This material is also used in the treatment of dry and moist epidermis, prophylactic action, prevention of kraurosis, dermatitis, eczema, psoriasis, neurodermatitis, herpes, subcutaneous infections (Heggers et al., 1993; Capasso et al., 1998). The application of fresh *Aloe* pith relieves pain, burning and itching and has antiseptic action. *Aloe vera* gel is used in the treatment of seborrhea, acne vulgaris and alopecia (Behl et al., 1993). *Aloe vera* is externally used for cicatrisation and internally as laxative. Hydro alcoholic extract is also part of some make-up products with cicatrisation effect. Several anthraquinones have been isolated from *A. vera* of which the most important are aloin and aloe-emodin (Shelton, 1991). The plant is reported to contain mono and polysaccharides, tannins, sterols, organic acids, enzymes, saponins, vitamins, aminoacids and minerals (Newall et al., 1996).

There are several reports about the antifungal activity of crude extractives of *Aloe vera* (Nebedum et al., 2009; Rosca-Casian et al., 2007; Subramanian et al., 2006; Shamim et al., 2004; Ali et al., 1999; Saks and Barkai-Golan, 1995), but there is very little information about the chemical nature of the active principles, which contribute towards antifungal activity of the plant. The aim of the present study was to have a comparative investigation of the antifungal activity of the extractives obtained by solvents of varying polarity and to understand the chemical nature of the active principles. Thus the activity of different extractives of *Aloe vera* was evaluated against *Colletotrichum gloeosporioides*, *Colletotrichum capsici* and *Fusarium solani* by poisoned food technique (Nene and Thapliyal, 2002; Nidiry and Babu, 2005). The activity of the extractives against *Cladosporium cucumerinum* was evaluated by Thin Layer Chromatographic (TLC) bioautography (Zhao et al., 1998). The activities of two compounds, namely aloin and aloe-emodin present in *Aloe vera* were evaluated against *C. gloeosporioides* and *C. cucumerinum*.

### MATERIALS AND METHODS

**Plant material:** *Aloe vera* L. Burm.f. (Liliaceae) leaves were harvested from the experimental plot of Indian Institute of Horticultural Research, Hessaraghatta, Bangalore, in April 2009, dried at 60°C and were powdered. The powdered plant material was Soxhlet extracted first with hexane, then with ethyl acetate and finally with methanol. The respective extractives were obtained by completely distilling out the solvents on a water bath. *Aloe* drug was obtained by drying the latex at 25°C.

**Tested material:** Hexane extractive, ethyl acetate extractive, methanol extractive of the dried leaves, *Aloe* drug (dried latex), Aloin (Barbaloin; 10-Glucopyranosyl-1,8-dihydroxy-3-[hydroxy-methyl]-9[10H]-anthracenone) from Sigma [Approx. 20%(HPLC)] and Aloe-emodin (1,8-dihydroxy-3-[hydroxy-methyl]anthracenone) from Sigma [Minimum. 95%(HPLC)].

**Used organisms:** *Cladosporium cucumerinum* IMI 249540 obtained from International Mycological Institute, UK, *Colletotrichum gloeosporioides* ITCC 4573 obtained from the Indian Type Culture Collection, Indian Agricultural Research Institute, New Delhi, India, *Colletotrichum capsici* and *Fusarium solani* isolated in Division of Plant Pathology, IIHR and maintained on a potato-dextrose-agar (PDA) medium.

Studied activity: Antifungal activity of Hexane, ethyl acetate, methanol extractives, aloe drug and aloin were studied by observing the mycelial growth inhibition of *Colletotrichum* species and *Fusarium solani* by poisoned food technique, surfactant Tween-80 being added at a level of 0.3% to the media in both the control and the treated samples. Phenol was used as a standard. In the case of aloe-emodin, the experiment was conducted by dissolving the required amount of the compound in 0.25 mL of acetone and incorporating to 30 mL of the medium, the same amount of acetone being added to the control also. The percent mycelial growth inhibition was calculated by the formula: where, C is the mycelial diameter of the control and T is the mycelial diameter of the treated samples (Nene and Thapliyal, 2002; Nidiry and Babu, 2005). Antifungal activity against *C. cucumerinum* was determined by TLC bioautography. In this case, the extractives, aloin and aloe-emodin were spotted on a TLC plate, eluted with ethyl acetate, sprayed with the inoculums of *C. cucumerinum* and observations were taken after an incubation period of 4 days.

## RESULTS AND DISCUSSION

The results show that the methanol extractive of *A. vera* exhibits highest antifungal activity against the mycelial growth of *Colletotrichum* species at both the concentrations. Non-polar extractives showed moderate activity. Aloe drug also showed antifungal activity against *Colletotrichum* species at both the concentrations. At low concentration (0.2%) of extractives, *Fusarium solani* did not show any inhibition. But at higher concentration (0.5%) it showed inhibition. Polar extractives (ethyl acetate and methanol) showed higher activity against the mycelial growth of *F. solani* compared to non-polar extractive (hexane).

Concentrations of the extractives expressed as a percentage of the compounds in PDA (w/v). \*Higher concentrations of aloe-emodin were not tried because of its poor solubility in acetone and water. #Observations were taken after an incubation of 5 days at  $27\pm 2^{\circ}\text{C}$ . \$Observations were taken after an incubation of 8 days at  $27\pm 2^{\circ}\text{C}$ . The average of two replications is shown. NI- No inhibition

Two compounds present in the extractives namely aloin and aloe-emodin showed moderate activity against *C. gloeosporioides*.

Activity of the extractives and the compounds against *C. cucumerinum* was determined qualitatively. Aloe-emodin, which had an  $R_f$  value of about 0.8, showed a very conspicuous inhibition spot. The inhibition spots corresponding to aloe-emodin were present in all extractives and the most conspicuous inhibition spot was observed in the case of ethyl acetate extractive.

The antifungal activity exhibited by the extractives of *Aloe vera* in the present study is consistent with the reports by earlier workers. It may be noted that Rosca-Casian et al. (2007) showed that hydroalcoholic plant extracts obtained from *A. vera* fresh leaves exhibited antifungal activity against the mycelial growth of *Botrytis gladiolorum*, *Fusarium oxysporum* f.sp. *gladioli*, *Heterosporium pruneti* and *Penicillium gladioli*. Saks and Barkai-Golan (1995) reported antifungal activity of the *Aloe vera* gel against *Penicillium digitatum*, *Alternaria alternata*, *Botrytis cinerea* and *Penicillium expansum*. Further, Subramanian et al. (2006), Nebedum et al. (2009) and Rodriguez et al. (2005) also reported antifungal activity of *Aloe vera* extractives against different phytopathogenic fungi. However, earlier investigators did not report any comparison of the efficacies of extractives obtained by different solvents. In the present study, we have reported the efficacies of different extractives obtained by different solvents indicating the chemical nature of the extractives and the active principles.

Present results show that polar extractives exhibit higher antifungal activity than non-polar extractives. The activity of the extractives is higher against *Colletotrichum* species than *Fusarium solani*. The antifungal activity of aloin and aloe-emodin against *C. gloeosporioides* and *C. cucumerinum* shows that they are two active principles, which contribute towards the antifungal property of the crude extractives. Both aloin and aloe-emodin are anthraquinone derivatives and antifungal activity of several anthraquinone derivatives in other plants has been reported (Agarwal et al., 2000; Singh et al., 2006). Further detailed studies may be required to understand the chemical nature of other active principles present in the plant.

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#### Appraisal of antifungal activity of Aloe vera

\*Rukhsana Bajwa, Sobiya Shafique and Shazia Shafique, Department of Mycology and Plant Pathology, University of the Punjab, Quaid-e-Azam Campus, Lahore, Pakistan.

\*E-mail: rukhsanabajwa\_mppl@yahoo.com

#### Abstract

In vitro studies were carried out to evaluate the antifungal activity of Aloe vera shoot extract in aqueous (polar) and organic (non-polar) solvents against few pathogenic species of genus *Alternaria* viz., *A. alternata*, *A. citri* and *A. tenuissima*. The assessments revealed that Aloe vera contained substantial antimicrobial efficacy. The shoot aqueous extracts caused significant inhibition in growth and biomass production of the three tested fungi. In case of n-hexane extraction the inhibitory effect was found to be variable with the applied concentration.

Keywords: Aloe vera, *Alternaria alternata*, *A. citri* and *A. tenuissima*.