



Chemical composition and larvicidal activities of the Himalayan cedar, *Cedrus deodara* essential oil and its fractions against the diamondback moth, *Plutella xylostella*

Abha Chaudhary^{1a}, Prabha Sharma^{2b}, Gireesh Nadda^{2c*}, Dhananjay Kumar Tewary^{2d}, Bikram Singh^{1e}

¹Natural Plant Products Division, CSIR - Institute of Himalayan Bioresource Technology, Himachal Pradesh, India

²Entomology and Pesticide Residue Analysis Laboratory, Hill Area Tea Science Division, CSIR - Institute of Himalayan Bioresource Technology, Himachal Pradesh, India

Abstract

Plants and plant-derived materials play an extremely important role in pest management programs. Essential oil from wood chips of Himalayan Cedar, *Cedrus deodara* (Roxburgh) Don (Pinales: Pinaceae), was obtained by hydrodistillation and fractionated to pentane and acetonitrile from which himachalenes and atlantones enriched fractions were isolated. A total of forty compounds were identified from these fractions using GC and GC-MS analyses. Essential oils and fractions were evaluated for insecticidal activities against second instars of the diamondback moth, *Plutella xylostella* L. (Lepidoptera: Yponomeutidae), using a leaf dip method. All samples showed promising larvicidal activity against larvae of *P. xylostella*. The pentane fraction was the most toxic with a LC_{50} value of 287 $\mu\text{g/ml}$. The himachalenes enriched fraction was more toxic ($LC_{50} = 362 \mu\text{g/ml}$) than the atlantones enriched fraction ($LC_{50} = 365 \mu\text{g/ml}$). LC_{50} of crude oil was 425 $\mu\text{g/ml}$ and acetonitrile fraction was $LC_{50} = 815 \mu\text{g/ml}$. The major constituents, himachalenes and atlantones, likely accounted for the insecticidal action. Present bioassay results revealed the potential for essential oil and different constituents of *C. deodara* as botanical larvicides for their use in pest management.

Keywords: atlantones, biopesticide, essential oils, himachalenes, insecticidal activity

Correspondence: c girish@ihbt.res.in, * Corresponding author

Editor: James Ottea was editor of this paper

Received: 20 October 2010, **Accepted:** 8 February 2011

Copyright : This is an open access paper. We use the Creative Commons Attribution 3.0 license that permits unrestricted use, provided that the paper is properly attributed.

ISSN: 1536-2442 | Vol. 11, Number 157

Cite this paper as:

Chaudhary A, Sharma P, Nadda G, Tewary DK, Singh B. 2011. Chemical composition and larvicidal activities of the Himalayan cedar, *Cedrus deodara* essential oil and its fractions against the diamondback moth, *Plutella xylostella*. *Journal of Insect Science* 11:157 available online: insectscience.org/11.157

Introduction

The diamondback moth, *Plutella xylostella* L. (Lepidoptera: Yponomeutidae), is an important and cosmopolitan pest of cruciferous crops (Harcourt 1962; Talekar and Shelton 1993; Gu et al. 2010). A major reason for the success of this pest is its remarkable ability to evolve insecticidal resistance (Cheng 1988; Sun et al. 2010). Several synthetic insecticides besides botanical and microbial control agents have been used for the control of this pest (Liu et al. 1982; Srinivasan and Krishnakumar 1982; Chaudhuri et al. 2001). The total annual cost for *P. xylostella* control throughout the world surpasses one billion US dollars (Talekar and Shelton 1993; Roux et al. 2007). The ill effects caused by chemical pesticides on health and environment, aside from resistance development in the pests, have fostered a need for the development of safer, lower-risk insecticidal agents. Natural plant products can be an excellent alternative source of novel insecticidal chemistries. With some exceptions, botanicals are considered to be less toxic to non-target species and more environmentally friendly because of their biodegradable nature (Copping 1996).

Plants are a virtually inexhaustible source of structurally diverse and biologically active substances; approximately 1800 plants have been reported to possess insecticidal properties (Jacobson 1982; Grainge et al. 1984). Plants are a good reservoir of eco-friendly allelochemicals. Extensive work has been done on bioactivity evaluation of extract/essential oil from various plants against important agricultural insect pests worldwide. Within the rich biodiversity of the Himalayas, there are abundant plant species, many of which are valued for their unique

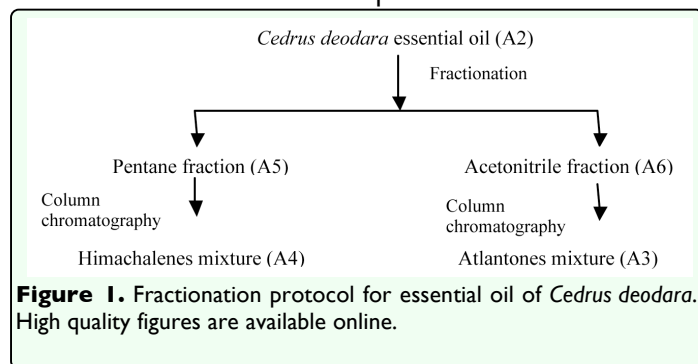
natural products and their biological and insecticidal properties (Tewary et al. 2005).

Himalayan Cedar, *Cedrus deodara* (Roxb. ex D. Don) (Pinales: Pinaceae), is found abundantly throughout the western Himalayas at altitudes of 1200-3000 m. Its essential oil has been reported to possess some activities against stored pests and houseflies (Singh et al. 1984; Singh and Rao 1985; Singh and Agrawal 1988; Singh et al. 1989). Its pesticidal activities are not reported against lepidopteron insect pests in the literature. Therefore, essential oil of *C. deodara* wood chip plant was selected for investigation against the diamondback moth. This study focuses on the chemical composition and larvicidal activities of *C. deodara* essential oil and its different chromatographic fractions against *P. xylostella*.

Materials and Methods

Insect cultures

P. xylostella used in this experimental study were collected from infested field crops and reared for more than 25 generations under laboratory conditions. Adult insects were allowed to lay eggs on one-week-old mustard plants grown in pots and caged inside wooden boxes. Larvae were removed from the mustard plants, transferred onto cabbage leaves, and kept in other boxes where development was completed. Adults were collected by aspirator and allowed to lay eggs on the mustard plants. Insects were reared and maintained at constant temperature of $25 \pm 2^\circ\text{C}$, relative humidity of $60\% \pm 15$, and photoperiod of 16:8 L:D. Second instar larvae were used in the experiments.



Plant material

Wood chips of *C. deodara* were collected in June 2007 from the forests of Mandi district in Himachal Pradesh, India, and voucher specimens (PLP 5969) were identified, processed, and deposited to the Herbarium of Institute of Himalayan Bioresource Technology (CSIR), in Palampur, India.

Essential oil extraction and fractionation

The air-dried wood chips (1.5 kg) of *C. deodara* were subjected to hydrodistillation for six hours using a Clevenger apparatus. The oil was dried over anhydrous sodium sulphate. The oil (50 ml) was fractionated between pentane and acetonitrile (50 ml each \times 3) (Figure 1). All the fractions were evaporated to dryness at 40° C under reduced pressure and stored in refrigerator at 4° C prior to analysis.

Chromatography and characterization of essential oil fractions

Aliquots of pentane and acetonitrile fractions were subjected to chromatography over silica gel and eluted sequentially with n-hexane/EtOAc gradients and finally with EtOAc. Fractions were tracked by thin layer chromatography and compounds with similar R_f values were pooled together to give sub-fractions. From pentane and acetonitrile fractions, mixtures of himachalenes and atlantones were isolated, respectively, which were identified by GC and GC-MS analyses.

Various fractions of essential oil were designated as A₂-A₆ (A₂: crude oil; A₃: atlantone enriched fraction; A₄: himachalene enriched fraction; A₅: pentane fraction; A₆: acetonitrile fraction).

Analysis of volatile compounds of essential oil and its fractions

Analysis of the samples was performed on GC 2010 Shimadzu Gas Chromatograph (Shimadzu, www.shimadzu.com) equipped with an FID detector and a carbowax phase BP-20 capillary column (30 m \times 0.25 mm i.d. with film thickness 0.25 μ m). Nitrogen was used as a carrier gas with a flow rate 1.0 ml/minute. Oven temperature was programmed from 40-220° C at 4° C/min with a four min hold at 40° C and a 15 min hold at 220° C. Injector and interface temperatures were each 250° C for both. Ion source temperature was 200° C. The 20 μ l sample was dissolved in 2 ml GC grade dichloromethane; sample injection volume was 2 μ l.

GC-MS analysis was conducted on a Shimadzu QP2010 GC-MS system with 2010 GC. A carbowax phase BP-20 capillary column (30m \times 0.25 mm i.d. with film thickness 0.25 μ m) was used with helium as a carrier gas at a flow rate of 1.1 ml/min on split mode (1:50), using the same conditions as above. Relative percentages were calculated from the FID from the automated integrator. Kovats indices (KI) of the compounds relative to a mixture of n-alkanes (C₈-C₂₃) were calculated. Identification of compounds was first attempted using the mass spectral libraries Wiley 7 and NIST 02 (McLafferty 1989; Stein 1990). Corroboration of the identification was then conducted by matching the mass spectra of compounds with those present in the literature (Adams 1995; Jennings and Shibamoto 1980), and finally by

matching the KI of the compounds reported on a column having an equivalent binding phase.

Insecticidal activity

Essential oil and its various fractions (A₂-A₆) were screened for insecticidal activity at higher concentrations (10000, 5000, 2500, and 1250 µg/ml) using leaf dip method. Briefly, 200 mg of the samples were dissolved in three ml acetone and then diluted to a volume of 20 ml in distilled water containing 0.05% Triton X-100 LR spreader (SD Fine-Chem Limited, www.sdfine.com). It was serially diluted with distilled water containing 0.05% Triton X-100 to obtain lower concentrations. Based on the preliminary toxicity results, stock solutions (3000 µg/ml) of samples were prepared using the above method for dose response bioassay studies. Eight different concentrations (3000-23.4 µg/ml) of test samples were prepared by serial dilution in distilled water containing 0.05% Triton X-100. The prepared concentrations were poured in glass Petri dishes. Distilled water containing 0.05% Triton X-100 and 15% acetone was used as control.

Three cabbage leaf disks (6 cm diameter) were cut and dipped in either individual test or control solutions for 30 seconds. The solution from the disks was allowed to dry at room temperature. Ten second instar larvae of *P. xylostella*, starved for 3-4 hours, were transferred to treatment and control leaf disks kept on the moist filter paper in Petri dishes. Petri dishes were then sealed using parafilm. Treated and control Petri dishes were kept at 25 ± 2°C, 60 ± 15% relative humidity, and a photoperiod of 16:8 L:D for observations. Moisture build up inside the Petri dishes, if any had accumulated, was blotted using tissue paper and Petri dishes were resealed. Observations on mortalities were recorded 48

hours after the treatment was given. Larvae that did not show movements when probed with a camel hairbrush were considered dead. The experiment was repeated three times with three replications and pooled data were analyzed.

Statistical analysis

Larval mortality was converted to percent mortality and corrected for control mortality using Abbott's formula (1925). Data were analyzed using EPA Probit Analysis Program version 1.5 for calculating LC_{50/90} values.

Table 1. Chemical constituents of essential oil and its fractions obtained from *Cedrus deodara* wood chips.

Compounds	KI	A5 (%)	A6 (%)	A4 (%)	A3 (%)
4-Acetyl-1-methylcyclohexene	1499	-	2.1	-	-
Longifolene	1503	0.91	-	-	-
Aromadendrene	1545	0.25	-	-	-
α-Himachalene	1579	13.29	2.56	20.33	-
α-Humulene	1617	0.48	-	-	-
γ-Himachalene	1627	11.28	2.29	17.95	-
β-Curcumene	1629	-	0.4	-	-
β-Himachalene	1651	27.78	6.85	52.61	-
3-Cyclohexene-1-methanol	1660	-	0.33	-	-
8-Cedrene-13-ol-acetate	1677	0.21	-	-	-
β-Vativenene	1702	0.5	-	-	-
cis-α-Bisabolene	1715	1.58	0.36	-	-
3-Methylacetophenone	1716	-	1.3	-	-
4,5-Dehydroisolongifolene	1755	0.23	-	-	-
α-Dehydro-ar-himachalene	1791	1.94	0.96	-	-
γ-Dehydro-ar-himachalene	1826	1.44	1.02	-	-
Vestitenone	1860	0.55	3	-	-
β-Himachalene oxide	1923	9.12	1.17	-	-
Calarene epoxide	1969	0.83	1.74	-	-
Nerolidol	1977	-	0.55	-	-
Carophyllene oxide	2002	0.25	0.5	-	-
(+)-8(15)-Cedrene-9-ol	2006	-	0.47	-	-
Aromadendrene oxide	2009	0.96	-	-	-
Longiborneol	2070	0.58	2.01	-	-
β-Bisabolol	2077	0.22	0.48	-	-
β-Atlantone	2097	0.94	2.71	-	1.95
(Z)-γ-Atlantone	2122	0.79	8.63	-	11.38
Himachalol	2129	2.04	4.51	-	-
m-Tolyldimethylactaealdehyde	2137	-	0.45	-	-
(E)-γ-Atlantone	2143	0.87	8.83	-	15.7
Deodarone	2152	0.53	4.18	-	-
Deodarone isomer	2156	-	3.7	-	-
Humulane-1,6-dien-3-ol	2161	-	4.37	-	-
(Z)-α-Atlantone	2172	3.4	5.23	-	4.99
(E)-α-Atlantone	2248	9.45	16	-	61.82
Longifolenaldehyde	2303	0.47	-	-	-
2-Butyl-1-methyl-1,2,3,4-tetrahydro-naphthalen-1-ol	2345	-	1.09	-	-
8-α-Acetoxyelemol	2349	-	0.47	-	-
7β,3α-Dihydroxy-1α-2,6-cyclohimachalane	2401	-	0.36	-	-
14-Hydroxy-9-epi-(E)-caryophyllene	2412	-	0.62	-	-
Total identification		90.89	89.24	90.89	95.74

A5: pentane fraction; A4: himachalene enriched fraction; A3: atlantone enriched fraction; A6: acetonitrile fraction; A2: crude oil (Chaudhary et al. 2009)

Results and Discussion

Chemical constituents of essential oil

Essential oil was obtained by hydrodistillation with a 0.98% yield (w/w on dry weight basis). Pentane, acetonitrile, atlantone enriched and himachalene enriched chromatographic fractions were obtained. A total of forty compounds were identified from these fractions using GC and GC-MS analyses. The identified constituents, percentage composition, and their KI values are shown in Table 1. In an earlier study, Chaudhary et al. (2009) reported 36 constituents in oil of woodchips of *C. deodara* using GC and GC-MS analyses. In the pentane fraction, 27 compounds were identified representing 90.89% of the constituents detected. A total of 11 sesquiterpene hydrocarbons and 16 oxygenated sesquiterpenes were identified constituting 59.68 and 31.21%, respectively. In the acetonitrile fraction, 31 compounds were identified representing 89.24% of the constituents detected assigning to three different classes: oxygenated monoterpene (3.73%), sesquiterpene hydrocarbons (14.44%), and oxygenated sesquiterpenes (71.07%). The major constituents in the pentane fraction were himachalenes (52.35%) and atlantones (15.45%). The other constituents were himachalene oxide (9.12%), himachalol (2.04%), α -dehydro-*ar*-himachalene (1.94%), *cis*- α -bisabolene (1.58%), and γ -dehydro-*ar*-himachalene (1.44%). The major constituents in the acetonitrile fraction were atlantones (41.40%) followed by himachalenes (11.70%). Further chromatography led to an increase in the percentage of himachalenes and atlantones from pentane and acetonitrile fraction to 90.89 and 95.74%, respectively.

Generally, the *Cedrus* oils contain high percentages of the himachalenes. In another

species, *C. atlantica*, essential oil is reported to contain 67% himachalenes as the major component (Boudarene et al. 2002). α -Pinene is commonly reported in *C. atlantica* needle oil, comprising more than 37% of the total essential oil. *C. libani* is rich in himachalenes (~ 42%) in wood extract, α -pinene (~ 24%), and caryophyllene (~ 7%) in the needle oil (Fleisher 2000). Himachalenes (68.52%), atlantones (15.02%), and himachalol (1.00%) in the fractions of oil were similar to those previously reported with variation in percent composition (Nigam et al. 1990). There is a significantly higher percentage of atlantone (~ 67%) from the extract of *C. deodara* as compared to other species, where they constitute less than 10% of the total oil or extract. Essential oil content could differ greatly even in the same genus, as well as in different plant parts (Duquesnoy et al. 2006; Salido et al. 2002; Cavaleiro et al. 2002). In our study, there was an absence of some major constituents like cedrene and cedrol, previously reported by Nigam et al. (1990). The chemical composition of an essential oil could also vary depending on geographical area, collecting season, distillation technique, stage of the plant part used for distillation, and presence of chemotypes and chemical races within the same species.

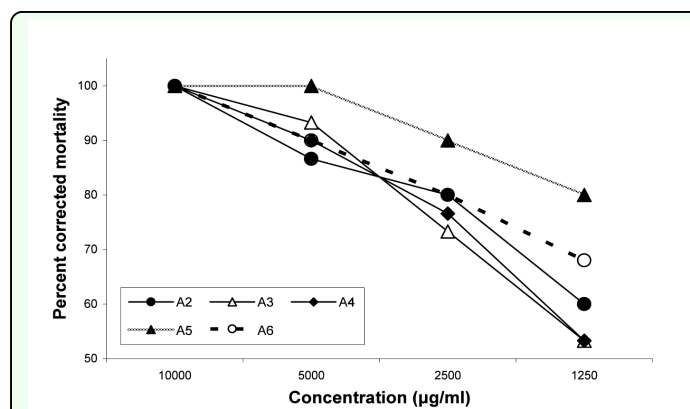


Figure 2. Preliminary screening of *Cedrus deodara* wood chip oil and its different fractions against larvae of *P. xylostella* (48 hours). High quality figures are available online.

Insecticidal activity

Mortalities of second instar larvae of *P. xylostella* that were exposed to higher concentrations of essential oil and fractions (A₂-A₆) of *C. deodara* during the preliminary screening are presented in Figure 2. The samples efficiently killed the larval stages, and activity increased with increasing concentrations. The highest concentration (10,000 µg/ml) resulted in ~ 100% mortality. The lethal concentration (LC₅₀ and LC₉₀) values along with other statistical parameters based on the dose response bioassay studies are presented in Table 2. The volatile constituents of pentane fraction A₅ exhibited maximum larvicidal activity with minimum LC₅₀ (287.06 µg/ml) and LC₉₀ (2253.33 µg/ml) values, whereas acetonitrile fraction was the least toxic (LC₅₀ = 815.48 µg/ml; LC₉₀ = 5720.00 µg/ml).

Larvicidal potential of the samples after 48 hours of exposure time was found in the following order: A₅>A₄>A₃>A₂>A₆ on the basis of their LC₅₀ and LC₉₀ values. Himachalenes and atlantones enriched fractions exhibiting LC₅₀ of 361.84 and 365.12 g/ml, respectively. The major constituents in the pentane fraction were himachalenes (52.35%) and atlantones (15.45%) (Figure 3). The other constituents were himachalene oxide (9.12%), himachalol (2.04%), α -dehydro-*ar*-himachalene (1.94%), *cis*- α -bisabolene (1.58%), and γ -dehydro-*ar*-himachalene (1.44%). It was observed that individual himachalenes and atlantones enriched fractions were less toxic than in the mixture.

In the present study, *C. deodara* essential oil and chromatographic fractions showed good larvicidal activities against *P. xylostella*. However, there is no available literature on the larvicidal potential of *C. deodara* oil and its fractions against this lepidopteron pest. Himalayan cedar wood oil and its constituents are reported to show insecticidal activities against some other insect pests like the Graham bean weevil (*Callosobruchus analis*), rice weevil (*Sitophilus oryzae*), housefly (*Musca domestica*), and the Chinese bean weevil (*Callosobruchus chinensis*) (Raguraman and Singh 1997; Singh et al. 1989; Singh and Agarwal 1988; Singh and Rao 1985). Further, *C. deodara* oil is reported to show toxicity against *Lymnaea acuminata* when combined with extracts of *Azadirachta indica* and *Embelia ribes* (Rao and Singh 2001). It is well known that the sensitivity of different insect species could be quite different for the same substance, and that insects vary widely in their responses to secondary plant products. Some plant-based compounds possess repellent and feeding

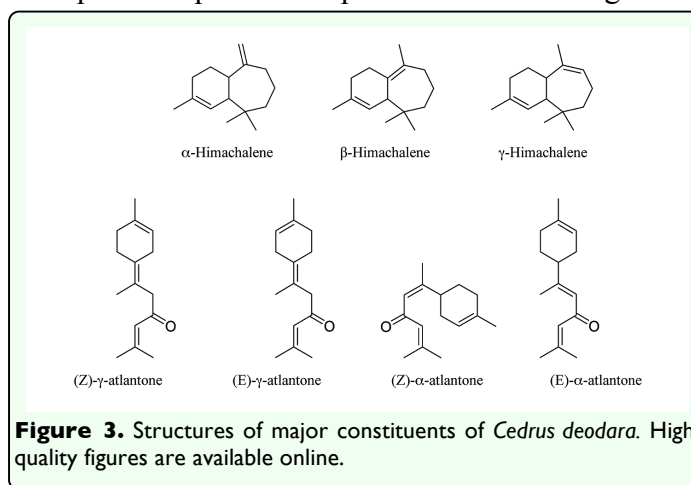


Figure 3. Structures of major constituents of *Cedrus deodara*. High quality figures are available online.

Table 2. LC₅₀ and LC₉₀ values of essential oil and its fractions from *Cedrus deodara* against *Plutella xylostella*

Test Samples*	LC ₉₀ (µg/ml)	LC ₅₀ (Fiducial Limits)		LC ₉₀ (µg/ml)	LC ₅₀ (Fiducial Limits)		Chi square	Slope	Intercept
		Lower (µg/ml)	Upper (µg/ml)		Lower (µg/ml)	Upper (µg/ml)			
A5	287.06	241.31	341.85	2253.33	1692.56	3210.21	1.194	1.432	1.4797
A4	361.84	299.44	439.89	3689.18	2615.25	5700.17	10.43	1.27	1.7483
A3	365.12	301.58	444.9	3836.84	2702.74	5982.32	3.121	1.254	1.7851
A2	424.82	349.26	522.02	4736.53	3262.93	7625.54	6.953	1.223	1.783
A6	815.48	682.6	991.5	5720	4098.4	8755.5	3.672	1.154	0.589

*A5: pentane fraction; A4: himachalene enriched fraction; A3: atlantone enriched fraction; A2: crude oil; A6: acetonitrile fraction

and/or oviposition deterrent properties (Ibrahim et al. 2001). A number of research results have also been published on the use of plant based compounds for controlling herbivorous insects.

The insecticidal potential of essential oils for developing promising insect control agents has been emphasized in a number of recent reports (Adebayo et al. 1999; Gbolade et al. 2000). In studies of evaluating essential oils against lepidopteron larvae, patchouli oil was found to be the most toxic to oblique banded leafroller (*Choristoneura rosaceana*) larvae ($LC_{50} = 2.8 \mu\text{l/ml}$ and $LD_{50} = 8.0 \mu\text{g/insect}$), whereas garlic oil was the most toxic to *Trichoplusia ni* larvae ($LC_{50} = 3.3 \mu\text{l/ml}$ and $LD_{50} = 22.7 \mu\text{g/insect}$), followed by patchouli oil and lemongrass oil (Machial et al. 2010). In another study, essential oils of *Thymus vulgaris* ($LC_{50} = 4.8 \text{ mg/ml}$), *Syzygium aromaticum* ($LC_{50} = 6.0 \text{ mg/ml}$), *Cymbopogon citrates* ($LC_{50} = 7.7 \text{ mg/ml}$), *Cinnamomum cassia* ($LC_{50} = 8.5 \text{ mg/ml}$), *Cymbopogon nardus* ($LC_{50} = 10.1 \text{ mg/ml}$) were found toxic to *T. ni* larvae in residual bioassays (Jiang et al. 2010). *Laurus azorica* and *Juniperus brevifolia* leaf essential oil caused 93.3% and 46.7% mortality of fourth-instar larvae of the armyworm, *Pseudaletia unipuncta*, and all essential oils significantly inhibited larval growth after five days of feeding on the treatment diet (Rosa et al. 2010).

Naturally occurring insecticide prototypes are in great demand at a global level to manage insect resistance and foster environmental health. In the present study, essential oil and fractions of *C. deodara* were found to possess larvicidal activities against *P. xylostella*. Therefore, these agents can be utilized for sustainable pest management. These fractions could be explored for their utilization in the botanical formulations either alone or in

different combinations. However, further studies should be conducted to evaluate cost, efficacy, and safety of essential oil and enriched fractions on a wide range of pests. Results of this study could be helpful in further research for selection/identification or synthesis of semi-synthetic, newer, and more selective larvicidal compounds to develop lower-risk pesticides for use in integrated pest management packages. The use of essential oil in pest management could be of both economic and ecological benefit.

Acknowledgements

First author is thankful to CSIR for providing the SRF fellowship. Authors are also grateful to the Director Dr. P.S. Ahuja, IHBT, Palampur, for his encouragement to carry out this research and to M/S Hari Industries for supplying plant material. Thanks to Mrs. Vijaylata Pathania and Mr. Ramesh Kumar for technical support, and to Mr. Kuldeep Chand for insect rearing.

References

-
- Abbott WS. 1925. A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology* 18: 265-267.
- Adams RP. 1995. *Identification of essential oil components by gas chromatography/mass spectroscopy*. Allured Publishing.
- Adebayo TA, Gbolade AA, Olaifa JI. 1999. Comparative study of toxicity of essential oils to larvae of three mosquito species. *Nigerian Journal of Natural Products and Medicine* 3: 74-76.
- Boudarene L, Rahim L, Baaliouamer A, Maklati BY. 2002. Analysis of Algerian essential oils from twigs, needles and wood of

- Cedrus atlantica* G. Manetti by GC/MS. *Journal of Essential Oil Research* 6: 531-534.
- Branco MC, Gatehouse AG. 2001. A survey of insecticide susceptibility in *Plutella xylostella* (L.) (Lepidoptera: Yponomeutidae) in the Federal district, Brazil. *Neotropical Entomology* 30: 327-332.
- Cavaleiro C, Salgueiro L, Barroso J, Figueiredo AC, Pedro LG, Fontinha SS, Bighelli A, Casanova J, Looman A, Scheffer JJC. 2002. Composition of the essential oil of *Juniperus cedrus* Webb & Berth grown on Madeira. *Flavour and Fragrance Journal* 17: 111-114.
- Chaudhary A, Kaur P, Singh B, Pathania V. 2009. Chemical composition of hydrodistilled and solvent volatiles extracted from woodchips of Himalayan Cedrus: *Cedrus deodara* (Roxb.) Loud. *Natural Product Communications* 4: 1257-1260.
- Chaudhuri N, Ghosh S, Ghosh J, Senapati, SK. 2001. Incidence of insect pests of cabbage in relation to prevailing climatic conditions of Terai region. *Indian Journal of Entomology* 63: 421-428.
- Cheng EY. 1988. Problems of control of insecticide resistant *Plutella xylostella*. *Pesticide Science* 23: 177-188.
- Copping LG. 1996. *Crop protection agents from nature*. The Royal Society of Chemistry.
- Duquesnoy E, Huu Dinh N, Castola V, Casanova J. 2006. Composition of a pyrolytic oil from *Cupressus funebris* Endl. of Vietnamese origin. *Flavour and Fragrance Journal* 21: 453-457.
- Fleisher A. 2000. The volatiles of the leaves and wood of Lebanon Cedar (*Cedrus libani* A. Rich) aromatic plants of the Holy Land and the Sinai. *Journal of Essential Oil Research* 12: 763-765.
- Gbolade AA, Oyedele AO, Sosan MB, Adewayin FB, Soyela OL. 2000. Mosquito repellent activities of essential oils from two Nigerian *Ocimum* species. *Journal of Tropical Medicinal Plants* 1: 146-148.
- Grainge MS, Ahmed WC, Mitchell, Hylin JW. 1984. Plant species reportedly possessing pest-control properties-A database. Resource Systems East-West Center.
- Gu X, Tian S, Wang D, Gao G. 2010. Interaction between short-term heat pretreatment and fipronil on 2nd instar larvae of diamondback moth, *Plutella xylostella* (Linn). *Dose-Response*, 8: 331-346.
- Guenther E. 1948. *The Essential Oil*, volumes 1-5. Van Nostrand Co.
- Harcourt DG. 1962. Biology of cabbage caterpillars in eastern Ontario. *Proceedings of the Entomological Society Ontario* 93: 61-75.
- Ibrahim MA, Kainulainen P, Aflatuni A, Tiilikkala K, Holopainen JK. 2001. Insecticidal, repellent, antimicrobial and phytotoxicity of essential oils: with special reference to limonene and its suitability for control of insect pests. *Agricultural and Food Science in Finland* 10: 243-259.
- Isman MB. 2000. Plant essential oil for pest and disease management. *Crop Protection* 19: 603-608.

Jacobson M. 1982. Plants, insect and man—their interrelationship. *Economic Botany* 36: 346-354.

Jennings W, Shibamoto T. 1980. *Qualitative analysis of flavour and fragrance volatiles by glass capillary gas chromatography*. Academic Press.

Jiang ZL, Akhtar Y, Zhang X, Bradbury R, Isman MB. 2010. Insecticidal and feeding deterrent activities of essential oils in the cabbage looper, *Trichoplusia ni* (Lepidoptera: Noctuidae). *Journal of Applied Entomology* (doi: 10.1111/j.1439-0418.2010.01587.x).

Liu MY, Tzeng YJ, Sun CN. 1982. Insecticide resistance in the Diamond back Moth. *Journal of Economic Entomology* 75: 153-155.

Machial CM, Shikano I, Smirle M, Bradbury R, Isman MB. 2010. Evaluation of the toxicity of 17 essential oils against *Choristoneura rosaceana* (Lepidoptera: Tortricidae) and *Trichoplusia ni* (Lepidoptera: Noctuidae). *Pest Management Science* 66: 1116-1121.

McLafferty FW. 1989. *Registry of mass spectral data*. John Wiley and Sons.

Nawaz MA, Muhammad M, Sami E, Richard GS, Meckenzi HNG. 2004. Microbial control of diamondback moth, *Plutella xylostella* L. (Lepidoptera: Yponomeutidae) using bacteria (*Xenorhabdus nematophila*) and its metabolites from the entomopathogenic nematode *Steinernema carpocapsae*. *Journal of Zhejiang University Science* 5: 1183-1190.

Nigam MC, Ahmad A, Misra LN. 1990. Composition of the essential oil of *Cedrus deodara*. *Indian Perfumer* 34: 278-281.

Raguraman S, Singh D. 1997. Biopotentials of *Azadirachta indica* and *Cedrus deodara* oils on *Callosobruchus chinensis*. *International Journal of Pharmacognosy* 35: 344-348.

Rao IG, Singh DK. 2001. Combinations of *Azadirachta indica* and *Cedrus deodara* oil with piperonyl butoxide, MGK-264 and *Embelia ribes* against *Lymnaea acuminata*. *Chemosphere* 44: 1691-1695.

Rosa JS, Mascarenhas C, Oliveira L, Teixeira T, Barreto MC, Medeiros J. 2010. Biological activity of essential oils from seven Azorean plants against *Pseudaletia unipuncta* (Lepidoptera: Noctuidae). *Journal of Applied Entomology* 134: 346-354.

Roux O, Gevrey M, Arvanitakis L, Gers C, Bordat D, Legal L. 2007. ISSR-PCR: Tool for discrimination and genetic structure analysis of *Plutella xylostella* populations native to different geographical areas. *Molecular Phylogenetics and Evolution* 43: 240-250.

Salido S, Altarejos J, Nogueras M, Sanchez A, Pannecouque C, Witvrouw M, Clercq E. 2002. Chemical studies of essential oils of *Juniperus oxycedrus* ssp. *Badia*, *Journal of Ethnopharmacology* 81: 129-134.

Shaaya SK, Kostjukovski M, Eilberg J, Sukprakarn C. 1997. Plant oils as fumigants and contact insecticides for the control of stored product insects. *Journal of Stored Products Research* 33: 7-15.

Shashi B, Singh J, Rao JM, Saxena AK, Qazi GN. 2006. A novel lignan composition from *Cedrus deodara* induces apoptosis and early nitric oxide generation in human leukemia Molt-4 and HL-60 cells. *Nitric Oxide* 14: 72-88.

Singh D, Agarwal SK. 1988. Himachalol and himachalene: insecticidal principles of Himalayan cedarwood oil. *Journal of Chemical Ecology* 14: 1145-1151.

Singh D, Rao SM, Tripathi AK. 1984. Cedarwood oil as a potential insecticidal agent against Mosquitoes. *Naturwissenschaften* 71: 265.

Singh D, Rao SM. 1985. Toxicity of cedarwood oil against pulse beetle, *Callosobruchus chinensis* Linn. *Indian Perfumer* 29: 201-204.

Singh D, Siddiqui MS, Sharma S. 1989. Reproduction retardant and fumigant properties in essential oils against rice weevil (Coleoptera: Curculionidae) in stored wheat. *Journal of Economic Entomology* 82: 727-733.

Srinivasan K, Krishnakumar NK. 1982. *Pest management in cabbage*. Annual Report, 1981 pp. 80-81. Indian Institute of Horticultural Research.

Stein SE. 1990. *National Institute of Standards and Technology (NIST) Mass Spectral Database and Software*. Version 3.02.

Sun J, Liang P, Gao X. 2010. Inheritance of resistance to a new non-steroidal ecdysone agonist, fufenozide, in the diamondback moth, *Plutella xylostella* (Lepidoptera: Plutellidae). *Pest Management Science* 66: 406-411.

Talekar NS, Shelton AM. 1993. Biology, ecology, and management of the diamondback moth. *Annual Review of Entomology* 38: 275-301

Tewary DK, Bhardwaj A, Shanker A. 2005. Pesticidal activities in five medicinal plants collected from mid hills of western Himalayas. *Industrial Crops and Products* 22: 241-247.