Toxicology Reports 3 (2016) 473–480

Contents lists available at ScienceDirect

Toxicology Reports

journal homepage: www.elsevier.com/locate/toxrep

Evaluation of toxicity of biorational insecticides against larvae of the alfalfa weevil

Gadi V.P. Reddy[a,*], Frank B. Antwi[a], Govinda Shrestha[a], Takashi Kuriwada[b]

[a] Montana State University, Western Triangle Agricultural Research Center, 9546 Old Shelby Rd., P.O. Box 656, Conrad, MT 59425, USA
[b] Kagoshima University, Faculty of Education, Korimoto 1-20-6, Kagoshima, 890-0065, Japan

ARTICLE INFO

Article history:
Received 14 April 2016
Received in revised form 4 May 2016
Accepted 4 May 2016
Available online 5 May 2016

Keywords:
Low risk insecticides
Insect pathogenic fungi
Efficacy
Lethal concentration
Mortality rate

ABSTRACT

The alfalfa weevil, *Hypera postica* (Coleoptera: Curculionidae), is a major pest of alfalfa *Medicago sativa* L. (Fabaceae). While *H. postica* usually causes the most damage before the first cutting, in summer of 2015 damaging levels of the pest persisted in Montana well after the first harvest of alfalfa. Although conventional insecticides can control *H. postica*, these chemicals have adverse effects on non-target organisms including pollinators and natural enemy insects. In this context, use of biorational insecticides would be the best alternative options, as they are known to pose less risk to non-target organisms. We therefore examined the six commercially available biorational insecticides against *H. postica* under laboratory condition: Mycotrol® ESO (*Beauveria bassiana* GHA), Aza-Direct® (Azadirachtin), Met52® EC (*Metarhizium brunneum* F52), Xpectro OD® (*B. bassiana* GHA + pyrethrins), Xpulse OD® (*B. bassiana* GHA + Azadirachtin) and Entrust WP® (spinosad 80%). Concentrations of 0.1, 0.5, 1.0, and 2.0 times the lowest labelled rates were tested for all products. However, in the case of Entrust WP, additional concentrations of 0.001 and 0.01 times the lowest label rate were also assessed. Mortality rates were determined at 1–9 days post treatment. Based on lethal concentrations and relative potencies, this study clearly showed that Entrust was the most effective, causing 100% mortality within 3 days after treatment among all the tested materials. With regard to other biorational, Xpectro was the second most effective insecticide followed by Xpulse, Aza-Direct, Met52, and Mycotrol. Our results strongly suggested that these biorational insecticides could potentially be applied for *H. postica* control.

1. Introduction

Alfalfa weevil *Hypera postica* (Gyllenhall) (Coleoptera: Curculionidae), is the most destructive insect pest of alfalfa *Medicago sativa* L. (Fabaceae) in the intermountain west of the United States [1]. *H. postica* not only decreases yield and quality of the first cutting, but can also harm subsequent cuttings [2]. Both larvae and adults damage terminals, foliage and new crown shoots, thereby lowering crop yield and quality [3]. However, the larvae caused the most damage [4]. During severe infestations, larvae can cause substantial defoliation, resulting in severe first cutting losses [5]. Heavily infested fields may appear silver or white, with most leaves skeletonized or consumed entirely [1]. If large numbers of adults or larvae survive until harvest, they damaged stems and crown buds, retarding regrowth [6]. A decrease in stem elongation occurred at a density of 30–100% of the smallest larval density [7]. Residual effects from severe damage decrease plant vigor, resulting in lower stand density and poor yields in subsequent harvests [2].

Although *H. postica* is native to Europe it was inadvertently introduced into the western United States in the early 1900s [8], and into the eastern United States in the late 1940s [9]. In Montana, alfalfa is the second most important crop after small grains [10]. Alfalfa growers in Montana first began to notice *H. postica* during spring 2013 when the weevil caused considerable damage and yield losses [10]. In addition, alfalfa weevils caused economic damage in irrigated fields in the Yellowstone and Missouri river valleys in Montana [10]. Insecticidal treatment are economical when a larval population average between 1.5–2.0 larvae/stem, or 20 larvae/sweep [11]. In 2014 and 2015, *H. postica* outbreak occurred in Valier, Montana. Even though *H. postica* does the most damage before the first cutting [12], considerable damage was also noticed even after the first harvest.

To date, other than classical biological control, insecticide applications and early harvesting are the most common management strategies for alfalfa weevil [13]. However, most of the chemical insecticides used to manage this pest are extremely

* Corresponding author.
E-mail address: reddy@montana.edu (G.V.P. Reddy).

http://dx.doi.org/10.1016/j.toxrep.2016.05.003

Table 1
Materials and application rates used for the laboratory bioassays against *Hypera postica* larvae.

Treatment	Chemical name	Trade name	Concentrations (ml/l)	Source
T1	Untreated control	–	–	–
T2	spinosad (*Saccharopolyspora spinosa*)	Entrust® WP	0.000091, 0.00091, 0.0091, 0.0455, 0.091, and 0.182	Dow Agro Science LLC, Indianapolis, IN
T3	*Metarhizium brunneum* F52	Met52® EC	0.072, 0.36, 0.72, and 1.44	Novozymes Biologicals, Salem, VA
T4	*Beauveria bassiana* GHA	Mycotrol ESO®	0.072, 0.36, 0.72, and 1.44	LAM International, Butte, MT
T5	Azadirachtin (extracts from *Azadirachta indica*)	Aza-Direct®	0.144, 0.72, 1.44, and 2.88	Gowan Company, Yuma, AZ
T6	*B. bassiana* GHA + pyrethrins	Xpectro® OD	0.25, 1.25, 2.5, and 5.0	LAM International, Butte, MT
T7	*B. bassiana* GHA + cold pressed Neem extract	Xpulse® OD	0.072, 0.36, 0.72 and 1.44	LAM International, Butte, MT

hazardous to bees [14,15], and other beneficial insects like the parasitoids *Bathyplectes curculionis* (Thomson) (Hymenoptera: Ichneumonidae) and *Oomyzus incertus* Ratzburg (Hymenoptera: Eulophidae) [16]. Increasing concerns for environmental safety and insecticide resistance arising from a frequent use of synthetic insecticides affect the long-term feasibility of the current strategy of alfalfa weevil management [17]. Consequently, many alfalfa growers in north central and central Montana are looking for more environmental friendly control methods for managing this destructive pest.

In this context, as a green alternative to synthetic insecticides, use of biorational insecticides would be the best alternative options because these insecticides are usually considered low-risk agents having the features of low mammalian toxicity as well as less impact on non-target organisms [18]. The biorational insecticides include the use of naturally derived compounds from plants or microbes such as spinosyns and azadirachtin, living organisms (insect pathogenic fungi) such as *Beauveria bassiana* (Bals.) Vuill (Ascomycota: Hypocreales) and *Metarhizium brunneum* (*anisopliae*) (Metsch.) Sorokin (Ascomycota: Hypocreales) or the combined formulation of these insecticides [18]. In recent years, a number of biorational insecticides are commercially available and have been used or tested against variety of pest species such as aphids [19], thrips [20], and coleopteran pests [21,22]. No attempts have been made so far to study the effects of these insecticides on *H. postica* control except the studies by Hedlund and Pass [23] and Sakurai et al. [24], who showed the infection of *H. postica* with *B. bassiana*, and *M. brunneum*. This study therefore aimed to evaluate the toxicity of biorational insecticides against *H. postica* under the laboratory conditions.

2. Materials and methods

2.1. Rearing of insects

H. postica larvae were collected from alfalfa fields in Valier, Montana, USA, using sweep nets in July 2015 and taken to the laboratory. Larvae were placed in collapsible cages (12 × 10 × 10 cm), fed alfalfa foliage, and held at 22 ± 2 °C, 70–80% RH and an approximately 14:10 h L:D photoperiod. Field-collected larvae were separated by instar as described by Harcourt [25]. The instars ranged from first to fourth instars. The first instar is light yellow or tan in color with a darker head and about 1 mm long while the second instar is yellowish-brown with their head deepening to black, third and

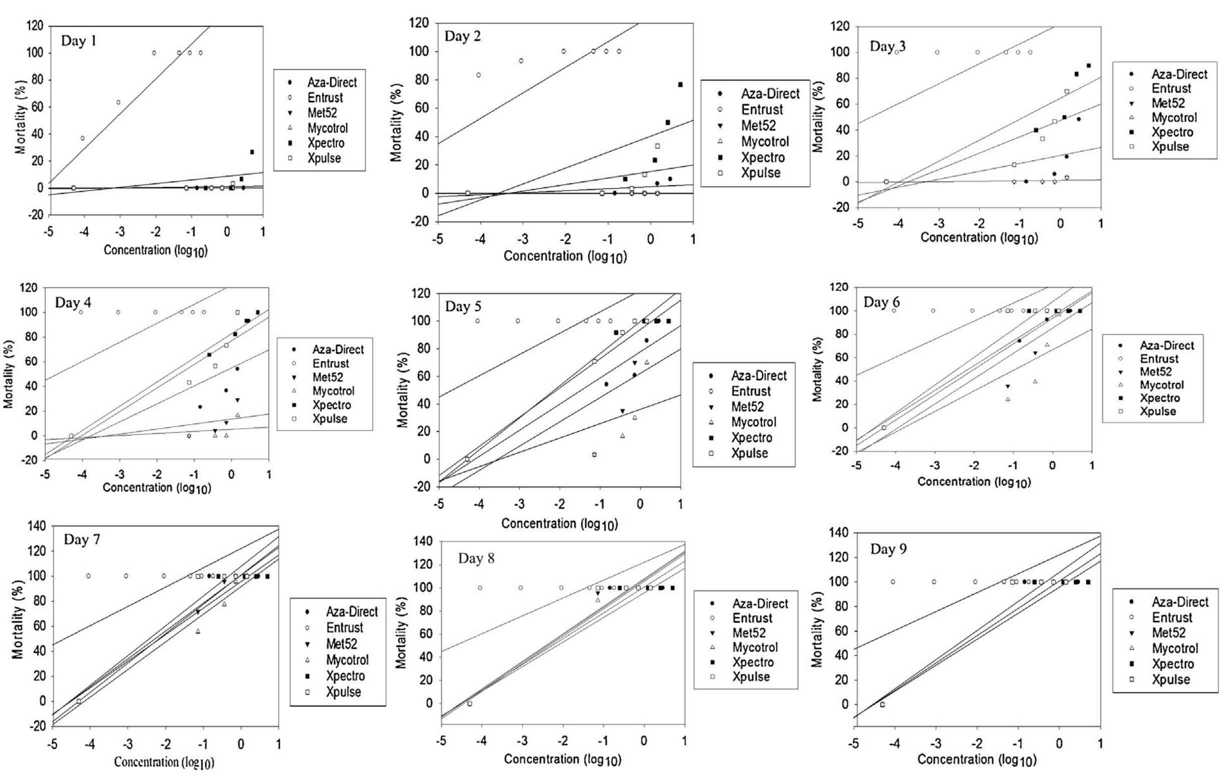

Fig. 1. Percentage mortality of 2nd instar larvae of *Hypera postica* treated with different concentrations (log) of biorational insecticides: Mycotrol® ESO (*Beauveria bassiana* GHA), Aza-Direct® (Azadirachtin), Met52® EC (*Metarhizium brunneum* F52), Xpectro OD® (*Beauveria bassiana* GHA + pyrethrins), Xpulse OD® (*Beauveria bassiana* GHA + Azadirachtin) and Entrust WP® (spinosad 80%) at days 1–9.