Efficacy of Cinnamon and Citronella Oil Vapours in the Control of *Callosobruchus chinensis* L. in Bulk Stored Green Gram

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ABSTRACT

Efficacy of Cinnamon bark oil vapour (*Cinnamomam verum* Presl.) and Citronella oil vapour (*Cymbopogon nardus* L.) were evaluated for the control of infestations of *Callosobruchus chinensis* L in bulk stored green gram. Both oils were used at 5ml and 10ml/5kg of seeds in 3L plastic baskets. Oviposition and damaged seeds were detected for up to two months of period. At the end of the storage period, the effect of the treatments on flavour, consumer acceptability and seed germination were evaluated. Both oils inhibited population growth of *C. chinensis* as compared to untreated seeds. Both oil vapours at 5 and 10ml/5kg were protected green grams from infestation by *C. chinensis* for 2 months period. But Cinnamon bark oil vapour was more effective than Citronella oil vapour in both concentrations used. No harmful effect was observed on the germination of oil vapour treated seeds. The oil vapour treatments conferred off-flavours to the green grams and product acceptability was compromised by treatment with Cinnamon bark oil vapour at 5ml/5kg.

KEYWORDS: *Callosobruchus chinensis* L., *Cinnamomam verum* Presl., *Cymbopogon nardus* L.

INTRODUCTION

Pulse beetles are a major problem in stored legumes in all tropical countries. While crops may infest in the field, infestations are often too low to detect at harvest. Pulse beetles breed rapidly in storage and the infested grains are unmarketable. Fumigation being the most effective method cannot be practice because of the primitive nature of the storage facilities in many of the villages. The use of Methyl Pirimiphos ether as a 0.25% solution sprayed on the storage bags containing seeds for consumption or as a 2% dust where seeds are used as planting material is the current method of control recommended by the Department of Agriculture in Sri Lanka [3]. Reducing the moisture content of seeds to less than 10% by the weight through proper drying could significantly reduce the pulse beetle infestation. But this is not practiced in Sri Lanka as pulses are sold by weight rather than volume [3]. As chemical insecticides cause several hazards, need arises to search for non-toxic grain protectants. Plant materials which are being traditionally used by some farmers are quite safe and appear to be the most promising grain protectants [1], [2].

Therefore, locally available and less toxic pest management alternatives such as use of botanicals with pesticidal effect against the pests are essential. Botanicals, which are traditionally produced and used by the farmers in the developing countries, appear to be quite safe and promising in the stored pest control. Research on oils as stored grain protectants has been conducted for a long time [10], [11]. Mixing with plant oils is an ancient method of protecting grains against insect attacks [9] but mixing oils with seeds reduce quality of seeds and consumer demand.

In the last two decades many studies have been undertaken to develop new botanical insecticides containing essential oil as an active ingredient [16], because essential oil is volatile and can be used as fumigants at relatively low dosage. However, very little information is available in Sri Lanka on effect of essential oils of indigenous plants against *Callosobruchus* spp. Therefore, present study was undertaken to investigate the effect of locally available two essential oil vapors on the *Callosobruchus chinensis* L. in bulk stored green grams and to test the effectiveness on germination and to evaluate the consumer demand after the treatments.

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MATERIALS AND METHODS

Effect of Oils on Pulse Beetle

The experiment was carried out at the laboratories of the Department of Agricultural Biology, Faculty of Agriculture, University of Ruhuna, Sri Lanka. The ambient temperature and the relative humidity during the experimental period were 27^0-30^0C and 70%-80% respectively.

Commercial product of Cinnamon bark oil (*Cinnamomam verum* Presl.) and Citronella oil (*Cymbopogon nardus* L.) were collected from the local market.

The Green gram (*Vigna radiata* Walp), variety MI-5 obtained from Field Crop Research and Development Institute, Mahailuppallama was cleaned and disinfested by keeping the seeds at 0^0C for 14 days. Seeds were oven dried to achieve constant moisture content of 11.3%. Plastic buckets with lids were used for the experiment containing 5kg of disinfected green gram seeds in each.

Pulse beetles were cultured in the Laboratory to obtain sufficient number of test insects for the experiment. This was done by placing seeds and adult pulse beetle in a glass jar covered with a piece of cheese cloth and were kept in the laboratory at ambient temperature and relative humidity. After 2 weeks of oviposition period, the parent adults were removed. Newly emerged insects (0-24 hours old) collected from above jars were used in the bioassay [14]. Ten pairs (10 males and 10 females) of *C. chinensis* were introduced in to each replicate. About 6 small holes were also made in each lid using a tiny pin to ensure aeration and humidity equilibrium and also to avoid a high concentration of oil vapor inside the apparatus [13]. Five kilo grams of undamaged disinfected green gram seeds and ten pairs of newly emerged adults were placed in each plastic bucket and covered with a lid containing a sponge treated with 5 ml and 10ml of oils in each to serve as vapor diffuser. The experiment was completely randomized with four replicates. In each treatment, oviposition and damaged seeds per treatment were recorded weekly for two months. Three random samples were taken at a time from each bucket.

Effect of Oils on Seed Germination

To test the effect of oils for the germination, seeds were tested for germination at the end of one and two months of treatment. Four hundred seeds were selected randomly and placed on 8 rolls of paper towel for germination (50 seeds on each) and rewetted with distilled water in an amount equal to 2.5 times the volume in the weight of the paper used in the test. 8 sets of untreated seeds were set up similarly as controls. The percentage of seeds that germinated was noted after 5 days. All germination experiments were conducted in temperature range from 27^0-$30\ ^0$C.

Sensory Analysis of the Grains

The sensory test was carried out to test the effect of oil on taste after one and two months of storage on treated and non-treated samples by using a 3 point scale (ISO) to assess the intensity of off-flavors. The scale used was; 0 = absent, 1 = trace, 2 = regular, 3 = strong. Each level of intensity converted into scores and the average of the judges replications submitted to an analysis of variance. DMRT was used for the multiple comparison between control and treated samples.

Seeds were randomly selected from each bucket and washed well. Pressure cooked for 20 minutes using constant amount of water for each sample. The cooked samples were presented on coded plates chosen at random.

Sensory test was carried out in two stages. In the first stage, untreated sample (control) and the samples treated with 10ml oil concentration were evaluated in two repetitions. In the second stage, the control sample and samples treated with 5ml oil concentration were evaluated. A panel with 20 judges was employed using a point scale

to detect the off-flavours, perceived intensity and consumer acceptability.

RESULTS

Effect of Oils on Pulse Beetle

Both oils significantly reduced oviposition at both concentrations tested when compared with control up to 2 months of period. The analyzed data revealed that there is a significant difference between two oils related to mean number of oviposition and damaged seeds. Therefore, low doses are cost effective. But there is no significant difference between two concentrations of cinnamon oil on both oviposition and damaged seeds. An increased effect at the higher dose was noted with Citronella oil (Table 1) (Figure 1).

Table 1: Effect of oils on the damage by *Callosobruchus chinensis* L.

Treatment	Mean number of damaged seeds
Cinnamon oil (5ml)	1.57^{d}
Cinnamon oil (10ml)	1.13^{d}
Citronella oil (5ml)	7.7^{b}
Citronella oil (10ml)	6.08^{c}
Un treated Control	27.35^{a}

Means with the same letter are not significantly difference at 5% level

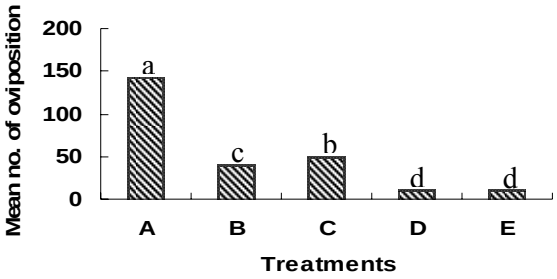

Figure 1: Effect of two volatile oils with two concentrations on ovoposition of *C. chinensis*

A: Control treatment B: Citronella oil (10ml) C: Citronella oil (5ml) D: Cinnamon oil (10ml) E: Cinnamon oil (5ml)

Means with the same letter are not significantly different at 5% level

Effect of Oils on Seed Germination

No loss of viability of the seeds was observed with all the treatments and control up to two months of storage. A small reduction in the percentage of germination but not significantly different was observed with time factor. This result was reported in many other reports [5], [15] (Table 2).

Sensory Analysis

No off-flavors were detected in the control samples after one and two months of storage. But Citronella oil (10ml) caused the significantly highest off-flavor and the sample treated with Cinnamon (5ml) showed very slight off flavor at the 5% level of probability according to DMRT (table 3).

At the end of the first month, high dose of citronella treated samples were rejected by all judges, however with the increment of storage period (two months) 10% of judges accepted for consumption. Therefore, it may be used as a treatment for planting materials. The sample treated with 5ml Cinnamon bark oil vapor presented a very slight off-flavor when compare to all

other treatments in both tested time periods (table 4, 5).

The results of the consumer acceptance test by the point scale showed that 30% of the judges detected off-flavors with 5ml Cinnamon bark oil vapour after 2 months storage. Among these judges, 20% considered the intensity of the off-flavors as a trace, 10% as regular, no one as strong and 70% did not detect off-flavors. Therefore, comparing with control, cinnamon treated samples were accepted by more judges at the end of the second month. It may due to the reduction of strength of the oil vapor (Table 4, 5).

Table 2: Rate of seed germination under different treatments at one month and two month

Treatment	Mean number of germinated seeds (after one month)	Mean number of germinated seeds (after two months)
Cinnamon (5ml)	89.3 [a]	88.6 [a]
Cinnamon (10ml)	88.6 [a]	88.6 [a]
Citronella (5ml)	88.7 [a]	88.6 [a]
Citronella (10ml)	88.6 [a]	87.3 [a]
Control	90.6 [a]	90.3 [a]

Means with the same letter are not significantly difference at 5% level

Table 3: Intensity of off-flavor in green gram treated with 5 and 10ml/kg Cinnamon and Citronella oil vapors *

Treatment	Intensity of off-flavour* after one month	after two month
Control	0.0[a]	0.0[a]
Cinnamon 5ml)	0.7[b]	0.4[ab]
Cinnamon (10ml)	1.0[b]	0.7[b]
Citronella (5ml)	0.7[b]	0.8[b]
Citronella (10ml)	2.1[c]	1.9[c]

Average followed by different letters differ significantly at the 5% level of probability according to DMRT

Table 4: Sensory test -mone month after storage*

Treatment	Appearance (%)			Detection of off flavor (%)		Intensity of off flavor (%)			Consumer Acceptance (%)	
	Good	Medium	Bad	Yes	No	Trace	Regular	Strong	Yes	No
Cinnamon (5ml)	20	60	20	60	40	50	10	0	85	15
Cinnamon (10ml)	40	60	00	60	40	30	20	10	70	30
Citronella (5ml)	60	30	10	70	30	70	00	00	70	30
Citronella (10ml)	0	20	80	90	10	20	20	50	00	100
Control	90	10	00	00	100	00	00	00	100	00

*Test carried out with 20 consumers

Table 5: Sensory test - two months after storage*

Treatment	Appearance (%)			Detection of off flavor (%)		Intensity of off flavor (%)			Consumer acceptance (%)	
	Good	Medium	Bad	Yes	No	Trace	Regular	Strong	Yes	No
Cinnamon 5ml)	80	10	10	30	70	20	10	0	95	05
Cinnamon (10ml)	30	60	10	40	60	20	10	10	80	20
Citronella (5ml)	60	40	00	40	60	20	00	20	80	20
Citronella (10ml)	30	70	00	90	10	30	20	40	10	90
Control	30	60	10	00	100	00	00	00	100	00

*Test carried out with 20 consumers

DISCUSSION

Cinnamon bark oil vapour is more efficient than Citronella oil vapour in the control of *C. chinensis*. Higher doses provide greater protection for a long period. Babu *et. al*. reported that immediately after application of different vegetable oils, oviposition of *C. maculatus* was reduced due to repellent action of the oils [4]. Stamopoulos reported that bitter almond and eucalyptus oil vapors strongly repel *Acanthoscelides obtectus* by reducing fecundity, egg hatchability and increasing L_1 mortality [13].

Khalequzzaman *et.al*. reported that adult emergence was completely prevented and the minimum grain loss was achieved by groundnut oil at 1% up to 66 days after treatment and treatments with groundnut and palm oils at 5 ml.kg-1 showed high acceptability by consumers and can be recommended for *C. chinensis* control in stored pigeon pea for approximately two months[8].

Other scientists reported that choice and no-choice tests with vapour form of thirteen essential oils revealed that most of them have a repellent action, reduce fecundity, decrease egg hatchability, increase neonate larval mortality and adversely influence offspring emergence against *Acanthoscelides obtectus* (Coleoptera: Bruchidae) [8], supporting results of this study that some essential oil vapors can be efficient on stored product pests. The mode of action of oils is partially attributed to interference in normal respiration, resulting in suffocation [12]. It is also thought oils exert some lethal action on developing embryos or first instar larvae by the reduction of gas exchange or direct toxicity by penetrated oil fractions [6]. Since Cinnamon bark oil 5ml/5kg preserve a desirable characteristics, it can be recommended for suppressing *Callosobruchus chinensis* in stored green gram for approximately two months.

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