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Efficacy Of Some Non-Conventional Insecticides Against Different Stages Of American Cockroaches (Periplaneta Americana L.) Using Contact Toxicity Method In Jeddah Governorate.

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ABSTRACT

Owing to the fact that using chemical insecticides cause many effects on human health and pollute the environment, non-conventional insecticides are being tested for the control of Periplaneta americana adults and nymphs. From this backdrop of knowledge, the present study was designed to investigate the insecticidal efficacy of three classes of non-conventional insecticides: commercial bacteria, botanicals and insect growth regulators. One preventative material from each class was selected to compare the toxicity. VectoBac from commercial bacteria, Camelliasinensis from botanicals and Admiral 10EC. The objective of this study was to determine which of these insecticides were most effective against P.americana adults and nymphs. They were tested for their LC_{50}'s and LC_{90}'s values under contact toxicity bioassay method, using different concentrations for each material. Admiral 10 EC was highly effective at all concentrations, while C. sinensis was least toxic among all for both adults and nymphs. Based upon the present experiment, insect growth regulators and commercial bacteria can be used against P. americana adults and nymphs as a safety method for control instead of chemical insecticides.
Keywords: Botanic, insect growth regulators, commercial bacteria, mortality, P.americana, toxicity.

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INTRODUCTION

Periplaneta americanais an important insects in medicine [1], and it is one of the largest common cockroach species [2]. They survive in warm weather with high moisture conditions as well as in unfavorable environments for humans (i.e., sewers and other human-made habitats) [3]. Theyspread bacteria, fungi, and other pathogenic microorganisms from infected areas [4], and cause allergies to human [5]. They transmit different pathogens and diseases [6], because they spends most of its time in sewage, sewer pipe which usually contains high density of pathogens [7], in addition, their nocturnal and filthy habits of eating their feces make them ideal carriers of numerous pathogenic microbes [8].All of these pathogens used as dangerous organisms targeting animal or human populations.A numerous pathogens are harmful to humans being and carried by cockroaches as well as they present in their faeces [9].More than 40 pathogenic and nonpathogenic bacterial species have been isolated from cockroaches. Moreover, 70 species of Gram positive and negative bacteria belonging to 37 genera were isolated from the surface and fecal pellets; including: Actinomyces randingae, Alcaligenes faecalis, Arthrobacter cumminnsii, Aureubacterium spp., Bacillus spp., Brevibacterium spp., Burkholderia vietnamiensis, Buttiauxella sp., Citrobacter sp., Corynebacterium spp., Enterobacter spp., Erwinia sp., Escherichia coli, Hafnia sp., Kigali sp., Klebsiella spp., Kluyvera sp., Kauri rosea, Leuconostoc sp., Micro bacterium spp., Micrococcus sp., Proteus spp., Providence ruttier, Pseudomonas spp., Rhodococcus australis, Rhodococcus rhodochrous, Salmonella typhimurium, Serratia spp., Shigella sp., Spingobacterium thalpophilum, Staphylococcus spp., Stenotrophomonas maltophillia, Streptococcus sp., Tsukamurella inchonensis, Vibrio metschnikovii, Xanthomonas spp., Entamoeba histolytica, Escherichia coli, Klebsiella pneumoniae, Mycobacterium leprae, Shigella dysenteriae, andSalmonellasp., including Salmonella typhi and Salmonella typhimurium, Serratia species, Staphylococcus aureus and Aeromonas sp.[10].Chemical control has been the most popular and effective method so far [11], but their control as insecticides is not a suitable because of several reasons; the most important of which is that they may develop resistance against certain frequently used insecticides [12]. Biological insecticides such as microbes, do not pose a disease risk to wildlife, humans, and other organisms not closely related to target insect [13].Essential groups of microbe's entomopathogens that parasitize insects are the bacteria, viruses, fungi, nematodes and protozoa, which have been used to control insect pests in the field [14]. Pathogenesis by microbial entomopathogens occurs by invasion through the integument or gut of the insect, followed by multiplication of the pathogen resulting in the death of the host e.g., insects [14]. Microbes is produced naturally acts as insecticide when it becomes attached to the cockroach body [15]. Different species of Bacillus can infect different groups of insect pests. The infection is mainly due to receptor sites on the gut wall such as Bacillus thuringiensis.Moreover, kurstaki acts on caterpillars of moths and butterflies, Bacillus thuringiensis var. israelensis is for larvae of flies such as fungus gnats and B. thuringiensis var. san diego is useful for larvae of beetles such as elm leaf beetles and Colorado potato beetle [16]. Botanical and their essential oils, are among the most efficient botanical insecticides, their low toxicity to wildlife and they are toxic to some adult cockroaches [17, 18]. Nerium oleander, Eucalyptus camaldulensis, Datura metel and Nicotiana tabacum were used against nymphs of P.americana.The essential oils Cymbopogon citratus, Cymbopogon nardus and Syzygium aromaticum (clove) were evaluated for repellent activity against adult P.americana [19].The essential oil components do have an effect on ootheca hatch, but they do not eliminate hatch. Follow up treatment would be necessary to prevent reinfestation by the hatched nymphs [20].Insect growth regulators (IGRs) are compounds that can regulate the growth of insect pests and play an important role in their control [21]. These active ingredients prevent insect larvae from developing into adults. Chemicalsinterferewith the normal function of insect juvenile hormone, which controls the growth, development and maturation of insects. The IGRs were discussed in terms of the potential effectiveness of lufenuron as a cockroach control agent and in relation to the substantial differences in susceptibility to the inhibitory effects of this compound on reproduction that were apparent between Blatta orientalis and Blattella germanica[22]. Pyriproxyfen, a pyridyl ether compound, which shows high juvenile hormone mimic (JHM) activity against insects, was evaluated for activities against B. germanica and the insects died off in less than a year[23]. The present work designed to investigate the insecticidal efficacy of some non-conventional insecticides on P.americana, and their susceptibility to different stages through laboratory bioassay using contact toxicity method.

MATERIALS AND METHOD

Experimental insect

P.americana was collected from dark and damp places (sewers) from different areas in Jeddah province by using food jars surrounded by dark cloth as a trap [24]. The strains were stored in the lab and used in this study. Traps were placed into main sewers. Cockroaches were collected every two days and placed in glass containers (30 × 60 × 30 cm). Then, they were thus kept under the laboratory condition of 25 ± 3 °C and 75 ± 5 % RH.

Insecticides

The present study was designed to investigate the insecticidal efficacy of three different non-conventional insecticides:V.Bas a commercial insecticidal bacteria, Green tea (Camelliasinensis),as a botanic and Admiral 10 EC as insect growth regulator. All insecticides were purchased from a local market of Jeddah, Saudi Arabia and used during bioassay. The choice of these insecticides was based on the fact that these formulations are available for everyone, low coast and have not much tested against different stages of P.americana in Jeddah governorate.The insecticides were tested against P.americana adult and nymphs by contact toxicity method, different concentrations were prepared and mortality percentages were recorded after 48 h.

Plant extraction

Green tea was extracted by hydro distillation according to [25]. 100 g of plant was submitted in 500 ml flask with water for 8-10 h. The volatile water were collected in another flask and stored at 4ºC for using in further experiments.

Contact toxicity bioassay

Contact toxicity bioassay was done according to [26], with some modifications against adults and nymphs. Contact toxicity mixture was improvised in the laboratory. Contact bioassays were conducted with previous method. Liquid mixture was then conducting by spraying different concentrations of the insecticide from inside plastic box and make sure that the insecticide covered all the sides. Three plastic boxes with 30 cockroaches (adults and nymphs) were used for each concentration.

Laboratory bioassay testing

The objective of this study was to test the potential use of the selected insecticides against different stages of P.americana. All tests were done according to[27],for laboratory testing of P.americana. Generally, following formula was used for preparing concentrations:

$$\frac{\text{(Concentration required in ml) x (Volume require in ml)}}{\text{(\% Concentration in ml) x 10}} = \text{………. ml}$$

Series of concentrations were then prepared by adding distilled water held in plastic (20 cmx 10 cmx 5 cm), with perforated lids for aeration. Three replicates were run at each concentration. Final test solution volume and 30 insects (adults and nymphs) were added to each plastic box. The plastic boxes were held at 25 ±1 ºC under a recommended 12:12 h light: dark photoperiod. Mortality was assessed at 48h.

Statistical analysis

This study was completely randomized design (CRD) in a factorial experiment.The data were statistically analyzed using analysis of variance (ANOVA). LC_{50} and LC_{90} were calculated according to Probit analysis program [28].All Malformations were captured using digital camera.

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RESULTS

Susceptibility to commercial V.B

Contact toxicity bioassay of V.Bagainst P.americana adults and nymphs were recorded in Table (1& 2) after 48 h. The relationship between the concentrations of V.B and mortality percentage is inverse relationship. V.B cause higher mortality by increasing the concentrations. According to the susceptibility level of adults and nymphs of P.americana after 48 h., of continuous exposure to residue of V.B, Fig. (1 and 2),showed that nymph stage were more sensitive to V.B by LC_{50}'s values (7.489 %) followed by adult stage (17.579 %) after 48h.

Fig 1: Comparison between the susceptibility of P.americanaadults against non-conventional insecticidesusing contact toxicity method after 48h.

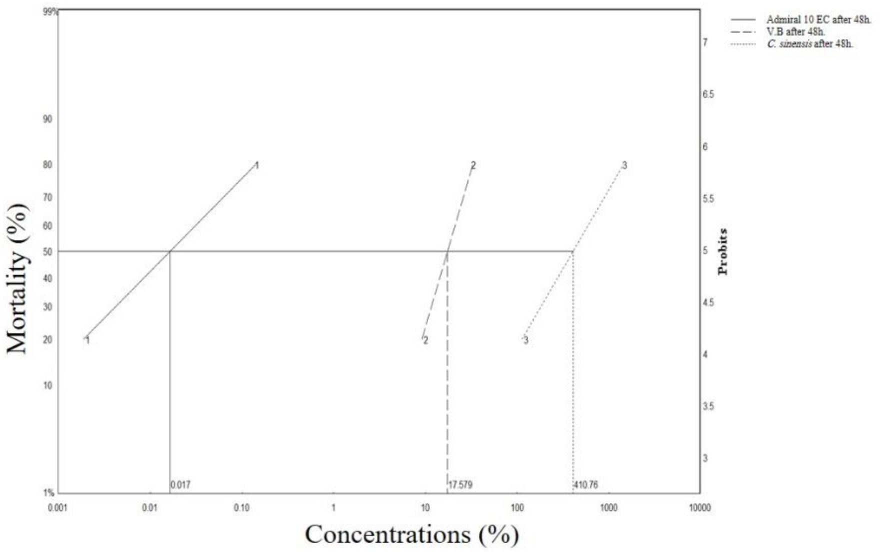
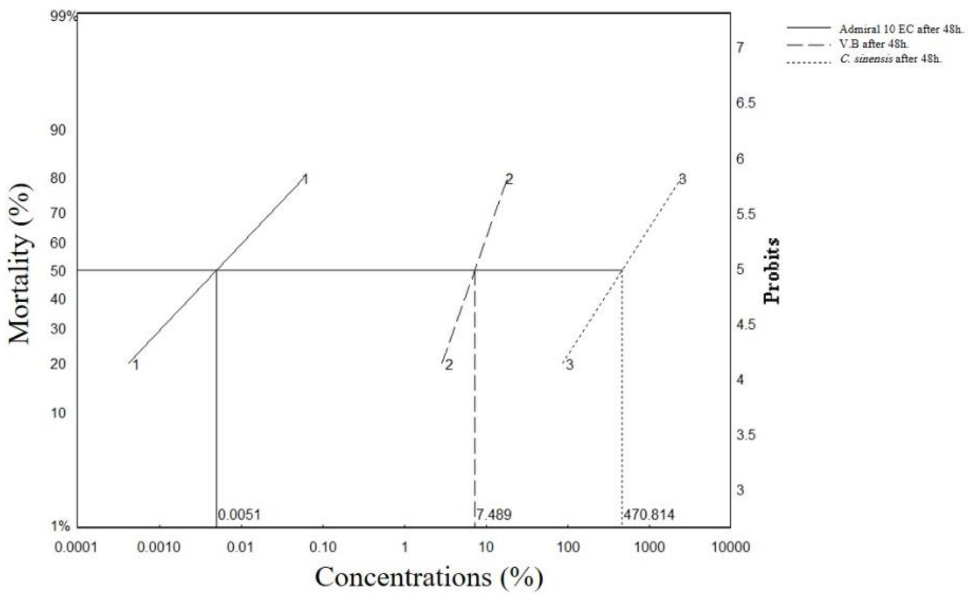


Fig 2: Comparison between the susceptibility of P.americananymphs against non-conventional insecticidesusing contact toxicity method after 48h.



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Susceptibility to botanic

Mortality of C. sinensis to P.americana adults and nymphs were recorded in Table (1 &2) and Fig. (1 and 2). There was positive correlation between mortality of C. sinensis, concentrations and exposure intervals, but the effectin the treatmentis weak for both adults and nymphs by LC_{50}'s values (410.76, 470.814) and LC_{90}'s values (2883.943, 6008.899) to adults and nymphs respectively after 48 h.

Table 1: Susceptibility adults of P.americana to non-conventional insecticidesusing contact toxicity methods after 48h.

Treatment		Parameters						
		Con. (%)	Mortality (%)	LC_{50} (%)	LC_{90} (%)	X^{2*}		Slope
						Calculated	Tabulated	
Commercial bacteria	V.B	10-50	30-96.66	17.579	46.593	9.71	11.1	3.028
Plant insecticides	C. sinensis	30-100	3.16.66	410.76	2883.943	0.659	7.8	1.514
Insect growth regulators	Admiral	0.001-0.5	3.33-0.5	0.017	0.448	7.212	11.1	0.896

LC_{50}=lethal concentration that kill 50% of the treated insects, LC_{90}= lethal concentration that kill 90% of the treated insects, U: upper limit, L: lower limit,
* X^2= Chi square, When tabulated $(Chi)^2$ larger than calculated at 0.05 level of significance indicates the homogeneity of results.

Table 2: Susceptibility nymphs of P.americana to non-conventional insecticides using contact toxicity methods after 48h.

Treatment		Parameters						
		Con. (%)	Mortality (%)	LC_{50} (%)	LC_{90} (%)	X^{2*}		Slope
						Calculated	Tabulated	
Commercial bacteria	V.B	3-30	23.33-93.33	7.489	30.645	5.0915	11.1	2.094
Plant insecticides	C. sinensis	10-100	3.33-23.33	470.814	6008.899	0.789	7.8	1.159
Insect growth regulators	Admiral	0.001-0.1	33.33-90	0.0051	0.219	6.329	9.5	0.784

LC_{50}=lethal concentration that kill 50% of the treated insects, LC_{90}= lethal concentration that kill 90% of the treated insects, U: upper limit, L: lower limit,
* X^2= Chi square, When tabulated $(Chi)^2$ larger than calculated at 0.05 level of significance indicates the homogeneity of results.

Susceptibility to insect growth regulators

Contact toxicity bioassay of Admiral 10 EC was recorded in Table (1 & 2) against P.americana adults and nymphs after 48h., and it seems clearly from the results that low concentrations exhibited high mortality to adults and nymphs. Required values, i.e. LC_{50}'s and LC_{90}'s are presented in Fig. (1 and 2). Data given summarized the susceptibility of both adults and nymphs to Admiral 10 EC. The results clearly showed that Admiral 10 EC was effective insecticide by LC_{50}'s (0.0051 %) against the nymphs comparing with adults which were LC_{50} (0.017 %). At LC_{90} level, data indicated that Admiral 10 EC (0.219 %) was effective insecticide against nymphs, while against adults gave(0.448 %) after 48 of exposure period.

Among all the non-conventional insecticides, Admiral 10 EC is the more effective insecticide and cause high mortality with low concentrations than commercial bacteria (by 1034.05, 1468.43 times), and botanic (by 24162.35, 92316.47 times), after 48 h., for adults and nymphs, respectively

DISCUSSION

The scientific community working in the field of insect pathology is experiencing an increasing academic and industrial interest in the discovery and development of new bioinsecticides as environmentally friendly pest control tools to be integrated, in combination or rotation, with chemicals in pest management programs **[29]**. The entomopathogenic bacteria domain has traditionally been well represented by members of the Bacillaceae family, such as B. thuringiensis, Lysinibacillus sphaericus, Paenibacillus spp. and Brevibacillus laterosporusLaubach [29, 30, 31]Btis famous commercial bacteria that secrete soluble toxins which are highly specific for many insects and have gained worldwide importance as an alternative to chemical insecticides [32]. Among non-conventional insecticides family. [33, 34],have reported that P.americana not significantly susceptible to Bt when orally administrated. An isolate of the bacterium Bt was also shown to induce cockroach mortality [33]. [35], reviewed that Bt accounts for 90% of the bioinsecticide market and it produces insecticidal toxins referred to as delta endotoxins and their formulations are widely used in the field against insects and has been a gradual development of insect resistance against Bt toxins.

Bt var israelensis (Bti) is a naturally occurring soil bacterium that can effectively kill mosquito [36]. Bti is much more effective against many species of mosquito and black fly larvae than any previously known bio-control agent [37], but a little data is available of Bti effects against different stages of P.americana. VectoBac (V.B) is examples of common trade names. V.B parasporal body is a gut poison, and the midgut epithelium of affected larvae is considered to be its initial site of action [38]. In addition, [39], reviewed that V.B is lethal to a wider variety of insects. In many studies, the V.B mode of action in P.americana has been studied. The current study found that V.B neuro effects on adults and nymphs. Similar effects were reported by [40], who found that V.B possessed both myotoxic and neurotoxic activity to P.americana, myotoxic effects were observed within 10–20 min whereas the onset of neurotoxic effects was considerably delayed. In another study [41], observed that V.B cause haemolytic and neurotoxic activities against P.americana.

Botanical insecticides based on plant material, plant extracts or natural products derived from plants, have been touted as potential alternatives to conventional insecticides because that the natural products have lesser environmental and human health impacts than many of conventional pesticides [42]. Research using plant extracts for controlling P.americana is limited. In the present study, Green tea (C. sinensis) was used against P.americana adults and nymphs by contact toxicity method. From the results obtained, C. sinensis extract showed weak mortality percentage. Similar to our finding using other botanicals, [43, 44, 45],found that higher concentrations of A. indica did not affect mortality to P.americana. Research using C. sinensis for controlling P.americana is very limited. Similarity to our results, [46], reported that there was a direct relation between the concentration and degree of lethal effectiveness of the active ingredient. [44], found that the larval exposure to crude extract at 250 ppm and 500 ppm for 24 h resulted in larval mortality rates of over 90 % in Anopheles gambiae and 75 % in Anopheles arabiensis. Aqueous plant extract of A. indica exhibited significant effect on feeding to tea mosquito bug [47].

The application of insect growth regulators (IGR) is one of the new biological control methods, and widely used to control pest insects [48], and they are one of the most promising alternatives to conventional insecticides for insect pest control [49]. In general, insect growth regulators induce a variety of reproductive, developmental and morphogenetic effects on insects, because of their hormonal activity [50]. However, the finding of the current study do not support the previous research.Our results found that application of Admiral 10 EC was lethal to P.americana adults and nymphs when applied in low concentrations. Much of the work regarding insect growth regulators efficacy have been done on B.germanica and other insects, however, very little data is available about the effect of insect growth regulator insecticides against different stages of P.americana in Jeddah governorate. But, many authors and authors have explain the lethal effect of these insecticides against pests and support the idea of our research. [51], indicated that hydroprene has significant biological activity through volatile action when against P.americana. [52], supported our finding, and he explain that insect growth regulator caused convulsions in P.americana when injected.

CONCLUSIONS

Commercial bacteria, botanicals and insect growth regulators can be suggested and used against different stages of P. americanaat all localities in Jeddah governorate by using contact toxicity method as a safety way for control

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