Kuwait J. Sci. **43 (3)** pp. 128-134, 2016

Heterotermesindicola (Wasmann) (Isoptera: Rhinotermitidae) responses to extracts from three different plants

Farkhanda Manzoor[1,*], Abeer Zafar[2], Irfana Iqbal[3]

[1]*Dept. of Zoology, Lahore College for Women University, Lahore, Pakistan*

[2]*Dept. of Environmental Science, Lahore College for Women University, Lahore, Pakistan*

[3]*Dept. of Zoology, Lahore College for Women University, Lahore, Pakistan*

[*]*Corresponding author: doc_farkhanda@yahoo.com*

Abstract

Termites *Heterotermes indicola* (Wasmann) are destructively dagamging insects that cause massive harm to timber, resulting in loss of billions of dollars in property damage. Various chemical antitermite solutions are available that cause environmental contamination and are also hazard to human health. Extracts from 3 plants *Cedrus deodara* (Pinaceae), *Betula utilis* (Betulaceae) *and Syzygium cumini* (Myrtaceae) were tested for repellent, antifeedant and their toxicity against *Heterotermes indicola* (Wasmann). Our results show that feeding and mean percentage mortality varied not only with the plant species, but alsowith the type of the extraction solvents. Hexane extract of *C. deodara* showed the highest termite mortality rate. *H.indicola* consumed more ethanolic extract treated/untreated filter paper than hexan treated untreated filter paper with various plant extracts.These results show that hexane extracts of the plants were comparatively more toxic and repellent against *H.indicola* (Wasmann), since insects consumed mimimum filter paper treated with all three plant extracts. Results show that for all three plants, hexane extracts had optimum anti-termite activity.

Keywords: Betulautilis; *Cedrusdeodara*; *Heterotermesindicola*; *Syzygiumcumini*; toxicity.

1. Introduction

Natural environment and human health has been seriously impacted by indiscriminate use of synthetic pesticides. Insecticides, organophosphates, carbamates and pyrethroidsare currently in use for plant protection (crops, vegetable, fruits) for the past several decades. The excessive use of commercial pesticides has resulted in contamination of human food chain, awide spread resistance/tolerance in targeted pest and noxious effect on field animals (Maehashi & Fujiyoshi, 1972; Bania & Polanowski, 1999). For example more than 27 insect species have become resistant to *Bacillus thuringiensis* toxins (Siegwart *et al.*, 2015). In addition, synthetic pesticides are reported to be hazardous for beneficial insects as well as for the plant itself where plant defense genes are suppressed by insecticides (El-Zahi & Abd-Elhady, 2013; Szczepaniec *et al.*, 2013; Lanka *et al.*, 2014). Another major hazard for the use of synthetic insecticides is the involuntary exposure of field wokers causing severe injury (Reiss *et al.*, 2015). Birth outcome and neuro-developmental abnormalities as a result of exposure to organophosphorus insecticides have been commonly reported in field workers (Reiss *et al.*, 2015).

Insecticides are not easily degradable materials and they remain in the environment for a long period of time. If insecticides are frequently altered, these can react with residual insecticides in the soil and have synergistic effects attenuating insecticidal activity and also modify soil chemistry and their toxicity (Ahmed *et al.*, 2015; Casida, 2015; Mohiddin *et al.*, 2015). In addition, an extensive use of chemical insecticides may impart resistance to termites causing extensive damage to wood. Termites resistance to insecticides has been explored by analysis of the differencially expressed genes (Zhang *et al.*, 2013). For this reason, bio-based substitutes are being explored for their eco-friendly nature.

At least 45 termites species belonging to four familis are reported world wide, but these species are difficult to characterize since there are little morphological differences in these species (de Figueiredo *et al.*, 2015; Garrick *et al.*, 2015). Mitochondrial cytochrome oxidase subunit II gene has been used in PCR to differenciate between termite species (Garrick *et al.*, 2015). Termites cause extensive damage to organic matter and annually these are reported to cause a billion dollar property damage (Kirker *et al.*, 2015).

The international scenario has led to a good deal of work about the establishment and development of plant based pesticides (Pathak *et al.*, 2000; Verma *et al.*, 2006; Siddiqui *et al.*, 2009). Thus, there is a great need to develop an environmentally sustainable system to control pests that are not as much dependent on chemical pesticides as the primary management tool (Chandler *et al.*, 2008).

An effective management of pest control has been investigated by plant-derived products such as essential oils. Insecticides of plant origin are reported to antagonize toxicity of chemical insecticides. Therefore, different plants and their derived products can be used as sustainable source of environmentally friendly botanical pesticides. Botanical insecticides/pesticides have several advantages over synthetic pesticides. Firstly these are eco-friendly, secondly these are easily available, also cost-effective and most importantly they are biodegradable (Dubey *et al.*, 2008).

Higher plants producea broad spectrum of secondary metabolites including polyphenols, tannins, quinones, alkaloids, essential oils, sterols, saponins etc. and these phytochemicals show diverse biological properties as insecticides/pesticides. Toxicity to termites depends on the type of the phytotoxicant, its maximal toxicant transfer, exposure time and toxicant recipient ratios (Neoh *et al.*, 2014). Keeping in view the significance of extracts as antitermitic agent, the present study was carried out to investigate the extracts of local plants possessing natural metabolites that can be used fortermite management in Pakistan.

2. Materials and methods

2.1. Preparation of plant extracts

The bark, sapwood and heart wood of *Cedrus deodara* (Pinaceae), *Betula utilis* (Betulaceae) *and Syzygium cumini* (Myrtaceae) were collected from Jallo forest near Lahore and brought to entomology research laboratory, Lahore College for Women University (LCWU), Pakistan. Fresh plant leaves were collected for identification of the wood by our taxonomist in LCWU and specimens labeled as SM-1200, were deposited in the university herbarium. Plant leaves and stems of *C. deodara, Z. cumini*and *B. utilis* were dried under shade at room temperature and powered using Wiley mill with 1mm mesh screen. Air tightglass bottles were used for storage of the ground samples. 20 g of the wood powder was soxhlet extracted with n-hexane followed by 95% ethanol. Since water maintenance and humidity are essential for survival of pests (Janei *et al.*, 2015), percentage of moisture content in soil placed in petri dishes was calculated using the following formula:

$$\%\ Soil\ moisturecontent\ = \frac{Weight\ loss\ in\ soil\ after\ drying}{Weight\ of\ moisted\ soil\ before\ drying} x\ 100$$

2.2. Forced feeding tests

In order to study the toxicity of plant extracts, termite species *H. indicola* (Wasmann) were used for bioassays. Mature termites were maintained in the laboratory at 26±2^{o}C with 80% relative humidity. Only active and healthy workers were used for experimental purposes. Bioassays were conducted by forced feeding tests (Hardy *et al.,* 2004; Habibpour, 2010). Sterilized glass petri dishes, measuring 5.5 Cm diameter with 2 g of sterilized moist soil, placed at the bottom of the petri dish, were used. Sterilized preweighed Whatmanfilter paper #1 was placed over the soil at the bottom of each petri dish. The filter paper was impregnated with the hexane and ethanolicplant extract solution of known concentration. The organic solvent was thoroughly evaporated with hot air blower, before placing it in the petri dishes. In control experiments, a preweighed filter paper, impregnated with pure solvent without the wood extract was placed in the petridishs, after eveaporating the organic solvent. Three replicates of each extract were used. Ten healthy termite worker insects were placed in each petri dish and maintained at at 26±2^{o}C and 80% relative humidity. Mortality of termites was observed for an average 7 days and dead individuals were removed on daily basis.

2.3. Repellency tests

Difference repellency test has been used to control pests (Bissinger *et al.*, 2016; Krause Pham & Ray, 2015). In our study, repellency of the three plant extracts in two different solvents, 95% ethanol, and hexane, was assessed. 2 g of soil was placed at the bottom of a petri dish and moistened with distilled water to maintain 20% humidity.

Whatman # 1 filter paper was cut into two halves and each half was weighed. One half of the filter paper was treated with hexane extract and the other half was treated with 95% ethanol extract of known concentrations as spelled out in section 2.2. Control petridishes received dried filter paper impregnated with only organic solvent. Ten healthy workers were placed in each petri dish and maintained at 26±2^{o}C and 80% relative humidity. The mean number of termites on extract-treated filter paper with reference to blank filter paper in choice feeding test were recorded and weight losses (mg) of extract treated and blank filter paper were also recorded after a period of 7 days.

2.4. Data analysis

The data were analyzed using one way ANOVA test and paired comparison t-test using graphpad prism version 4.00 for windows. Results with $P<0.05$ were considered statistically significant.

3. Results and discussion

The powdered palnt material was soxhlet extracted in two different solvents. A known volume and concentration of the extract was used in bioassays. Weight of extractable material from each plant, in different solvents is given in (Table 1). *S. cumini* yielded 1.8% ethanolic extract, while it yielded 1.55% extract with n-hexane. The percentage of other extracts is shown in Table 1. The ethanolic extract for all plants yielded greater percentage of extactable material, presumeably most secondary products were polar and were soluble in ethanol. In addition some primary metabolites such as sugars and amino acids could also be present in ethanolic extract of plants.

Table 1. Plant extracts and their weights in different solvents

Plant species	Solvent	Weight of Extract (mg)	Yield (%)
Syzygium cumini	Ethanol	360.0	1.8
	Hexane	310.0	1.55
Betula utilis	Ethanol	640.0	3.2
	Hexane	400.0	2
Cedrus deodara	Ethanol	1630.0	8.15
	Hexane	660.0	3.3

Table 2 shows the mean percentage mortality of *H. indicola* (Wasmann) workers treated with ethanolic and hexane extracts. With hexane extract, highest mortality (66.66±3.05) was recorded for *C. deodara.* However, the ethanolic extract was relatively less toxic for pests. Table 2 shows repellency of different extracts for *H. indicola*. These results suggest equal toxicity for ethanolic extracts of *B.utilis* and *S.cumini*, while n-hexane extracts of these two plants were more toxic than ethanolic extracts and were also unequal in their toxicity. When tested for repellency, the ethanolic extract of *C. deodara* and *B. utilis* showed equal repellency, but *S. cumini* had higher repellency than the other two plants. Repellency linearly increased in hexane extracts of the three plants, *C. deodara* being the most repellent (Table 2). Differences in toxicity and repellency to termites may be due to the crude extract composition extracted in different organic solvents.

Table 2. Percentage mortality ($\overline{X}$±SEM) of *Heterotermes indicola* (Wasmann) when exposed to ethanol or hexane extract of *Cedrus deodara, Betula utilis* and *Syzygium cumini* and mean number of termites ($\overline{X}$±SEM) on extract-treated filter paper with reference to untreated filter paper in choice feeding test.

[1]Plant Species	[2]Extraction Solvent Mean %age Mortality ($\overline{X}$±SEM)		Repellency score ($\overline{X}$±SEM) Mean # of termites	
	Hexane	Ethanol	Ethanol	Hexane
Cedrus deodara	66.66±3.05	61.66±2.90	2.00±0.88	6.00±2.02
Betula utilis	56.66±3.5	51.55±3.71	2.00±0.88	5.00±2.30
Syzygium cumini	56.66±3.20	48.88±3.71	3.00±0.80	4.00±1.73
Control	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00

[1]Mean of three replicates, 10 worker termites in each.
[2]Extracts prepared by sequential extraction of plant with hexane, ethanol

Figure 1 shows the weight of mean consumption of the blank filter paper compared with the extract treated filter papers for all three plants. Filter paper consumption could be taken as a measure of toxicity and replellency by the respective extact.The consumption (mg) were noticed as (28.0±0.003), (28.0±0.008) and (30±0.008) in filter paper treated with n-hexane extracts of *C.deodara, B.utilis* and *S.cumini* respectively.The consumption of untreated filter paper (control) was 32.6 mg. While filter paper treated with *C.deodara, B.utilis* and *S.cumini* with alcoholic extract showed maximum filter paper consumption as shown in Figure 1. Indicating minimal toxicity and repellency of the components present in ethanolic extract.

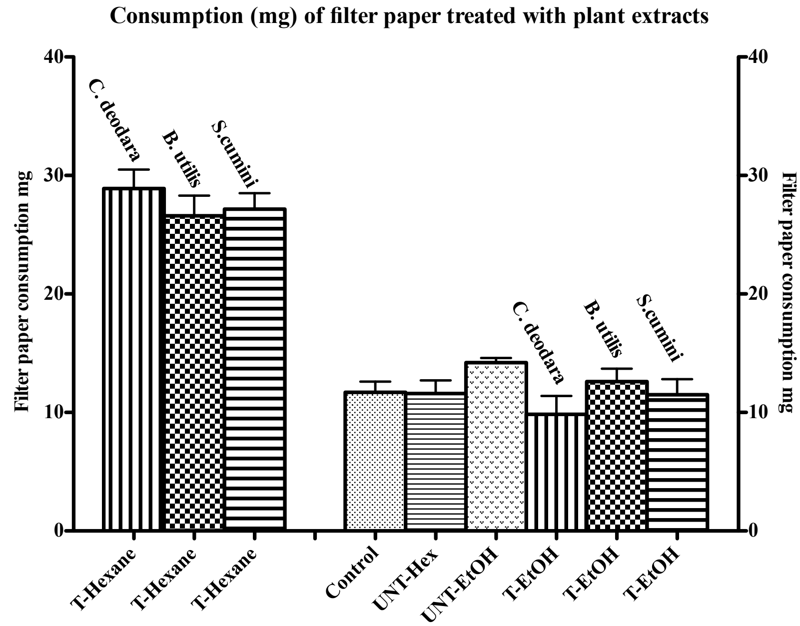

Fig. 1. Mean weight of filter paper consumed (mg) after extract-treated[a] and untreated filter paper in choice feeding tests[b] for one week by *Heterotermes indicola* (Wasmann). UNT = untreated with extract; T = treated with extract; Hex = hexane extract; EtOH = ethanol extract

Our results show that *C. deodara* (Pine) has higher pesticidal activity compared with *B. Utilis* and *S. cumini,* n-hexane extracts. A hydro distillation and GC analyses of *C. deodara* yielded 34 components representing 98.3% of the pine needle oil. It is known that pine oil contains 45.8% β-himachalene as its major volatile component, which shows LD50 1.08mg/mL against larvae of dimond balck moth (Reddy *et al.*, 2015). The other minor components of this plant have been identified as α-terpenol (1), limonene (2), linalool (3), anethole (4), eugenol (5), and bicyclic sesqueterpene cryophyllene Figure 2. A considerable antioxidant/prooxidant activity of the crude extract of *C. deodara* has been reported and it may cause pesticidal activity of this plant (Zeng *et al.*, 2012).

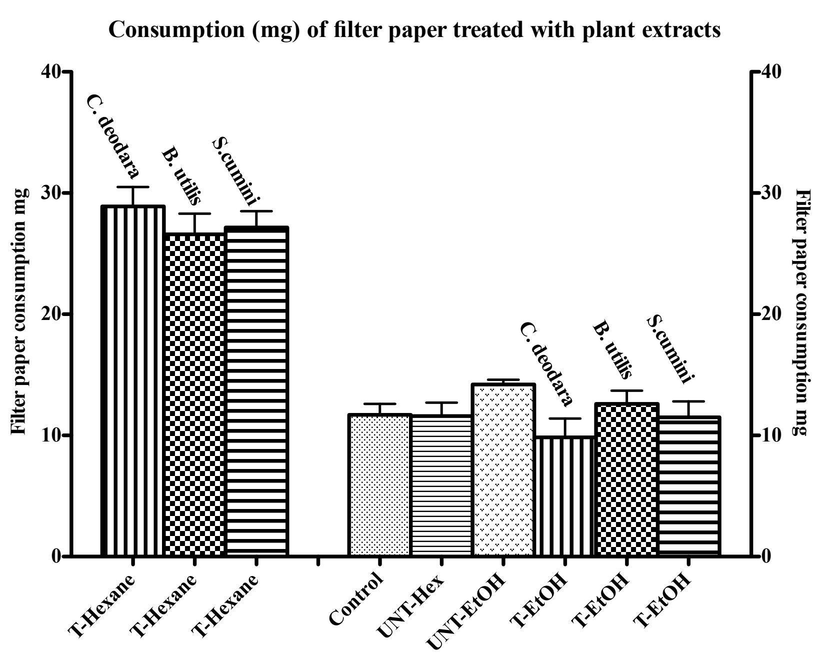

Fig. 2. Mean weight of filter paper consumed (mg) after extract-treated[a] and untreated filter paper in choice feeding test[sb] for one week by *Heterotermes indicola* (Wasmann). UNT = untreated with extract; T = treated with extract; Hex = hexane extract; EtOH = ethanol extract

A phytochemical analysis of *B.utilis* by GC/HPLC and GC/MS has shown the presence of at least five marker threprapeutic compounds in this plant.These are identified as botulin, lupeol, oleanolic acid, 3-acetyloleanolic acid, and β- sitosterol (Khan *et al.*, 2012). The third plant included in this study was *S.cumini* shows antileishmanial activity and cytotoxicity (Ribeiro *et al.*, 2014). This plant is known to yield four ellagic acid derivatives with antiplasmodial activity while its antidiabetic activity has also been reported (Simoes-Pires *et al.*, 2009; Tripathi & Kohli, 2014). The plant is rich in anthocycnins, ellagic acid, isoquercetin, Kaemferol, myrecetin and lignin derivatives (Mir *et al.*, 2009).

Survival and feeding responses of termites to plant extracts is becoming increasingly important in the wood preservation industry because there has been a greater focus in decreasing the potential leaching of chemicals into the environment. From the results obtained in this study, it is concluded that *C.deodara* n-hexane extract had higher termiticidal potential compared with *B. utilis* and *S. cumini* (Table 2). The later two extracts showed reduced mortality of insects. Our results confirm that the termicidal activity in all these pants was present in volatile oil fraction extracted with n-hexane that primarily contained terpenoid and related molecules (Kusumoto *et al.*, 2009).

4. Conclusion

Plant extracts showed antitermitic activity due to their repellant action and toxicity due to their terpenoid and volatile components. The results of this study show that for extraction of antitermitic components n-hexane is a suitable solvent. The antitermitic coumpounds being phytochemicals, have an excellent potential as environmentally safe, cost effective and degradeable compounds and can be reliably used indiscriminately for different types of wood preservation.

References

Ahmed, M.A., Vogel, C.F. & Matsumura, F. (2015). Unique biochemical and molecular biological mechanism of synergistic actions of formamidine compounds on selected pyrethroid and neonicotinoid insecticides on the fourth instar larvae of Aedes aegypti (Diptera: Culicidae). Pesticide Biochemistry and Physiology, **120**(1):57-63.

Bania, J. & Polanowski, A. (1999). Bio-insecticides and insect defense mechanisms. Postepy Biochemii Journal, **45**(2):143-150.

Bissinger, B.W., Kennedy, M.K. & Carroll, S.P. (2016). Sustained efficacy of the novel topical repellent TT-4302 against mosquitoes and ticks. Medical and Veterinry Entomology, **30**(1):107-111. doi:10.1111/mve.12151

Casida, J.E. (2015). Golden age of RyR and GABA-R diamide and isoxazoline insecticides: common genesis, serendipity, surprises, selectivity, and safety. Chemical Research in Toxicology, **28**(4):560-566.

Chandler, D., Davidson, G., Grant, W., Greaves, J. & Tatchell, G. (2008). Microbial biopesticides for integrated crop management: an assessment of environmental and regulatory sustainability. Trends in Food Science & Technology, **19**(5):275-283.

De Figueiredo, R.E.C., Vasconcellos, A., Policarpo, I.S. & Alves, R.R. (2015). Edible and medicinal termites: a global overview. Journal of Ethnobiology and Ethnomedicine **11**(2):29. doi:10.1186/s13002-015-0016-4

Dubey, N., Srivastava, B. & Kumar, A. (2008). Current status of plant products as botanical pesticides in storage pest management. Journal of Biopesticides, **1**(2):182-186.

El-Zahi, Z.S. & Abd-Elhady, H.K. (2013). Insect predators and control of *Aphis gossypii* comparing to certain insecticides under caged-cotton plants conditions. Pakistan Journal of Biological Sciences: PJBS **16**(5): 233-238.

Garrick, R.C., Collins, B.D., Yi, R.N., Dyer, R.J. & Hyseni, C. (2015). Identification of Eastern United States Reticulitermes Termite Species via PCR-RFLP, Assessed Using Training and Test Data. Insects, **6**(2): 524-537. doi:10.3390/insects6020524

Habibpour, B. (2010). Laboratory evaluation of Flurox, a chitin synthesis inhibitir, on the termite, Microcerotermes diversus. Journal of Insect Science, **10**:2. doi:10.1673/031.010.02201.

Hrdy, I., Kuldova, J. & Wimmer, Z. (2004). Juvenogens as potential agents in termite control:laboratory screening. Pest Management Science, **60**(10):1035-1042. PMID 15481831.

Janei, V., Lima, J.T. & Costa-Leonardo, A.M. (2015). Rehydration after water stress in Forager workers of *Coptotermes gestroi* (Wasmann) (Blattaria: Rhinotermitidae). Neotropical Entomology, **44**(3):301-307. doi:10.1007/s13744-015-0285-6.

Khan, I., Sangwan, P.L., Dhar, J.K. & Koul, S. (2012). Simultaneous quantification of five marker compounds of *Betula utilis* stem bark using a validated high-performance thin-layer chromatography method. Journal of Separation Science, **35**(3):392-399.

Kirker, G.T., Bishell, A.B. & Lebow, P.K. (2015). Laboratory evaluations of durability of Southern pine pressure treated with extractives from durable wood species. Journal of Economic Entomology, **109**(1):259-266. doi:10.1093/jee/tov286.

Krause, Pham, C. & Ray, A. (2015). Conservation of olfactory avoidance in Drosophila species and identification of repellents for *Drosophila suzukii*. Scientific Reports, **5**:11527. doi:10.1038/srep11527.

Kusumoto, N., Ashitani, T., Hayasaka, Y., Murayama, T. & Ogiyama, K. *et al.* (2009). Antitermitic activities of abietane-type diterpenes from *Taxodium distichum* cones. Journal of Chemical Ecology, **35**(6):635-642.

Lanka, S.K., Stout, M.J., Beuzelin, J.M. & Ottea, J.A. (2014). Activity of chlorantraniliprole and thiamethoxam seed treatments on life stages of the rice water weevil as affected by the distribution of insecticides in rice plants. Pest Management Science, **70**(2):338-344.

Maehashi, H. & Fujiyoshi, N. (1972). Transient bacillemia by bio-insecticides in chronic feeding test on mice (*Bacillus moritai* series no. 2). *Nihon Eiseigaku Zasshi,* **27**(3):342-346. PMID:4677423.

Mir, Q.Y., Ali, M. & Alam, P. (2009). Lignan derivatives from the stem bark of *Syzygium cumini* (L.) Skeels. Natural Product Research,

23(5):422-430.

Mohiddin, G.J., Srinivasulu, M., Maddela, N.R., Manjunatha, B. & Rangaswamy, V. *et al*. (2015). Influence of the insecticides acetamiprid and carbofuran on arylamidase and myrosinase activities in the tropical black and red clay soils. Environmental Monitoring and Assessment, **187**(6):388. doi:10.1007/s10661-015-4631-2.

Neoh, K.B., Yeoh, B.H. & Lee, C.Y. (2014). Mortality patterns in *Coptotermes gestroi* (Blattodea: Rhinotermitidae) following horizontal transfer of nonrepellent and repellent insecticides: effects of donor:recipient ratio and exposure time. Journal of Economic Entomology, **107**(4):1563-1572. PMID:25195449.

Pathak, N., Mittal, P., Singh, O., Sagar, D.V. & Vasudevan, P. (2000). Larvicidal action of essential oils from plants against the vector mosquitoes *Anopheles stephensi* (Liston), Culex quinquefasciatus (Say) and *Aedes aegypti* L. International Pest Control, **46**(2):53-55.

Reddy, S.G., Kirti Dolma, S., Koundal, R. & Singh, B. (2015). Chemical composition and insecticidal activities of essential oils against diamondback moth, *Plutella xylostella* (L.) (Lepidoptera: Yponomeutidae). Natural Product Research, 1-5. PMID:26264423.

Reiss, R., Chang, E.T., Richardson, R.J. & Goodman, M. (2015). A review of epidemiologic studies of low-level exposures to organophosphorus insecticides in non-occupational populations. Critical Reviewes in Toxicology, **45**(7):531-641.

Ribeiro, T.G., Chavez-Fumagalli, M.A., Valadares, D.G., Franca, J.R. & Lage, P.S. *et al*. (2014). Antileishmanial activity and cytotoxicity of Brazilian plants. Experimental parasitology, **143**:60-68.

Siddiqui, B.S., Ali, S.T., Rajput, M.T., Gulzar, T. & Rasheed, M. *et al*. (2009). GC-based analysis of insecticidal constituents of the flowers of *Azadirachta indica* A. Juss. Natural Product Research, **23**(3):271-283.

Siegwart, M., Graillot, B., Blachere Lopez, C., Besse, S. & Bardin, M. *et al*. (2015). Resistance to bio-insecticides or how to enhance their sustainability: a review. Frontiers in Plant Science, **6**:381. doi:10.3389/fpls.2015.00381.

Simoes-Pires, C.A., Vargas, S., Marston, A., Ioset, J.R. & Paulo, M.Q. *et al*. (2009). Ellagic acid derivatives from *Syzygium cumini* stem bark: investigation of their antiplasmodial activity. Natural product communications, **4**(10):1371-1376.

Szczepaniec, A., Raupp, M.J., Parker, R.D., Kerns, D. & Eubanks, M.D. (2013). Neonicotinoid insecticides alter induced defenses and increase susceptibility to spider mites in distantly related crop plants. PloS one, **8**(5):e62620.

Tripathi, A.K. & Kohli, S. (2014). Pharmacognostical standardization and antidiabetic activity of *Syzygium cumini* (Linn.) barks (Myrtaceae) on streptozotocin-induced diabetic rats. Journal of Complementary & Integrative Medicine, **11**(2):71-81.

Verma, P.R., Subburaju, T. & Balakrishnan, N. (2006). Larvicidal activity of *Artemisia nilagirica* (Clarke) Pamp. and *Ocimum sanctum* L.: a preliminary study. Journal of Natural Remedies, **6**(2):157-161.

Zeng, W.C., Zhang, Z., Gao, H., Jia, L.R. & He, Q. (2012). Chemical composition, antioxidant, and antimicrobial activities of essential oil from pine needle (*Cedrus deodara*). Journal of Food Science, **77**(7):C824-829.

Zhang, Y., Zhao, Y., Qiu, X. & Han, R. (2013). Differentially expressed genes of *Coptotermes formosanus* (Isoptera: Rhinotermitidae) challenged by chemical insecticides. Journal of Economic Entomology, **106**(4):1863-1870.

Submitted **:** 03/03/2016
Revised **:** 29/03/2016
Accepted **:** 31/03/2016

تأثير مستخلصات ثلاثة نباتات مختلفة على حشرة النمل الأبيض (التيرميد)

[1]فاراخندا مندور، [2]عبير زافار، [1]عرفان إقبال
[1]قسم علم الحيوان – كلية العمال للبنات – جامعة لاهور – باكستان
[2]قسم العلوم البيئية – كلية البنات – جامعة لاهور – باكستان

الملخص العربى

تعتبر حشرة النمل الأبيض (تيرمايد) من الحشرات المدمرة التى تسبب اضرار بالغة لجزوع الأشجار الخشبية ولأخشاب البناء تسبب خسائر ببلايين الدولارات. ويكمن حل هذه المشكلة فى استخدام عدة مواد كيماوية مضادة لهذه الحشرات متوفرة حاليا فى الأسواق ولكنها تضر بالبيئة وتلوثها علاوة على خطورتها على الصحة العامة للبشر. ولذا فقد تم اختبار ثلاثة مستخلصات نباتية لأشجار الصنوبر والبتولة والكافور واستخدمت كمواد طاردة وسامة لهذه الحشرة. وبينت نتائج تجاربنا البحثية أن نسبة التأثير المميت اختلفت ليس فقط مع انواع النباتات المستخدمة بل ايضا مع انماط مذيبات الاستخلاص التى تم استخدامها. وكان اعلى معدل لموت هذه الحشرات مع مستخلص الهكسان لنباتات الصنوبر نتيجة لاستهلاك حشرات النمل الأبيض كميات اكثر من المستخلص الكحولى (إيثانول) فى معاملة اوراق الترشيح المستخدم مقارنة بمستخلصات الهكسان لمختلف الأصناف النباتية. وتوضح هذه النتائج مدى سمية مستخلصات الهكسان التى تفوقت فى سميتها وقدرتها الطاردة عن باقى المذيبات الأخرى المستعملة حيث أن الحشرة استهلكت اقل كمية من ورق الترشيح المعامل بكل المستخلصات من النباتات الثلاثة المذكورة. وتشير النتائج الى أن مستخلصات الهكسان كانت الأنسب والأكفأ كمضادات لحشرة النمل الأبيض (انتى تيرمايد).