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A Remedy for Biodeterioration: *Zingiber officinale* Roscoe. Oil

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Abstract

In our study, bioactivity of essential oil extracted from Ginger (*Zingiber officinale* Roscoe.) that belongs to family Zingiberaceae was analyzed. Ginger rhizome oil was found to be effective against a range of bio-deteriogens. The Minimum Inhibitory Concentration (MIC) value was determined by Poisoned Food Technique and Volatile Activity Assay. The study demonstrates that essential oil had a wide spectrum of antifungal activities against common biodeteriogens *Aspergillus niger* van Tiegh., *Penicillium chrysogenum* Thom., *Alternaria alternata* (Fr.) Keissl and *Fusarium roseum* Link. Minimum inhibitory concentration (MIC) of oil was found to be effective, showing results at 1.0 µL/cm^{-3}of oil in respect to all fungi. At higher concentration of 2.0 µL/cm^{-3} of oil was fungicidal in action. 2.0 µL/cm^{-3} was proved appropriate concentration for commercial trial of ginger oil as preservative for stored pulses and nuts which were used as test commodities.

Keywords: *Zingiber officinale*, Essential oil, GC-MS Analysis, MIC.

INTRODUCTION

Deterioration by fungi always has been a major problem of stored food commodities and control of these fungal biodeteriogens in the public health sector continues to place heavy reliance upon the use of chemicals. Plants are a very rich source of bioactive natural products. They are a vast cornucopia of defense chemicals, comprising repellents, feeding deterrents, growth inhibitors, sterilants, toxicants and anti-microbial agents. In addition, the volatile substances obtained from higher plants have proved their usefulness in controlling biodeterioration and therefore are considered to have a bright future (Sharma, 1998).

Zingiber officinale Roscoe. (Family Zingiberaceae) is well known in Asia. The ginger has been listed in Generally Recognized as Safe (GRAS) document of USFDA and has antimicrobial and antimycotoxigenic effects (Tatsadjieu *et al.,* 2009) because of its aroma and taste has been used for culinary purposes from ages. It has also been reported in use for medicinal purposes for more than 2500 years. Ginger rhizome is widely used as an ingredient in food, pharmaceutical, cosmetic and other industries. Ginger is also known to possess antioxidant properties. Ginger contains a unique flavour derived from both nonvolatile and volatile oils.

Ginger is found to be rich in bioactive compounds and various researches have been carried out to explore the beneficial properties of ginger and its extracts (Grzanna *et al*., 2005). Essential oils have been largely employed for their properties already observed in nature, i.e. for their antibacterial, antifungal and insecticidal activities.Essential oils of ginger are of interest because of its richness in various functional compounds mostly terpene, monoterpene and sesquiterpenes, which gives the oil its biological activity (Daferera *et al*., 2002).

Mycotoxins are well known for their hazardous effects. The primary disease associated with mycotoxins intake is hepatocellular carcinoma (HCC, or liver cancer). This disease is the third-leading cause of cancer death globally (INCHEM, 2012) with about 550,000–600,000 new cases each year. Mycotoxins occurring the food commodities are secondary metabolites of filamentous fungi, which contaminate many types of food crops throughout the food chain (Reddy *et al.,* 2010). Mycotoxins are toxic substances produced mostly as secondary metabolites by fungi that grow on seeds and feed in the field, or in storage. The major mycotoxin producing fungi are species of *Aspergillus*, *Fusarium* and *Penicillium*.

Management of fungal contamination of harvested seeds /grains is based on physical (aeration, cooling and rapid drying) and chemical treatment with ammonia, food preservative even with pesticides (Atanda *et al*., 2007). Therefore, the use of herbal pesticide has become indispensible tool against the various synthetic pesticides causing health problems, environmental pollution, pathogen resistance to chemicals and consequent pest resurgence. Although different synthetic antimicrobials have been successfully commercialized in recent years they encounter major problem not only due to adverse side effects on consumers but also for the development of resistance by micro-organisms (Tolouee *et al*., 2010).

An alternative way to resolve these problems utilization of regenerative resources such as plants and their products for newer strategies of pest pathogen management is the demand of this century. Plants are very rich source of bioactive organic chemicals. Essential oils are natural products extracted from plants and fruits, which have been illustrated to be inhibitory, against a wide range of food spoiling microbes, depending upon their concentration, method of testing and active constituents present (Fisher and Phillips, 2008). The main advantage of essential oils was that they could be used in any food and are considered generally recognized as safe.

In the present commutation was conduct to analyze composition of essential oil and their antifungal activity against fungi causing storage rot and also study of effect of MIC of *Zingiber officinale* oil against mycelial growth of *Aspergillus niger* van Tiegh., *Penicillium chrysogenum* Thom, *Alternaria alternata* (Fr.) Keissler and *Fusarium roseum* Link were studied.

MATERIALS AND METHODS

A survey was done for isolation of storage pathogens from different grain shops and storage godowns at Lucknow (India). Pulses and nuts were selected as stored commodities. Ginger rhizomes were purchased from a local market in Lucknow (India). Ginger rhizomes were cleaned with running tap water to remove soil and dust then washed with 2% aqueous sodium hypochlorite solution and rinsed.

Extraction of ginger oil

The mature and healthy ginger rhizomes (5 kg) were purchased from the local market of Lucknow, India. They were properly washed thinly grated and hydro-distilled for 6-7 hrs. in a Clevenger apparatus. The oils were dried over anhydrous sodium sulphate. To isolate the oil from the aqueous portion, the oil was extracted with solvent ether in a separating funnel. The ether was removed at reduced pressure which resulted in an oily residue that was added to the oil collected earlier. The essential oil obtained was kept in sealed glass tube at 4˚C until analysis.

GC-MS analysis

Gas chromatographic analysis followed by mass spectra was carried out in Perkin Elmer Autosystem XL Packed mode. Column used for analysis was OV-1, 100% Methyl gum (10 feet). The conditions were as follows; Temperature programming from 4°C-220°C, hold at 75°C for 20 minute. Injection temperature 250°C and detector temperature was 255°C. Carrier gas was N2 at a flow rate 14 ml/min.

The identification of individual compound is based on their retention time's relatives to those of authentic samples and matching spectral beaks availably with NIST mass spectral libraries.

Determination of minimum inhibitory concentration (MIC)

Antifungal activity was tested against *Aspergillus niger*, *Penicillium chrysogenum*, *Alternaria alternate* and *Fusarium roseum* by volatile activity assay and poisoned food technique.

Poisoned food technique

The fungi-toxicity of the oil was evaluated against the test fungi separately by the poisoned food technique of Grover and Moore (1962). PDA (20ml) was poured into sterilized petridishes and measured amounts of oil were added. The assay plates were rinsed carefully to ensure even distribution of the oil in the medium. For control sets, the medium was supplemented with the same amount of distilled water instead of oil. After the medium solidified, inocula of the test fungi were placed in the centre of each assay plate, which were incubated at 28±2°C. The growth of the test fungi were recorded after 30 days and percent inhibition was computed after comparison with the control.

Volatile activity assay

Tests for volatile activity were carried out in 90 mm petriplates containing 20 ml of solidified potato dextrose plate. 5 mm diameter disc of inoculum of the test species cut from the periphery of an actively growing culture on PDA plates was placed on the agar in each petriplate and then petriplates were kept in inverted position. In the lid of each petriplate a sterilized cotton swab was placed on to which a different concentration of oil was poured. The petriplates were sealed by parafilm to check the release of volatile oil. For each corresponding control an equal amount of water was poured on sterilized cotton swab. The petriplate were kept at 28±2 ˚C for 30 days. Fungitoxicity was expressed in terms of radial growth.

Efficacy of ginger oil during storage

The effect of storage on the toxicity of the oil was determined by storing a stock of the oil in an air tight glass vials at room temperature. The fungal toxicity of the oil taken from the stock

at regular six months interval was tested at the MIC of respective fungi by the volatile assay and observations on mycelial growth were recorded.

Efficacy of ginger oil treatment against the biodeteriogens of pulses and nuts

The storage grains as pulses and nuts were obtained from the market and brought to the laboratory. The efficacy of *Z. officinale* oil as a preservative of pulses and nuts against fungal spoilage was determined as follows: 1kg of the commodities was placed separately in presterilized plastic containers of 2000cc capacity. Different amounts of ginger oil were soaked separately in sterilized cotton swabs as to obtain final concentrations of 0.2 µL/cm^{-3} and 2µL/cm^{-3} with respect to the volume of the containers. One swab of each concentration was placed in sterilized perforated polythene bag which was introduced into each plastic container for each concentration was prepared. A control set was run parallel to each treatment set uniform unsoaked sterile cotton swabs. All the sets were stored at room temperature ranging between 20-40 ˚C and relative humidity between 57 to 87% for a period of six months. Thereafter, fungal infestation of the stored commodities of both treatments and controls was determined by the protocols of Seed health testing (Vishunavat, 2007).

Statistical analysis

Experiments were carried out in triplicates. Data were expressed as means of three replicates. Statistical analysis was performed with Microsoft excel 2007. Difference on statistical analysis of data were considered Significant at $p \leq 0.05$. ANOVA was made based on the diameter of the radial growth among fungal isolates.

RESULTS

We selected 10 survey sites (shops and storage godown), four major species group *Alternaria alternate*, *Aspergillus niger*, *Fusarium roseum*, *Penicillium chrysogenum*, and (Table 1) were responsible for deterioration of pulses and nuts, showed notable loss in economic value due to mycotoxin.

Phytochemistry of ginger oil

The main monoterpenene ($C_{10}H_{16}$) compound was camphene which was 23.9% followed by sesquiterpene ($C_{15}H_{24}$) zingiberene 12.2%, whereas 1, 8- cineole reported as major component (27.9%) which was an oxygenated ether compound. Other hydrocarbons were α- pinene 7.2%, α-farnesene 1.1%, α- seliene 0.9%, ar- curcumene 0.8%, camphor 0.4%, tricyclene 0.1%. The main oxygenated compounds were geranial 2.8, nerolidol 1%, neral 0.9%, elemol 0.9 %, zingiberenol 0.9%, octanel 0.1%.(Table 2 and Fig. 1).

Bioactivity of ginger oil against growth of test fungi

The data obtained from the *In vitro* experiment which was done by poisoned food technique and assay of volatile activity indicated that essential oil had displayed a significant bioactivity on different tested fungus (Table 3 and Table 4). It was observed that increasing concentrations of ginger oil have suppressed the fungal growth. Minimum inhibitory concentration (MIC) by volatile activity assay was 1 µL cm^{-3} and 2 µL cm^{-3} by poisoned food technique.

Pulses and nuts were selected due to its high nutritional value and economic importance among all stored commodities. Starting concentration ranges of oil were 0.2 µL cm^{-3}, 0.4 µL cm^{-3}, 0.6 µL cm^{-3}, 0.8 µL cm^{-3} , 1 µL cm^{-3} and 2 µL cm^{-3} concentration, although 0. 6 µL cm^{-3} has displayed notable reduction in radial growth during *In vitro*, therefore this was the start up concentration for in vivo trials. Our *In vitro* tests were proved base for in vivo trials which was done on small commercial level.

When the fungal population from untreated commodities and treated commodities at 0.2

µL cm^{-3}, 0.5 µL cm^{-3}, 1 µL cm^{-3} and 2 µL cm^{-3} of oil respectively were compared (CFU/g) after six months of storage, in treated pulses and nuts a notable decrease in CFU was recorded (Fig. 2). At 2 µL cm^{-3}, no fungal flora was observed. This volatile antimicrobial capability of oil has been maintained continuously till six months (Fig. 3)

In statistical analysis, null hypothesis is not accepted that there is significant difference between the treatment of oil concentration on fungal growth by poisoned food technique and vapour toxicity method. Therefore, we found that values were not significantly different at the level of 5% significance.

As the null hypothesis is rejected, for further interpretation on the difference between different treatment means Duncan's multiple range test was performed.

DISCUSSION

As this study was designated to find out the possibility of utilizing volatile constituents of *Z. officinale* as remedy for pulses and nuts against fungal deterioration, *Alternaria alternate, Aspergillus niger, Fusarium roseum*, and *Penicillium chrysogenum*, were selected as the test organisms since they were found to be the most common bio-deteriogens during the survey of selected sites. A perusal of Table 1 shows the percentage loss by various saprophytic as well as parasitic fungi were found associated with pulses and nuts examined.

In present study, monoterpenes hydrocarbons ($C_{10}H_{16}$) were responsible for the fungicidal activity of ginger oil. Among which, 1,8-cineole account for 27.9% which was the major component followed by zingiberene 12.2%. In literature, many variations have been found in chemical composition of ginger oil. Agrawal *et al.,* (2001) reported curcumene as the major constituent in the fresh ginger rhizomes, while Menut *et al*., (1994) identified citral as the main constituent of ginger oil.

In other previous report on ginger oil (Singh *et al.*, 2005), α-zingiberene was found to be the major constituent. These differences in the chemical composition of the oil from the same plant/plant part could be due to the environmental, developmental, genetic or some other factors. Yield and composition of oil differ widely with the production conditions (Blair *et al*., 2001), variety, cultivars or population (Galambosi and Peura, 1996) and on climatic and soil factors.

Here, we are reporting 1, 8- cineole as major component responsible for antimycotic activity. The marked antimicrobial activity of essential oils from spices and herbs is believed due to its Phenolic compounds (Singh *et al*., 2008). From GC-MS studies it is clear that the ginger essential oil contain considerable amounts of compounds (1, 8- cineole, camphene and zingiberene) which might be responsible for the observed antimicrobial potency. This cytotoxic property is of great importance in the applications of essential oils not only against certain human or animal pathogens or parasites but also for the preservation of agricultural or marine products. Ginger essential oil is indeed effective against several mycotoxin producing fungi of stored commodities. It appears that the fungicidal/ fungitoxic nature of the oil is due to monoterpenes camphene and zingiberene. However, it is likely that the overall efficiency of essential oil result from the synergistic action of all constituents. The lowest concentration of an antimicrobial agent that will inhibit the visible growth of a microorganism is known as the MIC. The determination of the MIC of oil is necessary for prescribing its appropriate dose. Clearly, unnecessarily high doses of oil increase wastage and may cause considerable harm to the quality of the commodity treated.

In our *In vivo* experiment it is clear that ginger oil has shown fungistatic nature at 1µL cm^{-3}, but at higher dose it becomes fungicidal (2µL cm^{-3}). There is also a variation was observed in MIC of oil that was 1µL cm^{-3} in volatile activity assay whereas 2µL cm^{-3} in case of poisoned food technique. Such variation may be due to the use of different test fungi or different techniques adopted. However, in the present work the MIC of ginger oil was effective against all test fungi viz. *Alternaria alternate, Aspergillus niger, Fusarium roseum*, and *Penicillium chrysogenum.*

A fungitoxicant may act as a fungistat or a fungicide inhibiting the growth of fungus temporarily or permanently respectively (Sharma and Tripathi, 2007). A fungicide should be able to retain its activity over a long period of shelf life. There was a notable decrease in fungal CFU of treated and untreated commodities. After six months, no fungal flora was observed at 2 µL cm^{-3}, and volatile antimicrobial capability of oil has been maintained.

Litrature Cited

Agrawal, M., Walia, S., Dhingra, S. and Khambay, B.P.S. 2001. Insect growth inhibition, and composition in commercially available dill cultivars in comparison to caraway. Ind. Crop. Prod. 14: 229–239.

Atanda, O.O., Akpan, I. and Oluwafemi, F. 2007. The potential of some spice essential oils in the control of *A. Parasiticus* Cfr 223 and aflatoxin production. Food Control, 18: 601-607.

Bakkali, F., Averbeck, S., Averbeck, D., and Idaomar, M. 2008. Biological effects of essential oils. A review. Food. Chem. Toxicol. 46: 446–475.

Blair, J., Aichinger, T., Hackal, G., Hueber, K., and Dachler, M. 2001. Essential oil content antifeedant and antifungal activity of compounds isolated/derived from *Zingiber officinale* Roscoe (Ginger) rhizomes. Pest Manag. Sci., 57: 289–300.

Daferera D. J., Tarantilis P.A. and Polissiou, M.G. 2002. Characterization of essential oil from Lamiaceae species by fourier transform raman spectroscopy. J. Agr. Food Chem. 50: 5503-5507.

Fisher, K. and Phillips, C. 2008. Potential antimicrobial uses of essential oils in food: is *Citrus* the answer? Trends Food Sci.Technol. 19: 156-164.

Galambosi, B. and Peura, P. 1996. Agrobotanical features and oil content of wild and cultivated forms of caraway (*Carum carvi* L). J. Essent. Oil Res. 8: 389–397.

Grover, R.K. and Moore, J.D. 1962. Toxicometric studies of fungicides against brown rot organisms *Sclerotinia fructicola* and S. laxa. Phytopathology. 52: 876-880.

Grzanna R., Lindmark L. and Frondoza C.G. 2005. Ginger an herbal medicinal product with antinflammatory action. J. Med. Food, 8: 125-132.

Inchem, I. 2012. Principles of evaluating chemical effects on the aged population: International programme on chemical safety environmental health criteria 144, World Health Organization, Geneva, (1993). Http://www.Inchem.Org/Documents/Ehc/Ehc/ Ehc144. Htm (Accessed On 19th June 2012) (1993).

Logrieco, A., Bottalico, A., Mul´E, G., Moretti, A. and Perrone, G. 2003. Epidemiology of toxigenic fungi and their associated mycotoxins for some mediterranean crops. Eur. J. Plant Pathol. 109: 645–667.

Menut, C., Lanaty, G., Bessiere, J.M., Kowdav, J. 1994. J. Essent. Oil Res. 6: 101–108.

Mishra, D. 1990. Seed protectant property of essential oil of *Zingiber officinale* Roscoe. Ind. Perfum. 34: 266–268.

Pandey, D.K., Tripathi, N.N., Tripathi R.D. and Dixit, S.N. 1982 . Fungitoxic and phytotoxic properties of the essential oil and of *Hyptis suaveolens* (L) Poit. Z. Pflakrankh. Pflaschutz., 89 (6): 344 - 349.

Reddy, K.R.N., Salleh, B., Saad, B., Abbas, H. K., Abel, C.A. and Sheir, W.T. 2010. An overview of mycotoxin contamination in Foods and its implications for human health. Toxin Review. 29: 3-26.

Sharma, N., Verma, U.K. and Tripathi, A. 2007. Bioactivity of essential oil from *Hyptis squveoleans* against storage mycoflora. In: Controlled Atmosphere & Fumigation in Stored Products. Proc. International Conference, 2004 August 8-13; Gold-Coast Australia. FTIC Ltd. Publishing, Israel. pp: 99-116.

Singh, G., Kapoor, I.P.S., Singh, P., de Heluani S. C., de Lampasona, P. M. and Catalan, A. N. C. 2008. Chemistry, antioxidant and antimicrobial investigations on essential oil and oleorasins of

Zingiber officinale. Food. Chem. Toxicol. 46: 3295-3302.
Singh, G., Marimuthu, P., Heluani, C.S. and Catalan, C. 2005, Antimicrobial and antioxidant potentials of essential oil and acetone extract of *Myristica fragrans* houtt (aril part). J. Food Sci. 70: 2.
Singh, G., Marimuthu, P., Murali, H.S., and Bawa, A.S. 2005 Antioxidative and antimicrobial potentials of essential oils and extracts isolated from various spice Materials. J. Food Safety. 25: 130–145.
Singh, G., Maurya, S., Catalan, C. and Lampasona, M.P. 2005 Studies on essential oils, part 42: chemical, antifungal, antimicrobial and sprout suppressant studies on ginger essential oil and its oleoresin. Flavour Frag. J. 20: 1–6.
Tatsadjieu, N.L. Dongmo, P.M.J.,Ngssoum, M.B., Etoa, F.X., and Mbofung, C.M.F. 2009. Investigation on the essential oil of *Lippia rugosa* from cameroon for its potential use as antifungal agent against *Aspergillus flavus* Link E.X. Fries. Food Control. 20: 161-166.
Tolouee, M., Alinezhad, S., Saberi, R., Eslamifar, A., Zad, S. J., Jaimand, K., Taeb, J., Rezaee, M. B., Kawachi, M., Shams-Ghahfarokhi, M. and Razzaghi-Abyaneh, M. 2010. Effect of *Matricaria chamomilla* L. flower essential oil on the growth and ultrastructure of *Aspergillus niger* van Tieghem. Int. J. Food Microbiol. 139: 127-133.
Vishunavat, K. 2007. Seed health testing –Principles and Protocols.

Tables

Table 1. Economic loss of pulses and nuts by common biodeteriogens.

S. No.	Fungi isolated	Pulses (%)	Nuts (%)
1.	*Alternaria alternata* (Fr.) Keissler	4.06±0.12	4.3±0.05
2.	*Aspergillus fumigatus Fres.*	0.8±0.05	0.76±0.03
3.	*A. flavus Link Fr.*	2.7±0.08	1.2±0.03
4.	*A. ochraceous Wilhelm*	0.8±0.1	1.2±0.05
5.	*A. nidulans ukawa*	0.9±0.06	0.8±0.05
6.	*A. terreus Thom*	0.7±0.03	0.63±0.12
7.	*A. niger van Tiegh.*	4.9±0.1	6.5±0.17
8.	*Cladosporium cladosporoides*	0.5±0.05	0.6±0.03
9.	*C. herbarum (fresen.)*	1.5±0.14	0.6±0.03
10.	*Fusarium roseum Link*	2.3±0.06	2.43±0.02
11.	*Penicillium expansum (Link) Thom*	0.8±0.06	2.9±0.06
12.	*P. citrinum Thom*	1.2±0.03	0.69±0.05
13.	*P. oxalicum Currie&thom*	1.4±0.13	0.43±0.08
14.	*P. chrysogenum Thom*	4.7±0.06	5.2±0.03
15.	*P. funiculosum Thom.*	0.43±0.08	0.69±0.07
16.	*Rhizopus stolonifer (Ehrenberg:Fr.) Vuillemin*	0.4±0.11	0.79±0.05
18.	*R. nigricans Ehrenb.*	0.5±0.02	0.86±0.03
19.	*Syncephalastrum racemosum(Cohn)*	0.2±0.05	0.76±0.03
20.	*Tricothecium roseum(Person)Link ex*	0.3±0.03	0.23±0.13
21.	*Yeast like fungi*	0.7±0.06	0.76±0.03

The values are mean of ten survey spot, corresponding standard errors are represented as ± value.

Table 2. GC-MS study of ginger oil (*Zingiber officinale*).

S. No.	Retention time	Area	Compound	Percentage
1.	4.580	0.09	0.1	0.1
2.	7.748	0.01	0.1	0.1
3.	19.241	0.09	0.4	0.4
4.	21.237	7.96	7.2	7.2
5.	22.087	31.40	23.9	23.9
6.	23.699	3.94	2.8	2.8
7.	24.836	0.06	0.9	0.9
8.	26.271	47.47	27.9	27.9
9.	29.844	0.10	0.05	0.05
10.	29.844	0.09	0.8	0.8
11.	33.661	0.25	1.1	1.1
12.	34.830	0.07	0.9	0.9
13.	36.707	5.81	12.2	12.2
14.	38.027	0.46	0.9	0.9
15.	48.229	0.16	0.9	0.9
16.	48.920	0.10	1.0	1.0

The values are mean of ten survey spot, corresponding standard errors are represented as ± value.

Table 3. Antifungal activity of ginger oil against fungi by poison food technique.

Conc. ($\mu L/cm^{-3}$)	Pathogens (radial diameter in mm)			
	A.niger	*P. chrysogenum*	*A. alternata*	*F. roseum*
Control	85^{b} ±1.0	66.67^{a} ±0.88	83^{a} ±2.0	85^{c} ±0.57
0.2	77.66^{e} ±1.66	55.33^{bc} ±1.76	72^{c} ±2.50	82^{d}±1.154
0.4	62.33^{a}±1.45	40.66^{b}±0.88	62.33^{b}±0.88	63.67^{d}±0.88
0.6	46^{a}±0.577	31.33^{c}±0.88	42.66^{d}±0.88	52.67^{c}±1.20
0.8	24.33^{a}±2.33	21^{a}±1.15	31^{b}±1.52	31.67^{d}±1.45
1.0	11.66^{b}±0.88	$11a^{b}$±1.0	12.66^{c}±0.33	14.67^{c}±1.76
2.0	-	-	-	-

*-, no growth found the in medium.

One way ANOVA was made, based on the diameter of the radial growth among fungal isolates. Data were found significantly different at level of 5% significance.
Values in the same column followed by different superscript's letters are significantly different (P<0.05) according to Duncan's Multiple Range Test. The values followed by same superscript letters are not statistically significant. Each value is mean of three replicates and ±SE are given along the mean values.

Table 4. Antifungal activity of ginger oil against fungi by vapour toxicity method.

Conc. ($\mu L/cm^{-3}$)	Pathogens (radial diameter in mm)			
	A.niger	*P. chrysogenum*	*A. alternata*	*F. roseum*
Control	85^{b}±2.34	63.8^{b}±1.65	84.6^{c}±1.66	86^{c}±0.40
0.2	70.2^{ab} ±2.13	53.4^{d}±1.44	63^{b} ±2.16	83.2^{a}±0.75
0.4	46.6^{d} ±4.52	32.2^{b} ±2.52	42.2^{a} ±0.85	54.4^{a} ±0.65
0.6	23.6^{c}± 2.53	13^{b}±1.73	22.6^{d}±1.61	31.2^{e}±0.75
0.8	12.6^{e}±2.10	7.6^{a}±0.65	11^{b}±0.91	10.4^{d}±0.65
1.0	-	-	-	-
2.0	-	-	-	-

*-, no growth found the in medium.

One way ANOVA was made, based on the diameter of the radial growth among fungal isolates. Data were found significantly different at level of 5% significance.
Values in the same column followed by different superscript's letters are significantly different (P<0.05) according to Duncan's Multiple Range Test. The values followed by same superscript letters are not statistically significant. Each value is mean of three replicates and ±SE are given along the mean values.

Figures

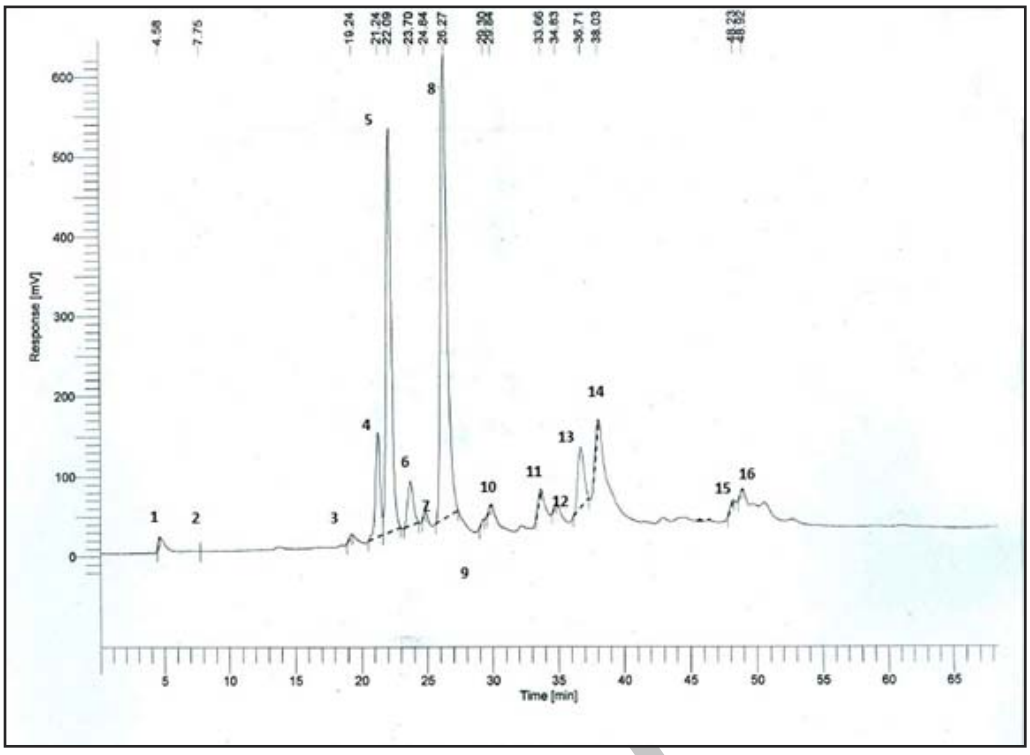


Fig. 1. Gas chromatogram of ginger essential oil.

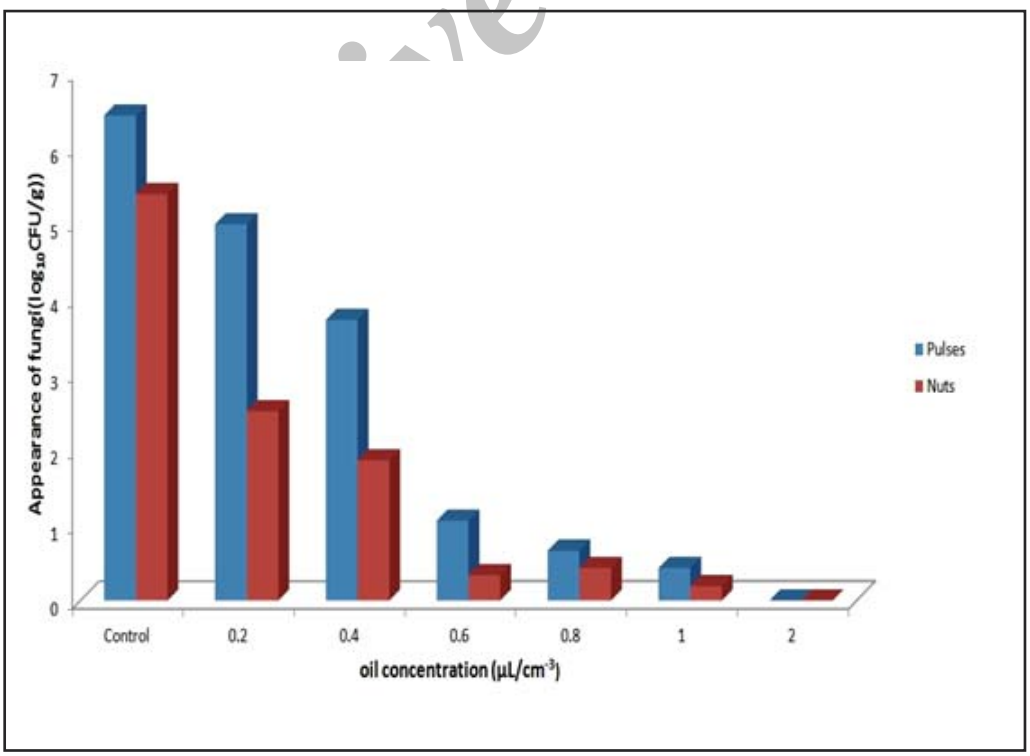
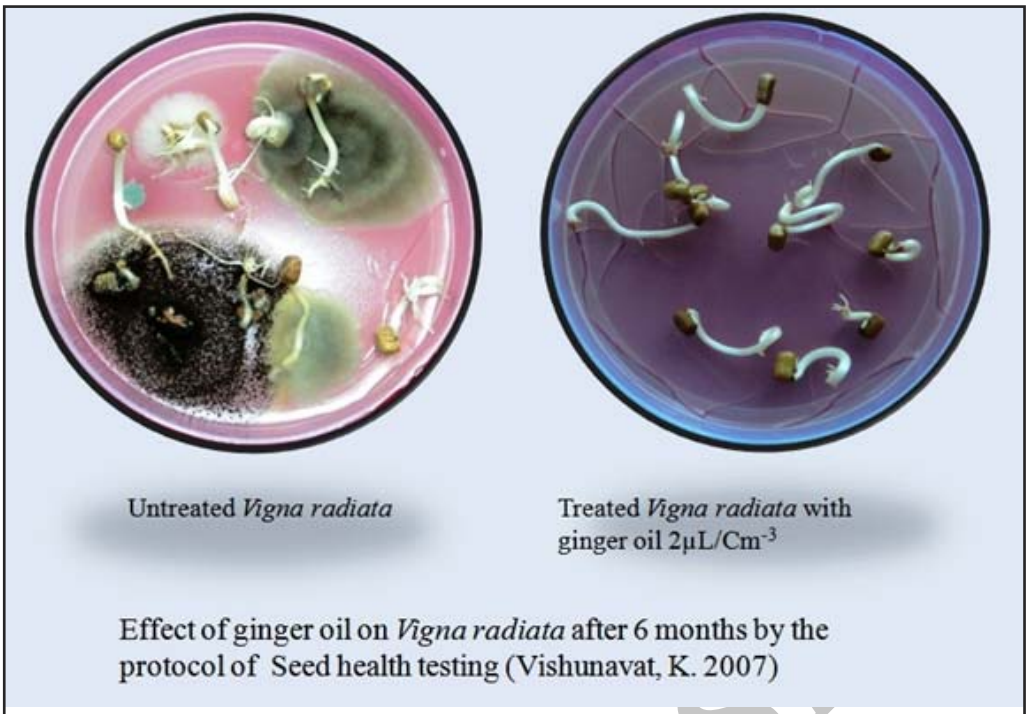


Fig. 2. Fungi isolated from treated and untreated commodities after duration of storage.



Vishunavat, K. 2007. Seed health testing –Principles and Protocols

Fig. 3. Volatile activity of *Zingiber officinale* oil after six month.