

Leaf dipping as an environmental screening measure to test chemical efficacy against *Bemisia tabaci* on poinsettia plants

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Received 6 August 2008; revised 5 January 2009; accepted 2 March 2009

ABSTRACT: The screening of potential chemicals for control of *Bemisia tabaci* on poinsettia (*Euphorbia pulcherrima* c.v. Lilo Pink) plants using a leaf dipping technique was investigated. All relevant *B. tabaci* lifestages (eggs, larvae and adults) were investigated. In leaf dipping tests, Certis spraying oil was the only compound to show potential to be used as a control agent against *B. tabaci* eggs, with 81% obtained mortality. Oberon resulted in no mortality of *B. tabaci* eggs in the reported experiments. Leaf dipping against second instar and adult *B. tabaci* proved more effective for all chemical products. Dipping techniques using Majestik, Certis spraying oil and Agri-50 E all resulted in high second instar larval mortality (93, 87 and 85.5 %, respectively). Certis spraying oil again resulted in a high efficacy against adult *B. tabaci* with 100 % mortality obtained. The potential of the various chemicals to be incorporated into integrated pest management strategies for the control of *B. tabaci* is discussed.

Keywords: Crop; Insecticide; Integrated pest management; Plant sap; Quarantine pest

INTRODUCTION

The sweetpotato whitefly, *Bemisia tabaci* Gennadius (Hemiptera:Aleyrodidae), remains a worldwide pest (Landa *et al.*, 1994; Nomikou *et al.*, 2001) causing yield losses in many economically important crops (Fransen, 1994; Gerling and Mayer, 1996; Gerling *et al.*, 1980). Within the UK, *B. tabaci* poses a threat primarily to protected vegetable crops due to the large number of virus diseases it is known to transmit (Alegbejo, 2000; Jones, 2003). The pest can also cause damage by direct feeding on plant sap and indirect damage through contamination of leaves by honeydew excretion (Naranjo *et al.*, 2002; Nomikou *et al.*, 2001). The species has been intercepted annually in the UK on imported plant material since 1987, and statutory action aimed at eradication is taken whenever it is found (Cheek and McDonald, 1994). Eradication of whitefly has normally relied on treatments using chemical insecticides. Within the UK the active ingredients currently used against *B. tabaci* are imidacloprid, spiromesifen and thiacloprid. However, the continued development of whitefly populations

resistant to chemical insecticides (Ahmad *et al.*, 2002; Cahill *et al.*, 1994; 1996; Cannon *et al.*, 2005), indicates that further chemicals and or biological agents must be investigated for their efficacy against *B. tabaci* (De Barro, 1995; Dennehy and Williams, 1997; Osborne and Landa, 1992; Prabhaker *et al.*, 1985). To continue providing horticultural consultants and protected ornamental and horticultural growers with sufficient information on which to base commercially viable control decisions, data on the efficacy of a wider range of insecticides for the control of all lifestages of *B. tabaci* (eggs, larvae and adults) is urgently required. The aim of the current study was to investigate the efficacy of various low toxicity, and potentially, integrated pest management (IPM) compatible insecticide products for their ability to control all relevant *B. tabaci* lifestages. Leaf dip treatments were used as they ensure good coverage of the leaf undersides, which is where the whitefly eggs and larvae stages are located (Buxton and Clarke, 1994).

The experiments were undertaken in the Plant Health Quarantine Entomology Laboratories based at the Central Science Laboratory, York, UK in 2006.

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MATERIALS AND METHODS

Products and insect cultures

Bemisia tabaci were cultured under quarantine conditions in perspex cages (60 × 60 × 80 cm) on poinsettia (*Euphorbia pulcherrima* c.v. Lilo Pink) plants at 23 ± 1°C following the method of Cuthbertson *et al.* (2005a, b; 2008). The entomopathogenic fungus *Lecanicillium muscarium* was supplied as Mycotal from Koppert Biological Systems Ltd., UK. The selected insecticidal products to test were as follows: Majestik (natural plant extract, 2.5 mL/100mL water, Certis UK); Agri-50E (alginate/polysaccharide, 300 µL/100 mL, Fargo Ltd, UK); Certis spraying oil (petroleum oil, 1 mL/100 mL water, Certis UK); Savona (fatty acids, 2 mL/100 mL water, Koppert Biological Systems Ltd, UK); Oberon (spiromesifen, 0.05 g/100mL water, Certis UK).

Leaf dipping

Three life stages of *B. tabaci* were tested against; eggs, second instar larvae and adults. Plants were infested following the method of Cuthbertson *et al.* (2003) and cohorts at the desired life-stages were obtained using the methods and data of Butler *et al.* (1983), Bethke *et al.* (1991), Wang and Tsai (1996) and Cuthbertson *et al.* (2003, 2007). Four separate insecticide dilutions (all UK recommended dose rates) of each chemical and fungal product were prepared for replication purposes. Two poinsettia leaves containing eggs were dipped into each dilution for 10 s then allowed to air dry, before being placed within sealed petri dishes for each individual dilution of each insecticide. This procedure was repeated with leaves infested with second instar larvae. For adult studies, leaves were dipped and again allowed to air dry. Five adult whitefly were then placed on the leaf and held in position by a clip cage modelled on those described by MacGillivray and Anderson (1957). These were maintained in sealed Petri dishes. All Petri dishes were incubated at 20 °C, 14:10 L:D for 48 h. Control tests for each lifestage were also carried out using 0.02 % solution of the non-ionic wetting agent Agral (Syngenta Crop Protection Ltd., UK; active ingredient: alkyl phenol ethylene oxide) and water as appropriate.

Assessment of efficacy

Following chemical insecticide treatment the numbers of live and dead *B. tabaci* adults and larvae were recorded after 48 h. In the case of all the fungal

treatments (larval death determined both by no movement and the presence of mycelial mass where *L. muscarium* was applied) and all the *B. tabaci* egg trials, dishes were incubated for 7 days in order to allow both the fungus to germinate (Cuthbertson *et al.*, 2005b) and eggs to potentially hatch (Bethke *et al.*, 1991; Butler *et al.*, 1983; Wang and Tsai, 1996). Treated eggs were noted as live (hatched larvae) or dead (unhatched). Data underwent the non-parametric Kruskal-Wallis rank sum test to compare the effect of the different insecticides.

RESULTS AND DISCUSSION

Mortality of Bemisia tabaci eggs following leaf dipping

There was a significant difference in mortality of eggs after leaf dipping with the different active ingredients (Fig. 1). Exposure to Majestik, Savona and Certis spraying oil was followed by egg mortalities (33, 46 and 81 %, respectively) that were all significantly higher than the water control (Majestik and water: Kruskal-Wallis chi-squared = 4.6, df = 1, *P* = 0.03; test; Savona and water: Kruskal-Wallis chi-squared = 6.4, df = 1, *P* = 0.002; Certis Spraying Oil and water: Kruskal-Wallis chi-squared = 8.5, df = 1, *P* = 0.004). However, no significant difference was observed for other applications compared to the water control. Oberon had no effect on the *B. tabaci* eggs in this study

Mortality of Bemisia tabaci second instar larvae following leaf dipping

The comparative analysis for second instar larvae indicated highly significant difference (Kruskal-Wallis test, *P* < 0.001) between chemical treatments and the water control (Fig. 2). All of the chemical treatments resulted in over 65 % mortality of larvae. Oberon caused 73 % larvae mortality, significantly higher than the water control (Kruskal-Wallis chi-squared = 12.0, df = 1, *P* < 0.001). There was significant difference in mortality caused by Agri-50E and Majestik (Kruskal-Wallis chi-squared = 0.26, df = 1, *P* = 0.6). Dipping the larvae infested leaves in the fungus produced significantly lower larvae mortality than the chemical treatments (Kruskal-Wallis test, *P* < 0.01), but was still significantly higher than the water control (Kruskal-Wallis chi-squared = 5.1, df = 1, *P* = 0.02).

Mortality of Bemisia tabaci adults following exposure to treated leaves

Dipping of the leaves and then testing for resultant efficacy against adult *B. tabaci* generated lower mortality than that of second instar, apart from the treatment of Certis spraying oil, where complete

mortality of adult *B. tabaci* was recorded (Fig. 3) significantly higher than the other treatments (Kruskal-Wallis test, $P < 0.001$). The other chemicals all resulted in significantly higher mortality of *B. tabaci* adults than the water control (e.g. Majestik and water: Kruskal-Wallis chi-squared = 9.1, $df = 1$, $P = 0.003$).

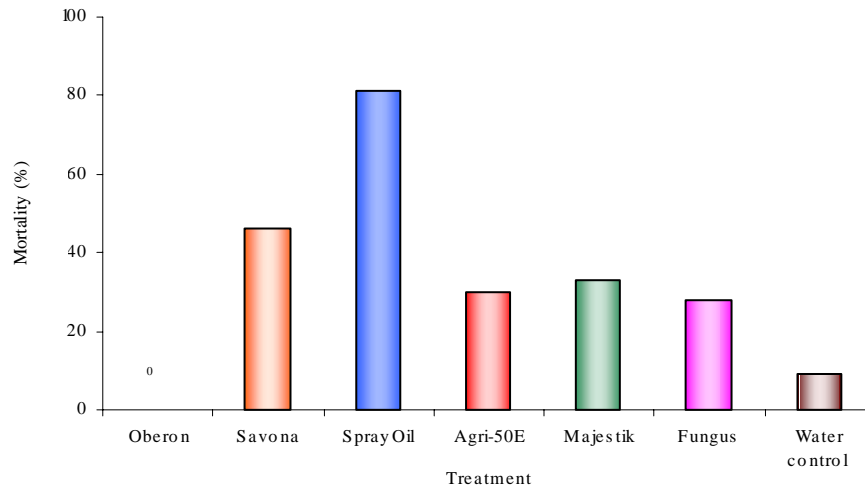


Fig. 1: The efficacy of insecticide treatments against *Bemisia tabaci* eggs following leaf dipping. Mortality assessed after 7 days

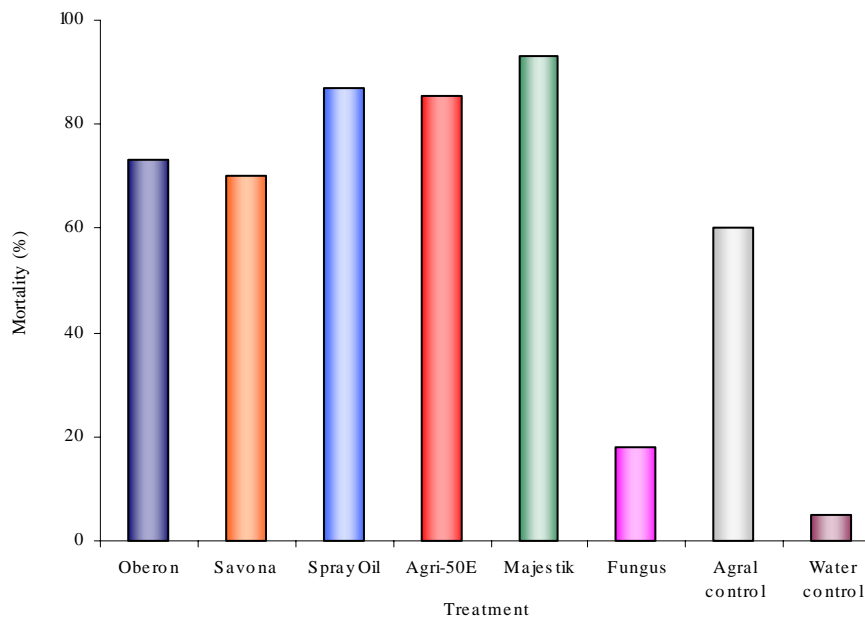


Fig. 2: The efficacy of insecticide treatments against *Bemisia tabaci* second instar larvae following leaf dipping; Mortality assessed after 48 h for all treatments except Mycotol (fungus) which was assessed after 7 days

Leaf dipping against *B. tabaci*

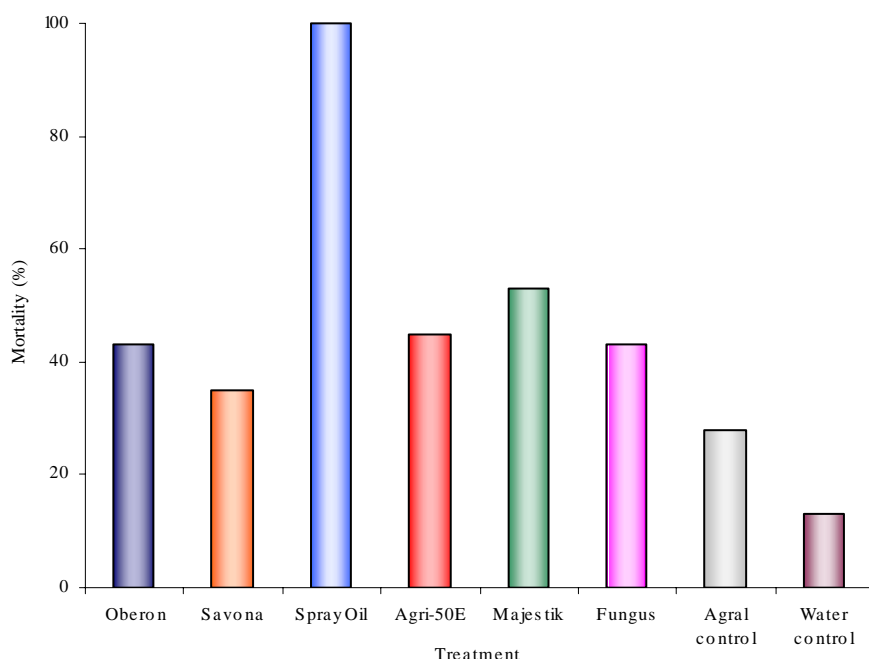


Fig. 3: The efficacy of insecticide treatments against *Bemisia tabaci* adults following leaf dipping; Mortality assessed after 48 h for all treatments except Mycotol (fungus) which was assessed after 7 days

This study, using leaf dipping as a screening method (Buxton and Clarke, 1994; Buxton *et al.*, 2008), investigated the potential of various low toxicity chemical and fungal products for their efficacy against various lifestages of *B. tabaci*. The collected information will be added to the knowledge base of potential chemicals already used against *B. tabaci* within IPM strategies in the United Kingdom (Buxton and Clarke, 1994; Cuthbertson *et al.*, 2005a).

Oberon, which has been reported as having low ovicidal properties (Nauen *et al.*, 2005), resulted in no mortality of *B. tabaci* eggs following leaf dipping. Indeed, all chemicals with the exception of Certis spraying oil produced low egg mortalities. The results from this study further confirm the difficulty of eradicating egg stages. Leaf dipping against second instar and adult life stages produced higher mortality. The chemicals Majestik, Savona, Agri-50E and Certis spraying oil all produced high mortalities of second instar larvae *B. tabaci*, such treatments could act as

a first line of defence in dealing with a *B. tabaci* outbreak. Buxton and Clarke (1994) also found spray oil to provide good control of both eggs and larval *B. tabaci* lifestages. In regards, to adult whitefly control Certis spraying oil showed great potential as total mortality of adult *B. tabaci* was achieved. It is thought that Certis Spraying Oil may act directly via a toxic action when the oil penetrates the insect cuticle (Martin and Woodcock, 1983) and indirectly by simply depriving the insects of oxygen (Buxton and Clarke, 1994).

Further work will now be to test the selected chemicals for their toxicity to non-target beneficial organisms with the aim of incorporating them into IPM strategies. Certis spraying oil is known to have only a moderate effect on the whitefly parasite *Encarsia Formosa* (Matteoni, 1993). However, the effects of these chemicals on the many predatory mite species' and entomopathogenic nematodes available for biological control of *B. tabaci* are unknown.

ACKNOWLEDGEMENTS

The authors thank Koppert Biological Systems Ltd, (UK) for supplying the formulation of *Lecanicillium muscarium*; Certis UK for the supply of chemical products used in this study and Mr Richard Natt (Fera Horticultural Manager) and his team for the provision of plant material. The work was funded by Plant Health Division, Defra. *Bemisia tabaci* were held under quarantine license number: PHL 251B/5328(02/2006) Amended (04/2006).

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This article should be referenced as follows:

Cuthbertson, A. G. S.; Blackburn, L. F.; Northing, P.; Luo, W.; Cannon, R. J. C.; Walters, K. F. A. (2009). Leaf dipping as an environmental screening measure to test chemical efficacy against *Bemisia tabaci* on poinsettia plants. *Int. J. Environ. Sci. Tech.*, 6 (3), 347-352.