African Journal of Biotechnology Vol. 8 (19), pp. 5128-5131, 5 October, 2009
Available online at http://www.academicjournals.org/AJB
ISSN 1684–5315 © 2009 Academic Journals

Full Length Research Paper

Larvaecidal effects of azadirachtin on the pine processionary moth

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Accepted 24 August, 2009

The effects of azadirachtin, a natural extract of the neem tree *Azadirachta indica* A. Juss, on larval growth and feeding activity of pine processionary moth (*Thaumetopoea pityocampa* (Schiff.)) was investigated in laboratory experiments conducted in 2007 - 2008. The organic insecticide NeemAzal-T/S (a commercial neem preparation) was applied as suspension in distilled water at concentration levels of 0.3, 0.5 and 1%. Comparing with the untreated leaves, the effect of the insecticide NeemAzal-T/S was tested on the 10th day after treatment. The results obtained in conducted experiments indicated that NeemAzal-T/S was effective against *T. pityocampa* larvae, significantly inhibiting larval growth and reducing feeding activity.

Key words: Azadirachtin, insecticide, *Thaumetopoea pityocampa*, larval growth, food consumption.

INTRODUCTION

The pine processionary moth *Thaumetopoea pityocampa* (Schiff.) (Lep. Thaumetopoeidae) is one of the most devastating caterpillars of pine forests particularly in Mediterranean, Aegean and Marmara regions of Turkey. Host plants of *T. pityocampa* are mainly *Pinus* sp. and *Cedrus libani.* Various control methods have been deployed against the pest *T. pityocampa* in many countries. However, chemical control methods, in particular, have a negative impact on the environment and specifically on many beneficial organisms. Thus, environmentally friendly methods of control are much in need (Roessler, 1989).

The neem tree, *Azadirachta indica* A. Juss, (Meliaceae) extracts have been used as a natural insecticide, recently (Koul et al., 1990; Ascher, 1993). Neem extracts have little impact on non-target organisms such as parasitoids, predators and pollinators (Naumann, 1996) and degrade rapidly in the environment (Barrek et al., 2004). Koul et al. (2004) indicated that the primary active ingredient of most neem-based pesticides has shown an excellent insecticidal activity against lepidopteron larvae. Azadirachtin (C35H44O16), a neem-seed extract, is the active insecticidal ingredients of this species. It is a slow acting naturally based anti-feeding insecticide (Sonata et al., 2005). The effects of azadirachtin on insects include feeding and oviposition deterrence, growth inhibition, fecundity and fitness reductions, low mammalian toxicity and low persistence in the environment (Schmutterer, 1990; Koul, 1992). For these reasons, it has generated enormous worldwide interest due to its potential as a new insect pest control agent (Butterworth and Morgan, 1968; Ley, 1994).

Several studies have reported the effect of azadirachtin on nutrition, growth and development in Lepidoptera (Barnby and Klocke, 1987; Isman, 1993; Koul et al., 1987; Osman, 1993). The main goal of the present work was to determine the effects of organic insecticide Azadirachtin (active ingredient in the NeemAzal-T/S) on some morphological characteristics and food consumption of *T. pityocampa* larvae under laboratory conditions (22 ± 1°C, 65 ± 5% RH and L: D 16:8 h photoperiod).

MATERIALS AND METHODS

This study was conducted between the years 2007-2008. On April 10, 2007, second larval instars of *T. pityocampa* were collected from infected *Pinus nigra* trees in Taşköprü (Kastamonu) Forest Enterprise forests (41°14'38"N-34°11'29"E) located in Western Blacksea Region of Turkey. The average altitude of sampling sites in this area was 1200 m.

In this study, the following treatments: NeemAzal-T/S (a comercial neem preparation with an Azadirachtin content) used as suspension in distilled water at concentration levels of 0.3, 0.5, 1%

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Table 1. Results of one-way ANOVA applied to the effects of NeemAzal-T on larval growth and feeding activity of *T. pityocampa*.

Sources of variation	Degree of freedom	Sum of squares	Mean square	F	Sig.
Head capsule width					
Con. lev.	3	5.111	1.704	16.875	0.0012
Error	8	0.808	0.101		
Total	11	5.918	0.538		
Head Capsule Length					
Con. lev.	3	5.521	1.840	11.211	0.0036
Error	8	1.313	0.164		
Total	11	6.834	0.621		
Food consumption					
Con. lev.	3	1734.003	578.001	1044.580	0.0001
Error	8	4.427	0.553		
Total	11	1738.429	160.488		
Faecal output					
Con. lev.	3	159.600	53.200	13.300	0.0023
Error	8	32.000	4.000		
Total	11	191.600	17.418		

Table 2. Results of Duncan's multiple range test on the head capsule width and length of *T. pityocampa* affected by NeemAzal-T/S.

Conc. level (%)	Head capsule width (mm)	Head capsule length (mm)	Food consumption (g)	Faecal output (g)
1	3.050a	2.890b	5.000a	8.200b
0.3	3.043a	3.160ab	10.400b	6.600b
0.5	1.603b	1.843c	4.800a	8.400b
Control (0.0)	3.213a	3.710a	34.000c	16.000a

Means followed by the same letter within each column and parameter are not significantly different ($P < 0.05$).

and the control (untreated plants) were investigated. The four treatments were replicated three times in a randomized block design. Thirty larvae in second instar were placed in each plot with 50 g fresh *P. nigra* twigs. Except control plants, the leaves were sprayed with NeemAzal-T/S at concentration levels of 0.3, 0.5, 1%. After that, each 50 g *P. nigra* twigs were placed in each of 12 small indoor cages (40 x 40 x 40 cm).

Following measurements: Head capsule width and length, food consumption (leaf weight eaten) and weight of larva faeces were carried out later on the 10th day after treatment. Food consumption of larvae was calculated by subtracting the leaf weight eaten from the 50 g fresh leaves.

All statistical analyses were performed using SPSS® 15.0 for Windows® software. Data were analyzed by using one-way ANOVA to examine the effects of NeemAzal-T/S on larval growth and feeding activity of *T. pityocampa* and means were separated using Duncan's multiple range test. Following the results of ANOVAs, statistical significances between the samples were indicated by probability values of $P < 0.05$. Correlation analysis was performed to test relationship between concentration level of NeemAzal-T/S and head capsule dimensions and relationship between head capsule length and width.

RESULTS AND DISCUSSION

The effect of NeemAzal-T/S used at azadirachtin concentration of 1, 0.3 and 0.5% on the larval growth and food consumption of *T. pityocampa* fed during 10 days with treated pine leaves in comparison to controls were analyzed with one-way analysis of variance (ANOVA). As shown in Table 1, the investigations with the second larval instar of *T. pityocampa* showed quite large differences between the experimental variants. There is a significant effect of NeemAzal-T/S on head capsule dimensions, food consumption and faecal output of *T. pityocampa* (Table 1).

The present study showed that the head capsule width and length were accordingly high in the control and head capsule growth was disrupted particularly by 0.5% NeemAzal-T/S (Tables 1 and 2). Duncan Test revealed that average head capsule width and length were significantly ($P < 0.05$) lower for the 0.5% concentration level

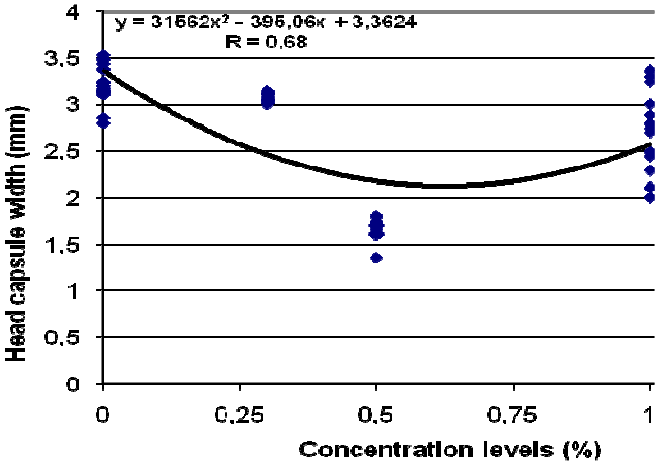

Figure 1. Relationship between concentration level of NeemAzal-T/S and head capsule width of *T. pityocampa* larvae later on the 10th day after treatment.

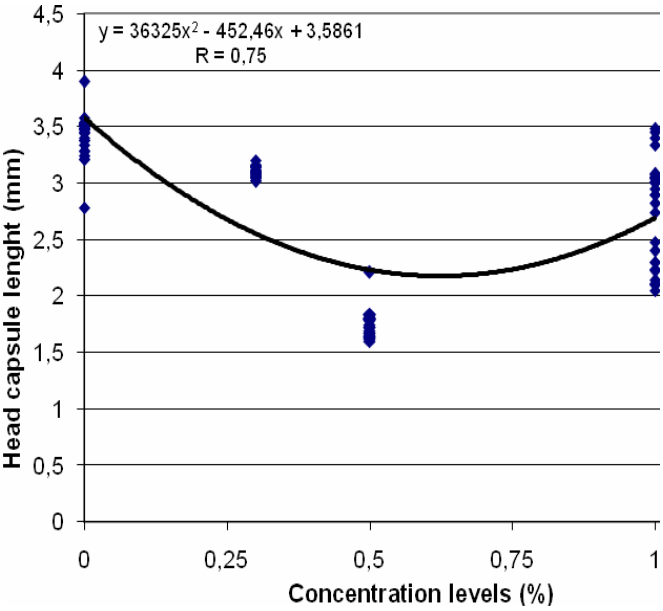

Figure 2. Relationship between concentration level of NeemAzal-T/S and head capsule lenght of *T. pityocampa* larvae later on the 10th day after treatment.

than for the other concentration levels and control plants (Table 2, Figures 1 and 2). Figures 1, 2 show that 0.5% concentration level is the best suitable dosage to disrupt the larval growth of *T. pityocampa*. Positive effects of neem-extracts on disrupting larval growth of insects have been reported by several authors, such as Koul (1984), Garcia and Rembold (1984) and Dorn et al. (1986). Breuer and De Loof (1998) also indicated that *T. processionea* treated with NeemAzal-T/S approximately maintains their mass and size from the beginning of the experiment.

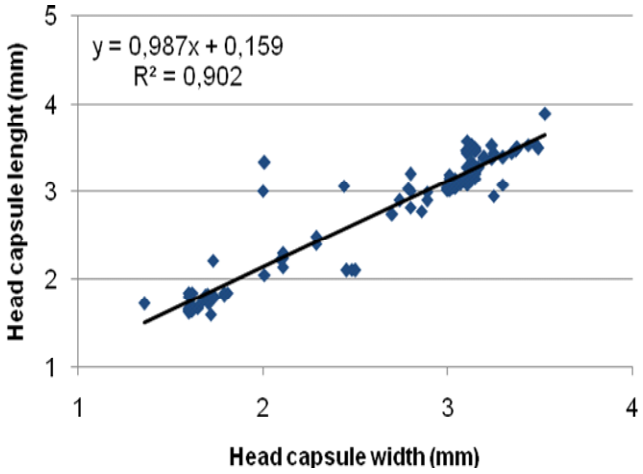

Figure 3. Relationship between head capsule length and width of *T. pityocampa* larvae.

Figure 3 shows the correlation between head capsule length and width of *T. pityocampa* larvae. As it is expected, there is a strong positive correlation (r^2 = 0.902) between head capsule length and width (Figure 3). Food consumption of larvae on untreated leaves was significantly higher than those fed on NeemAzal-T/S treated leaves. NeemAzal-T/S showed the highest antifeedant activity at concentration level of 0.5%. Similar results have been reported by Nicol and Schmutterer (1996). The authors found that the feeding activity of gypsy moth larvae in the treated stands decreased rapidly.

The faecal production of larvae feeding on untreated twigs (control) was significantly higher than those feeding on the other NeemAzal-T/S treated twigs. Reduced amount of faecal output of larvae fed on NeemAzal T/S-treated leaves can be a direct consequence of the reduced food intake (Table 2). Similar results were also found by Breuer and De Loof (1998) for *T. processionea.* Breuer and De Loof (1998) reported that the faecal production was accordingly high in the control (untreated oak twigs) and in contrast, caterpillars feeding on oak twigs sprayed with different concentrations of NeemAzal-T/S produced significantly lower quantities of faeces in the first four days and almost none after this period.

Conclusions

The objective of this research was to assess the efficacy of azadirachtin on larval growth (determined by measuring head dimensions) and feeding activity (determined by food consumption and faecal output) of *T. pityocampa*. Results show that azadirachtin inhibits larval growth and reduces the feeding activity of *T. pityocampa*. In conclusion, this study has revealed that azadirachtin has antifeedant and growth inhibition properties against *T. pityocampa*.

ACKNOWLEDGEMENT

The authors would like to thank Dr. Sezgin Ayan for his assistance and invaluable comments on the early version of the manuscript.

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