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ADAPTATION OF CITRONELLA GRASS OIL (*CYMBOPOGON WINTERIANUS JOWITT*) TECNOLOGIES AS AN ALTERNATIVE METHOD FOR COCKROACHES (*BLATTELLA GERMANICA* L.) REPELLANT

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ABSTRACT
Cockroaches (*Blattella germanica* L.) are considered major pests of homes, restaurants and other structures which contaminate food and eat utensils. This study was aimed to adapt citronella grass oil as an alternative method for cockroach repellent. Cockroaches were reared in the dark and hot area of insect cages set at 25 ± 2^0C. In the first experiment, different concentrations of citronella oil (5, 10, 15 and 20 µl) were mixed with 1ml of 70% alcohol and poured into Petri dishes. For the second experiment however, similar aliquots were applied on filter papers with 3 replications. Twenty adult cockroaches were then introduced in to the centre of the cages. After 10-15 min, number of cockroaches at each treated and controls were counted and repellency was computed. The third experiment was done by spraying similar aliquots of citronella oil directly on the cockroaches in the cages. In all cases, Water and alcohol alone each at 1ml were used as controls. The results of this study clearly demonstrated that, repellence activity of citronella oil against cockroach was effective in all experiments. Spraying method using 20µl/ml showed 87.77% repellency followed by 15µl/ml, 10µl/ml and 5µl/ml with 76.66%, 72.22% and 66.11% repellency respectively.
Key word: Citronella grass, insect, repellency bioassay, cockroaches

INTRODUCTION

Cockroaches (*Blattella germanica* L.) are insects of the order *Blattodearia, family Blattellidae* and are one of the most common household pests (Baldwin and Fasulo, 2005). They are considered major pests of schools, homes, restaurants, hospitals, warehouses, offices and other structures, especially those that handle food and provide warm, moist, cracks and crevices. These insects can contaminate food and eat utensils. Cockroaches have been found all over the earth in every kind of building, shelter, cave, mine, pit, nest, in basements, ducts, telephones, sewers, radios, televisions, computers, planes, ships, boats, microwaves, septic tanks, walls and musical instruments (Rust *et al.*, 1991). They have existed > 300 million years ago. Currently, about 3,500 species of cockroach are found worldwide (Esmail *et al.* 2011).

Cockroaches are insects, flattened from top to bottom, usually with two pairs of wings folded flat over the back. Most species rarely fly but they walk very fast. The color usually varies from light brown to black (Roth and Willis, 1960). The cockroach lifecycle actually has three developmental stages. These stages are known as the egg, the nymph, and the adult stages. There is no pupal stage (Koehler, 1993).

Many people, particularly children's health are exposed to infectious diseases that are transmitted by insects, specifically cockroach and flies annually. They cause allergic and compromised health in humans. They are also one of the mechanical vectors to transmission of pathogen to human (Kinfu and Erko, 2008). Some pathogens such as bacteria, protozoans and viruses have been found on cockroach bodies. Cockroaches are also the responsible cause to human physiological stress, gastroenteritis, dysentery, diarrhea, allergic response, skin rashes, watery eyes, sneezing, congestion of nasal passages and asthma (Wang and Bennett, 2009; Nalyanya *et al.*, 2009). Therefore the control is very important. However, excessive and irregular application of chemical pesticides has lead to resistance of cockroaches to insecticides (Nasirian *et al.*, 2009).

Recently, as an alternative pest control technology, essential oil and their constituents have attracted attention because of their low toxicity and their high volatility (Li and Zou, 2001). Citronella grass (*Cymbopogon winterianus* L.) is an aromatic grass belonging to the Poaceae family. It is assumed to have originated from Indonesia. Citronella oil was selected as a positive control in all performed repellency tests because it is a renowned plant-based natural insect repellent, registered for this use in the United States and considered as a biopesticide with a non-toxic mode of action. It is used alone or in combination with cedar wood, lavender, peppermint, clove, eucalyptus, and garlic in a number of commercial insect repellent products (Katz, 2008). Local production of insect repellent would remove the high cost of importation in developing countries. Several studies have investigated the behavioral mode of action of repellents through structure-activity studies of contact versus spatial repellency (Paluch *et al*., 2009). The repellent development from plants is extremely fertile due to wealth of insecticidal compounds found in plants as defenses against insects (Harrewijn *et al*., 1995).

Plant-based repellents are still extensively used in this traditional way throughout rural communities in the tropics because for many of the poorest communities it is the only means of protection from mosquito bites (Moore *et al*., 2002).

Citronella grass is one of the most effective natural repellents among the cultivated plants. Its essential oil is widely used in commercial production of natural repellents that can be observed on the market. It is safe for skin and can be applied during a long time without any damage to the user. The objective of this research was to adapt citronella grass oil as alternative method to cockroach repellant.

MATERIALS AND METHODS

Citronella oil extraction

Fresh and disease free leaves of citronella grass (500 gm) were harvested from experimental fields at Wondo Genet Agricultural research Center. The oil was obtained by hydro-distillation of *C. winterianus* for three hrs using Clevenger apparatus and the extracted oil was stored in a refrigerator at 4°C for further use.

Collection and Maintenance of Cockroaches

Cockroaches were collected around a house and kitchen using bottle with air entrance. The collections were made in the evenings and early in the mornings. According to (Cochran,1983), the cockroaches were reared in the cages set at 25 ± 2^0C, 40-50 RH and 12hr photoperiod. Cockroaches were reared without any restriction and were free to access water and food. Adult Cockroaches were used to investigate repellence activity of the oil.

Repellency bioassay

In this study, three experiments were conducted as described by (Negahban *et al*., 2006). In the first experiment, insect repellent activity of citronella oil was tested by mixing different concentrations (5, 10, 15 and 20 µl) of the oil with 1 ml of 70% alcohol. Water and 70% alcohol each at 1 ml were used as control. Each solution was dispensed into Petri dish with three replications. Then, the different treatments were carefully arranged in alternative patterns in the insect cages. Twenty adult Cockroaches were then introduced in to the centre of the cages and after 10-15 min, number of Cockroaches at each treated and controls was counted and repellency was computed using the formula of (Liu *et al*.,2006) as:

$$\mathbf{R = (C - E) / T \acute{} 100}$$

Where C is the number of insects in the control, E is the number of insects in oil treated chamber and T is the number of total insects released in the central chamber.

For the second experiment, similar aliquots (5, 10, 15 and 20 µl) dissolved in 1 ml of 70% alcohol was applied on filter paper (6.5 cm diameter). Filter papers treated with 1 ml alcohol and water alone was used as control. Similar replications and treatment arrangement as for the first experiment was used. Then, twenty adult Cockroaches were released in to the centre of the cages and after 10-15 min, number of Cockroaches at each treated and control was counted and the repellency was computed using the above formula.

The third experiment was done by spraying similar aliquots (5, 10, 15 and 20 µl) of citronella oil on the cockroaches. Water and alcohol each at 1ml were used as control. Three replications were used. Twenty adult Cockroaches were released into Petri dish inside the cages. Then, each solution was directly sprayed onto the cockroaches inside the Petri dishes and after 10-15 min, number of

cockroaches at each treated and control was counted and repellence activity of the oil was computed using the above formula. Difference between means was assessed using the least significance difference (LSD) test at $P < 0.05$.

RESULT

The relative repellency of citronella oil against cockroach in all treatments is shown in Table 1. There was a significant ($P < 0.05$) overall difference in repellence activity among the treatments. At the highest concentration (20 µl/ml), the mean number of adult cockroaches was very low in all experiments, indicating the better repellence activity of citronella oil at higher concentration. The repellence activities were 70%, 66.11% and 87.77% in the first, second and third experiments respectively (Table 1). Among all concentrations investigated, 20µl/ml was more effective than other concentrations followed by 10, 5 and 1 µl/ml in all the experiments. The present results suggested that mode of application of treatments was the pre-dominant reason for the improved repellence activity of the essential oil. When compared among the different methods of treatments application, the most effective repellence activity (87.77%) was observed in the third experiment (spraying method) using 20µl/ml (Table 1).

Table 1 Percentage repellency of citronella grass essential oil against cockroach

Concentration (µl/ml)	Experiment 1	Experiment 2	Experiment 3	mean
5	48%[d]	50.33%[d]	66.11%[d]	54.88
10	57%[c]	54.44%[c]	72.22%[c]	61.22
15	63%[b]	61.11%[b]	76.66%[b]	66.92
20	70%[a]	66.11%[a]	87.77%[a]	74.62
Mean	77.33	74.40	100.92	
CV	2.02	1.65	1.64	
LSD	2.42	1.92	2.48	

Means with the same letter within the column are not significantly different at p < 0.05% by Duncan's Multiple Range test.

DISCUSSION

The present study results clearly demonstrated that the use of citronella oil for cockroach repellent had a considerable practical significant. In this study, spraying method using 20µl/ml oil was proved to be more effective in repelling cockroaches. However, significantly better repellence activities were recorded in all other concentrations compared to the controls. Likewise, Wasuwat *et al.*, (1990) reported that repellency of a cream containing 14% citronella oil was about two hours against *Aedes aegypti* (L.). On the other hand, Suwonkerd and Tantrarongfoj, (1994) showed that a repellent cream containing less than 10% citronella oil provided only two hours protection against *Anopheles minimus*. A 10% citronella oil formulation was found to repel against this species for at least four hours under laboratory conditions. According to Katz, (2008) the components present in the oil of Citronella species are responsible for the desirable repellent characteristics of the plants against mosquitoes, when applied at the rate of 20 µl/cm2.

It was found that burning an unscented candle had the same effects on reducing the biting rate of mosquitoes in the field as a citronella candle. On the other hand, the potential of citronella oil was reported to reduce pest damage and to increase quality of Chinese kale (Zehnder, 2004). Similarly, Neem oil when formulated as 2% in coconut oil provided complete protection for 12 hours from *Anopheles* mosquitoes (Sharma *et al.*, 1993). *Eucalyptus*-based repellent product, contains a mixture of *p*-menthane-3,8-diol (PMD), isopulegone and citronellol. *Eucalyptus* oil itself, the principal ingredient of which is PMD, provided protection comparable to *N,N*-diethyl-*m*-toluamide (DEET) in repelling *Anopheles* mosquitoes in field studies (Trigg, 1996).

Harris and Dent (1999) indicated that biopesticides are effective in controlling pests that have developed resistance to chemical pesticides and leaving little or no toxic residues thus are commonly harmless to beneficial insects and other non-target organisms. Biopesticides such as Neem products have been reported to reduce the infestation of various insect pests in tea. In recent years, several

essential plant oils had been found to have repellent properties. Such plants included citronella, cedar, verbena, pennyroyal, geranium, lavender, pine, cinnamon, rosemary, basil, thyme, allspice, garlic and peppermint (Mohinder, 2001).

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