
Mentha and Citrus oil against Post Harvest Deterioration of Wheat during Storage

Upadhyay, R. S.¹, Mishra, A. K.² and Singh, R.^{1*}

¹Bio-Prospects Research Lab, Department of Botany, K.N. GOVT. P.G. College, Gyanpur Bhadohi - 221304 Uttar Pradesh, India; ²Regional Higher Education Officer, Gorakhpur, Uttar Pradesh, India.

Upadhyay, R. S., Mishra, A. K. and Singh, R. (2018). Mentha and citrus oil against post harvest deterioration of wheat during storage. International Journal of Agricultural Technology 14(4):613-620.

Abstract Severe deterioration of wheat seeds were taken by different mycotoxin producing fungi as well as insects during its storage and for control of such post harvest losses and several synthetic chemicals are commonly used. But the use of many synthetic chemicals has been cautioned due to their residual toxicity, carcinogenicity, teratogenicity and other adverse effects on different biological system of animals and human beings after consumption. In this attempts were made to study the efficacy of Mentha and Citrus oil against post harvest management of stored wheat. Out of 08 fungi isolated from stored wheat sample, *Aspergillus flavus* was found most dominant, and followed by *Aspergillus niger* and *Aspergillus fumigatus*. These mycoflora were treated with different concentration of Mentha and Citrus oils. 500 and 1000 µl/ml of Mentha and citrus oil respectively, which found effective against them. The effect of these two oils were also affected to two insects *Sitophilus oryzae* and *Tribolium castaneum* which commonly associated with stored wheat samples. It was found that at 500 µl/ml concentration of Mentha oil and at 1000 µl/ml concentration of Citrus oil, infestation of stored wheat samples with insects was completely checked. Seeds of wheat exhibited significantly increasing in percent germination after storage that was provided with fumigation by both the oils, which inoculated by fungi.

Keywords: Essential oils, *Aspergillus flavus*, *Sitophilus oryzae*, *Tribolium castaneum*, wheat

Introduction

Wheat (*Triticum aestivum* L; Hindi-Gehu or Gehoon) is an important cereal crop of India belonging to family poaceae. It is cultivated in China, India, USA, Australia, Canada, France, Argentina and Germany. Globally, wheat is the leading source of vegetable protein in human food having higher protein content than either maize or rice, the other major cereals. (Kumar *et al.*, 2011; Cauvain, 2003). Wheat is grown in India over an area of about 266.92 lakh hectares with a production of 721.40 Lakh tonnes. Paradoxically, the developing countries, in which from 50 to 80 percent of the population is engaged in agriculture, have the lowest agricultural output (Kumar and Kalita,

* **Corresponding Author:** Singh, R.; **E-mail:** rashmiknpg@gmail.com

2017). Their people are living on substandard diet and they have the highest population growth rates (2.64%).

Severe invasion of post harvest commodities take place whenever environmental conditions become favorable for the proliferation of microorganisms. Almost all insect pests of stored grains have a remarkably high rate of multiplication and within a few months they may destroy 10-15% of the grains and contaminate the rest with undesirable odours and flavours. Of the many stored grain insects, the most predominant and widely distributed pests are red flour beetle (*Tribolium castaneum* H), rice weevil (*Sitophilus oryzae* L.) and pulse beetle (*Callosobruchus maculatus* F.) (Srivastava and Subramnian, 2016).

It is a common observation that invasion of insects is almost always accompanied by fungi. Fungi represent one of the main causes of post harvest losses. In addition to causing rot, they can also contaminate food with highly toxic chemicals known as mycotoxins. It is estimated that each year 30-40% of the world wide food production is contaminated by mycotoxins with consequent adverse effects on population, health and economy (Tzatzarakis *et al.*, 2000).

Attempts to control post harvest disease have been carried out by different physical and chemical treatments. Physical treatments are capital intensive and the requisite manpower is lacking or inadequate in most developing tropical countries. Moreover, physical treatment has been used with limitations. While chemical pesticides have played an important role in increasing the food production in India, their indiscriminate use has led to several environmental problems including development of resistance in insects to insecticides, resurgence of non-target pests, pesticide residues in food, fodder and feed, destruction of beneficial insects like honeybees, pollinators parasites and predators. Persistent residues of DDT and BHC have been detected in foodgrains, vegetables, fruits, oil, butter, fish, meat, milk and milk products and even in mother's breast milk. (Dhaliwal and Singh, 2000).

Plants are very rich sources of bioactive chemicals such as phenolics, polyphenols, quinones, flavones, flavonoids, flavonols, tannins, cumarins, terpenoids, lectins and polypeptides. Some plants yield fraction of essential oils, which have inhibitory effects on microorganisms. They are highly enriched with terpenoids. They are volatile, biodegradable, eco-friendly and are easily available in local environment (Shukla, 2013).

Keeping these views in mind the present study has been undertaken to find out the potentiality of some volatile plant products (Mentha oil and Citrus oils) as ecofriendly, natural pesticides against fungi (*Aspergillus flavus* and *Aspergillus niger*) and insects (*Sitophilus oryzae* and *Tribolium castaneum*)

causing heavy deterioration of wheat during storage (Azam *et al.*, 1994, Shukla, 2013).

Material and Methods

Fumigation of stored wheat samples by the oils for management of deterioration caused by fungi

To find out the practical applicability of Mentha oil and Citrus oils as fumigant for protection of food commodities from biodeterioration, experiments were designed to fumigate the wheat samples separately with the oils by the method adopted by; Shaaya *et al.* (1997); Pandey (2003); Singh (2004) and Yadav (2007). The Fumigation of wheat seeds samples with the oils was done in different sets. Requisite amount of the Mentha and Citrus oils were introduced separately in the closed plastic containers (Volume ½ liter) containing 250 gm of wheat seeds samples (moisture content $13 \pm 1\%$) by soaking in a cotton piece which were already placed in perforated polythene tube, so as to procure concentration of 250, 500 & 1000 $\mu\text{l/ml}$ for Mentha and Citrus oils respectively (v/v). Each container was made air tight. In another set the wheat sample stored as such in plastic container without any fumigation referred as control set.

In the agar plate technique, wheat samples were plated in pre-sterilized petriplates containing Potato dextrose agar medium and incubated at $25 \pm 1^\circ\text{C}$. Each plate contained ten seeds and in all 3 replicates (30 seeds) were kept for each set. Fungal colonies appearing on the seeds were isolated regularly after two days until the complete mycoflora had been isolated and transferred to Potato dextrose agar slants. The isolates were then examined and identified (Founder, 1968; Moore and Jaclow, 1979).

After the analysing of fungi associated with wheat samples of treatment and control sets, the percent occurrence of each fungus were observed by following formula (Agrawal *et al.*, 1980) with some modification.

$$\text{Percent Occurrence of fungus} = \frac{\text{No. of colonies of the fungus in the respective set}}{\text{Total No. of colonies of all the fungal species in control set}} \times 100$$

Fumigation of stored wheat samples by the oils for management of deterioration caused by Insects

The efficacy of mentha and citrus oils to protect wheat by *Sitophilus oryzae* and *Tribolium castaneum* was investigated. The wheat samples were fumigated by the usual Fumigation methods of Pandey (2003). Different lots,

each containing 250 gm of wheat samples (moisture content $13 \pm 1\%$) were taken separately in closed plastic containers (volume $\frac{1}{2}$ liter). The insects viz. *Sitophilus oryzae* and *Tribolium castaneum* in wheat (20 adults of each insects) were introduced. Required concentration of oils were separately introduced in the containers by soaking in cotton pieces which were already placed in perforated polythene tubes so as to procure concentration of 250 & 500 for Mentha and 500 & 1000 for Citrus oils (v/v). Each container was made air tight with the help of their lid. In Control sets contained wheat samples infested by respective insects (without any treatment with the oils). After 4 months of storage at laboratory conditions (Temp. 9°C to 43°C and RH 30 to 90%) the analysis of insects associated with wheat samples of treated and control sets were made.

Fumigation of wheat samples for phytotoxic activity of test oils on germination during storage period of 04 months

Phytotoxicity of test oils on wheat was tested with respect to germination after fumigation for a storage period of 04 months. The experiments were designed separately for both fungi and insects in the same way as described in previous experiment except the uninoculated treatment in case of fungi.

Results

Result indicated that the total 08 fungi appeared on wheat samples in uninoculated control and *Aspergillus flavus* was found to be the dominant fungus followed by *A. niger* and *A. fumigatus*. From the observation, it was noted that 500 and 1000 $\mu\text{l/ml}$ concentration of Mentha and Citrus oil respectively, which was found effective against *A. flavus*, *A. niger*, *A. fumigatus* and all the other mycoflora (Figure 1).

It was shown that in control set severe infestation of *Sitophilus oryzae* and *Tribolium castaneum* which were observed from wheat samples. The presence of these insects was completely checked in the wheat samples fumigated with Mentha and Citrus oils at 500 and 1000 $\mu\text{l/ml}$ concentration, respectively (Table 1).

The results indicated that seeds of wheat exhibited significant increase in percent germination after storage that provided with fumigation by both the oils with inoculated by fungi as well as insects. This is strongly indicative of relative non-phytotoxic nature of these oils when used as fumigants in storage for management of fungi and insects (Figure 2).

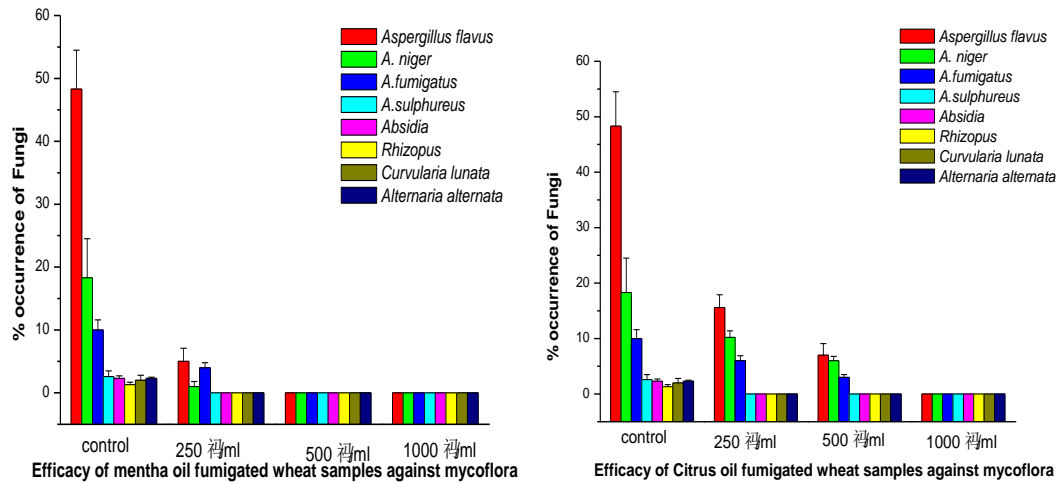


Figure 1. Mycoflora analysis of Mentha and Citrus oils fumigated wheat samples after 4 months of storage

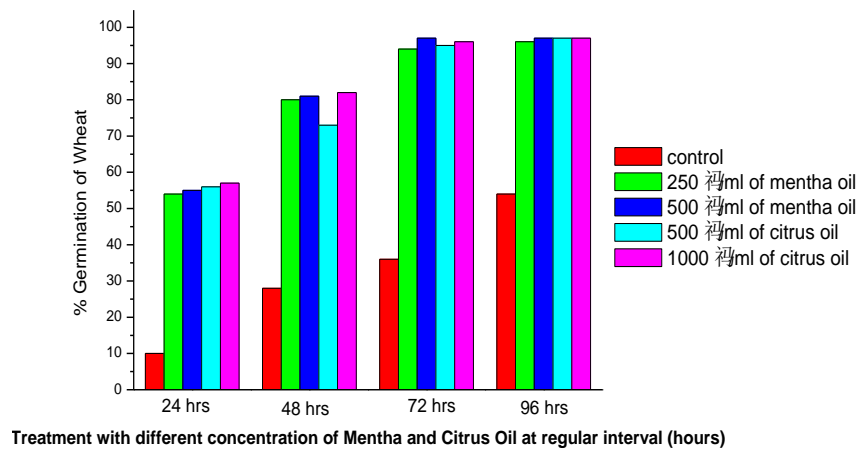


Figure 2. Treatment with different concentration of Mentha and Citrus Oil at regular interval (hours)

Table 1. Analysis of effect of oils fumigated samples of Wheat on infestation by insects after 04 months of storage

Oils	Infestation of Wheat by <i>Sitophilus oryzae</i>				Infestation of Wheat by <i>Tribolium castaneum</i>			
	Control	Fumigated Treatment			Contro	Fumigated Treatment		
		1000 µl/ml	500 µl/ml	250 µl/ml	1	1000 µl/ml	500 µl/ml	250 µl/ml
Menth a oil	++	×	—	+	++	×	—	+
Citrus oil	++	—	+	×	++	—	+	×

Unit- Uninoculated, **Ino.-** Inoculated, + = Poor infestation, ++ = Severe infestation
— = No infestation, × = Concentration not considered.

Discussion

Postharvest losses of stored wheat are enormous due to fungal and insects deteriorations. Stored grain infestation is a very serious problem as it causes economic damage and deteriorates the quality of food grains and food products. Plant essential oils have been used to prevent insect infestation (Isman, 2006) and are being deliberated as an alternative to conventional pesticides because of their low toxicity. More ever they possess fungicidal and insecticidal properties. So these can be used as compare to synthetic/ chemical products.

Therefore, in the present study it was thought desirable to find out the efficacy of some higher plant products (essential oils) in control of biodeterioration of wheat seeds during storage. However, different test fungi of stored food commodities viz. *Aspergillus flavus* (Singh, 2004); *Aspergillus niger* (Pandey, 2003); *Penicillium expansum* and *P. digitatum* (Agrawal, 2003); *Alternaria alternata* (Yadav, 2007) have been taken by various workers during screening of essential oils.

Liu *et al.* (2001) have determined the minimum inhibitory concentration of the oils against test fungus. In the present investigation the MIC of Mentha oil as well as Citrus oils was found 500 µl/ml and 1000 µl/ml (Table 1) respectively against test fungi i.e. considerably lower as compared to several other plant products.

In the present study it was thought desirable to find out the effect of increased inoculum density of the test fungi for the antifungal potency of the Mentha and Citrus oils. It was observed that these, plant products showed the capacity to withstand their fungitoxicity even at high dose of fungal inoculum

thereby indicating the possibility of their exploitation even for the food commodities heavily infested with the test fungi.

Besides fungi several insects also severely deteriorate the stored food commodities. Thus a post harvest chemical control measure should have efficacy against stored insect pests apart from the storage fungi. A perusal of previous literature shows that several plant products have shown pronounced insect repellent/insecticidal activity *in vitro* as well as *In Vivo* condition (Liu *et al.*, 2001b). The Mentha and Citrus oils exhibited insect repellent as well as insecticidal activity against common storage pest of wheat *Sitophilus oryzae* & *Tribolium castaneum* *in vitro* conditions (Table 1).

An ideal fungicide should be pathocidal but not phytocidal, i.e. it must be free from ill effects on the host plants. A number of synthetic fungicides have been found to show phytotoxic effects chiefly on seeds germination, speed of germination index, seedling growth and biomass of test plant. However, the natural fungitoxicants like Lawsone (Tripathi *et al.*, 1978) and Isoalntolactone (Mishra *et al.*, 2000) were found to be non-phytotoxic. In the present study the oils of Mentha and Citrus did not show any adverse effects on the germination, seedling growth, and general health and morphology of wheat on recommended level thereby exhibiting their non-phytotoxic nature on recommended concentration and method adopted (Figure 2).

The study thus reveals that the oils may be exploited as ideal pesticides for control of deterioration of stored food commodities from fungi as well as from insects and it may provide the complete protection of wheat seeds during storage

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(Received: 15 January 2018, accepted: 20 May 2018)