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Original article

Toxic effects of some insecticides, herbicides, and plant essential oils against *Tribolium confusum* Jacquelin du val (Insecta: Coleoptera: Tenebrionidae)

Syed Muhammad Zaka [a], Naeem Iqbal [b], Qamar Saeed [a,*], Ahmad Akrem [c], Muazzama Batool [a], Alamgir A. Khan [d], Adeel Anwar [e], Muhammadi Bibi [f], Shahbaz Azeem [a], Dur-e-Najaf Rizvi [g], Rehana Bibi [g], Khalid Ali Khan [h,i], Hamed A. Ghramh [h,i,j], Mohammad Javed Ansari [k,l], Sohail Latif [m]

[a] Department of Entomology, Faculty of Agricultural Sciences & Technology, Bahauddin Zakariya University, 60800 Multan, Pakistan
[b] Department of Plant Protection Faculty of Agricultural Sciences, Ghazi University, 32200 Dera Ghazi Khan, Pakistan
[c] Institute of Pure & Applied Biology Botany Division, Bahauddin Zakariya University, Multan 60800, Pakistan
[d] Department of Agricultural Engineering, Muhammad Nawaz Shareef University of Agriculture, Multan 60800, Pakistan
[e] Department of Agronomy, PMAS-Arid Agriculture University, Rawalpindi 46000, Pakistan
[f] Department of Botany, PMAS-Arid Agriculture University, Rawalpindi 46000, Pakistan
[g] Department of Entomology, PMAS-Arid Agriculture University, 46000 Rawalpindi, Pakistan
[h] Unit of Bee Research and Honey Production, Faculty of Science, King Khalid University, P.O. Box 9004, Abha 61413, Saudi Arabia
[i] Department of Biology, Faculty of Science, King Khalid University, P.O. Box 9004, Abha 61413, Saudi Arabia
[j] Research Center for Advanced Materials Science (RCAMS), King Khalid University, P.O. Box 9004, Abha 61413, Saudi Arabia
[k] Bee Research Chair, Plant Protection Department, College of Food and Agriculture Sciences, King Saud University, PO Box 2460, Riyadh 11451, Saudi Arabia
[l] Department of Botany, Hindu College Moradabad, 244001, India
[m] Research Farm Crops, Baldmas District Kotli, Azad Jammu and Kashmir, Pakistan

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ABSTRACT

Cereals are staple food for many countries and are grown on millions of hectares of land, but much of the harvest is wasted due to losses by pests. To minimize these losses, many pesticides are used which are damaging to the environment and human health. There are debates to get rid of these chemicals but they are still in use at large scale. An alternative control strategy for insect pests in storage houses is the use of botanicals. In this study, four plant essential oils, two plant extracts, two herbicides, and two insecticides were used against *Tribolium confusum* and the comparison of toxicity was made by calculating LC_{50} and LT_{50} values. LC_{50} values were higher for abamectin (2.09–10.23 mg/L) and cypermethrin (3.41–11.78 mg/L) insecticides followed by neem essential oil (7.39–19.24 mg/L) and citrus extract (10.14–24.50 mg/L). However, LC_{50} values were maximum in case of jaman plant extract (22.38–176.42 mg/L) followed by two herbicides, Logran (19.66–39.72 mg/L) and Topik (29.09–47.67 mg/L) However, LC_{50} values were higher for topic herbicide (24.098 ppm) and jaman essential oil (16.383 ppm) after four days of treatment. Abamectin and cypermethrin insecticides, neem essential oil and citrus plant extract also killed adults of *T. confusum* quicker as compared other essential oils, extracts and herbicides. Results revealed that botanical formulations being environmentally safe could be used instead of highly hazardous pesticides for stored products' pests. This study also elaborates the non-host toxicity of herbicides commonly applied in our agroecosystem.

1. Introduction

The world population will reach to 10 billion till 2050. To feed this increasing population more food is to be produced. One method to secure high food reserves is reducing losses by applying crop protection chemicals (Carvalho, 2017). These chemicals are very important in the cropping system to minimize food losses caused by insects,

* Corresponding author.
E-mail address: qamarsaeed@bzu.edu.pk (Q. Saeed).

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weeds, microbial diseases, and other pests (Ghorab and Khalil, 2016). These chemicals are meant for toxicity and not only cause severe pollution of the environment, but also cause an imbalance of microorganisms in the ecosystems which can degrade some toxic compounds (Garbi et al., 2006). Their residues often remain in the soil, air, water and in food commodities (Ccanccapa et al., 2016). These chemicals are converted into more toxic forms from the parent one during metabolism after entering into the body (De Gerónimo et al., 2014; Köck-Schulmeyer et al., 2014).

Excessive use of insecticides on cash crops causes the killing of important fauna, including honey bees, amphibians, small mammals, birds, and human intoxication (Köhler and Triebskorn, 2013; Paoli et al., 2015; WHO, 2017). Soon after the start of using these synthetic chemicals it was realized that they are causing toxicity not only at local but also at the global scale (Carson, 2002). Fungicides and weedicides are the main sprayed chemicals on vegetables, which are a source for acute poisoning for avian and mammalian species (Garron et al., 2009). Some pesticides (endrin, chlordane, dieldrin, heptachlor, hexachlorobenzene thiobencarb, propanil) have persistent organic pollutants (POPs) which resist degradation, remain in the environment for years and disrupt food chains. Moreover, these compounds are able to bioaccumulate and can reach up to a bioconcentration of more than 70,000 fold compared to the original concentration (Kim et al., 2017; Sapari and Ismail, 2012). Herbicide application is the primary source of soil and water pollution by surface runoff upon receiving rains soon after sprays (Miller and Spoolman, 2014). From water and soil these chemicals may volatilize during temperature changes and enter in air (Jiang and Cai, 1990). Increased applications of insecticides result in the development of resistance in insects and hence results into pest resurgence (Damalas and Eleftherohorinos, 2011). Moreover, humans are most likely to develop diseases after pesticide intoxication which include cancer, asthma, diabetes, Parkinson's disease, leukemia, endocrine disorders, and many others (Kim et al., 2017).

To minimize the effects of chemicals on non-target organisms, there is a need of time to develop some safe pesticides to attain maximum food security. An alternate approach is the use of botanicals against pests. Currently the botanicals comprise only 1% of the pesticides market (Debashri and Tamal, 2012). These botanicals may become a potential solution to synthetic chemicals, and they do not have severe side effects as they are biodegradable and hence do not interfere with ecosystems (Rajendran and Sriranjini, 2008). In this study, four essential oils and two plant extracts of botanical origin were tested against *Tribolium confusum* Jacquelin Duval (Tenebrionidae: Coleoptera) which is one of the major pests of stored grain products. The toxicity of these tested compounds was compared with two conventional insecticides and herbicides that are used extensively in our agro-ecosystems.

2. Material and methods

2.1. Insect rearing

A homogenous culture of the insects was obtained by rearing *T. confusum* on wholemeal wheat flour with 6% brewer's yeast, added to increase fecundity of female beetles. The cultures were maintained at 30 ± 5 °C and 40% ± 5% RH. The sex determination of insects was done by the method described by Halstead (1963). Seven days old adults were starved for 24 h before treatment.

2.2. Preparation of plant material

Azadirachta indica (neem), *Ricinus communis* (castor), *Eruca sativa* (arugula), *Eucalyptus globulus* (eucalyptus), *Syzygium cumini* (jaman), and *Citrus reticulata* (mandarin seeds were obtained from the local market. The essential oils of neem, castor, arugula, and eucalyptus were made by drying their seeds at 60 °C, grinding in electrical grinder and then dissolving into 80% ethanol solution by the ratio of 1: 4 (seed powder: solvent) respectively. While, in case of citrus and jaman, the peel and seeds were oven dried at 60 °C for 48 h and then grounded and mixed into solvent for the said ratio. The extracts were prepared by following the methodology of Manzoor et al. (2016). The concentrations of insecticides and herbicides were prepared in distilled water by serial dilution by following the methodology of Iqbal and Saeed (2013).

2.3. Bioassays

Four concentrations of 0.05, 0.025, 0.012, and 0 mg/L (ppm) were used for bioassays with four plant essential oils, two plant extracts, two insecticides (abamectin, cypermethrin) and two herbicides (Topik® clodinafop-propargyl, Logran® butafenacil + triasul furon). The experiment was plotted in Complete Randomized Design while having five replications for each treatment (each essential oil and extract). Petri dishes of 5 cm diameter were treated for surface film bioassay as explained by Busvine (1967). The Petri dishes were allowed to dry up for 1 h and five adults of *T. confusum* were released in each petri dish. Mortality was recorded after 24, 48, 72, and 96 h of exposure.

2.4. Statistical analyses

To compare the toxicity of the botanicals and the traditional insecticide at different doses against time, as well as the toxicity of different chemicals (LC_{50} and LT_{50}) values were calculated using probit analysis by SPSS Software (Version 10.0 for windows, SPSS Inc., Chicago, USA).

3. Results

3.1. Determination of lethal concentration against T. confusum (LC_{50})

The results showed that mortality (LC_{50}) of *T. confusum* was directly proportional to the time elapsed after treatment (Table 1). Abamectin was regarded as the most toxic insecticide with the lowest LC_{50} values after day 1 (10.23 mg/L), day 2 (8.32 mg/L), day 3 (3.80 mg/L), and day 4 (2.09 mg/L) against *T. confusum*. The second most toxic toxin was cypermethrin insecticide having LC_{50} values 11.78 mg/L, 9.58 mg/L, 4.80 mg/L, and 3.41 mg/L after 1, 2, 3, and 4 days of exposure. Among the plant essential oils and extracts, neem essential oil and citrus extract were found to be the most toxic chemicals having LC_{50} values ranging from 7.39 to 19.24 mg/L and 10.14 to 24.50 mg/L, respectively. However, the least toxic chemical was jaman plant extract having comparatively higher LC_{50} values ranging from 22.38 to 176.42 mg/L followed by the two herbicides, Logran (LC_{50} values ranging from 19.66 to 39.72 mg/L) and Topik (LC_{50} ranging from 29.09 to 47.67 mg/L) (Table 1).

3.2. Determination of lethal time against T. confusum (LT_{50})

The lethal time (LT_{50}) was reduced with the increase in concentration of pesticide. Abamectin was observed as the most potent pesticide in term of lethal time and killed 50% of *T. confusum* adults in 1.20 days at 0.05 mg/L, 2.11 days at 0.025 mg/L and 2.98 days at 0.012 mg/L concentrations. After abamectin, the most fast-acting chemical was cypermethrin having lower LT_{50} values ranging from 1.17 to 3.63 days at 0.012 to 0.05 mg/L concentrations. The LT_{50} values of plant derived botanicals were also lower or at par or with

Table 1
Toxicity (LC_{50}) of different plant extracts and pesticides against *Tribolium confusum*.

Treatment	DAT*	LC_{50} (mg/L) (95% FL)	Slope (±SE)	X^2**	DF***	P
Argula	1	38.01 (11.297–127.882)	1.36 (0.19)	0.727	1	0.39
	2	24.473 (9.952–60.181)	0.98 (0.26)	0.932	1	0.33
	3	13.234 (4.733–37.003)	1.05 (0.23)	0.931	1	0.33
	4	11.988 (3.248–15.034)	1.38 (0.17)	0.937	1	0.33
Castor	1	25.34 (20.26–69.33)	1.17 (0.22)	0.964	1	0.33
	2	19.16 (16.06–45.53)	1.35 (0.73)	0.869	1	0.35
	3	18.02 (11.65–48.77)	1.12 (0.22)	0.701	1	0.40
	4	15.32 (8.22–18.40)	1.58 (0.14)	0.853	1	0.36
Eucalyptus	1	33.503 (9.068–123.786)	0.87 (0.29)	0.956	1	0.33
	2	22.321 (7.615–65.424)	1.05 (0.23)	0.947	1	0.33
	3	16.784 (5.185–22.428)	1.51 (0.16)	0.755	1	0.38
	4	12.591 (2.844–15.994)	1.57 (0.15)	0.692	1	0.41
Neem	1	19.24 (9.70–22.99)	2.42 (0.11)	0.84	1	0.36
	2	13.12 (4.55–15.48)	1.90 (0.12)	0.898	1	0.34
	3	9.35 (1.59–7.06)	1.43 (0.16)	0.785	1	0.38
	4	7.39 (1.61–3.56)	3.34 (0.08)	0	0	0.00
Citrus	1	24.50 (14.52–30.294)	2.08 (0.13)	0.875	1	0.35
	2	16.01 (12.95–19.830)	2.25 (0.12)	0.753	1	0.39
	3	12.06 (5.41–18.32)	2.37 (0.10)	0.54	1	0.46
	4	10.14 (6.49–14.65)	1.43 (0.16)	0.911	1	0.34
Jaman	1	167.42 (33.85–828.06)	1.14 (0.35)	0	0	0.00
	2	42.76 (33.45–146.81)	0.98 (0.27)	0.798	1	0.37
	3	26.92 (19.04–77.61)	1.17 (0.23)	0.705	1	0.40
	4	22.38 (14.12–43.89)	1.12 (0.22)	0.939	1	0.33
Topik	1	47.67 (15.03–141.45)	1.73 (0.17)	0.908	1	0.34
	2	36.72 (13.69–66.75)	1.36 (0.20)	0.999	1	0.32
	3	34.93 (11.37–54.63)	0.87 (0.29)	0.806	1	0.37
	4	29.09 (9.66–102.54)	0.75 (0.32)	0.826	1	0.36
Logran	1	39.72 (34.69–66.75)	1.73 (0.17)	0.908	1	0.34
	2	26.14 (19.76–88.03)	1.36 (0.20)	0.999	1	0.32
	3	24.93 (15.37–54.63)	0.93 (0.26)	0.794	1	0.37
	4	19.66 (14.59–28.71)	1.35 (0.18)	0.609	1	0.44
Abamectin	1	10.23 (5.94–20.41)	1.86 (0.14)	0.842	1	0.36
	2	8.32 (5.46–29.35)	1.32 (0.18)	0.588	1	0.44
	3	3.80 (1.37–10.54)	1.03 (0.22)	0.872	1	0.35
	4	2.09 (1.32–3.28)	2.88 (0.10)	0	0	0.00
Cypermethrin	1	11.78 (3.26–35.69)	0.88 (0.26)	0.901	1	0.34
	2	9.58 (3.49–26.29)	1.05 (0.22)	0.857	1	0.35
	3	4.80 (1.99–11.52)	1.19 (0.19)	0.677	1	0.41
	4	3.41 (1.83–6.32)	1.75 (0.14)	0.826	1	0.36

Note: LC_{50} = lethal concentration to kill 50% of population.
* DAT = days after treatment.
** = chi square.
*** = degree of freedom; SE = standard error; P = probability; FL = fiducial limits.

the synthetic pesticides. The results revealed that neem essential oil killed adults of *T. confusum* quicker with lower LT_{50} values among the plant based botanicals (1.99–4.17 days at 0.012–0.05 mg/L concentrations). Among the plant derived toxins, jaman plant extract took more time to kill 50% of adults at all concentrations i.e. 10.86 days, 7.02 days, 5.04 days at 0.012, 0.025 mg/L and 0.05 mg/L concentrations, respectively. Topik herbicide was regarded as the most slow-acting toxins in term of lethal time to kill 50% of the adults of *T. confusum* having LT_{50} values of 11.41 days, 10.70 days, 6.49 days at 0.012, 0.025, and 0.05 mg/L concentration (Table 2).

4. Discussion

Insecticides are extensively used in grains storage and field application to secure maximum food. They are formulated as more toxic with lower degradation and increased potency. Using such chemicals for insect control in food commodity is highly dangerous (Thatheyus and Selvam, 2013). In this study insecticide cypermethrin and abamectin were used against *T. confusum* and their results were compared with the toxicity values of plant-based essential oils and extracts. The results suggested although abamectin and cypermethrin insecticides caused higher mortalities in less time but the plant essential oils also showed promising results. Moreover, plant botanicals could be proposed as environmental and human health safe pesticide and this is also confirmed by Khan and Ahmed (2003).

Plant extracts have attained importance due to their insecticidal potential of a future insecticide market. In our present study, the essential oil of *A. indica, R. communis, E. sativa, E. globules*, plant extracts of *S. cumini*, and *C. reticulate* were also extensively toxic against the beetle and proved to be good alternates of toxic chemicals for control of stored grain pests as described by Bekele et al. (1996), Bouda et al. (2001), and Hassanali et al. (1990). Plant extracts pose repellence or killing by showing mortality at all life stages against various stored-product insects Desmarchelier (1994).

El-Nahal et al. (1989) studied the toxic effect of essential oil on adults of five stored-product insect species: showing sensitivity against *Callosobruchus chinensis*, *Sitophilus granarius* and *Sitophilus oryzae*, *T. confusum* and *Rhyzopertha dominica*. In their study, time

Table 2
Lethal time (LT_{50}) of different plant extracts and pesticides against *Tribolium confusum*.

Treatment	Conc. (mg/L)	LT_{50} (95% FL) (days)	Slope (±SE)	X^2*	DF**	P
Argula	0.05	2.64 (1.34–5.19)	1.34 (0.15)	0.862	2	0.65
	0.025	5.12 (2.18–12.02)	1.09 (0.18)	0.915	2	0.63
	0.012	7.05 (3.48–14.24)	1.41 (0.15)	0.969	2	0.62
	Control	48.65 (13.93–169.85)	1.02 (0.27)	0.968	2	0.62
Castor	0.05	2.32 (1.08–4.96)	1.19 (0.16)	0.883	2	0.64
	0.025	6.55 (2.45–17.49)	0.95 (0.22)	0.841	2	0.66
	0.012	11.41 (4.01–27.00)	1.03 (0.21)	0.933	2	0.63
	Control	166.90 (27.53–1011.13)	0.66 (0.39)	0.911	2	0.63
Eucalyptus	0.05	3.02 (1.08–3.77)	1.46 (0.14)	0.905	2	0.64
	0.025	5.39 (1.99–20.52)	0.79 (0.25)	0.919	2	0.63
	0.012	10.48 (3.51–31.31)	0.87 (0.24)	0.95	2	0.62
	Control	48.65 (13.93–169.85)	1.02 (0.27)	0.968	2	0.62
Neem	0.05	1.99 (0.54–1.82)	1.59 (0.13)	0.134	1	0.71
	0.025	4.92 (1.33–2.75)	2.65 (0.08)	0.771	2	0.68
	0.012	4.17 (2.25–4.47)	2.87 (0.07)	0.91	2	0.63
	Control	21.67 (8.62–54.49)	1.36 (0.20)	0.361	1	0.55
Citrus	0.05	2.43 (0.80–2.56)	1.61 (0.12)	0.923	1	0.34
	0.025	4.51 (1.36–4.62)	1.49 (0.13)	0.989	2	0.61
	0.012	5.39 (3.32–8.75)	2.14 (0.11)	0.411	2	0.81
	Control	16.35 (7.59–35.19)	1.92 (0.17)	0.618	1	0.43
Jaman	0.05	5.04 (2.41–6.77)	1.85 (0.11)	0.951	2	0.62
	0.025	7.02 (3.76–13.12)	1.64 (0.13)	0.948	2	0.62
	0.012	10.86 (4.39–14.08)	1.89 (0.12)	0.911	2	0.63
	Control	91.33 (21.18–393.76)	0.95 (0.32)	0.873	2	0.65
Topik	0.05	6.49 (1.88–16.03)	0.85 (0.24)	0.952	2	0.62
	0.025	10.70 (4.44–17.03)	1.03 (0.21)	0.933	2	0.63
	0.012	11.41 (4.01–27.00)	1.58 (0.15)	0.879	2	0.64
	Control	563.87 (63.32–5.21.63)	0.65 (0.48)	0.928	2	0.63
Logran	0.05	3.63 (1.49–9.39)	0.95 (0.21)	0.919	2	0.63
	0.025	8.73 (3.63.37.783)	0.82 (0.25)	0.972	2	0.62
	0.012	9.95 (4.367–22.697)	1.23 (0.18)	0.914	2	0.63
	Control	166.91 (27.53–1011.10)	0.66 (0.39)	0.911	2	0.63
Abamectin	0.05	1.20 (0.63–2.25)	1.48 (0.14)	0.509	1	0.48
	0.025	2.11 (1.52–2.93)	2.93 (0.07)	0.512	2	0.77
	0.012	2.98 (2.05–4.33)	2.59 (0.08)	0.931	2	0.63
	Control	202.80 (32.71–1257.28)	0.71 (0.40)	0.266	1	0.61
Cypermethrin	0.05	1.17 (0.67–2.04)	1.74 (0.12)	0.967	2	0.62
	0.025	2.51 (1.36–4.62)	1.49 (0.13)	0.989	1	0.32
	0.012	3.63 (1.57–8.39)	1.09 (0.18)	0.977	2	0.61
	Control	27.67 (9.99–76.67)	1.27 (0.23)	0.313	1	0.58

LT_{50}= lethal time to kill 50% of population.
* = chi square.
** = degree of freedom; SE = standard error; P = probability; FL = fiducial limits.

elapsed after treatment was an essential factor for toxicity studies, which supports our results. Aslam et al. (2002) investigated the insecticidal potency of five difference spices, i.e. clove (*Syzygium aromaticum*), black pepper (*Piper nigrum*), cinnamon (*Cinnamomum zeylanicum*), red pepper (*Capsicum annum*), and black cardamom (*Amomum subulatum*) against pulse beetle (*Callosobruchus chinensis*) a stored grain insect pest. In their study, number of days to 100% mortality after treatment was an essential factor for toxicity studies, our results are also comparable to their findings.

This study supports that the plant essential oils and extracts could give a good control measure against stored grain insect pests as results demonstrate the mortality level of plant products treated units and insecticide treated units. This is an alternative environmentally safe and health ensuring measure to treat stored grains instead of hazardous chemicals as plant extracts being natural, are biodegradable and do not interfere with the ecosystem. Molecules present in plants which are responsible for poisonous, repellency, ovicidal, and antifeedant effects are terpenes, terpenoids, cyanates, sulfur compounds, alkaloids and others (Coats et al., 1991). Essential oils of plant have many different chemicals which pose insecticidal effect, for example, cineole in eucalyptus oil, limonene in citrus, azadirachtin in neem (Rajendran and Sriranjini, 2008). These components being biomolecules are nature friendly and could be used as an insecticide to get rid of side effects of the synthetic chemicals, i.e., insecticides, fungicides, and weedicides as this study confirms the direct toxicity of weedicides to the insects.

5. Conclusions

Plant essential oils have caused higher mortalities of *T. confusum*. Plant essential oils can provide better alternatives to highly toxic and hazardous chemicals for the management of stored grain pests. In addition, their use will be safer to the public health and the environment.

Conflict of interest

The authors disclose that there is no any conflict of interest in connection with submitted manuscripts.

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