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Further evaluation of the southern ladybird (*Cleobora mellyi*) as a biological control agent of the invasive tomato–potato psyllid (*Bactericera cockerelli*)

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HIGHLIGHTS

- There was no prey preference between psyllid and aphids, but an aversion to whitefly.
- Buckwheat effected ladybird longevity.
- Ladybird did significantly effect psyllid densities.
- There was a positive effect on potato tuber number and weight.

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ABSTRACT

The southern ladybird (*Cleobora mellyi* Coleoptera: Coccinellidae) is a voracious predator of the invasive tomato–potato psyllid (TPP) (*Bactericera cockerelli* Hemiptera: Triozidae) in New Zealand. We examined important aspects of the southern ladybird's ecology to obtain further insight into its potential as a biocontrol agent of TPP in potato crops. We found that the southern ladybird did not prefer TPP over either *Myzus persicae* Sulzer or *Macrosiphum euphorbiae* Thomas in choice tests, but avoided consumption of greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood). Ladybird longevity was tested under the conditions of low prey provision, a floral resource (buckwheat), and a combination of buckwheat and low density of TPP, over a 3 month period. There was no difference in longevity between ladybirds supplied with TPP only or buckwheat only. However, those with access to TPP and buckwheat lived longer than those with only TPP. In a glasshouse microcosm study, the ladybird was able to significantly reduce TPP densities after 3 weeks, and maintain the reduced numbers for 7 weeks. A species-level trophic cascade was found for both number and weight of potato tubers. These results indicate that the southern ladybird has potential as a biological control agent of the invasive tomato–potato psyllid in New Zealand.

1. Introduction

The 'new association' approach to biological control is based on the premise that a natural enemy introduced to manage a pest which has not coevolved with it could be a more effective biocontrol agent than one that is coevolved (Hokkanen and Pimentel, 1989; Eilenberg et al., 2001; Irvin and Hoddle, 2010). A meta-analysis showed this new association approach to biological control can be more successful than traditional classical biological control (Laing and Hamai, 1976). Recent studies continue to find supporting evidence for the new association approach (Lomer et al., 2001; Quimby et al., 2003; Goolsby et al., 2005). However, the debate continues as to whether the species most likely to be effective new association biological control agents are polyphagous, thus potentially resulting in non-target effects (Carruthers and Onsager, 1993; Roderick and Navajas, 2003). This study offers insight into a new species association between a recently established invasive species, tomato potato psyllid (TPP), *Bactericera cockerelli* Šulc (Hemiptera: Triozidae), and the southern ladybird, *Cleobora mellyi* Mulsant (Coleoptera: Coccinellidae).

TPP is native to Texas and Mexico, where it is considered a pest species causing substantial losses in tomato and potato crops (Liu et al., 2006). This psyllid has become an invasive pest species in parts of southern, central, western, and northwestern USA (Al-Jabr, 1999; Liu et al., 2006; Swisher et al., 2012). TPP was first

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detected in New Zealand in May 2006 and spread rapidly throughout the country (Teulon et al., 2009; Thomas et al., 2011). This herbivore is a major pest of solanaceous crops, inducing a plant condition known as psyllid yellows (Munyaneza et al., 2007; Sengoda et al., 2010). Tomato potato psyllid also vectors the bacterium *Candidatus* Liberibacter solanacearum syn. Psyllaurous (CLso), the purported causal agent of zebra chip disease in potatoes (Munyaneza et al., 2007; Munyaneza, 2010), which causes substantial yield loss (Munyaneza et al., 2007, 2008; Liefting et al., 2008). This bacterium is also vectored to important crops such as tomato, capsicum, eggplant and tamarillo (Liefting et al., 2009).

The management of TPP has focused primarily on insecticide use, presently the only approach capable of providing efficient management of zebra chip disease by killing TPP (Goolsby et al., 2007; Munyaneza, 2010). However, the long term use of chemical control alone is unsustainable due to factors such as the evolution of insecticidal resistance and disruption of integrated pest management programs. The need to reduce reliance on chemical controls has resulted in research on reduced insecticide applications (Anderson et al., 2013), use of entomopathogens (Lacey et al., 2009, 2011) and natural enemies as biological control options (Al-Jabr, 1999; Walker et al., 2011; O'Connell et al., 2012). In New Zealand, resident natural enemies have failed to control TPP numbers in potato crops during mid and late summer when conditions ares ideal for TPP development (Walker et al., 2011).

The southern ladybird is endemic to Australia (Slipinski, 2007) and feeds predominantly on the larvae of chrysomelids and hemipterous insects such as aphids and psyllids (Baker et al., 2003; Slipinski, 2007; Murray et al., 2008). This ladybird was originally introduced to New Zealand in the 1970s and 1980s as a biological control agent for the eucalypt tortoise beetle, *Paropsis charybdis*, a major defoliator of *Eucalyptus* spp. (Baker et al., 2003; Berndt et al., 2010). Previous research on this ladybird investigated predator behavior and rates of TPP consumption (O'Connell et al., 2012). Adults and larvae of the southern ladybird were voracious predators of TPP, consuming up to 100 TPP nymphs over 24 h in a laboratory bioassay, but were impeded by the dense trichomes on tomato leaves (O'Connell et al., 2012).

Assessment of predator voracity is important in determining a biological control agent's potential to reduce prey numbers (Lucas et al., 1997). However, the presence of alternative prey may affect voracity toward the target if these are preferred by the predator (Lucas et al., 1997). New Zealand has several phloem-feeding herbivores in potato crops with potential to serve as alternative prey for the southern ladybird. In particular, aphids are a major pest in potato crops (Stufkens and Teulon, 2001; van Toor et al., 2008) with species including the green peach aphid, *Myzus persicae* Sulzer, and the potato aphid, *Macrosiphum euphorbiae* Thomas, (Stufkens et al., 2000).

Ladybirds may also utilize alternative foods such as pollen and nectar when prey is scarce (Majerus, 1994; Coll and Guershon, 2002; Wäckers et al., 2008; Lundgren, 2009). Buckwheat, *Fagopyrum esculentum* Polygonaceae, has been widely used as a floral resource to provide pollen and nectar to natural enemies such as lacewings, hoverflies, and parasitoids (Landis et al., 2000; Robinson et al., 2008; Jonsson et al., 2009). Buckwheat may increase local natural enemy numbers (Stephens et al., 1998), parasitism rates (Stephens et al., 1998; Berndt et al., 2002) and extend the longevity of natural enemies (Irvin et al., 2006; Robinson et al., 2008).

Little is known about ladybird-psyllid population dynamics because psyllids are often considered less important prey than other herbivores (Hodek and Honěk, 2009). In systems where psyllids is the predominant insect pest, investigations have examined consumption rates, behavior, prey choice, life-cycles and developmental rates of the ladybirds (e.g. Mehrnejad and Jalali, 2004; Pluke et al., 2005; O'Connell et al., 2012). Some studies have gone on to show ladybirds can significantly impact psyllid numbers on host plants (e.g., Michaud, 2004), or as part of the predatory insect guild (Westigard et al., 1968; Qureshi and Stansly, 2009).

We used a combination of glasshouse and laboratory experiments to examine prey preference of the southern ladybird for common hemipteran pests of potatoes and investigated the effect of a floral resource and TPP availability on southern ladybird longevity. We also examined predator-prey dynamics between the southern ladybird and TPP on potato plants in a microcosm experiment.

2. Materials and methods

2.1. Prey preference of adults and larvae

Prey preference of adult and larval southern ladybirds were examined in three treatments utilizing immature prey only: (1) TPP + green peach aphid; (2) TPP + whitefly; (3) TPP + potato aphid. To standardize ladybird hunger levels, all individuals were starved for 24 h before testing. All prey species were reared on potato plants (cv. Desiree) for multiple generations. Ladybirds were obtained from Bioforce Ltd (Auckland, New Zealand) and maintained on a mixture of all four prey species to ensure prior experience with these prey. Third and fourth instar ladybird larvae were used in test prey preference.

Tests were conducted in Petri dishes (8.5 cm × 2.5 cm) in a temperature-controlled room (24 ± 2 °C). The Petri dishes were modified by removing the ribs under the lid and a moist filter paper was placed in the bottom of each. Two similar-sized potato leaflets from insect-free plants were placed in each dish opposite each other. One leaflet was randomly assigned 10 TPP and 10 individuals of the alternative prey were placed on the other leaflet. After prey placement, a single ladybird was placed in the centre of the dish and left to forage for 3 h. At the end of the experiment, adult ladybirds were euthanized and dissected to determine their sex.

2.2. Ladybird longevity

We used newly-emerged (<4 days), unmated adults to determine the longevity of adult ladybirds on a floral resource and/or TPP. All ladybirds were provided with TPP and green peach aphids ad libitum prior to the experiment. The experiment was a 2 × 2 factorial design with eight replications of three treatments plus a control: (1) water only, (2) buckwheat only, (3) TPP only, (4) TPP + buckwheat. Water was provided in each replicate in a 1.5 ml microcentrifuge tube with a moist wick, replaced every 3 days. One ladybird of unknown sex was placed into each experimental container. Buckwheat seed was sourced from (King Seeds, Katikati, New Zealand) and grown as single plants in 500 ml pots.

The experiment was carried out using plastic containers (18.5 cm × 10.5 cm), each erected on a bamboo pole. This enabled the containers with buckwheat (cv. Katowase) treatments to be vertically adjusted depending on the height of the most apical buckwheat inflorescences. Each experimental container had a foam plug in the bottom with a slit to allow for a buckwheat stem, while sealing the container. There was a small foam plug in the side to allow TPP to be added. The most apical inflorescence was enclosed on each buckwheat plant, without leaves. The buckwheat was replaced weekly to ensure a constant supply of nectar and pollen. A mixture of third, fourth and fifth TPP nymphs were provided daily to appropriate treatments on potato (cv. Desiree) leaflets placed in a 3.0 cm diameter plastic dish at the bottom of the experimental container; leaflets from the previous day were removed.