Annals of Plant Sciences
ISSN: 2287-688X
OPEN ACCESS
www.annalsofplantsciences.com

Original Research Article

Antifungal activity of essential oil and leaf extract of *Adenocalymma alliaceum* and its role in management of stem end rot disease of mango fruits caused by fungal pathogen *Botryodiplodia theobromae*

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Received: June 20, 2015; **Accepted:** July 11, 2015.

Abstract: Study was carried out for evaluation of antifungal activity of essential oil and extract of plants for control of stem end rot of mango. To investigate antifungal activity leaves of 20 angiospermic taxa were extracted separately in water, acetone, ethyl acetate, ethyl alcohol, methanol and petroleum ether and screened against *Botryodiplodia theobromae* that cause stem end rot of mango fruits. Plant *Adenocalymma alliaceum* leaf extract in water, acetone, ethyl alcohol, petroleum ether, ethyl acetate and methanol was found to exhibit strongest antifungal activity. The essential oil of *A. alliaceum* was also found fungi toxic against other fruit rotting fungi. The volatile fraction of the leaves of *A. alliaceum* was isolated in the form of essential oil and was standardized through physicochemical and fungitoxic properties. The minimum inhibitory concentration (MIC) of the oil was found to be 100ppm. The oil was thermostable in nature and had a long shelf life of 2 years. The oil was found to withstand high inoculum density. An *in vivo* trial on mango fruits (Dashehari) was conducted with aqueous extract and essential oil of *A. alliaceum*. Aqueous extract enhanced the shelf life of mango fruits up to 5 days. Fumigation of the fruits with the oil of *A. alliaceum* enhanced the shelf life up to 8 days by protecting them from fungal rotting.

Key words: Antifungal; Garlic creeper; essential oil; fungicidal; postharvest diseases; stem end rot

Introduction

Mango is the largest sub-tropical fruit crop in India where it occupies an area of 1.23 million hectare with a production of 10.99 million tones (57.18 % of the world production) (Negi, 2000). About 17 to 37% mango is spoiled by mechanical and physical damage, transportation and other post harvest handling practices (Madan and Ullasa, 1993). Biodeterioration during storage caused by microbial activities are equally responsible for the loss. Fungi have been identified as major cause of post harvest loss. On an average 17.7 % mangoes are decayed due to fungal diseases in transit, storage and marketing (Sharma *et al.,* 1994). An important disease that causes spoilage of mango is stem end rot caused by *Botryodiplodia theobromae* Pat. (Alam and Nahar, 1990). This is responsible for 26.7% decay out of the total diseased fruits (Sharma *et al.,* 1994).

Chemical fungicides provide the primary means for controlling post harvest fungal decay of fruits. Continuous use of fungicides has been criticized after awareness in general public regarding contamination of perishables (fruits and vegetables) with fungicidal residues and their consequences as well as about proliferation of resistance in the pathogen population. Recently, the exploitation of natural products to control decay and prolong storage life of fruits has received more and more attention (Tripathi and Dubey, 2004). Biologically active natural products have the potential to replace synthetic fungicides. Exploitation of some natural products, such as flavour compounds (Wilson *et al*., 1987; Utama *et al*., 2002; Stadelbecher and Prasad, 1974), acetic acid (Molys *et al*., 1996; Chu *et al*., 2001), jasmonates (Droby *et al*., 1999), glucosinolates (Mari *et al*., 2003), essential oils (Bishop and Reagon, 1998; Bellerbeck *et al*., 2001; Hidalgo *et al*., 2002; Tripathi *et al*., 2004) and plant extracts (Mohamed *et al*., 1994; Rana *et al*., 1999; Mohapatra *et al*., 2000; Tripathi and Dubey, 2003) are well documented.

Keeping all these points into consideration an attempt has been made to screen some local angiospermic plants as biocontrol agent against the post harvest

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fungal diseases of mango fruits. The *Adenocalymma alliaceum* Mart (Bignoniaceae) commonly called garlic creeper was found to have pronounced activity against *B. theobromae*. The plant is a climber and both the leaves and flowers of this plant have pungent garlic like smell. The extract and essential oil of *A. alliaceum* were selected for detailed antifungal study in the present investigation.

Material and Methods

Fungal cultures

The cultures of fungi *Botryodiplodia theobromae* Pat, *Botrytis cinerea* Pers *ex* Fr., *Ceratocystis paradoxa* (Dade) C. Moreau, *Colletotrichum gloeosporioids* Penz., *Monilinia fructicola* (Wint.) Honey, *Penicillium digitatum* (Pers) Sacc., *P. expansum* Link *ex* S.F Gray, *P. italicum* Wehmer and *Phomopsis citri* Fawe were obtained from Indian Agricultural Research Institute, New Delhi. The Cultures of *Aspergillus niger* Van Tiegh and *Rhizopus stolonifer* (Ehren. *ex* FR.) Lind was isolated from the infected mango fruits in laboratory. All the fungal cultures were maintained on PDA medium. The Czepeks agar medium ($NaNO_3$,3.0g; K_2HPO_4, 1.0 g; $MgSo_4.7H_2O$, 0.5g; KCl,0.5g; $FeSO_47H_2O$, 0.1; Sucrose 30 g; Agar 15 g and distilled water 1L) was used throughout the investigation. Five mg streptopenicillin (a mixture of streptomycin and penicillin) was added to the medium to prevent bacterial contamination.

Aqueous extract preparation and antifungal testing

Leaves of 20 angiospermic taxa viz. *Adenocalymma alliaceum, Adhatoda vasica, Allamanda cathartica, Annona squamosa, Callistemon lanceolatus, Catharanthus roseus, Celosia cristata, Clerodendrum indicum, Jatropha gossipifolia, Justicia betonica, Lawsonia inermis, Leucas aspera, Ocimum canum, Plumeria rubra, Polygonum glabrum, Prunus persica, Rauvolfia serpentina, Solanum nigrum, Tecoma stans* and *Vitex negundo* were collected from the Banaras Hindu University Campus, Varanasi and were screened against *B. theobromae* as well as other fruit rotting fungi. The leaves were washed thoroughly in tap water, blotted dry and kept at room temperature. For the preparation of aqueous extract, leaves were crushed with equal amounts (1:1 W/V) of distilled sterile water in grinder. The resulting homogenate was passed through several folds of muslin cloth. The filtered extract was then used for experiments. Five mg streptopenicillin was added to the extract in order to check bacterial contamination. The extracts were then assayed for the activity against test fungus by modified paper disc technique (Conner and Beachat, 1984).

Extraction in organic solvents and antifungal testing

Ten gram leaves of each plant species were extracted separately in five different organic solvents viz. acetone, ethyl alcohol, ethyl acetate, methanol and petroleum ether by macerating them to pulp in a pestle and mortar. Ten ml of each solvent was added separately to pulp in conical flasks, left overnight at room temperature and filtered through Whatman's filter paper NO.1.The filtrates were assayed separately against the test pathogen by the modified paper disc technique.

Isolation of volatile fungitoxic fraction (Essential oil) from the leaves of A. alliaceum

Since the leaves of *A. alliaceum* were having strong aromatic odor, the volatile fungitoxic fraction of leaves was isolated by hydro distillation through Clevenger's apparatus. The isolated fraction showed two distinct layers-an upper oily layer and a lower aqueous layer. Both the layers were separated and the moisture from the oily layer was removed by adding anhydrous sodium sulphate.

Antifungal testing and Minimum inhibitory concentration (MIC) of the A. alliaceum oil

Fungitoxicity of the extracted oil was tested by the poisoned food technique (Pandey *et al.*, 1982; Perrucci *et al.*, 1994) by using Czepeks agar medium. The concentration of the essential oil was prepared by dissolving the requisite amounts in 0.5ml of 0.1% Tween-80 and then mixing with 9.5 ml of Czepeks agar medium to produce 500ppm, 400ppm, 300ppm, 200ppm, 100ppm concentrations. The control sets were prepared similarly using equal amounts of sterilized distilled water in place of oil.

Nature of toxicity of the extracts and essential oil of A. alliaceum

The nature of toxicity (fungitoxic/ fungicidal) of the extracts (leaves extracted in water, acetone, ethyl acetate, ethyl alcohol, methanol and petroleum ether) and essential oil was tested against the test fungus following Thompson (1989). The inhibited fungal discs of the extracts and oil treated sets were re-inoculated in to fresh medium and revival of their growth was observed.

Range of fungitoxicity of the extracts and oil of A. alliaceum

The range of fungitoxicity of the extracts (in water, acetone, ethyl acetate, ethyl alcohol, methanol and petroleum ether) and essential oil of *A. alliaceum* was tested against 10 most common fruit rotting fungi viz. *Aspergillus niger, Botrytis cinerea*, *Ceratocystis paradoxa*, *Colletotrichum gloeosporioids*, *Monilinia fructicola*, *Penicillium digitatum, P. expensum*, *P. italicum*, *Phomopsis citri* and *Rhizopus stolonifer* by poison food technique.

Fungitoxic properties of the oil and effect of storage on fungitoxicity of leaves and oil of A. alliaceum

The effect of increased inoculum densicity of the test fungus on fungitoxicity of the oil was studied following Moleyar and Pattisapu (1987). The effect of storage on the fungitoxicity of leaves of *A.alliaceum* was studied by keeping 2kg leaves at room temperature (28$\pm$1ºC) in a sterilized paper bag for 30 days. The fungitoxicity of the leaves was tested after regular interval of 2 days. The effect of storage and temperature on the fungitoxicity of the oil was determined at its MIC by poisoned food technique.

Physicochemical properties of the oil

The oil was standardized through GLC and physicochemical properties viz. specific gravity, specific rotation, refractive index, solubility in different organic solvents, acid number, saponification value, ester value, phenolic content and carbonyl content following Chowdhury and Kapoor (2000).

Comparison of the fungitoxicity of the oil of A. alliaceum with some prevalent synthetic fungicides

The efficacy of the oils was compared with some fungicides, viz. benzimidazole (benomyl), diphenylamine, phenylmercuric acetate (ceresan) and zinc dimethyl dithiocarbamete (zirum) by the usual poisoned food technique.

In vivo testing of the aqueous extract of A. alliaceum in control of post harvest stem end rot of mango

Mature healthy fruits of medium size (Dashehari) obtained from local market were used for the experiment. The fresh mango fruits surface disinfested with 0.1% sodium hypochlorite solution and washed in sterile distilled water. The pathogenicity of the fungus was tested as suggested by Garcha and Singh (1980). To prepare spore suspension, spores were harvested from 7 day old culture and suspended in sterile distilled water and a wetting agent .01 %Tween 80. Fruits were wounded on two sides at a depth of 1.5 mm by puncturing them with a cork borer. Each wound site was then inoculated with 40 µl of spore suspension (10^5 spores/ml) of *B. theobromae*. The inoculated fruits were kept in desiccators. The fruits of treatment sets were given dip treatment in aqueous extract of *A. alleaceum* (1:1w/v) for 5 min, 15 min and 30 min. The fruits of control were dipped in water. Six fruits were taken in each control and treatment sets and the experiment was repeated thrice. The initiation of rotting of the fruits was observed. The observation is based on the mean values.

In vivo testing of the oil of A. alliaceum in control of post harvest stem end rot of mango

The *in vivo* efficacy of oil of *A. alliaceum* was tested by fumigating the inoculated fruits. The inoculation was done by the same method as used with extract. In treatment sets the requisite amount of oil were introduced in desiccators by soaking in a piece of cotton so as to give concentration of 100 ppm (V/V).The initiation of rotting of the fruits was observed. Six replicates were kept for treatment and control sets and the experiment was repeated thrice.

Results

Antifungal activity of angiospermic taxa

Out of 20 plant species screened against *B.theobromae*, the leaf extracts of *A. alliaceum* (in water, acetone, ethyl alcohol, ethyl acetate, methanol and petroleum ether) exhibited strongest activity inhibiting mycelial growth of the test fungus completely (100% activity). As the *A. alliaceum* leaves extract in water and in five organic solvents found to have antifungal activity and therefore it was selected for further investigations.

Besides *A. alliaceum* five other plants viz. *Adhatoda vasica* (ethyl acetate), *Allamanda cathartica* (ethyl acetate, methanol), *Lawsonia inermis* (ethyl acetate), *Plumeria rubra* (ethyl acetate) and *Prunus persica* (ethyl acetate) were also found to show antifungal activity against the test pathogen but, their activity was found to be confined to only one or two organic solvents. None of them was found to be active in aqueous extract. The ethyl acetate was found to be most desirable solvent for extraction because in these solvent 6 plants have shown 100% activity. The antifungal activity of these 5 plants was static in nature (Table 1).

Table 1: Antifungal activity test for different angiospermic taxa against B. *theobromae*

Angiospermic Plants	Solvents used					
	Water	Acetone	Ethyl acetate	Ethyl alcohol	Methanol	Petrolium ether
Adenocalymma alliaceum (Mart) (Bignoniaceae)	100	100	100*	100	100*	*100*
Adhatoda vasica Nees (Acanthaceae)	0	5.00	100	12	25	*5*
Allamanda cathartica Linn (Apocynaceae)	10	90	100	95	100	*15*
Annona squamosa Linn(Annonaceae)	0	0	0	0	0	*0*
Callistemon lanceolatus Dc(Myrtaceae)	0	10	60	50	10	*5*
Catharanthus roseus Linn G.Don (Apocynaceae	0	0	0	0	0	*0*
Celosia cristata Linn (Amaranthaceae)	0	0	0	10	0	*0*
Clerodendrum indicum (Linn.) Ktze(Verbenaceae)	0	0	0	0	0	*0*
Jatropha gossipifolia Linn (Euphorbiaceae)	20	25	50	45	55	*15*
Justicia betonica Linn (Acanthaceae)	0	0	0	0	0	*0*
Lawsonia inermis Linn.(Lythraceae)	50	70	100	95	80	*40*
Leucas aspera (Willd.)Spreng.(Labiateae)	0	0	0	0	0	*0*
Ocimum canum Sims (Labiateae)	5	24	50	35	30	*10*
Plumeria rubra Linn.(Apocynaceae)	20	50	100	40	50	*0*
Polygonum glabrum Willd.(Polygonaceae)	0	0	0	0	0	*0*
Prunun persica (L.)Stockes (Rosaceae)	60	30	100	80	90	*50*
Rauvolfia serpentina (Linn.Benth.*exkurz*) (Apocynaceae)	0	0	0	0	0	*0*
Solanum nigrum Linn.(Solanaceae)	0	0	0	0	0	*0*
Tecoma stans (Linn) H.B.ε K (Bignoniaceae)	0	0	0	0	0	*0*
Vitex negundo Linn. (Verbenaceae)	*5*	*0*	*25*	*0*	*0*	*0*

*Cidal nature of extracts

Effect of storage on fungitoxicity of A. alliaceum leaves

The fungitoxicity of leaves persisted for 8 days in aqueous extract while the toxicity in organic solvents was remained up to 25 days in ethyl acetate, 10 days in methanol and alcohol and 5 days in acetone and 2 days in petroleum ether. The characteristic aroma of the leaves was disappeared after 5 days of storage.

Volatile fungitoxic fraction from A. alliaceum leaves

The volatile fungitoxic fraction was obtained in the form of pale yellow colored essential oil with pungent, unpleasant, characteristic aroma. The recovery of the oil from the plant was 0.5 %.

Minimum Inhibitory Concentration of the oil and nature of toxicity

The oil completely inhibited mycelial growth of the test fungus at 100ppm indicating its MIC to be 100 ppm. At 50ppm it inhibited growth of the fungus by 85 % (Table 2).

Nature of toxicity of the extracts and essential oil of A.alliaceum

The toxicity of the extracts was found to be static in water, acetone, petroleum ether and absolute alcohol, showing very weak hyaline mycelial growth on the medium. Whereas, it was cidal in methanol and ethyl acetate extracts (Table 1). The fungitoxicity of the essential oil was found to be cidal in nature at 100ppm and hypertoxic at 200ppm concentrations (Table 2).

Table 2: Fungitoxic properties of *A. alliaceum* essential oil

Inhibitory action		**Fungi toxicity (cidal/static)**		**Inoculums density**		**Thermo stability at one hr**		**Storage effect**	
MIC (ppm)	MGI (%)	MC (ppm)	MGI (%)	Fungal discs	MGI (%)	Temperature (^{0}C)	MGI (%)	Months	MGI (%)
50	85	100	100	2	100	10	100	2	100
100	100	100	100*	4	100	20	100	4	100
200	100			8	100	30	100	6	100
300	100			16	100	40	100	8	100
400	100			32	100	50	100	10	100
500	100			64	100	60	100	12	100
								14	100
								16	100
								20	100
								24	100

MIC-Minimum Inhibitory Concentrations; MC-Minimum Concentration; MGI-Mycelial Growth Inhibition; *- Cidal nature

***Range of fungitoxicity of the extracts and oil of A. alliaceum* against fruit rotting fungi**

The extracts and essential oil of *A. alliaceum* exhibited a broad range of fungitoxicity inhibiting growth of all the 10 most common fruit rotting fungi viz. *Aspergillus niger, Botrytis cinerea*, *Ceratocystis paradoxa*, *Colletotrichum gloeosporioids*, *Monilinia fructicola*, *Penicillium digitatum, P. expensum*, *P. italicum*, *Phomopsis citri* and *Rhizopus stolonifer.*

Fungitoxic properties of the oil

It has been observed that the oils inhibited the fungal growth of the treatment sets containing even 64 discs of the test fungus indicating the potency of the oil to withstand high inoculum density (Table 2). It was found that the oil had long shelf life. The oil remained active up to two years. The oil was thermostable in nature as it remained fungitoxic at different temperatures between 10 and 80 ºC (Table 2).

Physicochemical properties of the oil

The GLC analyses of the essential oil indicated it to be a mixture of 7 major and 15 minor components. The various physichochemical properties viz. specific gravity, specific rotation, refractive index, solubility in different organic solvents, saponification value, ester value, phenolic content and carbonyl percentage of the oil are recorded in Table 3.

Table 3: Physicochemical properties of *A. alliaceum* essential oil

Parameters	**Observations**
Yield of oil	0.5 %
Colour	Pale yellow
Odour	Pungent
Specific gravity at 28 ºC	0.6832
Specific rotation	+23.0º
Refractive index	1.621
Solubility	
Acetone	Soluble (1:1Conc)
Absolute alcohol	Soluble (1:1Conc)
90 % alcohol	Soluble(1:1Conc)
Ethyl acetate	Soluble (1:1Conc)
Benzene	Soluble (1:1Conc)
Chloroform	Soluble (1:1Conc)
Hexane	Soluble (1:1Conc)
Methanol	Soluble (1:1Conc)
Acid number	2.83mg
Saponification value	58.21mg
Ester value	45.35mg
Phenolic content	nil
Carbonyl percentage	2.00

Comparative efficacy of oil

MIC of synthetic fungicides benzimidazole,diphenylamine, phenylmercuric acetate and zincdimethyl dithiocarbamate was found to be 200, 800, 500 and 400ppm respectively which was higher as compared to *A. alliaceum* oil (100ppm).Therefore the oil was more potent than the synthetic ones.

In vivo efficacy of the aqueous extract of A. alliaceum in control of postharvest stem end rot of mango

The fruits of control sets were completely covered with water soaked brown appearances with ash colored blackish mycelial growth after 6 days of storage showing complete rotting of the fruits. The fruits given 5 min dip treatment did not show any enhancement of shelf life and showed similar symptoms of rotting after 6 days. The initiation of rotting in fruits which were given 15 min and 30 min. dip treatments in aqueous extract of *A. alliaceum* leaves was started after 8 days and 11 days respectively. Therefore the respective enhancement of shelf life was up to 2 days and 5 days in each type of dip treatments (Table 4).

Table 4: In *vivo* trial on mango fruits with aqueous extract and essential oil of *A. alliaceum*

Treatment with aqueous Extract (1:1W/V)	Initiation of rotting (in days)	Enhancement (in days)
Control	6	*0*
5min dipping	6	*0*
15min dipping	8	*2*
30 min dipping	11	*5*
Essential oil		
Control	6	*0*
Fumigated at 100ppm	*14*	*8*

In vivo efficacy of the A. alliaceum oil in control of post harvest stem end rot of mango

The oil treated fruits were found to enhance the shelf life as there was no fungal growth on the treated fruits as compared to control fruits. Though, the initiation of rotting was started after 14 days of storage showing 8 days of enhancement of shelf life but the treated fruits peel showed browning coloration (Table 4).

Discussion

The preservative nature of some plant extracts has been known for centuries and there has been renewed interest in the antimicrobial properties of extracts from aromatic plants. Some plants extracted in different solvents have shown inhibitory action against different storage fungi (Gujar and Talwankar, 2012; Sattar et al., 2014: Gawade et al., 2014). In the present study the extracts of *A. alleaceum* in water, acetone, ethyl acetate, ethyl alcohol and methanol were found to exhibit strong toxicity against *B. theobromae*. The activity was static in water, acetone, absolute alcohol and petroleum ether and the revived mycelium showed weak and hyaline mycelium. The extracts in ethyl acetate and methanol were found to be cidal as the reinoculated fungal discs were not revived when transferred to fresh medium. The occurrence of antifungal activity in water and all the organic solvents indicated the strong potency of *A. alleaceum* leaf extract to be exploited as strong botanical fungicide in the management of post harvest fungal pathogen of fruits during storage and transit.

Investigations on the success of essential oils as biodegradable and ecofriendly fungitoxicants have shown the possibilities for their exploitation as natural fungicides (Dixit *et al*. 1995; Tripathi *et al*. 2004). The fungicidal activity of oils obtained from *Ocimum, Thymus, Origanum, Anethum, Eucalyptus, Foeniculum* and *Citrus* against several post harvest pathogens reveals the marked fungicidal activity of carvacrol (in thyme, origanum oil) and p-anisaldehyde (oxidation products of anethol, found in anise oil) (Caccioni and Gizzardi 1994; Gujar and Talwankar, 2012). Certain volatile aromatic components produced by fruits during ripening also showed antifungal activity. Acetaldehyde has been found effective in postharvest protection of apples (Stadelbacher and Prasad 1974) and stone fruits (Caccioni *et al*. 1994). Hexanal and benzaldehyde, produced by stone fruit metabolism, also have a fungistatic/fungicidal activity when utilized in postharvest treatment against *Monolinia laxa* and *Rhizopus stolonifer* (Caccioni *et al*. 1995).

In the present investigation the volatile fungitoxic principle of *A. alliaceum* was obtained in the form of essential oil. The yield was 0.5 %. The oil was yellow in color with strong unpleasant smell. The unpleasant, although characteristic aroma of the plant is due to the presence of a combination of alkenyl sulphide and thiosulfonates, coproducts formed from the akylcystein sulfoxides, precursors in the intact plant material (Zoghbi *et al.,* 1984). Although the leaves of the *A. alliaceum* (garlic bush) quickly produce the pungent, garlic like odor upon handling, the aroma disappears after several days of drying.

The oil was found to be fungicidal in nature and toxic at 100ppm and hypertoxic at 200ppm concentration. The oil was found to withstand high inoculum density as the oil has shown the antifungal activity in the treatment sets containing the 64 fungal discs. This is another potential of the oil to be exploited as botanical fumigant. The oil remained fungitoxic for up to 2 years having long shelf life .The oil was thermostable in nature as it can withstand up to 80ºC temperature without losing toxicity. The GLC analyses showed it to be a mixture of seven major and fifteen minor components indicating that these components were synergistically responsible for the toxicity in the oil. The oil exhibited a wide range of activity at 100 ppm against 10 most common Indian fruit rotting fungi namely *Aspergillus niger, Botrytis cinerea, Ceratocystis paradoxa, Colletotrichum gloeosporioids, Monilinia fructicola, Penicillium digitatum, P. expansum, P. italicum, Phomopsis citri* and *Rhizopus stolonifer*. On comparing the MIC of the oil with some synthetic fungicides the oil was more active than the suynthetic pesticides as the MIC of the oil was found to be low (100ppm) as compared to synthetic fungicides.

Several fungitoxicants of plant origin have been found to be non injurious to the treated commodities and some have shown enhancement in their shelf life. In the present investigation the extracts and essential oils proved good fungitoxic activity with enhancement of shelf life of mangoes during storage. The fruits were given dip treatments in aqueous leaf extract (1:1 w/v).This treatment for 15 min enhanced the shelf life up to 2 days, while the dipping of fruits for 30 min enhanced the shelf life up to 5 days. The oils were used as fumigants at 100ppm (MIC). The fumigated fruits of treatment sets showed enhanced shelf life up to 8 days. The aqueous extracts did not show any adverse symptoms on the fruit peel. Though the essential oils enhanced the shelf life of fruits but the color of the fruits was turned to brown.

Therefore, use of these substances as antimicrobial agents can be an interesting field of investigation as the toxicity to mammals is quite low, and their degree of volatility allows their use for fumigation in cold storage or for active packaging. The extract of *A. alliacea* with strong fungitoxicity, fungistatic (in aqueous, petroleum ether and acetone) and fungicidal (in ethyl acetate and methanol) nature, long shelf life and broad fungitoxic spectrum could be recommended as botanical fungitoxicant .The essential oil with low MIC in comparison to synthetic ones, fungicidal nature against the test fungus as well as against other common fruit rotting fungi, thermostable nature and the efficacy to withstand high inoculum density is an important fungitoxicant of the *A. alliaceum*. It has the potential to be used as botanical fungicidal fumigant.

In vivo trials with the oil showed a negative effect in the form of brown coloration on peel of fruits. The similar kind damage of citrus fruits treated with thyme and cinnamon essential oil was reported by Plaza *et al.,* (2004). Therefore the potential use of essential oils to control postharvest diseases requires a detailed examination of their biological activity and dispersion in fruit tissues, and the development of a formula which inhibits the growth of pathogens at nonphytotoxic concentrations. Investigation on the mode of action and practical applicability of such plant products is still required so as to recommend their formulation in control of postharvest diseases.

Acknowledgements

The author is grateful to the Department of Science and Technology, New Delhi, for providing financial assistance and IARI New Delhi for providing fungal cultures.

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Source of support: Department of Science and Technology, New Delhi, India
Conflict of interest: None Declared