



## Toxicity of Some Extracts of Common Plants Towards Three Species of *Pheidole* Ants Under Laboratory Conditions

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### ABSTRACT

The toxicity of some common plant extracts towards workers of three species of *Pheidole* ants was evaluated under laboratory conditions. Methanolic extracts of nine plants indigenous in Egypt, were examined for their potential use as alternatives to synthetic chemical pesticides. Extracts of the tubers of *Allium sativum* L., the seeds of *Anethum graveolens* L., *Coriandrum sativum* L., *Trigonella foenum-graecum* L and *Nigella sativa* L., flowers of *Calndula officinalis* L., and leaves of *Mentha viridis* L., *Rosamarinus officinalis* L and *Eucalyptus citriodora* Hook were tested in this way. Bioassays were performed on mature workers of three species of *Pheidole* ants; *P. jordanica* (Saulcy), *P. laticeps* (Mayr) and *P. sinatica* (Mayr) (Hymenoptera: Formicidae) also common in Egypt. All crude methanolic plant extracts exerted toxic effects on workers of the three *Pheidole* species. The crude methanolic extract of *A. sativum* was the most effective, as indicated by the highest percentage mortality of ant workers, followed by extracts of *T. foenum-graecum*, while crude extract of *E. citriodora*, showed the least activity. The results indicate the potential use of some of these plant extracts as a source of safe alternatives to insecticides.

### INTRODUCTION

Ants are the dominant components of the terrestrial ecosystem throughout the world (Holway *et al.* 2002; Mahalakshimi and Channaveerappa, 2016). They are not only annoying pests, but they can also invade houses; destroy electrical equipment, damage agricultural products and spoil stored foods, with resulting economic loss (Pimentel *et al.* 2005; Chaudhari *et al.*, 2013). The genus *Pheidole* is among the largest ant genera that belong to the subfamily Myrmicinae. The genus is widespread and ecologically dominant across the globe (Wilson, 2003; Presty and Karmaly, 2016). It probably includes more than a thousand species that form a serious threat through their potential impact on biodiversity, ecosystems, agricultural production or society. Many *Pheidole* species are ecologically important seed consumers having an impact on valued plant species in several arid and semi-arid ecosystems around the world (Whitford *et al.*, 1981; Hölldobler and Wilson, 1990; Pirk *et al.*, 2009).

Unfortunately, due to their complex social lives, foraging habits, nesting sites, and fecundity, ants can be quite difficult to control (BASF, 2010). For many decades, ant control practices have relied upon the use of conventional insecticides. However, their negative effects on native biota, non-target organisms including humans, as well as the residual impact of insecticides on the environment (Lester, 2008; Simberloff, 2001) make them increasingly unattractive for this purpose of ant control. Recently, the use of some plant products has drawn attention to their use as pesticides (Begum *et al.*, 2011). Plant products are reported to be more effective, less expensive, biodegradable and safer for human health and the environment (Behal, 1998; Isman, 2006; Chaudhari *et al.*, 2013). Plant extracts, sometimes called botanicals, can be easy to process and apply by farmers in developing countries (Belmain *et al.*, 2001; Isman, 2006 and Regnault-Roger *et al.*, 2012). Botanicals are the most promising sources for new pest control products and some are under trials for their biological activity against pests, such as ants. The unique modes of action of some plant extracts or pure compounds from them can be manifest in various ways, including toxicity, mortality, growth inhibitor, inhibition of chitin biosynthesis, suppression of reproductive activity and fertility as well as reduction of fecundity and longevity (Jannet *et al.*, 2001; Begum *et al.* 2011). These advantages aid their possibilities to be a part of pest management programs for pests such as *Spodoptera litura* Fab. (Baskar *et al.* 2011), *Helicoverpa armigera* Hub. (Vendan *et al.*, 2009), as well as against some ant species (Dos Santos *et al.*, 2013; Chaudhari *et al.*, 2013; Souza *et al.*, 2017). The present research had the objective of evaluating the efficiency of extracts from nine familiar plant species as possible sources of control of *Pheidole* ants.

## MATERIALS AND METHODS

### Experimental Area:

All experiments were conducted in the laboratory of the Zoology Department of El-Minia University, El-Minia, Egypt during the summer of 2017 and 2018 under the prevailing environmental conditions of  $32^{\circ}\pm 1^{\circ}\text{C}$  and  $65\pm 5\%$  relative humidity, typical of the Nile valley.

### Ant Nesting Sites and Species Identification:

For identification, specimens of ant workers taken from three nesting sites were preserved in 70% ethyl alcohol in test tubes and sent for accurate identification to Dr. Brian Tayler, an expert of ant taxonomy at the University of Nottingham, U.K. The ant workers were identified as from the three species: *Pheidole jordanica* (Saulcy), *Pheidole laticeps* (Mayr) and *Pheidole sinatica* (Mayr).

The nest sites of the three species used were characterized by specific vegetation cover of each species; *i.e.* nests of *P. jordanica* were found under a *Acacia nilotica* tree in the Farm of the Botany Department, El-Minia University, nests of *P. laticeps* were located under a *Ficus nitida* tree and *P. sinatica* were nested under a *Delonix regia* tree. *F. nitida* and *D. regia* trees are located on the Nile bank near to the El-Minia Governorate, El-Minia City, Egypt. The ant nests were located in the soil with colonies comprised of a few queens, males, and several hundreds of workers and brood of all stages.

### Ant Collections and Maintenance:

The ant nests were collected from their natural nest sites and moved to the laboratory. For each species, the soil containing the ant nest was carefully transferred to a plastic container and left for a few days until dry. A few drops of water were poured on one small area of the soil to attract ants to that moist area. The dry soil was removed from the other side each day. The remaining moistened soil containing the ants was transferred to a plastic bottle which served as an artificial nest. A small hole was made at the lower side of the bottle to serve as an exit and entrance for the ants. The bottle was placed in a plastic container with an

internal diameter of 25 cm and a height of 60 cm to serve as a foraging area. The inner wall of this container was coated with Vaseline to prevent ants from escaping. To prevent drying, a few drops of water were added to the soil as needed. The ants were fed on tiny drops of honey on a sheet of paper, as well as portions of cooked tuna fish, small pieces of cooked chicken, and freshly killed insects. Colonies were fed *ad libitum* with 10 % sugar water using a glass test tube plugged with a piece of cotton wool.

**Plant Materials:**

Samples of nine plant species, locally available, were selected to assess their effectiveness against the three ant species. The selected plants including four species collected from the Agricultural Farm at El-Minia University: dill (*Anethum graveolens* L.), lemon-scented gum (*Eucalyptus citriodora* Hook.), spearmint (*Mentha viridis* L.), and rosemary (*Rosmarinus officinalis* L.). The other five plant species were purchased as commercially available products from El-Minya shops: garlic (*Allium sativum* L.), pot marigold (*Calendula officinalis* L.), coriander (*Coriandrum sativum* L.), black cumin (*Nigella sativa* L.), and fenugreek (*Trigonella foenum-graecum* L.). A list of the plant species with their family, common name, part of the plant used, dry material weight, crude extract weight and percentage of crude extract yield used in bioassays are presented in Table 1.

**Preparation of Crude Plant Extracts:**

For *Allium sativum*, fresh bulbs (200 g.) were used to prepare an extract. The bulbs were chopped into slices in an electric blender; the slices were macerated with 900 ml methanol (95%) at room temperature for one day. For the other plants, 50 g of dry material was used. For spearmint, rosemary and lemon-scented gum, fresh leaves were used; for pot marigold, flowers were collected, and seeds were taken from dill, coriander, fenugreek, and black cumin. All these materials were washed under running water for 20 min., then left for seven days to dry under room conditions and weighed several times to obtain a constant weight to ensure dryness. The dried materials were powdered in an electric blender. A sample of 50 g of powder of each plant species was macerated (with 300 ml of methanol (95%) at room temperature for two days and filtered using two thicknesses of filter paper. In the case of *Allium sativum*, the plant material was separated into portions and each portion extracted with fresh portions of methanol, the combined methanol extracts were concentrated to dryness, at 40°. The yield of methanolic extract for each plant was weighed (Table 1).

**Table (1):** List of selected plant species, family, common name, plant part used, dry material weight, crude extract weight and percentage of crude extract yield used in bioassays

Family	Species	Common name	Used part of the plant	Dry material weight (g)	Crude extract weight (g)	Crude yield (%)
Alliaceae	<i>Allium sativum</i> L.	Garlic	Bulb	200	21.8	10.9
Apiaceae	<i>Anethum graveolens</i> L.	Dill	Seed	50	13.14	26.3
	<i>Coriandrum sativum</i> L.	Coriander	Seed	50	5.59	11
Compositae	<i>Calendula officinalis</i> L.	Pot marigold	Flower	50	4.99	10
Fabaceae	<i>Trigonella foenum-graecum</i> L.	Fenugreek	Seed	50	7.79	15.6
Lamiaceae	<i>Mentha viridis</i> L.	Spearmint	Leaves	50	5.82	11.7
	<i>Rosmarinus officinalis</i> L.	Rosemary	Leaves	50	7.28	14.6
Myrtaceae	<i>Eucalyptus citriodora</i> Hook	Lemon scented gum	Leaves	50	10.05	20
Ranunculaceae	<i>Nigella sativa</i> L	Black cumin	Seed	50	14.3	28.6

Crude Yield (%) = (dried weight of the produced methanol extract / dried weight of powdered test plant) × 100.

**Laboratory Bioassay of Ant Workers:**

The toxic effect of the dried extracts of the selected plants was evaluated against workers of the three ant species. For each plant extracted, the total dried extract (Table 1) was dissolved in 25% aqueous sugar solution (10 ml.) (regarded as a concentration of 100%) and

various dilutions prepared by adding more of the 25% sugar solution, to obtain serial dilutions of 100%, 75%, 50%, and 25%. These diluted solutions were freshly prepared one hour before the assays.

Each laboratory bioassay was performed with a group of ten mature workers of each species randomly picked from their artificial nests, and introduced with a camel-hair brush into a plastic Petri dish (9 cm diameter) lined with filter paper. Treatments and untreated controls (25% aqueous sugar solution only) were replicated three times. Petri dishes were closed and kept under laboratory conditions. The lid of each Petri dish was punctured by a tiny hole, through which a little of the plant extract could be added with a micropipette without removing the lid to prevent ant disturbance.

Groups of workers were daily provided with 50  $\mu$ L of one of the extract concentrations placed on a glass microscope slide in the center of the plastic Petri dish. The workers were provided with distilled water only in the 24 hours before starting each bioassay. To record the percentage mortalities, Petri dishes were daily observed for three days till the end of the assay. The dead ants were recognized if they gave no response to stimulation by touch with a camel-hair brush.

The percentage of mortality was calculated and data were corrected for the control mortality using Abbott's formula (Abbott, 1925).

Percentage mortality = (Number of dead workers / Number of alive workers)  $\times$  100

Corrected Percentage of mortality = [(T - C) / (100 - C)]  $\times$  100

Where, T = % mortality in tested concentration, and C = % mortality in control

#### Statistical Analysis:

The percentage mortality values were corrected with Abbott's (1925) formula and then data were statistically analyzed by one-way analysis of variance (ANOVA) using SPSS (Statistical Package for the Social Sciences) software, followed by LSD test at a significant level of  $P < 0.05$ . A standard Probit analysis (Raymond, 1985) was used to estimate LC50 and LC90 values of the crude extracts of the tested plant materials against each ant species over the entire inspection period.

## RESULTS

All plant extracts tested for toxic activities against ant workers of the three *Pheidole* species were found to have substantially toxic effects at most of the tested concentrations and exposure periods of 24, 48 and 72 hours (Table 2). The mortality rates increased in the period of the experiments. The highest concentration of 100% crude extract had pronounced insecticidal activity (up to 100% accumulated mortality 72 hours (post-treatment) for *A. sativum* extract, followed by 96.66% mortality for *T. foenum-graecum* extract. The crude extract of *R. officinalis* also produced high mortality at 93.3% in *P. laticeps* workers and 90.5% mortality in workers of the two other species after 72 h of treatment. The accumulated mortality rates caused by the crude extracts of the tested plants ranged between 86.66% and 63.33% in workers of the three ant species post-treatment. Statistical analysis showed that there were significant differences among tested crude plant extracts, at concentration of 100%, in their mortality rates in *P. laticeps* workers 24 h post-treatment ( $F = 57.49$ ;  $P \leq 0.05$ ), 48 h ( $F = 38.3$ ;  $P \leq 0.05$ ) and 72 h ( $F = 35.8$ ;  $P \leq 0.05$ ) post-treatment; in *P. jordanica* workers 24 h post treatment ( $F = 40.75$ ;  $P \leq 0.05$ ), 48 h ( $F = 30.26$ ;  $P \leq 0.05$ .) and 72 h ( $F = 32.31$ ;  $P \leq 0.05$ ) post-treatment; and *P. sinatica* workers 24 h post-treatment ( $F = 45.99$ ;  $P \leq 0.05$ ), 48 h ( $F = 45.09$ ;  $P \leq 0.05$ ) and 72 h ( $F = 29.98$ ;  $P \leq 0.05$ ) post-treatment.

The effectiveness of tested materials using a concentration of 50% was decreased significantly with most of the tested plant extract, whereas *A. graveolens* extract maintained its high efficiency at 83.33% accumulated mortality in the three *Pheidole* species. The

extracts of *T. foenum-graecum*, *A. sativum* and *N. sativa* had moderate effectiveness at 70.5, 66.63 and 63.33 accumulative mortalities in workers of the three ant species, respectively. The least effectiveness was recorded in *R. officinalis* extract at 26.66% mortality 72 hours post-treatment.

**Table (2):** Mortality percentage (Mean ± SE) in mature workers of three *Pheidole* species treated with four sequential concentrations of certain plant extracts 24, 48 and 72 hours post treatments

Plant species	Crude extract concentration, % (w/v)	Mean mortality (%) ± SE at hours after treatment								
		<i>Pheidole laticeps</i>			<i>Pheidole jordanica</i>			<i>Pheidole sinatica</i>		
		24	48	72	24	48	72	24	48	72
<i>Allium sativum</i> <i>Anethum graveolens</i> <i>Calndula officianalis</i> <i>Corianabrum sativum</i> <i>Eucalyptus citriodora</i> <i>Mentha viridis</i> <i>Nigella sativa</i> <i>Rosamarinus officinalis</i> <i>Trigonella foenum-graescum</i> Control	100%	96.66±6.6g	100±0e	100±0d	95.66±3.3g	96±6.6e	100±0d	96.66±6.6g	96±3.3e	100±0d
		76.66±3.3ef	83.33±3.3de	90±5.7cd	77.66±3.3ef	83.66±3.3de	90±5.7cd	76.66±3.3ef	83.66±3.3de	90±5.7cd
		56.66±6.6cd	73.33±3.3cd	86.66±3.3cd	55.66±3.3cd	73.33±6.6cd	86.66±3.3cd	56.66±6.6cd	73.33±6.6cd	86.66±3.3cd
		43.33±3.6bc	53.33±3.3bc	63.33±6.6b	41.33±6.6bc	54.33±3.3bc	63.33±6.6b	43.33±3.6bc	54.33±3.3bc	63.33±6.6b
		36.66±3.3b	36.66±3.3b	63.33±3.3b	35.33±3.3b	37.66±3.3b	66.66±3.3b	36.66±3.3b	37.66±3.3b	66.66±3.3b
		46.66±3.3bc	53.33±3.3bc	86.66±6.6cd	47.66±6.6bc	56.66±3.3bc	86.66±6.6cd	46.66±3.3bc	56.66±3.3bc	86.66±6.6cd
		66.66±3.3de	86.66±6.6de	93.33±3.3cd	64.66±6.6de	86.66±3.3de	90±5.7cd	66.66±3.3de	86.66±3.3de	90±5.7cd
		53.33±3.3bcd	70±5.7cd	83.33±3.3cd	55.33±3.3bcd	67.66±6.6cd	86.33±3.3cd	53.33±3.3bcd	67.66±6.6cd	86.33±3.3cd
		86.66±3.3fg	96.66±6.6e	96.66±6.6cd	86.66±6.6fg	93.66±3.3e	96.66±3.3cd	86.66±3.3fg	97.66±6.6e	96.66±3.3cd
		0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
		0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
<i>Allium sativum</i> <i>Anethum graveolens</i> <i>Calndula officianalis</i> <i>Corianabrum sativum</i> <i>Eucalyptus citriodora</i> <i>Mentha viridis</i> <i>Nigella sativa</i> <i>Rosamarinus officinalis</i> <i>Trigonella foenum-graescum</i> Control	75%	70±5.7e	76.66±3.3d	83.33±6.6e	70±5.7e	77.33±3.3d	83.66±3.3e	70±5.7e	77.33±3.3d	83.66±3.3e
		50±5.7cde	76.66±6.6d	86.66±6.6de	50±5.7cde	76.33±6.6d	86.66±3.3de	50±5.7cde	76.33±6.6d	86.66±3.3de
		43.33±6.6bcd	50±5.7bc	66.66±3.3cde	43.33±6.6bcd	50±5.7bc	66.66±3.3cde	43.33±6.6bcd	50±5.7bc	66.66±3.3cde
		23.33±3.3b	43.33±3.3b	46.66±3.3b	22.33±3.3b	43.33±6.6*	46.66±3.3b	23.33±3.3b	43.33±6.6b	46.66±3.3b
		26.66±3.3bc	33.33±3.3b	56.66±3.3bc	27.66±6.6bc	33.33±6.6b	60±5.7bc	26.66±3.3bc	33.33±6.6b	60±5.7bc
		40±5.7bcd	46.66±3.3bc	73.33±3.3cde	40±5.7bcd	50±5.7bc	70±5.7cde	40±5.7bcd	50±5.7bc	70±5.7cde
		43.33±3.3bcd	73.33±3.3d	73.66±3.3cde	46.33±3.3bcd	76.66±6.6d	73.66±3.3cde	43.33±3.3bcd	76.66±6.6d	73.66±3.3cde
		23.33±3.3b	36.66±3.3b	60±5.7bcd	22.33±6.6b	34.66±3.3b	56±3.3bcd	23.33±3.3b	34.66±3.3b	56±3.3bcd
		56.66±3.3de	66.66±6.6cd	76.33±6.6d	56.33±6.6d	65.66±3.3cd	73.33±6.6de	56.66±3.3de	65.66±3.3cd	76.33±6.6d
		0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
		0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
<i>Allium sativum</i> <i>Anethum graveolens</i> <i>Calndula officianalis</i> <i>Corianabrum sativum</i> <i>Eucalyptus citriodora</i> <i>Mentha viridis</i> <i>Nigella sativa</i> <i>Rosamarinus officinalis</i> <i>Trigonella foenum-graescum</i> Control	50%	36.36±3.3d	53.36±6.6cd	66.63±6.6ef	36.36±6.6d	53.36±6.6cd	66.63±6.6ef	36.36±3.3d	53.36±6.6cd	66.63±6.6ef
		26.66±3.3bcd	56.66±6.6cd	83.33±6.6f	28.66±0.3bcd	55.66±3.3cd	83.33±6.6f	26.66±3.3bcd	55.66±3.3cd	83.33±6.6f
		13.66±0.33 <sup>bc</sup>	36.63±3.3bcd	43.33±3.3bcd	12.66±0.6 <sup>bc</sup>	37.63±3.3bcd	40±5.7bcd	13.66±0.33 <sup>bc</sup>	37.63±3.3bcd	40±5.7bcd
		16.33±3.3 <sup>bc</sup>	26.36±3.3 <sup>b</sup>	36.33±3.3 <sup>bc</sup>	16.33±3.3 <sup>bc</sup>	23.66±3.3 <sup>b</sup>	36.33±3.3 <sup>bc</sup>	16.33±3.3 <sup>bc</sup>	25.66±3.3 <sup>b</sup>	36.33±6.6 <sup>bc</sup>
		16.33±0.33 <sup>bc</sup>	23.66±0.3 <sup>b</sup>	36.66±3.3 <sup>bc</sup>	16.66±3.3 <sup>bc</sup>	26.66±3.3 <sup>bc</sup>	36.66±3.3 <sup>bc</sup>	16.33±0.33 <sup>bc</sup>	26.66±3.3 <sup>bc</sup>	36.66±3.3 <sup>bc</sup>
		26.66±0.66bcd	33.66±3.3bcd	53.33±6.6cde	26.66±3.3bcd	30±5.7bcd	56.33±6.6cde	26.66±0.66bcd	30±5.7bcd	56.33±6.6cde
		33.66±3.3cd	60±5.7d	63.33±3.3def	33.33±6.6cd	60±5.7d	63.33±3.3def	33.66±3.3cd	60±5.7d	63.33±3.3def
		6.66±0.33 <sup>a</sup>	16.63±0.3 <sup>b</sup>	26.66±6.6b	6.66±0.33 <sup>a</sup>	16.63±0.3 <sup>b</sup>	26.66±6.6b	6.66±0.33 <sup>a</sup>	16.63±0.3 <sup>b</sup>	26.66±6.6b
		30±5.7bcd	53.36±3.3cd	70±5.7ef	30±5.7bcd	53.36±6.6cd	70±5.7ef	30±5.7bcd	53.36±6.6cd	70±5.7ef
		0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
		0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
<i>Allium sativum</i> <i>Anethum graveolens</i> <i>Calndula officianalis</i> <i>Corianabrum sativum</i> <i>Eucalyptus citriodora</i> <i>Mentha viridis</i> <i>Nigella sativa</i> <i>Rosamarinus officinalis</i> <i>Trigonella foenum-graescum</i> Control	25%	16.66±3.3b	26.63±3.3c	46.63±3.3c	13.66±3.3b	26.66±6.6c	46.66±3.3c	16.66±3.3b	25.66±6.6c	46.66±3.3c
		0 <sup>a</sup>	0 <sup>a</sup>	6.63±0.33 <sup>a</sup>	0 <sup>a</sup>	6.63±0.33 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	6.63±0.33 <sup>a</sup>
		6.66±0.33 <sup>b</sup>	10±0 <sup>b</sup>	16.63±0.3 <sup>b</sup>	6.33±6.6 <sup>b</sup>	13±3.3b	16.66±0.33 <sup>b</sup>	6.66±0.33 <sup>b</sup>	13±3.3b	16.66±0.3 <sup>b</sup>
		0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
		10±0 <sup>b</sup>	10±0 <sup>b</sup>	13.33±0.3 <sup>b</sup>	6.33±0.33 <sup>b</sup>	13.33±6.6b	13.33±0.3 <sup>b</sup>	6.66±0.33 <sup>b</sup>	13.33±6.6b	13.33±0.3 <sup>b</sup>
		6.66±0.33 <sup>b</sup>	16.63±0.3b	30±5.7bc	6.66±0.33 <sup>b</sup>	16.66±6.6b	26.63±3.3bc	6.66±0.33 <sup>b</sup>	16.66±6.6b	26.63±3.3bc
		6.63±0.33 <sup>b</sup>	16.66±0.3b	26.66±3.3b	6.33±0.33 <sup>b</sup>	16.36±0.3b	26.33±6.6b	6.63±0.33 <sup>b</sup>	16.36±0.3b	26.33±6.6b
		0 <sup>a</sup>	0 <sup>a</sup>	6.66±0.33 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	5±0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	6±0 <sup>a</sup>
		6.66±0.33 <sup>b</sup>	10±0 <sup>b</sup>	16.66±0.3 <sup>b</sup>	5.66±0.66 <sup>b</sup>	10±0 <sup>b</sup>	16.33±0.6 <sup>b</sup>	6.66±0.33 <sup>b</sup>	10±0 <sup>b</sup>	16.33±0.6 <sup>b</sup>
		0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
		0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>

Means followed by different letters are significantly different (P < 0.05). S.E. = standard error

A considerable decrease in efficiency was recorded in all tested plant extracts using a concentration of 25%, since the maximum lethal effect against workers of the three tested ant species estimated at 46.66% for the extract of *A. sativum*, followed by 30% for the extract of *M. viridis* on *P. laticeps* and 26.66% for the same extract on both *P. jordanica* and *P. sinatica*. *N. sativa* extracts at a concentration of 25% achieved about 26.33% accumulated mortality in workers of the three ant species. The results also revealed that there were no significant differences between the efficiency of other plant extracts tested at a concentration of 25% and untreated control, while significant differences were obtained between *A. sativum* extract and control at all tested periods post-treatment.

The same trend of significances was observed among tested crude plant extracts with concentration of 75% in their mortality rates in *P. laticeps* workers 24 h post treatment (F= 13.25; P≤ 0.05), 48 h (F=16.42; P≤0.05) and 72 h (F= 40.48; P≤0.05) post treatment; in *P. jordanica* workers 24 h post treatment (F= 13.43; P≤0.05, 48 h (F= 17.76.; P≤0.05) and 72 h (F= 22.24; P≤0.05) post treatment; and *P. sinatica* workers 24 h post treatment (F= 10.84; P≤0.05.), 48 h (F= 19.66; P≤0.05) and 72 h (F= 35.02; P≤0.05) post treatment.

With regard to the concentration of 50%, significance differences existed among tested crude plant extracts in their mortality rates in *P. laticeps* workers 24 h post-treatment (F=7.94 P≤0.05), 48 h (F= 25.2; P≤0.05) and 72 h (F= 23.4; P≤0.05) post-treatment; in *P. jordanica* workers 24 h post-treatment (F= 7.67; P≤0.05), 48 h (F= 16.77; P≤0.05) and 72 h

( $F=13.67$ ;  $P\leq 0.05$ ) post-treatment; and *P. sinatica* workers 24 h post-treatment ( $F= 9.17$ ;  $P\leq 0.05$ ), 48 h ( $F= 10.28$ ;  $P\leq 0.05$ ) and 72 h ( $F= 10.68$ ;  $P\leq 0.05$ ) post-treatment, while no significant differences were observed between *R. officinalis* extract and control in mortality rates in workers of the three tested ant species only 24 h post-treatment.

However, no significant differences were observed among most of the tested crude plant extracts with a concentration of 25% and control in their mortality rates in workers of *P. laticeps* workers 24 h post-treatment ( $F= 5.42$ ;  $P\leq 0.05$ ), 48 h ( $F= 19$ ;  $P\leq 0.05$ ) and 72 h ( $F= 19.4$ ;  $P\leq 0.05$ ) post-treatment; in *P. jordanica* workers 24 h post-treatment ( $F= 3.03$ ;  $P\leq 0.05$ ), 48 h ( $F= 13.3$ ;  $P\leq 0.05$ ) and 72 h ( $F= 15.75$ ;  $P\leq 0.05$ ) post-treatment; and *P. sinatica* workers 24 h post-treatment ( $F=2.08$   $P\leq 0.05$ ), 48 h ( $F= 5.62$ ;  $P\leq 0.05$ ) and 72 h ( $F= 9.31$ ;  $P\leq 0.05$ ) post-treatment

## DISCUSSION

The botanical extracts offer promising materials for further study and separation to find alternatives to chemical insecticides. Such materials may act as effective insecticides against vegetable pests (Visetson and Milne, 2001), repellents (Hori, 2003) and antifeedants (Park *et al.*, 2003). Similar results have been reported of extracts with a specific mode of action against leaf cutting-ants such as *Ricinus communis* L. (Bigi *et al.*, 2004), *Sesamum indicum* L. (Morini *et al.*, 2005), *Cedrela fissilis* Vell (Bueno *et al.*, 2005), *Helietta puberalla* RE Fr. (Almeida *et al.*, 2007), *Citrus* sp. (Fernandes *et al.*, 2002), *Raulinoa echinata* R.S. Cowen (Biavatti *et al.*, 2005), *Simarouba versicolor* St. Hil. (Peñaflor *et al.*, 2009), *Jatropha curcas* L. and *Ricinus communis* L. (Alonso and Santos, 2013), and *Esenbeckia pumila* (Souza *et al.*, 2017). Araújo *et al.*, (2008) reported that *Ruta graveolens* L. and *Ageratum conyzoides* L. extracts induced mortality of *Atta sexdens* workers through a topical application at a concentration of 1 mg. mL<sup>-1</sup>. Tangchitphinitkan *et al.* (2007) evaluated the efficiency of three Thai herbs against the adult workers of the pharaoh ant (*Monomorium pharaonis* L.). They demonstrated that the tuba root extracts (*Derris elliptica* Benth.) had a LC<sub>50</sub> against the adult workers at 0.22% w/v, whereas yam bean seed extracts (*Pachyrhizus erosus* L.) had a LC<sub>50</sub> against adult workers at 0.35% w/v and tea seed cake extracts (*Camellia* sp.) had a LC<sub>50</sub> against the adult workers at 0.55% w/v after 24 h exposure, respectively. Mashaly *et al.* (2014) assessed the three Saudi plants, harmful (*Rhaza stricta*), boxthorn (*Lycium shawii*) and artemisia (*Artemisia inculta*), in a minced meat bait against workers of the samsun ant (*Pachycondyla sennaarensis*). They found that among the plant extracts tested, the plant extract of boxthorn at a concentration of 0.3 mg per gram of food exhibited the highest toxicity to samsun ants, causing 20.30% mortality per day and 100% mortality rate of all ants during 4.9 days.

The results of the present study were also consistent with those of Visetson 2001 and Visetson and Milne 2001 who demonstrated the effectiveness of neem (*Azadirachta indica*) seed kernel extracts against *Callosobruchus maculatus* F. and derris (*D. elliptica*) extracts against *Plutella xylostella* L.

Results obtained by Pandey *et al.* (2000) demonstrated that the essential oil from *E. citriodora* was highly toxic to *Meloidogyne incognita* beetles and inhibited the growth of root-knot nematode at 250 ppm. However, Al-Jaber (2006) found that the extract of *Mentha viridis* had but a week effect against the beetle *Tribolium castaneum* at 1% concentration. De Sousa *et al.* (2005) reported that *C. sativum* essential oils killed 53.99% of *Callosobruchus maculatus* (Fabricius) at a concentration of 2.5% (w/w). Hazrat and Soaib (2012) also showed that the essential oil from *C. sativum* is effective against mosquito larvae. Oparaeke *et al.* (2007) revealed that garlic bulb extracts significantly reduced the populations of the *Maruca vitrata* (Legume pod borer and *Clavigralla tomentosicollis* (Brown pod-sucking bug).

**In conclusion**, this study confirmed that among extracts of nine plants, a simple methanol extract of *Allium sativum* particularly, showed potential as an effective toxin to cause up to 100% mortality in ant workers of the tested three *Pheidole* species. However, further investigations are necessary to demonstrate field application, as well as the possibility of attempts to use these compounds as slow-acting agents in ant baits to allow workers to transfer the toxic baits to other individuals and brood inside the ant nests to achieve effective control.

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#### REFERENCES

- Abbott, W. S. (1925). A method of computing the effectiveness of an insecticide. *J. Econ. Entomol.*, 18: 265-267.
- Al-Jabr, A. M. (2006). Toxicity and repellency of seven plant essential oils to *Oryzaephilus surinamensis* (Coleoptera: Silvanidae) and *Tribolium castaneum* (Coleoptera: Tenebrionidae). *Sci. J. King Faisal Univ. (Basic and Applied Sciences)*, 7(1) 49-60.
- Almeida, R.; Peñafior, M.; Simote, S.; Bueno, O.; Hebling, M.; Pagnocca, F. and da Silva, M. (2007). Toxicity of substances isolated from *Helietta puberula* RE Fr. (Rutaceae) to the leaf-cutting ant *Atta sexdens* L. (Hymenoptera: Formicidae) and the symbiotic fungus *Leucoagaricus gongylophorus* (Singer) Möller. *BioAssay*, 2:1-8.
- Alonso, E. C. and D. Y. A. C. Santos (2013). *Ricinus communis* and *Jatropha curcas* (Euphorbiaceae) seed oil toxicity against *Atta sexdens* rubropilosa (Hymenoptera: Formicidae). *J. of Econo. Entomol.* 106: 742-746.
- Araújo, M. S.; Della Lucia, T. M.; Moreira, M. D. and Picanço, M. C. (2008). Toxicidade de extratos hexânicos de plantas às operárias de *Atta laevigata* e *Acromyrmex subterraneus subterraneus* (Formicidae: Attini). *Revista Brasileira de Agrociência*, 14: 106-114.
- BASF (2010). Pest Control Solutions controlling and preventing ant infestations. BASF Pest Control Solution - The Chemical Company, 5 pp. <https://pestcontrol.basf.us/>.
- Baskar, K.; Maheswaran, R.; Kingsley, S. and Ignacimuthu, S. (2011). Bioefficacy of plant extracts against Asian army worm *Spodoptera litura* Fab. (Lepidoptera: Noctuidae). *J. of Agricultural Techno.* 7: 123- 131.
- Begum, N.; Sharma, B. and Pandey, R. S. (2011). Toxicity potential and anti-AchE activity of some plant extracts in *Musca domestica*. *J. Biofertil. Biopestic*, 2 (108) 1-6.
- Behal, S. R. (1998). Effect of some plant oils on the olfactory response of the larvae of rice-moth *Corcyra cephalonica* Stainton. *Ann. of plant prot. Sci.*, 6(2) 146-150.
- Belmain, S. R.; Neal, G. E.; Ray D. E. and Golop P. (2001). Insecticidal and vertebrate toxicity associated with ethno botanicals used as post-harvest protectants in Ghana. *Food Chem. and Toxicol.*, 39: 287-291.
- Biavatti, M. W.; Westerlon, R.; Vieira, P. C.; Silva, M. F. G. F.; Fernandes, J. B.; Peñafior, M. F. G. V.; Bueno O. C. and Ellena J. (2005). Leaf-cutting ants toxicity of limonexic acid and degraded limonoids from *Raulinoa euchinata*. X-ray structure of epoxy-fraxinellone. *J. of the Brazilian Chem. Soc.* 16: 1443-1447.
- Bigi, M. F. M. A.; Torkomian, V. L. V.; Groote, S. T. C. S.; Hebling, M. J. A.; Bueno, O. C.; Pagnocca, F. C.; Fernandes, J. B.; Vieira P. C. and Silva M. F. G. F. (2004). Activity of *Ricinus communis* (Euphorbiaceae) and ricinine against the leaf-cutting ant *Atta*

- sexdens rubropilosa* (Hymenoptera: Formicidae) and the symbiotic fungus *Leucoagaricus gongylophorus*. Pest Management Sci. 60: 933-938.
- Bueno, F. C.; Godoy, M. P.; Leite, A. C.; Bueno, O. C.; Pagnocca, F. C.; Fernandes, J. B.; and Silva, M. F. G. F. (2005). Toxicity of *Cedrela fissilis* to *Atta sexdens rubropilosa* (Hymenoptera: Formicidae) and its symbiotic fungus. Sociobiology. 45: 389-399.
- Chaudhari, P. S.; Jangam, S. S.; Chaudhari, S. V.; Baheti, K. G.; Patil, S. U. and Jadhav, G. B. (2013). Herbal plants as an ant repellent. International J. of Bioassays. 2(6) 883-886.
- De Sousa, A. H.; Maracajá, P. B.; da Silva, R. M. A.; de Moura, A. M. N. and de Andrade, W. G. (2005). Bioactivity of vegetal powders against *Callosobruchus maculatus* (Coleoptera: Bruchidae) in caupi bean and seed physiological analysis. Revista de Biologia e Ciências da Terra, 5(2) 96-103.
- Dos Santos, J. C.; Zanetti, R.; de Oliveira, D. F.; Gajo, G. C. and Alves, D. S. (2013). Plant-Derived Products for Leaf-Cutting Ants Control. In Insecticides-Development of Safer and More Effective Technologies. IntechOpen.
- Fernandes, J. B.; David, V.; Facchini, P. H.; Da Silva, M. F. D.; Rodriguesfilho, E. and Vieira, P. C. (2002). Extrações de óleos de sementes decitros e sua atividade sobre a formiga cortadeira *Attasexdens* e seu fungo simbiote. Quimica Nova, 25: 1091-1095.
- Hazrat, B. and Soaib, A. H. (2012). Plants secondary metabolites for mosquito control. Asian Pac. J. Trop. Dis. 12: 166-168.
- Hölldobler, B. and Wilson, E. O. (1990). The ants. Harvard University Press.
- Holway, D. A.; Lach, L.; Suarez, A. V.; Tsutsui, N. D. and Case, T. J. (2002). The causes and consequences of ant invasions. Annu. Rev. of Ecol. and Syst. 33: 181- 233.
- Hori, M. (2003). Repellency of essential oils against the cigarette beetle, *Lasioderma serricorne* (Fabricius) (Coleoptera: Anobiidae). Appl. Entomol. Zool. 38: 467-473.
- Isman, M. B. (2006). Botanical insecticides, deterrents and repellents in modern agriculture and increasingly regulated World. Annu. Rev. of Entomol. 51: 45-66.
- Jannet, H. B.; H-Skhiri, F.; Mighri, Z.; Simmonds, M. S. J. and Blaney, W. M. (2001). Antifeedant activity of plant extracts and of new natural diglyceride compounds isolated from *Ajuga pseudoiva* leaves against *Spodoptera littoralis* larvae. Industrial Crops and Products, 14(3) 213-222.
- Lester, P. J. (2008). Integrated pest management: an under-utilized tool for conservation and the management of invasive ants and their mutualistic Hemiptera in the Pacific. Pacific Conservation Bio. 14: 246-247.
- Mahalakshimi, B. R. and Channaveerappa, H. (2016). Diversity of ant species (Hymenoptera: Formicidae) in the campus of maharani's science college for women: a mini model of habitat persistence. I.J.P.A.Z. 4(3) 277-281.
- Mashaly, A. M. A.; Farooq, M.; Hozzein, W.; Al-Qahtani, A. M. and Al-Mekhlafi, F. A. (2014). Assessment of toxic baits for the control of the samsam ant, *Pachycondyla sennaarensis* (Formicidae: Ponerinae). African Entomology, 22(1) 204-209.
- Morini, M. S. C.; Bueno, O. C.; Bueno, F. C.; Leite, A. C.; Hebling, M. J. A.; Pagnocca, F. C.; Fernandes, J. B.; Vieira, P. C. and Silva, M. F. G. F. (2005). Toxicity of sesame seed to leaf-cutting ant *Atta sexdens rubropilosa* (Hymenoptera: Formicidae). Sociobiology, 45: 195-204.
- Oparaeke, A. M.; Dike, M. C. and Amatobi, C. I. (2007). Effect of application of different concentrations and appropriate schedules of aqueous garlic (*Allium sativum* L.) bulb extracts against *Maruca vitrata* and *Clavigralla tomentosicollis* on cowpea, *Vigna unguiculata* (L.) Walp. Arch. of Phytopathol. and Plant Prot., 40(4) 246-251.
- Pandey, R.; Kalra, A.; Tandon, S.; Mehrotra, N.; Singh, H. N. and Kumar, S. (2000).

- Essential oils as a potent source of nematicidal compounds. *J. Phytopathol.* 148: 501–502.
- Park, J. D.; Ahn, Y. J.; Park, I. K.; Lee, S. G. and Choi, D. H. (2003). Insecticidal activities of constituents identified in the essential oil from leaves of *Chamaecyparis obtusa* against *Callosobruchus chinensis* (L.) and *Sitophilus oryzae* (L.). *J. of Stored Product Res.* 39: 375-384.
- Peñaflor, M. F. G. V.; Almeida, R. N. A.; Simote, S. Y.; Yamane, E.; Bueno, O. C.; Hebling, M. J. A.; Fernandes, J. B.; Vieira, P. C.; Silva, M. F. G. F. and Pagnocca, F. C. (2009). Toxicity of substances isolated from *Simarouba versicolor* St. Hil. (Simaroubaceae) to the leaf-cutting ant *Atta sexdens* L. (Hymenoptera: Formicidae) and the symbiotic fungus *Leucoagaricus gongylophorus* (Singer) Möller. *Bioassay*, 4: 1-7.
- Pimentel, D.; Zuniga R. and Morrison D. (2005). Update on the environmental and economic costs associated with alien-invasive species in the United States. *Ecol. Econom.*, 52(3) 273–288.
- Pirk, G. I.; di Pasquo, F. and Lopez de Casenave, J. (2009). Diet of two sympatric *Pheidole* spp. ants in the central Monte desert: implications for seed-granivore interactions. *Insect. Soc.*, 56: 277-283.
- Presty, J. and Karmaly, K. A. (2016). Distribution, ecology and collection methods involved in sampling of five species of genus *Pheidole* westwood (Formicidae: Myrmicinae) from kerala. *Inter. J. of Plant, Animal and Enviro. Sci.* 6(2) 43-45.
- Raymond, M. (1985). Presentation d'un programme basic d'analyse log-probit pour micro-ordinateur. *Cah. ORSTOM Ser. Entomol. Med. Parasitol*, 23: 117-121.
- Regnault-Roger, C.; Vincent, C. and Arnason, J. T. (2012). Essential oils in insect control: low-risk products in a high-stakes world. *Annu. Rev. of Entomol.* 57: 405- 424.
- Simberloff, D. (2001). Eradication of island invasives: practical actions and results achieved. *Trends in Ecolo. and Evolu.* 16: 273-274.
- Souza, B. R.; Coelho, G. M.; Rocha, E. C.; Jesus, F. G.; Menezes, A. C. and Araújo, M. S. (2017). Topical Toxicity of *Esenbeckia pumila* Extracts on Leaf-Cutting Ants *Atta laevigata* and *Acromyrmex balzani*. *J. of Agricultural Sci.* 9(12) 248-258.
- Tangchitphinitkan, P.; Visetson, S.; Maketon, M. and Milne, M. (2007). Effects of some herbal plant extracts against Pharaoh ant, *Monomorium pharaonis* (Linnaeus). *Current appl. Sci. and techno.* 7(2-1) 155-159.
- Vendan, S. E.; Baskar, K.; Paulraj, M. G. and Ignacimuthu, S. (2009). Antifeedant and larvicidal effects of *Hydnocarpus alpine* Wt. (Flacourtiaceae) extracts against the larvae of *Helicoverpa armigera* Hub. (Lepidoptera: Noctuidae). *Ecofriendly insect pest management*. Elite Publishing House, New Delhi, India, 210-216.
- Visetson, S. (2001). Effect of Azadirachtin from Various Thai Neem Extracts on Some Detoxification Enzyme Activity in *Callosobruchus maculatus* F. 20th ASEAN / 2nd APRC Seminar on Postharvest Technology, Chaingmai Thailand, pp. 34-46.
- Visetson, S. and Milne, M. (2001). Effects of root extract from derris (*Derris elliptica* Benth) on mortality and detoxification enzyme levels in the diamondback moth larvae (*Plutella xylostella* Linn.). *Kasetsart J. of Nat. Sci.* 35: 157-163.
- Whitford, W. G.; Depree, D. J.; Hamilton, P. and Ettershank, G. (1981). Foraging ecology of seed-harvesting ants, *Pheidole* spp. in a Chihuahuan desert ecosystem. *Am. Midl. Nat.* 105: 159-167.
- Wilson, E. O. (2003). *Pheidole* in the New World: a dominant, hyperdiverse ant genus (Vol. 1). Harvard University Press.

## ARABIC SUMMARY

## فاعلية بعض المستخلصات النباتية ضد ثلاث أنواع من نمل الفيدولي تحت الظروف المعملية

أسماء صابر علي<sup>١</sup> - محمود فضل علي<sup>٢</sup>

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تم اختبار فاعلية المستخلصات النباتية لتسع نباتات وهي الثوم، الشبث، الأقحوان، الكزبره، الكافور الليموني، النعناع، حبة البركه، حصا اللبان والحلبه ضد شغالات ثلاث انواع من نمل الفيدولي وهم فيدولي جوردانیکا، فيدولي لايسبس وفيدولي سيناتيكا تحت الظروف المعملية في اطباق بتري مع ورقة ترشيح مبلله وتركيزات مختلفه من المستخلصات النباتية لفترات زمنية مختلفه. ووضحت النتائج ان المستخلصات النباتية لها تأثير سمي ضد شغالات النمل وهذا التأثير يختلف باختلاف نوع النبات وتركيزه والفترة الزمنية التي تتعرض لها شغالات النمل لهذه المستخلصات، وكان المستخلص النباتي لنبات الثوم هو الأكثر فاعلية وتأثير سمي على شغالات النمل في الأنواع الثلاثة يليه نبات الحلبه، الشبث، حبة البركه، الكزبره، النعناع، الأقحوان، حصا اللبان، ثم نبات الكافور الليموني. ومن هذا يتضح لنا قدرة المستخلصات النباتية كمبيد حشري آمن على البيئه وبديل للمبيدات الحشريه التي لها تأثير سيئ على البيئه.