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REPELLENT PROPERTIES OF PLANT OIL VAPOURS ON PULSE BEETLE (*CALLASOBRUCHUS MACULATUS* L.) (COLEOPTERA: BRUCHIDAE) IN STORED GREEN GRAM (*VIGNA RADIATA* WALP.)

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ABSTRACT

The repellent effects of ten oils, Domba (*Calophyllum inophyllum* L), Batu (*Solanam indicum* L), leaf oil and bark oil of Cinnamon (*Cinnamomum verum* Presl.), Mustard oil (*Brassica juncea* Cross.), Neem oil (*Azadiracta indica* A.Juss), Mee oil (*Maduka longifolia* Koenig.), Castor oil (*Ricinus communis* L.), Citronella oil (*Cymbopogon nardus* L.) and Sesame oil (*Sesamum indicum* L.) were tested for pulse beetle (*Callosobruchus maculatus* L.) in the laboratory conditions. Data were recorded on distribution, oviposition and adult emergence. Ventilated containers each with five pairs of newly emerged adults with 20 green gram seeds were exposed to different oil vapours at the rate of 200µml. Each container was fixed to a device that provided a tunnel for pulse beetle to escape from or enter into any container. Citronella oil, Neem oil, Cinnamon leaf oil and Cinnamon bark oil vapours recorded significantly lowest number of pulse beetles after infestation and their oviposition and adult emergence indicating the highest repellent action and toxic effects. Mustard oil, Domba oil, Mee oil, Castor oil and Batu oil show indications of higher repellent effect at 4DAT on the distribution of the *C. maculatus.* Mustard oil and Domba oil showed a lower rate oviposition than that of Mee oil, Castor oil and Batu oil. All the treatments except Sesame oil had significantly reduced adult emergence at 18DAT. Sesame oil showed positive effect on distribution, oviposition and adult emergence and no repellent activity against the *C. maculatus.*

***Key words**: C. maculatus*, Green gram, Oil vapours, Repellent effects

INTRODUCTION

Green gram (*Vigna radiata* Walp) is a pulse crop producing protein rich edible seeds which is widely grown in the dry zone of Sri Lanka. This crop is subjected to significant losses from storage pests both in the field before harvest and in storage. *Callosobruchus maculatus* and *Callosobruchus chinensis* are the most serious common pests of stored green gram and cowpea *Vigna unguiculata* L. Walp (Dobie 1981). Since grains are already infested in the field by pulse beetle, signs of natural infestation are visible before storage. Larvae feed inside the seeds and without specific protection, 80% of the grain is lost after 6 months of storage (Caswell 1961).

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. Reduction the moisture content of seeds to less than 10% by the weight through proper drying could significantly reduce the pulse beetle infestation. However, this is not practiced in Sri Lanka as pulses are sold by weight rather than volume (Anonymous 1986). As chemical insecticides cause several hazards, need arises to search for non-toxic grain protectants. Therefore, locally available and less toxic pest management alternatives such as the 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 pest control (Rajapakse *et al*. 1998; Rajapakse *et al*. 2002; Jilini 1988). Mixing with plant oils is an ancient method of protecting grains against insect attacks (Pereira 1983). However it has been recorded that mixing oils with seeds reduce quality of seeds and consumer demand.

During the past two decades many studies have been undertaken to develop new botanical insecticides containing essential oil as an active ingredient, (Weaver *et al.* 1995) because essential oil is volatile and can be used as fumigants at relatively low dosage. However, very few studies were undertaken in Sri Lanka to evaluate the repellent action of locally available essential oils against *Callosobruchus* spp. Therefore, present study was conducted to investigate the repellent action of locally available essential oil vapours on the *Callosobruchus* spp. in stored green grams.

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MATERIALS AND METHODS

The experiment was conducted 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 was 27-30^{0}C and 70%-80% respectively.

Plant oils

Commercially available Domba oil (*Callophyllum inophyllum* L), Batu oil (*Solanam indicum* L), Cinnamon leaf oil, Cinnamon bark oil (*Cinnamomam verum* Presl.), Mustard oil (*Brassica juncea* Cross), Citronella oil (*Cymbopogon nardus* L.), Neem oil (*Azadirecta indika* A. Juss), Mee oil (*Maduka longifolia* L.), Castor oil (*Ricinus communis* L.) and Sesame oil (*Sesamum indicum* L.) were collected from the local market.

Preparation of seed

Seeds of green gram (*Vigna radiata* Walp), variety MI-5 obtained from the Field Crop Research and Development Institute, Mahailuppallama were used as a feed of pulse beetle in this study.Seeds were cleaned and disinfected by keeping the seeds at 0^{0}C for 14 days prior to use.

Rearing of the test insects

Pulse beetles were collected from green gram seeds stocks in the market and cultured them on green gram (*Vigna radiata* Walp) in a glass jar covered with a piece of cheese cloth in the laboratory under ambient temperature and relative humidity. After 2 weeks of oviposition period, the culture adults were removed from the culture. Newly emerged insects (0-24 hours old) collected from above culture were used in the bioassay as described by Strong *et.al.* 1968. Five pairs (5 males and 5 females) of *C. maculatus* were introduced in to the each replicate.

The apparatus was made out of plastic cups and straws as shown in figure 1 following Stampoulos (1991) was used to evaluate the repellent effect of treatments. A piece of sponge was glued on the inside of each lid of the plastic cup to serve as vapor diffuser. About 6 small holes were made at equal distance in each lid and side wall of cups using a tiny entomological pin to ensure aeration and humidity equilibrium and also to avoid a high concentration of oil vapor inside the apparatus. One hole was made on side wall of each plastic cup in periphery was fixed with the cups in the centre with 6 holes by using 6cm long plastic straws. The plastic straws formed tunnels to connect the centre cup with surrounding cups

Twenty undamaged green gram seeds and five pairs of newly emerged adults were placed in each plastic cup and covered with a piece of paper that kept no space for pulse beetles to escape. A volume of 200µml oil from each treatment was pipetted and applied into the sponge, glued inside of each cup. The two independent experiments were conducted with five oil vapours per each of the above apparatus and each experiment was completely randomised with ten replicates.

The number of pulse beetle in each cup was recorded by 1, 2, 3 and 4 days after treatment (DAT). After 5 days, all the adult pulse beetles in the apparatus were removed and emerging young ones were counted at 6 and 10 days after treatment. Subsequently emergence of adult pulse beetles was counted daily from 14 to 18 DAT. The data were analysed using one-way ANOVA and student's t-test.

RESULTS AND DISCUSSION

The results showed that the majority of insects moved towards the control. Citronella, Cinnamon leaf, Cinnamon bark and Neem oils recorded significantly the least number of pulse beetles (Table 1, 2) showing repellent and toxic effects. Caster, Musterd, Domba, Mee and Batu oil have also shown significantly lower number of beetles. No significant difference was observed between treatment of Sesame oil and the control.

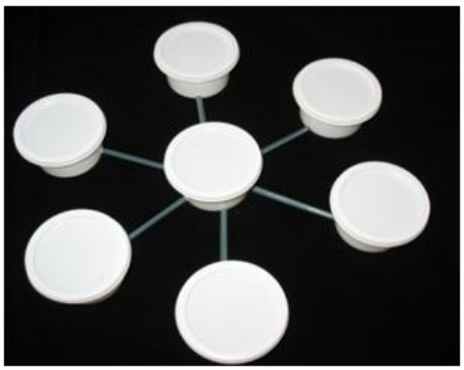

Figure 1: The apparatus used for the evaluation of repellent properties of plant oils

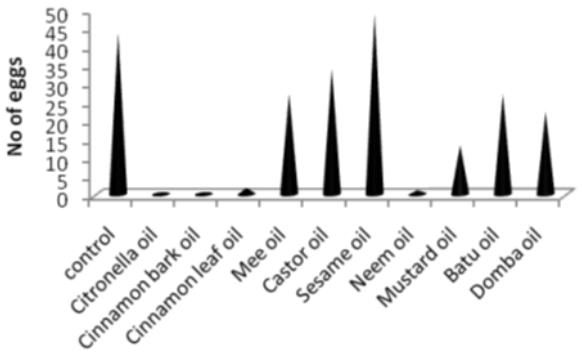

Figure 2: Rate of oviposition of pulse beetles under different treatments (means ± SD of 10 replicates)

Table 1: Number of pulse beetles associated with treatment cups at 1, 2, 3 and 4 DAT in experiment I (mean of 10 replicates)

Treatment (oils)	No. of insects 1DAT	2DAT	3DAT	4DAT
Control	3.9[c]	4.3[c]	5.1[c]	4.6[c]
Citronella oil	0.0[a]	0.0[a]	0.1[a]	0.6[a]
Cinnamon leaf oil	0.5[a]	0.1[a]	0.0[a]	0.0[a]
Mee oil	1.3[b]	1.1[b]	1.3[b]	1.9[b]
Caster oil	1.2[b]	1.1[b]	1.2[b]	1.4[b]
Sesame oil	3.2[c]	3.4[c]	2.6[c]	3.1[c]

DAT-Days After Treatment
Means with same letter in each column are not significantly difference at 5% level

Table 2: Number of pulse beetles associated with treatment cups at 1, 2, 3 and 4 DAT in experiment II (mean of 10 replicates)

Treatment (oils)	No. of insects 1DAT	2DAT	3DAT	4DAT
Control	3.4c	3.4c	2.6c	3.1c
Neem oil	0.8a	1.0a	1.3a	0.9a
Cinnamon bark oil	0.0a	0.1a	0.0a	0.0a
Musterd oil	2.1b	2.2b	1.3b	1.4b
Batu oil	2.6bc	1.8b	1.6b	1.9b
Domba oil	1.2b	1.7b	1.5b	1.6b

DAT-Days After Treatment
Means with same letter in each column are not significantly difference at 5% level

Oviposition

All the treatments, except Sesame oil showed significantly lower rate of emergence when compared to control. Citronella oil indicated no sign of any oviposition. Cinnamon leaf and bark oil and Neam oil treatments showed the least oviposition rate at 6 DAT followed by Mustard oil and Domba oil respectively (Figure 2). Sesame oil showed positive effect on oviposition of pulse beetle has no repellent activity against the *C. maculatus.*

Adult Emergence

All the treatments except the Sesame oil showed significantly lower rate of adult emergence at 18 DAT (Table 3, 4). Citronella oil, Cinnamon leaf oil and bark oil recorded the least number of adult emergence at 14 DAT. Emergence of pulse beetle was significantly lower with the treatments of neem, mustard, cinnamon leaf and bark oils at 18 DAT than at 14,15,16 and 17 DAT.

The results clearly showed that the tested plant oils in this study have affected to reduce the number of adults of pulse beetles initially placed in the cups, their rate of oviposition and development upto adult stage. The tested oils have volatile properties and they give rise vapours in the treatment cups. Citronella oil, Cinnamon leaf oil and Cinnamon bark oil were stronger than that of other oils tested in this study. Thus they have high value for utility as the most promising natural oils for the control of pulse beetle damage in stored products of pulse crops.

Many reports have provided evidences on many volatile essential oils with pesticidal, repellent or toxic properties and usages of some of them (Rani and Osmani 1984). However, essential oils may not serve as contact poisons due to their poor ability to penetrate the insect's integument. Lale (1991) suggested that insects may acquire lethal amounts of essential oil only by contact or by topical application. When tested against different species of pulse beetle, *C. maculatus*, *C. chinensis* and *C. analis* attack in *Vigna radiata*, neem oil allowed no adult emergence, reduced oviposition and prevented insect development (Yadav 1985; Sujatha and Punnaiah 1985). Khaire *et. al*. (1992) showed that adult emergence of *C. maculattus* was completely prevented on cowpea by neem oil at all levels at 100 days.

Babu *et al.* (1989) reported that castor oil 10 ml/kg admixed with mung bean and stored for 18

Table 3: Rate of adult emergence of pulse beetles under different treatments in experiment I (means of 10 replicates)

Treatment	Adult Emergence 14 DAT	15 DAT	16 DAT	17 DAT	18 DAT
Control	19.4[c]	6.7[b]	4.6[b]	2.0[c]	0.9[a]
Citronella oil	0.0[a]	0.0[a]	0.0[a]	0.0[a]	0.0[a]
Cinnamon leaf oil	0.0[a]	0.0[a]	0.0[a]	0.0[a]	0.0[a]
Mee oil	11.7[b]	4.2[b]	4.0[b]	1.7[b]	0.5[a]
Castor oil	11.0[b]	6.4[b]	3.9[c]	3.0[c]	0.7[a]
Sesame oil	16.6[c]	6.1[b]	4.2[b]	1.4[b]	1.5[a]

Mean with same letter in each column are not significantly difference at 5%level

Table 4: Rate of adult emergence of pulse beetles under different treatments in experiment II (means of 10 replicates)

Treatment	Adult Emergence 14 DAT	15 DAT	16 DAT	17 DAT	18 DAT
Control	14.3[c]	5.1[c]	6.8[c]	9.1[d]	10.7[c]
Neem oil	2.0[b]	2.2[b]	1.2[b]	1.0[b]	0.0[a]
Cinnamon bark oil	0.1[a]	0.0[a]	0.0[a]	0.2[a]	0.0[a]
Mustard oil	0.1[a]	0.1[a]	0.5[a]	0.6[a]	0.0[a]
Batu oil	5.3[c]	5.2[c]	4.3[c]	3.2[c]	2.1[b]
Domba oil	2.1[b]	2.8[b]	1.5[b]	1.3[b]	1.0[b]

Mean with same letter in each column are not significantly difference at 5%level

months prevented the emergence of F1 adult *C. chinensis* following artificial inoculation with adult insects. Khalequzzaman *et .al*. (2007) 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 $5mlkg^{-1}$ showed high acceptability by consumers and can be recommended for *C. chinensis* control in stored pigeonpea for approximately two months.

Papachristos and Stamopoulos (2002) reported that choice and no-choice tests with vapour form of thirteen essential oils, 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), supporting our results that some essential oil vapours can be efficient for repel of stored product pests.

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