Effects of bacterial and jasmonic acid treatments on insect pests of canola

Kate Bergen[1], W. G. Dilantha Fernando[2], Neil J. Holliday[1]

[1]Department of Entomology, [2]Department of Plant Science, University of Manitoba
Winnipeg, MB, Canada R3T 2N2 Email: D_Fernando@Umanitoba.ca

Abstract

Two bacterial strains, *Pseudomonas chlororaphis* (PA23) and *Bacillus amyloliquifaciens* (BS6) have been shown to control fungal pathogens in canola, through various mechanisms including induced systemic resistance. Chemical changes in other plant systems due to induced systemic resistance have been shown to control certain insect pests. Jasmonic acid, a signalling molecule required for induced systemic resistance, has shown potential for use in the control of insect pests on other crops. The effects of the two bacterial strains and jasmonic acid on insect pests of canola were investigated in the field. Initial laboratory experiments carried out using diamondback moth (*Plutella xylostella*) have shown that jasmonic acid affects oviposition preference and larval feeding rates. Field experiments were carried out in 2006 to investigate the response of numerous insect pests to jasmonic acid and the bacterial treatments. Flea beetle damage, and numbers aphids, diamondback moths, lady beetles, lygus bugs, root maggots, and other species were assessed. No consistent treatment effects were seen, and potential explanations are discussed.

Introduction

Canola (oilseed rape) is the second most economically important crop in Canada, with an average annual production of 6.2 million tonnes (Canola Council of Canada 2005). There are many important insect pests of canola that can cause significant losses in yield. Due to concerns over the environmental and health issues involved in chemical control, there is a great deal of interest in alternatives to chemical control strategies. For the control of plant pathogens, one option that has arisen in the past few decades is the use of bacterial biocontrol agents. Strains of certain plant growth promoting rhizobacteria (PGPR) have been shown to control plant pathogens through multiple mechanisms (van Loon *et al*. 1998). Our research investigated the use of bacterial biocontrol agents in the control of insect pests of canola.

Two PGPR strains, *Pseudomonas chlororaphis* (PA23) and *Bacillus amyloliquifaciens* (BS6) have been investigated for the ability to control fungal pathogens in canola (*Brassica napus*). These two strains are able to control the pathogen *Sclerotinia sclerotiorum* (Fernando *et al.* 2006) and have shown potential to control the blackleg pathogen (*Leptosphaeria maculans*) (Ramarathnam and Fernando 2006). Fungal infection is controlled through the production of various bacterial metabolites, as well as through induced systemic resistance (ISR), a form of plant-mediated resistance that can help prevent or reduce the effects of subsequent pathogen challenge by activating and enhancing plant defences (van Loon *et al*. 1998). There is potential that activation and enhancement of the canola's natural defensive capacity through induced resistance could provide protection against insect pests. Jasmonic acid is the signalling molecule involved in the activation of ISR (Blechert *et al*. 1995), and its application can increase defensive compounds in canola that are active against insect pests (Bodnaryk and Rymerson 1994). Jasmonic acid was added as a treatment as a positive control for induced systemic resistance.

In 2006, we performed field studies to determine the effects of the two bacterial strains and of jasmonic acid on insect pests of canola. The chemical changes in the plants due to the activation of ISR may have the potential to control insect pests of canola in various ways. To investigate the effects of the treatments on canola, plots were sampled by beat tray and sweep net for a variety of insects, including aphids, bertha armyworm, diamondback moth and lygus bugs. Root samples were also taken to compare levels of damage by cabbage root maggots for each of the treatments

Materials and Methods

Experiments were carried out during the summer of 2006 on canola (*Brassica napus*) at the University of Manitoba's Carmen Research Station. The 42×19 m field was divided into 20 test plots of 2×6 m surrounded by a buffer. The treatments were assigned based on a randomized complete block design. Plots receiving the bacterial treatments were sprayed with 250×10^8 cfu per m^2 (3L per plot), which was diluted in 3L of water to allow all plants to be sprayed to runoff. Jasmonic acid (Sigma, Oakville, Ontario) was dissolved in methanol to produce a solution with a concentration of 0.5 millimolar, which was sprayed to runoff (6L per plot). Control plots were sprayed to runoff with 6L of water. Plants were treated in the early evening to avoid high evaporation rates during the heat of the day. Treatments were applied using a backpack sprayer containing the desired amount of each treatment for one plot.

The canola was to receive 2 applications of the treatments, one at the cotyledon stage and one at 30-50% bloom. Cotyledon and 1st true leaf samples were taken just before the first application of the treatments and one week after to compare levels of flea beetle damage. Due to poor emergence in the canola, a second field had to be planted, and this field was used for the second treatment application and subsequent sampling.

Beat cloth, sweep net and root samples were taken 1 and 3 weeks after the second application of treatments (July 26 and August 9). Total numbers of each insect species or family were recorded and log transformed for the analysis. Plants were pulled from each of the plots to assess root maggot damage. Root maggot pupae were collected at the end of the season to compare levels of parasitism between the treatments.

Results

Flea beetle damage

The cotyledon and 1st true leaf damage were analyzed by analysis of variance, and no significant treatment effects were found. A repeated measures analysis to compare changes between the pre- and post-treatment samples also showed not treatment effects.

Root Maggot Damage

Root samples were taken on the 3 occasions and brought back to the lab or processed in the field. Maggot damage was rated on a scale of 0–4 (Dosdall *et al*. 1994) (Fig. 1). No significant treatment effects were found when each sampling date was analyzed individually or when a repeated measures analysis was done to detect treatment effects on temporal pattern.

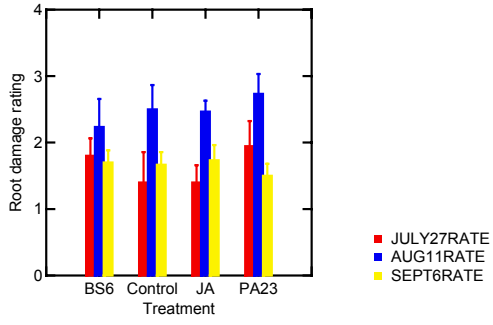

Figure 1. Mean (± SEM) root maggot damage ratings (Dosdall et al. 1994) for each sample date

Beat Cloth

Aphids, cabbage butterflies, diamondback moth larvae, lygus bugs, flea beetles, lady beetles and thrips were all present in sufficient numbers for statistical analysis. The only significant treatment effects were for flea beetles (Phylotreta cruciferae and P. striolata) on 26 July ($F=6.95$, $df = 3,12$, $P=0.031$) (Fig. 2); however, a Dunnett's test showed that none of the treatments were significantly different from the control.

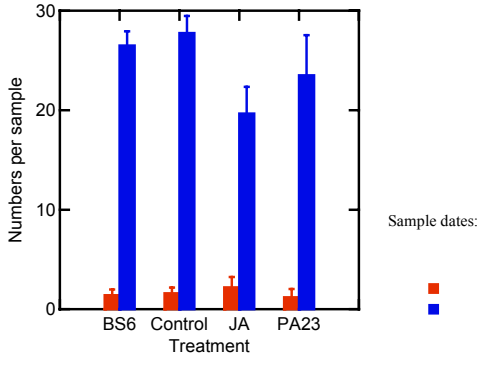

Figure 2. Mean (± SEM) number of flea beetles for July and August beat tray samples

Sweep Net

Aphids, diamondback moth larvae, lacewings, lygus bugs, flea beetles, lady beetles, thrips and several species of Diptera were present in sufficient numbers for statistical analysis. There were no significant treatment effects for any of the insects for 26 July or 9 August samples or an effect of treatment on the pattern of change between the sample dates.

Pupal Parasitism

Pupae collected from the field were placed in vials of moist vermiculite and sand (1:1) and placed in an incubator at 20°C with a 16:8h light:dark photoperiod. After 6 weeks, very few insects had emerged, so the remaining pupae were placed at 1°C for 12 weeks to complete diapause development.

Discussion

Our results so far show no consistent effects of treatment on any of the insects sampled or on flea beetle damage. Although there are relatively few studies that have shown the potential to control insect pests through the use of bacterial

biocontrol agents, it has been shown that some bacteria induce changes in chemical compounds that can negatively affect insect herbivores (Zehnder *et al* 1997). Work has also been done on the use of jasmonic acid to control insects in tomato. Thaler *et al* (2001) found that numbers of all major herbivores were significantly reduced on plants treated with jasmonic acid compared with control plants, and jasmonic acid treated plants showed significant increases in polyphenol oxidase and proteinase inhibitors that may reduce the growth and development of insects (Duffey & Stout 1996). Jasmonic acid has also been shown to affect insect pests of canola. Bodnaryk and Rymerson (1994) found that treating canola with jasmonic acid reduced flea beetle feeding and damage. Based on previous studies on the use bacterial biocontrol agents and jasmonic acid, we had predicted that treatment effects would be seen in the field, although this was not the case.

In the laboratory, we conducted experiments to examine whether the treatments affected adult diamondback moth (*Plutella xylostella*) oviposition preference and larval feeding and growth rate. From the oviposition experiment we found that jasmonic acid treated plants received significantly greater numbers of eggs than the controls. From the larval experiment we found that larvae feeding on jasmonic acid treated plants had significantly lower relative growth rates and consumed a significantly lower percent of the total leaf area.

Based on the laboratory results, it was expected that the jasmonic acid treatment would have had some affect on *Plutella xylostella*. Our field experiment was more likely to detect effects on oviposition preference than on feeding rate, as we examined numbers of diamondback moths rather than the damage they caused. The timing of the second application of treatments appears to have been after eggs had been laid, as there were many large larvae 1 week after the application and few 3 weeks after the application (Fig. 3). Thus the second application probably could not influence oviposition pattern in our study. Had the first and second applications both been made to the same plants, as had originally been planned, we might have seen evidence for oviposition preference; however, poor emergence prevented this. The experiment will be carried out again in 2007, and we will ensure that the plots receive 2 applications of the treatments.

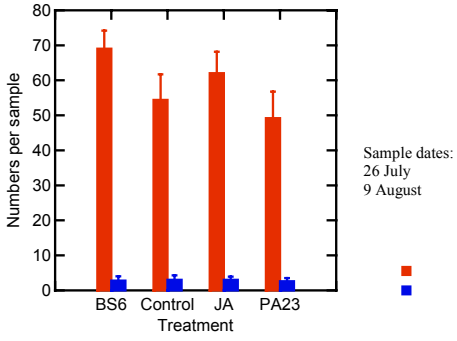

Figure 3. Mean (± SEM) number diamondback moths in July and August beat tray net samples

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