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Efficacy of Some Essential Oils against Post-Harvest Fungal Diseases of Kiwifruits

Habung Yami[1], Pramila Tripathi[2], A.K. Shukla[3@]
[1]Department of Botany, Rajiv Gandhi University, Itanagar – 791112
[2]Department of Botany, DAV-PG College, Kanpur - 208001
[3]Department of Botany, Indira Gandhi National Tribal University, Amarkantak - 484887

@Corresponding author

Abstract

A study on evaluation of essential oils of plant species (*Litsea cubeba, Piper mullesua* and *Pogostemon cablin*) was carried out for their antifungal activity against phytopathogenic fungi viz. *Alternaria alternata, Botrytis cinerea, Fusarium oxysporum* and *Penicillium expansum* following Poisoned food technique method. Different concentration of oil such as 125ppm, 250ppm, 500ppm, 1000ppm and 5000ppm were taken to evaluate the effect. There was 100% inhibition in the growth of phytopathogenic fungi at 5000 and 1000ppm concentration by essential oil of *Litsea cubeba* and *Pogostemon cablin.* The decrease in colony diameter or growth of fungus was corresponding to the concentration of oil. In comparison essential oil of *P. mullesua* was reported less effective against phytopathogenic fungi. Minimum inhibitory concentration for oil of *Litsea cubeba* and *P. cablin* was found 500ppm. At higher concentration essential oil of both species were reported fungicidal in nature. Results depict that *Litsea cubeba* and *P. cablin* essential oil may be used as botanical pesticides to control post-harvest fungal pathogens after further investigations.

Keywords: Essential oil, fungicidal, Phytopathogenic fungi, *Litsea, Piper, Pogonatum.*

Introduction

Kiwifruit [*Actinidia deliciosa* (A. Chev.) C.F. Liang and A.R. Ferguson var. *deliciosa* Hayward] is a climacteric and susceptible fruit to fungal decays in postharvest stage. Many fungi are associated with post harvest fruit rots of kiwifruit (Pennycook 1985; Hawthorne *et. al*., 1982). Botrytis gray mold rot caused by *Botrytis cinerea* is the most important and can directly invade the fruit or enter through wounds. Other fungal pathogens *Phomopsis mali*, Botryosphaeria *dothidea* and *Diaporthe actinidiae* have also been reported to cause post harvest fruit rots of kiwifruit (Koh *et. al.,* 2003). Post harvest and storage pathogens of kiwifruit most often have been controlled by use of synthetic fungicides (Eckert and Ogawa 1988). In the recent years, pesticide residues on horticultural products, especially

the fungicides used in postharvest stage, are of major concerns to the horticultural industry. Also, the increase in consumer's awareness about hazards of pesticide residues on fresh products and their demand to non-residue products, human health, and environmental pollution are considered.

Essential oils or volatile oils are very complex mixture of compounds whose constituents of the oils are mainly monoterpenes and sesquiterpenes. Generally, the action of essential oils is the result of the combined effect of both their active and inactive compounds (Tripathi and Shukla, 2007). Plants contain a wide range of bioactive secondary metabolites which include alkaloids, flavonoids, tannins, saponins, phenols, phlobatannins quinones, lecitins, polyphenols, glycosides, terpenoids, polypeptides and steroids (Edeoga *et al*., 2005; Enyiukwu and Awurum, 2013). Biologically active essential oils represent a rich potential source of an alternative and perhaps environmentally more acceptable disease management compounds (Tripathi and Shukla, 2009). It has long been recognized that some essential oils have antimicrobial properties (Boyle 1955). With a broad range of natural fungicidal plant volatiles, numerous opportunities exist to explore their usefulness in controlling post-harvest diseases. Present study was aim to evaluate for antifungal activity of essential oils against phytopathogenic fungi of kiwifruits.

Materials and Methods

Isolation and identification of phytopathogenic fungi

Isolation of post harvest pathogens of kiwifruits were carried out from infected fruits on rose Bengal agar and potato dextrose agar medium (Johnson and Curl, 1972). Infected kiwifruits were randomly collected from market. Fruits were surface sterilized by 4% sodium hypochlorite and then by 75% alcohol and finally with sterilized distilled water. Small pieces of fruit were cut and placed in the petriplates containing sterilized medium and incubated at 27°C for 7-10 days. Identification of fungal pathogens was done on the basis of morphological, cultural and microscopic characteristics as detailed in available literature (Barnett and Hunter 1972, Domsch *et al*., 1980). In process of culture the isolated fungal pathogens were cultivated on Potato Dextrose Agar (PDA) medium and Peptone Dextrose Rose Bengal Agar medium were used. Potato dextrose agar medium (39 gm of Hi–PDA medium dissolved in 1000 ml of distilled water) was used throughout the investigation. The medium was autoclaved and cooled to 40°C ±2°C. Thirty milligram of streptomycin was added to it and mixed thoroughly so as to prevent bacterial contamination. Similarly Peptone Dextrose Rose Bengal Agar (31.55 gm of Hi-RBA medium dissolved in 1000 ml of distilled water) medium was prepared to maintain the fungal culture.

Plant material collection and essential oils extraction

Plants were collected from different parts of Arunachal Pradesh during the study period. Identification of plants was done by the plant taxonomist in the Department of Botany, Rajiv Gandhi University, Itanagar as well as by the Scientist from Regional Centre of Botanical Survey of India at

Itanagar. Herbarium was preserved and voucher specimens were deposited in the department. Extraction of essential oils was carried out from some locally available larger number of angiospermic taxa namely *Acorus calamus*, *Artimesia nilogerica, Erigeron canadensis, Eupatorium odoratum, Litsea cubeba, Mesua ferrae, Mikania cordata, Piper mullesua and Pogostemon cablin* etc. Subsequently on getting results potent 3 plants were taken for detailed study. An amount of 250 gm of fresh leaves of each plant were cut separately into small pieces and were thoroughly washed with sterilized water. The volatile fractions were isolated by hydro distillation through Clevenger's apparatus. Leaves of the plants were used for extraction of essential oils. The isolated fractions of plant parts exhibited two distinct layers an upper oily layer and the lower aqueous layer. Both the layers were separated and the essential oils were stored in clean glass vials after removing water traces with the help of capillary tubes and anhydrous sodium sulphate (Guenther, 1972).

Antifungal activity assay

Fungitoxic activities of the essential oils were tested by the poisoned food technique of Grover and Moore (1962) and Perrucci *et al.,* (1994). Potato dextrose agar medium (39 gm of Hi–PDA medium dissolved in 1000 ml of distilled water) was used throughout the investigation. The medium was autoclaved and cooled to 40°C ±2°C. Thirty milligram of streptomycin was added to it and mixed thoroughly so as to prevent bacterial contamination. A requisite amount of the oil was dissolved separately in 0.5ml of 0.01 percent of aqueous solution of Tween –80 in presterilized Petri plates (7cm. diam.). While using Tween-80 as solvent care was taken in designing the experiments to evaluate the true effect of essential oils on the pathogenic fungi. PDA medium (9.5 ml) was pipetted to each Petri plate and was mixed so as to obtain the requisite concentrations viz. 5000ppm, 1000ppm, 500ppm, 250ppm and 125ppm. For control sets, requisite amount of sterilized water in place of the oil was added to the medium. Discs of test fungi (5 mm diam) were cut with the help of sterilized cork borer from the periphery of a seven day old culture and were inoculated aseptically to the center of each Petriplate of treatment and control sets. The petriplate were incubated at 27± 1°C for six days in incubation chamber. Measurement of colony diameters of the test fungus in treatment and control sets were done in mutually perpendicular directions and were recorded in terms of percent mycelial inhibition using the following formula

$$\text{Percentage of mycelial inhibition} = \frac{dc-dt}{dc} \times 100$$

Where dc =mean colony diameter of control sets
dt = mean colony diameter of treatment sets

Standardization of essential oils through fungitoxic properties

The standardization of essential oils was done through fungitoxic properties viz. minimum inhibitory concentration and nature of toxicity (Thompson, 1989).

Minimum inhibitory concentration (MIC)

To find out the minimum inhibitory concentration at which the oil showed absolute fungitoxicity (complete inhibition of growth of test fungi), experiments were carried out by the usual poisoned food technique. Different concentrations of the oils were prepared by dissolving separately their requisite amount in 0.5 ml of 0.01 per cent of aqueous solution of Tween-80 and then mixing with 9.5 ml potato dextrose agar medium. The medium of control sets contained requisite amount of sterilized water was added in 0.5 ml Tween-80 in place of oils. As usual the prepared plates were inoculated upside down aseptically with the assay disc of the test fungi to the center of petriplate of treatment and control sets. The petriplates were incubated at 27± 1° C for six days in BOD incubator. Diameters of fungal colony of treatment and control sets were measured in mutually perpendicular directions on the seventh day and percentage inhibition calculated.

Nature of toxicity

Nature of toxicity (funtgistatic / fungicidal) of essential oils against the fungi was determined as suggested by Thompson (1989). Requisite amount of the oil was dissolved separately in 0.5 ml of 0.01 per cent of aqueous solution of Tween-80 and mixed with 9.5 ml potato dextrose agar medium to get final concentrations. Sterilized water was used in control sets in place of the oils. The plates were inoculated upside down aseptically with fungal disc (5mm diam.) taken from the periphery of a seven day old culture of the test fungi and were incubated for six days at 27± 1°C. On seventh day the inhibited discs were taken out from the plates, washed with sterilized water and reinoculated aseptically to plates containing fresh potato dextrose agar medium. The revival of the growth of the fungal discs was observed and the per cent inhibition of growth of the test fungi were calculated on the seventh day with respect to control sets.

Results

Evaluation of Essential Oils against Different Fungi

Essential oils extracted from different plant species were evaluated to visualize their effect on the growth of four phytopathogenic fungi viz. *Alternaria alternata, Botrytis cinerea, Fusarium oxysporum* and *Penicillium expansum* following Poisoned food technique method. Different concentration of oil such as 125ppm, 250ppm, 500ppm, 1000ppm and 5000ppm were taken to evaluate the effect. Simultaneously, a control was also maintained by inoculating culture disc on the medium without adding any oil.

Litsea cubeba

Essential oil of *Litsea cubeba* inhibited the growth of all phytopathogenic fungi at higher concentrations. The decrease in colony diameter or growth of fungus was corresponding to the concentration of oil. On *F. oxysporum* effect of oil was also found to be severe and 100% inhibition was observed at 5000ppm concentration and at 1000ppm even on the 15th day of observation. In case of *B. cinerea*, effect of *L. cubeba* essential oil was quite remarkable, at 5000 and 1000ppm

concentration 100% inhibition was recorded even after 15 days of incubation. Essential oil of *L. cubeba* shows drastic effect on the growth of fungus colony at higher concentration 5000 and 1000ppm. At lower level of concentration *i,e* 125, 250, and 500ppm of oil colony growth was recorded for all the four phytopathogenic fungi but it always remain lesser than the control. The effect of essential oil was corresponding to their concentration.

Table1: Effect of *Litsea cubeba* essential oil on the phytopathogenic fungi.

Period	***Alternaria alternata***					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5th	0.00±0.00	0.00±0.00	0.80±0.00	1.10±0.60	2.20±0.10	4.50±0.10
7th	0.00±0.00	0.00±0.00	0.80±0.00	2.90±0.50	2.65±0.15	5.90±0.00
9th	0.00±0.00	0.00±0.00	1.20±0.10	3.05±0.45	3.65±0.55	6.50±0.10
11th	0.00±0.00	0.00±0.00	1.50±0.10	3.05±0.45	3.65±0.55	6.80±0.10
13th	0.00±0.00	0.00±0.00	1.90±0.05	3.05±0.45	3.65±0.55	6.80±0.10
15th	0.00±0.00	0.00±0.00	1.90±0.05	3.05±0.45	3.65±0.55	7.00±0.10
	Botrytis cinerea					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5th	0.00±0.00	0.00±0.00	2.25±0.25	3.35±0.15	4.25±0.15	4.50±0.10
7th	0.00±0.00	0.00±0.00	4.90±0.30	5.75±0.25	5.05±0.95	5.90±0.00
9th	0.00±0.00	0.00±0.00	5.60±0.40	5.90±0.10	5.25±1.05	6.50±0.10
11th	0.00±0.00	0.00±0.00	6.10±0.10	6.65±0.35	7.00±0.00	6.80±0.10
13th	0.00±0.00	0.00±0.00	6.20±0.20	6.75±0.25	7.00±0.00	6.80±0.10
15th	0.00±0.00	0.00±0.00	6.20±0.20	6.75±0.25	7.00±0.00	7.00±0.10
	Fusarium oxysporum					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5th	0.00±0.00	0.00±0.00	0.00±0.00	2.45±0.05	3.30±0.10	4.00±0.00
7th	0.00±0.00	0.00±0.00	1.40±0.00	4.20±1.00	5.00±1.00	4.50±0.10
9th	0.00±0.00	0.00±0.00	1.60±0.00	4.75±1.25	5.00±1.00	4.70±0.10
11th	0.00±0.00	0.00±0.00	1.90±0.10	5.25±0.75	5.00±1.00	5.40±0.10
13th	0.00±0.00	0.00±0.00	2.00±0.00	5.85±0.15	5.05±0.95	6.30±0.10
15th	0.00±0.00	0.00±0.00	2.20±0.10	6.00±0.00	5.20±1.00	7.00±0.10
	Penicillium expensum					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5th	0.00±0.00	0.00±0.00	1.45±0.25	2.25±0.15	1.95±0.35	4.00±0.00
7th	0.00±0.00	0.00±0.00	2.35±0.25	2.5±0.40	2.55±0.85	4.50±0.10
9th	0.00±0.00	0.00±0.00	2.80±0.60	3.35±0.15	2.55±0.85	4.70±0.10
11th	0.00±0.00	0.00±0.00	3.15±0.75	3.65±0.45	2.55±0.85	5.40±0.10
13th	0.00±0.00	0.00±0.00	3.50±0.90	4.00±0.70	2.60±0.80	6.30±0.10
15th	0.00±0.00	0.00±0.00	4.05±0.55	4.05±0.75	2.70±0.80	7.00±0.10

Piper mullesua

Essential oil of *P. mullesua* also inhibits the growth of four phytopathogenic fungi. In case of all four fungi i.e. *P.expansum*, *F. oxysporum*, *B.cinerea* and *A. alternata* the effect of oil was quite similar. At initial days of growth there was remarkable inhibition. In case of *P. expansum* percent inhibition ranges between 11 and 60 while in case of *F. oxysporum* it was 28.5 to 71%. Growth of *A. alternata* was inhibited 38 to 62% at different concentration of oil. Effect of oil on growth of *B. cinerea* was similar to *A. alternata*. In general growth of tested fungi was noticed at all concentration of oil treatment. None of the phytopathogenic fungi was inhibited cent percent.The effect of essential oil was corresponding to their concentration.

Table2: Effect of *Piper mullesua* essential oil on the phytopathogenic fungi

Period	***Alternaria alternata***					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5th	1.35±0.05	1.30±0.00	1.40±0.00	1.40±0.00	1.55±0.05	4.50±0.10
7th	1.85±0.05	1.80±0.00	1.90±0.00	1.90±0.00	2.20±0.30	5.90±0.00
9th	2.35±0.05	2.00±0.20	2.40±0.00	2.40±0.00	2.75±0.25	6.50±0.10
11th	2.85±0.05	2.30±0.50	2.90±0.10	2.90±0.10	3.10±0.20	6.80±0.10
13th	3.10±0.10	2.50±0.70	3.20±0.10	3.20±0.10	3.40±0.10	6.80±0.10
15th	3.60±0.20	3.15±0.75	3.75±0.05	3.80±0.10	3.80±0.10	7.00±0.10
	Botrytis cinerea					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5th	1.65±0.15	2.25±0.35	2.35±0.25	3.75±0.05	3.55±0.55	4.50±0.10
7th	1.95±0.40	2.85±0.65	2.50±0.10	4.25±0.05	3.85±0.85	5.90±0.00
9th	2.30±0.50	3.20±1.00	2.65±0.15	4.45±0.15	3.95±0.85	6.50±0.10
11th	2.30±0.50	3.75±1.45	2.65±0.15	4.45±0.15	3.95±0.85	6.80±0.10
13th	2.30±0.50	4.00±1.60	2.65±0.15	4.45±0.15	3.95±0.85	6.80±0.10
15th	2.30±0.50	4.00±1.60	2.65±0.15	4.45±0.15	3.95±0.85	7.00±0.10
	Fusarium oxysporum					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5th	1.15±0.05	1.45±0.05	1.40±0.10	1.75±0.05	2.20±0.20	4.00±0.00
7th	1.45±0.05	1.75±0.05	1.75±0.05	2.30±0.20	2.75±0.05	4.50±0.10
9th	1.75±0.05	2.20±0.20	2.35±0.25	2.95±0.05	3.30±0.10	4.70±0.10
11th	2.15±0.20	2.65±0.15	2.75±0.15	3.45±0.05	3.80±0.10	5.40±0.10
13th	2.50±0.10	3.20±0.10	3.40±0.10	3.85±0.05	4.50±0.10	6.30±0.10
15th	3.20±0.20	3.70±0.10	3.90±0.10	4.30±0.10	4.70±0.10	7.00±0.10
	Penicillium expensum					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5th	1.45±0.05	1.90±0.10	2.50±0.30	2.95±0.05	3.55±0.05	4.50±0.10
7th	1.95±0.35	2.50±0.30	2.70±0.30	3.05±0.55	4.25±0.05	5.90±0.00
9th	2.30±0.50	2.85±0.65	2.75±0.25	3.85±0.65	4.45±0.15	6.50±0.10

11th	2.30±0.50	3.05±0.75	3.15±0.35	3.85±0.65	4.55±0.05	6.80±0.10
13th	2.40±0.40	3.30±0.90	3.15±0.35	3.85±0.65	4.55±0.05	6.80±0.10
15th	2.40±0.40	3.30±0.90	3.15±0.50	4.00±0.80	4.55±0.05	7.00±0.10

Pogostemon cablin

Essential oil of *P. cablin* was also found to inhibit the growth of all four phytopathogenic fungi. In case of *P. expansum* and *A. alternata* at 5000 and 1000ppm concentration inhibition in growth was 100%. At 500ppm also the growth was negligible. In case of *F. oxysporum* and *B. cinerea* 100% growth was inhibited at 5000ppm concentration. The decrease in colony diameter or growth of fungus was corresponding to the concentration of oil. At lower concentration of oil i.e. 125, 250 and 500ppm colony growth of fungus always remain lesser than the control.

Table3: Effect of *Pogostemon cablin* essential oil on the phytopathogenic fungi

Period	***Alternaria alternata***					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5^{th}	0.00±0.00	0.00±0.00	0.55±0.05	0.70±0.00	0.80±0.00	4.50±0.10
7^{th}	0.00±0.00	0.00±0.00	0.85±0.05	0.80±0.10	1.25±0.05	5.90±0.00
9^{th}	0.00±0.00	0.00±0.00	0.85±0.05	1.00±0.00	1.55±0.05	6.50±0.10
11^{th}	0.00±0.00	0.00±0.00	1.10±0.10	1.40±0.10	1.65±0.05	6.80±0.10
13^{th}	0.00±0.00	0.00±0.00	1.10±0.10	1.40±0.10	1.90±0.10	6.80±0.10
15^{th}	0.00±0.00	0.00±0.00	1.25±0.05	1.90±0.10	2.10±0.10	7.00±0.10
	Botrytis cinerea					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5^{th}	0.00±0.00	0.50±0.00	1.30±0.10	1.65±0.15	2.40±0.00	4.50±0.10
7^{th}	0.00±0.00	1.80±0.00	2.05±0.25	2.70±0.20	3.90±0.00	5.90±0.00
9^{th}	0.00±0.00	2.90±0.70	3.10±1.30	5.15±0.15	4.30±0.10	6.50±0.10
11^{th}	0.00±0.00	3.10±0.90	3.45±1.65	5.45±0.45	4.30±0.10	6.80±0.10
13^{th}	0.00±0.00	3.20±1.00	3.85±2.05	5.55±0.55	4.35±0.15	6.80±0.10
15^{th}	0.00±0.00	3.20±1.00	4.05±2.25	5.55±0.55	4.35±0.15	7.00±0.10
	Fusarium oxysporum					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5^{th}	0.00±0.00	1.45±0.05	1.40±0.00	1.30±0.00	2.45±0.05	4.00±0.00
7^{th}	0.00±0.00	2.05±0.05	2.15±0.05	2.95±0.05	3.50±0.30	4.50±0.10
9^{th}	0.00±0.00	3.15±0.15	3.20±0.50	4.55±0.55	4.60±1.40	4.70±0.10
11^{th}	0.00±0.00	3.70±0.20	3.55±0.85	4.70±0.70	4.85±1.65	5.40±0.10
13^{th}	0.00±0.00	4.20±0.20	3.95±1.25	4.85±0.85	5.00±1.80	6.30±0.10
15^{th}	0.00±0.00	4.55±0.15	4.25±1.55	4.85±0.85	5.00±1.80	7.00±0.10

	Penicillium expensum					
Days	**5000ppm**	**1000ppm**	**500ppm**	**250ppm**	**125ppm**	**Control**
5th	0.00±0.00	0.00±0.00	0.45±0.05	0.50±0.00	0.50±0.00	4.00±0.00
7th	0.00±0.00	0.00±0.00	0.75±0.05	0.80±0.10	0.95±0.05	4.50±0.10
9th	0.00±0.00	0.00±0.00	0.75±0.05	1.00±0.00	1.30±0.10	4.70±0.10
11th	0.00±0.00	0.00±0.00	0.75±0.05	1.00±0.00	1.40±0.20	5.40±0.10
13th	0.00±0.00	0.00±0.00	0.75±0.05	1.00±0.00	1.55±0.35	6.30±0.10
15th	0.00±0.00	0.00±0.00	0.75±0.05	1.00±0.00	1.70±0.50	7.00±0.10

Minimum Inhibitory concentration (MIC)

Inhibitory evaluation of essential oils against phytopathogenic fungi showed the effective results. Essential oil of *L. cubeba* and *Pogostemon cablin* were found fungicidal at 5000ppm and 1000ppm concentrations and minimum inhibitory concentration was reported 500ppm. The essential oil of *Piper mullesua* was inhibitory at higher concentration for all the phytopathogenic fungi.

Table 4. Minimum inhibitory concentration of essential oils against pathogenic fungi

MIC of oils against fungi				
Essential oils of plants				
			F. oxysporum	***P. expansum***
Litsea cubeba	500ppm	500ppm	500ppm	500ppm
Piper mullesua	Higher Conc.	Higher Conc.	Higher Conc.	Higher Conc.
Pogostemon cablin	500ppm	500ppm	500ppm	500ppm

Nature of toxicity

L. cubeba and *P. cablin* essential oils were found fungi toxic for all the phytopathogenic fungi. At higher concentration both the oils were fungicidal. But at lower concentrations oils were fungistatic. While, essential oil of *P. mullesua* was reported fungistatic in nature.

Table 5. Toxicity nature of Essential oils on phytopathogenic fungi

Essential oils	***A. alternata***	***B. cinerea***	***F. oxysporum***	***P. expansum***
Litsea cubeba	Fungicidal at 1000ppm	Fungicidal at 1000ppm	Fungicidal at 1000ppm	Fungicidal at 1000ppm
Piper mullesua	Fungistatic	Fungistatic	Fungistatic	Fungistatic
Pogostemon cablin	Fungicidal at 1000ppm	Fungicidal at 5000ppm	Fungicidal at 5000ppm	Fungicidal at 1000ppm

Discussion

The essential oil of L. cubeba was found to inhibit the growth of all the phytopathogenig fungi at 5000 and 1000ppm concentrations. Si *et. al*., (2012) reported 59 compounds from *L. cubeba* oil out of which dominant components were monoterpenes (94.4–98.4%), represented mainly by neral and geranial (78.7–87.4%), and D-Limonene was in lesser constituent (0.7–5.3%). Several components were only detected in certain regions and compounds such as o-cymene and eremophilene have never before been reported in EOLC. Su *et. al*., (2012) identified main components in oil as β-caryophyllene (13.0%), τ-cadinol (11.1%), α-cadinol (8.6%), α-humulene (7.5%), α-pinene (7.0%), globulol (6.6%), and β-eudesmol (6.1%). The anti-wood-decay fungal activity of the oil showed that the oil was inhibitory for wood-decay-fungi species and compounds were determined to be τ-cadinol, α-cadinol, and β-eudesmol. Yang (2010) through preliminary bioassay study showed *L. cubeba* oil has good fungicidal activities against *Sclerotinia sclerotiorum, Thanatephorus cucumeris, Pseudocer-cospora musae* and *Colletotrichum gloeosporioides* at the concentration of 588 and 272 μM, and the essential oil has good fungicidal activities against *T. cucumeris* and *S. sclerotiorum*, with IC_{50} values of 115.58 and 151.25 μg/mL, repectively.

Essential oil of *Piper mullesua* also inhibits the growth of four phytopathogenic fungi. In case of all four fungi i.e. *P.expansum, F. oxysporum, B.cinerea* and *A. alternata* the effect of oil was quite similar. In case of *P. expansum* percent inhibition ranges between 11 and 60 while in case of *F.oxysporum* it was 28.5 to 71%. Growth of *A. alternata* was inhibited 38 to 62% at different concentration of oil. Myristicin, asarinin, sesamin and fargesin were found in *Piper mullesua* (Srivastava *et. al*., 2000).

Essential oil of *Pogostemon cablin* was also found to inhibit the growth of all four phytopathogenic fungi. In case of *P. expansum* and *A. alternata* at 5000 and 1000ppm concentration inhibition in growth was 1000%. At 500ppm also the growth was negligible. *Pogostemon cablin* essential oil consist about 70 compounds and out of those Patchoulol, pogostol, seychellene, nor-patchoulinol, patchoulipyridine, methylchavicol, limonene, pinine, p-methoxy cinnamaldehyde, Germacrene and Norpatchoulenol are the main chemical constituents (Chakrapani *et. al.,* 2013, Karimi, 2014). Kocevski *et. al*. (2013) reported antifungal activity of *Pogostemon cablin* essential oil against *A. flavus* at 1500ppm concentration. The Pogostemon cablin (Patchouli) is the most prominent member of family Lamiaceae which has been used against the common cold and as an antifungal agent in traditional medicine (Wu *et. al*., 2004).

Essential oils are mainly conjugated to phenolic compounds which accumulate in some plant cells, and have positive effects on pathogen control (Plotto, *et. al*., 2003). It is known that oxidation products of phlorsidzin (an *o*-dihydroxyphenolic compound) inhibit fungal growth, and are thought to inhibit growth of the apple scab fungus, *Venturia inaequalis* (Asghari Marjanlo, *et. al.,* 2009). Reports indicated that essential oils containing carvacrol, eugenol and thymol (phenolic compounds) had the highest antibacterial performances (Kim *et. al.,* 1995). Thus, our findings revealed that exogenous

essential oils may have a positive role in preservation of kiwi fruit and reduce decay. In conclusion it is stated that *Litsea cubeba* and *P. cablin* essential oil may be used as botanical pesticides to control post-harvest fungal pathogens after further investigations.

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