UNIVERSITI PUTRA MALAYSIA

TOXICITY OF METHANOLIC CRUDE EXTRACTS OF AZADIRACHTIN AGAINST Plutella xylostella (L.) (Lepidoptera: Plutellidae)

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BY

NOR SHUHAIDAH NORIZAN

A project report submitted to Faculty of Agriculture, Universiti Putra Malaysia, in fulfillment of the requirement of PRT 4999 (Final Year Project) for the award of the degree of Bachelor of Agricultural Science

FACULTY OF AGRICULTURE

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SERDANG, SELANGOR DARUL EHSAN

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This project report entitled Toxicity of Methanolic Crude Extracts of Azadirachtin against *Plutella xylostella* (L.) (Lepidoptera: Plutellidae) is prepared by Nor Shuhaidah bt Norizan and submitted to the Faculty of Agriculture in fulfillment of the requirement of PRT 4999 (Final Year Project) for the award of the degree of Bachelor of Agricultural Science.

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ABSTRACT

Leaf mustard is the most popular leaf vegetables in Malaysia, occupying more than 3,000 ha and heavily sprayed with synthetic insecticides against its insect pest *Plutella xylostella.* The use synthetic insecticides can result resistance problem, residues exceeding the MRL, health hazard to consumers, crops and ecosystem. The use of the botanical insecticides could solve these problems due to their bioactive compounds are fairly complex, making it more difficult for the pest to develop resistance and easily degraded. The toxicity of the methanolic crude extracts *Azadirachta excelsa*, *Melia azedarach* and Neem oil against *P. xyloxtella* was evaluated by leaf dip bioassay using early 3rd instars larvae in the laboratory. A minimum of 6 concentrations (0.75, 1.00, 1.50, 2.00 and 2.50ppm) for each treatment and 6 replications arranged in Randomized Complete Block Design are utilized to obtain the LC_{50} values for each extracts. The mortality and leaf consumed was recorded at 24, 48 and 72 hours after treatment and the data were subjected to probit analysis. The amount of leaf consumed by the larvae was also measured and the data were subjected to analysis of variance (ANOVA). The methanolic extract of *A. excelsa* showed the higher toxicity with lower LC_{50} value of 0.590 ppm and the lowest leave consumed by the larvae compared with *M. azedarach* (0.794 ppm) and Neem oil (1.716 ppm). *Azadirachta excelsa* contained an active compound known as Marrangin (azadirachtin L) that are more toxic than azadirachtin A and B in Neem oil and *M. azedarach.*

ABSTRAK

Sawi adalah sayuran daun yang paling popular di Malaysia, menduduki lebih daripada 3,000 ha dan banyak disembur dengan racun serangga terhadap serangga perosak *Plutella xylostella* itu. Penggunaan racun serangga boleh menyebabkan masalah rintangan, sisa melebihi MRL, bahaya kepada kesihatan pengguna, tanaman dan ekosistem. Penggunaan racun serangga botani boleh menyelesaikan masalah-masalah ini disebabkan oleh sebatian bioaktif mereka adalah agak kompleks, menjadikannya lebih sukar untuk perosak untuk membangunkan rintangan dan mudah dihina. Ketoksikan mentah metanol ekstrak *Azadirachta excelsa*, *Melia azedarach* dan minyak Neem terhadap *P. xyloxtella* telah dinilai oleh bioesei rendaman daun menggunakan larva awal instar ketiga di dalam makmal. Sekurang-kurangnya 6 kepekatan (0.75, 1.00, 1.50, 2.00 dan 2.50ppm) bagi setiap rawatan dan 6 replikasi disusun dalam Reka Bentuk Rawak Lengkap digunakan untuk mendapatkan nilai LC50 bagi setiap ekstrak. Kadar kematian dan daun yang digunakan dicatatkan pada 24, 48 dan 72 jam selepas rawatan dan data tertakluk kepada analisis probit. Jumlah daun dimakan oleh larva juga diukur dan data tertakluk kepada analisis varians (ANOVA). Ekstrak metanol *A. excelsa* menunjukkan tahap ketoksikan yang lebih tinggi dengan nilai LC50 rendah 0.590 ppm dan yang paling rendah meninggalkan kesan dimakan oleh larva berbanding dengan *M. azedarach* (0,794 ppm) dan minyak Neem (1.716 ppm). *Azadirachta excelsa* terkandung sejenis bahan aktif yang dikenali sebagai Marrangin (Azadirachtin L) yang lebih toksik daripada Azadirachtin A dan B dalam minyak Neem dan *M. azedarach*.

CHAPTER 1

INTRODUCTION

Plutella xylostella (L.) (Lepidoptera:Plutellidae) or commonly known known as diamondback moth (DBM) is a cosmopolitan and oligophagous pest of cruciferous crops (Thorsteinson, 1953; Ahmad et al., 2009). It is the most important pests of cruciferous crops throughout the world, causing direct damage to cabbage with losses as much as 100% (Castelo-Branco & Gatehouse, 2001). The insect feeds on numerous cruciferous plants that contain mustard oils and glucosides and attacks the crop from the nursery stage and can cause up to 52% loss in marketable yield in cabbage. Srinivasan (1984) reported that 90–92% loss could occur if cabbage plants were left unprotected and Lingappa et al., (2000) reported that losses vary from 30 to 100%.

In Malaysia, *P. xylostella* has become resistant to all groups of conventional insecticides in some major Cruciferae family production areas including in India, Cameron Highlands and Thailand. Resistance is "a genetic change in response to selection by toxicants (i.e., insecticides) used to control *P. xylostella* in the field." (Sawicki,1987). By 1981, *P. xylostella* had become resistant to more than 36 insecticides across multiple chemical classes including chlorinated hydrocarbons, carbamates, organophosphates and pyrethroids (Miyata et al., 1986). By 1990, *P. xylostella* was resistance to abamectin, benzophenylureas, and various strains of *Bacillus thuringiensis* (Sun, 1990).

The damages become serious from the abuse of chemicals by spraying excessively, mixtures of chemicals, increased dosage and frequency of spraying. It has led to the resistance and absence of effective natural enemies by the use of broad-spectrum insecticides such as Ambush (permethrin) and Dibrom (naled) (Chua & Ooi, 1986). This is primarily because of its genetic elasticity, and as a consequence the *P. xylostella* has become one of the most difficult pests to control in the past 50 years worldwide (Attique et al., 2006; Sarita et al., 2010b & Gong et al., 2010).

The rapid widespread and frequent use of newer insecticides is usually followed by the rapid evolution of resistance (Amit et al., 2004). In South Asia, substantial resistance to new insecticides has appeared within as little as two years of first use (Wright, 2004) and in Hawaii after only 30 months (Mau & Gusukuma-Minuto, 2004). There is frequent "burn out" of new products such as spinosad, avermectin and indoxacarb in the intensive cruciferous crop production areas in Cameron Highland and Thailand (Wright, 2004).

Plant-derived extracts and phytochemicals have long had been a subject of research in an effort to develop alternatives to conventional insecticides but with reduced health and environmental impacts. In this review we compare bioactivities of some plants extracts against the cruciferous crops pest, *P. xylostella*. There is a great variety of families of plants that possess potent anti-insect compounds.

From the Meliaceae family, strong insecticide molecules have been isolated with the limonoid azadirachtin obtained from *A. indica* being the most potent and studied (Govindachari, 1992; Schmutterer, 1995). *Melia azedarach*, commonly named is china berry is known to be toxic to higher animals. However, its application in pest control is still limited. A limonoid has been isolated from this tree as the most potent substance for controlling insects. The others Meliacaea family is *A. excelsa* and limonoids are known to be excellent antifeedants for many pest species with no deleterious effects on humans, animals or beneficial insects (Mordue & Blackwell, 1993). A methanolic extract of *A. excels*a wood inhibited growth, feeding and was toxic to the larvae of *Crocidolomia binotalis* (Ng et al., 2003).

The objective of this study was to evaluate of *P. xylostella* larvae against Neem oil and methanolic crude extracts of *A. excelsa* and *M. azedarach*.

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