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Commentary

Control of western flower thrips (*Frankliniella occidentalis*) pupae with *Metarhizium anisopliae* in peat and peat alternative growing media

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Abstract

The entomogenous fungus *Metarhizium anisopliae* V275 was more efficacious than chemical insecticides (imidacloprid, fipronil) in killing pupae of the western flower thrips (70–90% versus 20–50%) in a range of horticultural growing media (peat, coir, bark and peat blends with 10% and 20% composted green waste). Premixing inoculum of *M. anisopliae* into the growing media gave better control than drench applications in coir and peat blended with 20% composted green waste. Use of *M. anisopliae* with sublethal dose of the insecticides gives slightly better control than the individual control agents but no clear cut additive or synergistic effects. Overall our study shows that *M. anisopliae* is efficacious in all growing media and compatible with conventional insecticides and offers much promise for the control of thrips as part of an integrated pest management program.

Keywords: *Metarhizium anisopliae*; *Frankliniella occidentalis*; Thrips pupae; Composts; Fipronil; Imidacloprid

1. Introduction

Western flower thrips (WFT), *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae), is one of the world's major pests causing damage to a wide range of economically important crops directly through feeding and indirectly through the transmission of harmful plant virus diseases (van Lenteren et al., 1992; Kirk and Terry, 2003). Thrips are difficult to control because of their high reproductive rate, cryptic habit (larvae hide in closed buds and pupate in soil) and resistance to many insecticides (van Lenteren and Loomans, 1998; Jensen, 2000; Herron and James, 2005). In addition to chemical insecticides, a range of biological agents are available for thrips control including arthropod predators and parasitoids, and insect pathogenic nematodes and fungi (Jacobson et al., 2001; Blaeser et al., 2004; Georgis et al., 2006; Xu et al., 2006). Whereas most attention has focused on the control of adults and larvae in the crop canopy little effort has been made to interrupt the life cycle by controlling the pupae.

The entomogenous, hyphomycete fungus *Metarhizium anisopliae* (Metsch) Sorokin has been studied extensively for the control of a wide range of pests, including WFT (Butt et al., 2001; Vestergaard et al., 1995; Maniania et al., 2002) and shows much promise for the control of subterranean pests (Zimmermann, 1992; Ansari et al., 2004). Unlike the canopy layer, the soil environment is less prone to dramatic fluctuations in temperature and humidity which can check fungal development. Indeed, Helyer et al. (1995) showed that *M. anisopliae* applied to peat-based media was effective in killing WFT pupae and helped reduce thrips populations. In light of growing pressure in the UK horticultural industry to reduce dependency on peat as a growing medium, we determined the efficacy of *M. anisopliae* for WFT pupal control in a range of alternative growing media.

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2. Materials and methods

2.1. Rearing of WFT

WFT were reared in ventilated plastic containers (29 cm × 29 cm × 16 cm) kept in a controlled temperature room set at 24 ± 2 °C, 50–60% RH and 16L:8D h photoperiod. Between 40 and 50 adult WFT were introduced into the containers and provided 3–4 pieces (8–9 cm length) of green bean (*Phaseolus vulgaris* L.) and 2–3 yellow chrysanthemum flowers. After three days, the egg infested beans were transferred to fresh ventilated plastic containers (28 cm × 20 cm × 10 cm). First instar larvae usually started to hatch 2 days later. Three days post-eclosion second instar larvae (L2) were collected for experimental use.

2.2. Fungal strain and maintenance

Metarhizium anisopliae strain V275 was used in this study, details of its selection, and steps to maintain its virulence are described in Shah et al. (2005). *M. anisopliae* strain V275, isolated from Codling Moth, *Cydia pomonella* L. (Lepidoptera: Tortricidae), was passed through mealworm, *Tenebrio molitor* L. (Coleoptera: Tenebrionidae) larva and isolated on Oatmeal dodine agar. Single spore colonies were transferred to Sabouraud Dextrose Agar (SDA) and culture incubated at 25 °C for 15 days in the dark, conidia obtained from 1st subculture were used for mass production of inoculum.

2.3. Production of conidia

Aerial conidia of *M. anisopliae* V275 was produced on broken Basmati rice (Jenkins et al., 1998) with slight modifications. The harvested conidia were air dried and 0.1 g suspended in 100 ml of 0.05% (v/v) aqueous Tween 80 and the number determined using a hemocytometer. The viability of the conidia was determined by the plate count technique on SDA (Goettel and Inglis, 1997) and always exceeded 98%.

2.4. Efficacy studies

Assays were done using 250 ml white opaque plastic pots (8 cm dia.) obtained from Tesco, UK. A 3 cm dia. hole was made into the centre of the lid to allow ventilation into the pot. To prevent thrips from escaping, thrips-proof nylon gauze (64 μm pore size) was glued onto the hole. A 5 cm × 4 cm yellow/blue sticky trap (AgriSense, UK) was attached to the inner part of the lid to trap emergent adults WFT. The efficacy of *M. anisopliae* V275 was determined in five different types of growing media kindly provided by Bord na Mona, Ireland. These included peat, coir, bark and peat blended with 10% and 20% composted green waste (CGW).

The following treatments were tested: *M. anisopliae* V275 was applied as a drench or premixed such that the final concentration was 1×10^{10} conidia/l of growing media. The chemical insecticides Provado® (Bayer a.i. 5% w/w imidacloprid) and Vi-Nil® (Certis, a.i. 1% fipronil) were applied as drench or premixed at the full concentration (FC) (65 ml and 1 g/l growing media) and pre-determined sublethal doses (SLD). A preliminary study showed that imidacloprid and fipronil SLD corresponded to 1% and 10% of the FC. Sublethal dose of insecticides were applied alone or combined with *M. anisopliae* V275. Control treatments were treated with water or 0.05% (v/v) Aqueous Tween 80. Preliminary study shows that there were no differences in WFT emergence in both control treatments so we included only one control (water) for data analysis.

For the drench application, 1×10^{10} conidia/l of growing media was diluted in 160 ml of Tween 80 and 20 ml of the homogeneous spore suspension was uniformly poured over the surface of the growing medium (125 ml volume per pot) and additional water was added to ensure the medium was at field capacity. For premixed applications, dried conidia were uniformly mixed into the growing media and water added so the compost reached field capacity. Similar adjustments were made for the chemical treatments. The final moisture levels of the growing media were ca. 45% (peat), 35% (coir), 25% (bark), 35% (10% CGW-peat blend) and 32% (20% CGW-peat blend).

Each pot was inoculated with 20 L2 WFT to a small piece of bean (2 cm length provided as a source of food) placed on the top of the growing media with the help of fine camel hair brush. The experiments were conducted in a controlled temperature (24 ± 2 °C, 50–60% RH and 16L:8LD photoperiods). Four days after the introduction of the L2, adult WFT started to emerge. These adhered to the sticky traps or were found on top of the growing media or the side of pot. Adult emergence was monitored using a binocular microscope over a 7 day period until no more WFT were observed. Adults trapped on sticky cards were incubated in Petri dishes lined with moist filter paper and examined under a binocular microscope to see if they were infected with *M. anisopliae*. The surviving adults

Fig. 1. Mean (± SE) mortality of western flower thrips with *M. anisopliae* (1×10^{10} conidia/l compost) alone, sublethal dose (SLD) or full concentration (FC) of imidacloprid or fipronil alone, or the combination of *M. anisopliae* with SLD of insecticides in 250-ml cup in different growing media. Ma-DR: *Metarhizium* applied as drench, Ma-PM: *Metarhizium* premixed, Imi-FC-DR: Imidacloprid applied as a drench at the FC, Fip-FC-PM: Fiprinol premixed at the FC, Imi-SLD-DR: Imidacloprid applied as a drench at the SLD, Fip-SLD-PM: Fiprinol premixed at the SLD, Imi-SLD+Ma-DR: Imidacloprid used at the SLD with *Metarhizium* applied as a drench, Imi-SLD+Ma-PM: Imidacloprid used at the SLD with *Metarhizium* premixed, Fip-SLD+Ma-DR: Fiprinol used at the SLD with *Metarhizium* applied as a drench, Fip-SLD+Ma-PM: Fiprinol used at the SLD with *Metarhizium* premixed. Means with same letter (11 days after treatment) in bars are not significantly different within each growing media by ANOVA and Tukey's test ($P < 0.05$). Data shown are corrected for control mortality.