

## Efficacy of a micro-encapsulated formulation compared with a sticky barrier for excluding ants from citrus canopies

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### Keywords

ant barriers, Inesfly IGR FITO<sup>®</sup>, *Lasius grandis*, *Linepithema humile*

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Received: April 9, 2010; accepted: July 1, 2010.

doi: 10.1111/j.1439-0418.2010.01572.x

### Abstract

Inesfly IGR FITO<sup>®</sup> is an insecticidal paint containing chlorpyrifos and pyriproxyfen incorporated in a micro-encapsulated formulation that confers the advantage of releasing active ingredients slowly. In this study, a 15-cm band of Inesfly IGR FITO<sup>®</sup> was painted around citrus trunks. The efficacy of this paint and a sticky barrier to exclude ants from foraging in citrus trees was evaluated in two citrus orchards during the season in two different ant communities, one dominated by *Lasius grandis* and the other by *Linepithema humile*. Field results demonstrated that a single application of Inesfly IGR FITO<sup>®</sup> at the beginning of the season was highly effective in excluding ants from canopies throughout the season. Inesfly IGR FITO<sup>®</sup> provides an efficient and more economical alternative than current ant exclusion strategies used in many perennial crops. Further studies should be performed to determine the effects of this strategy on other pests and on beneficial arthropods in citrus.

### Introduction

Ants have long been associated with outbreaks of citrus pests (DeBach 1951; James et al. 1999). Ants protect honeydew-producers pests such as aphids and soft scales from predators and parasitoids and in return they feed on the sugar-rich secretion, honeydew, produced by these hemipterans (Way 1963; Buckley 1987). Furthermore, in their trail from the nest to the hemipteran colony in the citrus canopy, ants may disrupt the activity of parasitoids and predators of other pests that do not produce honeydew such as armored scales (Bartlett 1961; James et al. 1997; Pekas et al. 2010a). Current strategies to control ants generally include the use of insecticides and physical barriers to prevent their access to canopies. Physical barriers based on sticky materials banding the trunk exclude ants rapidly and effectively from foraging in the canopies. However, this method requires monthly replacements of bands, which is laborious and expensive (Vega and Rust 2001; Vincent et al. 2001; Pekas et al. 2010a). Insecticide

treatments include broad spectrum insecticide surface sprays, toxic baits and insecticide-treated trunk barriers. The first option primarily kills the aboveground foragers, but the colony itself is usually not eliminated (Davis and Van Schagen 1993). Moreover, this is not a selective strategy and may impact natural enemies (Smith et al. 1996). Broadcast application of toxic baits is generally considered the most efficient method to control multiple colonies over a large area in different crops (Stanley 2004; Greenberg et al. 2006; Daane et al. 2008). Toxic baits are carried into the nest where they usually kill the brood and sterilize or kill the colony queen(s), which eliminates the entire colony (Williams 1993). However, their action is not immediate and does not last the entire season (Rust et al. 2004; Daane et al. 2007, 2008). In addition, important technical details such as bait station design, seasonal periods of deployment and bait insecticide formulations have yet to be properly determined for Mediterranean citrus ants. Finally, insecticide-treated trunk barriers, as physical barriers, exclude ants from foraging in trees instantaneously and, moreover, kill

foraging workers that come into contact with the insecticide. Nevertheless, the effectiveness of insecticide-treated trunk barriers is limited by high temperature, irrigation and ground cover (Rust et al. 2000) and they usually degrade within 2–3 months, increasing the need for repeated applications (Tollerup et al. 2006). Only James et al. (1998) did achieve a prolonged ant exclusion using plastic controlled-release bands impregnated with chlorpyrifos.

Inesfly IGR FITO<sup>®</sup> (Industrias Químicas Inesba S.L., Paiporta, Spain) is an insecticidal paint (active ingredients 3.0% chlorpyrifos and 0.063% pyriproxyfen) in a micro-encapsulated formulation. Both insecticides are widely used to control insect pests and have been previously used against ants in citrus (Klotz et al. 2003) and other perennial crops (Banks and Lofgren 1991). The micro-encapsulated formulation is a polymer composed of two parts: (i) vinyl acetate that gives the polymer its consistence and its capacity of carrying active ingredients and (ii) versaic acid ester that provides resistance to water and sunlight. Inesfly IGR FITO<sup>®</sup> confers the advantage of releasing active ingredients slowly, so paint does not have to be applied frequently and its effect can last from six to twelve months (López et al. 1999; Mosqueira et al. 2005; Días and Jemmio 2008; Amelotti et al. 2009; Llácer et al. 2010). To our knowledge, no formulation of this polymer has been previously tested to exclude ants from tree canopies.

The objective of this study was to evaluate the efficacy and persistence of the paint Inesfly IGR FITO<sup>®</sup> as insecticide-treated trunk barrier compared to a Tangle-Trap sticky barrier to exclude two different ant communities found in Mediterranean citrus orchards from foraging in tree canopies.

## Materials and Methods

### Study sites

The study was conducted in two Clemetine (*Citrus sinensis* var. 'clemenules') orchards located at La Pobla de Vallbona (UTM X713419 Y4390371; 145 m altitude) and Bétera (UTM X722481 Y4385219; 35 m altitude) (Valencia, Spain) in 2009. Both orchards were drip irrigated and a cover crop was maintained. The orchard of La Pobla de Vallbona (8.24 ha) was sown with a mono-specific sod of *Festuca arundinacea* Schreber (Poacea) that was mowed mid-spring. The ant community of this orchard was dominated by *Lasius grandis* Forel (Formicinae) and *Pheidole pallidula* (Nylander) (Myrmicinae) (Vanaclocha et al. 2005), the native ant species commonly found in Spanish citrus agroeco-

systems (Alvis 2003; Vanaclocha et al. 2005; Cerdá et al. 2009). The orchard in Bétera (3.49 ha) preserved a natural cover crop (the five most abundant plant species were *Convolvulus arvensis* L., *Conyza Canadensis* L., *Hordeum leporinum* L., *Amaranthus retroflexus* L., *Avena* sp.) that was mowed twice during spring and once at the beginning of fall. The ant community was dominated by the exotic ant *Linepithema humile* (Mayr) (Dolichoderinae) (Vanaclocha et al. 2005).

### Ant-exclusion and ant activity

In each orchard, 12 blocks were selected. Each block consisted of four adjoining trees in a square distribution in which ants had been previously observed. Four of these blocks were randomly selected for treatment with Inesfly IGR FITO<sup>®</sup>, four with the physical barrier Tangle-Trap<sup>®</sup> (Bioestimulantes Agrícolas S.L., Massalfassar, Spain) and the other four were left untreated. Inesfly IGR FITO<sup>®</sup> was applied by painting a 15-cm wide band of the trunks with a 170–200 g/m<sup>2</sup> paint deposit. Tangle-Trap<sup>®</sup> was sprayed monthly over 15 cm width adhesive bands Tesapack<sup>®</sup> Universal (Tesa Tape S.A., Argentona, Spain), placed around the trunk of the trees. Each barrier was placed 20 cm high above the soil line. Trees were pruned before barrier applications to prevent ants from using alternative routes into the canopies. Ant activity was evaluated monthly from April to December 2009 by counting during 2 min the number of ants ascending and descending through a 15-cm wide imaginary band 5 cm above barriers (Pekas et al. 2010a). Ant activity on the selected trees was assessed previously to barrier applications, to confirm homogeneity among the 12 selected blocks.

### Statistical analysis

Ant activity data were analysed monthly using one-way ANOVA and mean values were compared using a Tukey's test ( $P < 0.05$ ). Ant activity was log-transformed [ $\log(x + 1)$ ] to correct for heterogeneity of variance. Trees were used as independent samples because no significant differences were found in ant activity among the 12 blocks of each orchard in the sample prior to the application of the barriers.

## Results

### Ant abundance

*Lasius grandis* was the most active ant in the orchard of La Pobla de Vallbona comprising 75.5% of the

individuals counted on citrus trunks over the year, followed by *P. pallidula* and *Formica rufibarbis* Fabricius (Formicinae) with 16.2% and 7.5% of individuals, respectively. *Linepithema humile* was the unique species observed in the orchard of Bétera (table 1).

### Seasonal activity and efficacy of barriers

There were no significant differences in ant activity among blocks in either orchard at the beginning of the season before the barriers were applied (La Pobla de Vallbona:  $F = 0.83$ ; d.f. = 11, 36;  $P = 0.62$ ; Bétera:  $F = 1.29$ ; d.f. = 11, 30;  $P = 0.28$ ). Ant activity at La Pobla de Vallbona, peaked in July, then decreased but continued through 24 October (fig. 1). Monthly applications of Tangle-Trap<sup>®</sup> and a single application of Inesfly IGR FITO<sup>®</sup> in April significantly reduced ant activity until September when ant activity in trees treated with Inesfly IGR FITO<sup>®</sup> increased and became similar to control trees. The most active ant in September was *P. pallidula* (129 ants) followed by *F. rufibarbis* (29 ants) whereas *L. grandis* activity was almost null (three ants). The Tangle-Trap<sup>®</sup> barriers totally excluded foraging activity of ants in citrus canopies until the end of the season. In Bétera, *L. humile* remained active until December, with higher activity from July to September (fig. 2). Inesfly IGR FITO<sup>®</sup> barriers acted as a complete ant barrier at Bétera. The Tangle-Trap<sup>®</sup> barriers acted as a non-continuous barrier during the experiment in this location. Finally, no phytotoxic effects due to Inesfly IGR FITO<sup>®</sup> were observed on the trunk or canopy of the assayed trees.

### Discussion

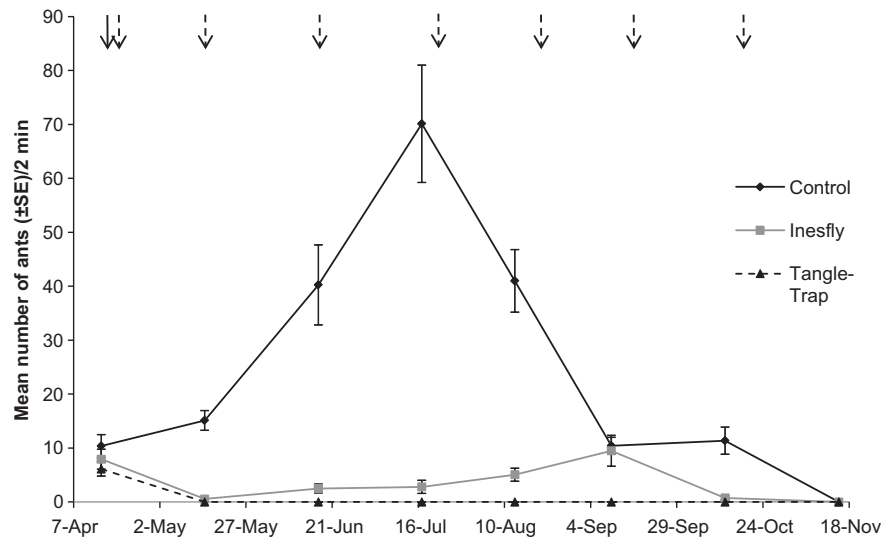
The seasonal activity of *L. grandis* described herein coincides with the published by Pekas et al. (2010b) and Alvis (2003). A single application of Inesfly IGR

FITO<sup>®</sup> at the beginning of the season and monthly applications of Tangle-Trap<sup>®</sup> exclude most ants from canopies throughout the season (fig. 1). The increase of ant activity in trees treated with Inesfly IGR FITO<sup>®</sup> at the end of the summer coincided with the increase of *P. pallidula* and *F. rufibarbis* which may benefit from the exclusion of *L. grandis*. This species and *P. pallidula* avoid nesting and foraging in the same trees (Pekas et al. 2010b). The number of Argentine ant specimens ascending and descending the trunk was much higher than that of the Mediterranean native ants from April to December (fig. 2). *Linepithema humile* was even able to elude Tangle-Trap<sup>®</sup> barriers. Despite this activity pattern, Inesfly IGR FITO<sup>®</sup> maintained the citrus canopies free of *L. humile*. The exclusion of ants began in April, when high densities of *L. humile* were already present in canopies. Considering this high activity at the beginning of the season, and the long persistence and high efficacy of this insecticide-treated trunk barrier, we would suggest growers apply this barrier at least one month earlier, in late February to early March as recommended by Markin (1970). Finally, Inesfly IGR FITO<sup>®</sup>, as a physical barrier, avoided ant transit immediately after application. This provides an advantage with respect to the use of baits that act more slowly.

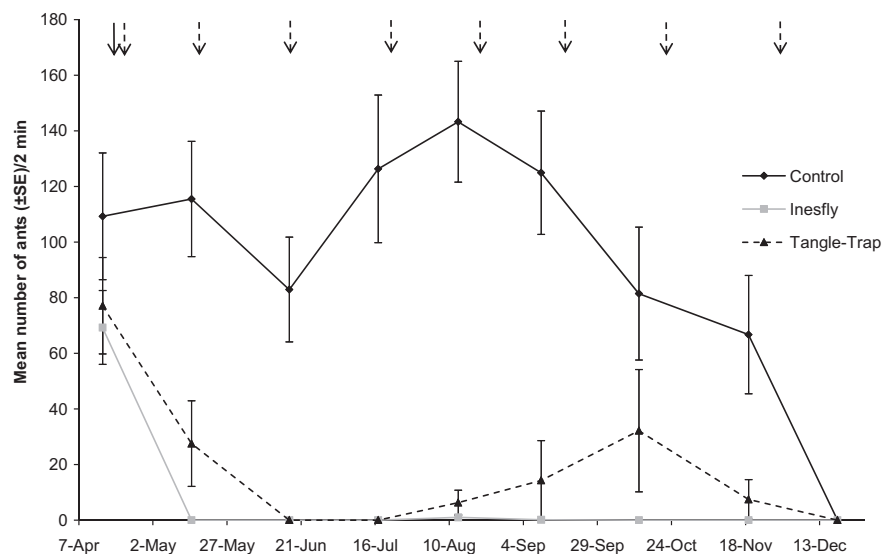
The present work demonstrates that Inesfly IGR FITO<sup>®</sup> can effectively exclude ants from citrus canopies throughout the season. Field results obtained proved that the micro-encapsulated formulation was effective in excluding ants from two different ant complexes in two citrus orchards, including *L. grandis*, the most abundant ant in Mediterranean citrus (Alvis 2003; Vanaclocha et al. 2005; Cerdá et al. 2009), and the invasive Argentine ant *L. humile*, the most damaging ant in other citrus areas such as Californian citrus (DeBach 1951; Vega and Rust 2001). Moreover, the estimated product cost of an Inesfly

**Table 1** Ant species and total number of ants counted in ant-allowed (control) and ant-excluded (Tangle-Trap<sup>®</sup> and Inesfly IGR FITO<sup>®</sup>) trees in two citrus orchards sampled from April to December 2009

Ant species	La Pobla de Vallbona			Bétera		
	Control	Tangle-Trap	Inesfly IGR FITO	Control	Tangle-Trap	Inesfly IGR FITO
<i>Linepithema humile</i> (Mayr)	0	0	0	9871	1323	17
<i>Lasius grandis</i> Forel	2492	0	38	0	0	0
<i>Pheidole pallidula</i> (Nylander)	399	0	144	0	0	0
<i>Formica rufibarbis</i> Fabricius	103	0	150	0	0	0
<i>Camponotus sylvaticus</i> (Olivier)	12	0	6	0	0	0
<i>Plagiolepis schmitzii</i> Forel	7	0	1	0	0	0
Total	3013	0	339	9871	1323	17



**Fig. 1** Seasonal activity of a Mediterranean citrus ant community on canopies of trees where Inesfly IGR FITO<sup>®</sup> and Tangle-Trap<sup>®</sup> were used as barriers and without barriers (control) along 2009 in La Pobra de Vallbona citrus orchard. Activity is presented as the mean ( $\pm$ SE) of the ants ascending and descending the tree trunk during 2 min. The arrows show the day that barriers were applied. For each sample date, the overlap of confidence intervals indicates non-significant differences among means (Tukey's test:  $P < 0.05$ ).



**Fig. 2** Seasonal activity of *L. humile* on canopies of trees where Inesfly IGR FITO<sup>®</sup> and Tangle-Trap<sup>®</sup> were used as barriers and without barriers (control) along 2009 in Bétera citrus orchard. Activity is presented as the mean ( $\pm$ SE) of the ants ascending and descending the tree trunk during 2 min. The arrows show the day that barriers were applied. For each sample date, the overlap of confidence intervals indicates non-significant differences among means (Tukey's test:  $P < 0.05$ ).

IGR FITO<sup>®</sup> treatment per tree (748.8 g/32 trees  $\times$  11€/1000 g = 0.26€/tree) was more economic than that of Tangle-Trap<sup>®</sup> applications per tree (6800 g/32 trees  $\times$  12.16€/283.5 g = 9.15€/tree). Further research should focus on secondary side effects of this methodology and on the possibility of implementation in large citrus areas.

## Acknowledgements

Two anonymous referees provided valuable comments to the manuscript. We are grateful to Aureli Marco and Martí Llavador for their help managing citrus orchards, Pablo Bru for technical assistance, and Dr Jesús López Ferrer for the insecticidal paints

supply and for technical collaboration (Agreement 7717 of 12-09-2007 between Industrias Químicas Inesba S.L. and IVIA). This research was partly funded by Conselleria d'Agricultura, Pesca i Alimentació from Generalitat Valenciana. M.J.B. and P.V. were recipient of a PhD grant from IVIA.

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