

Microencapsulated Bait: Does It Work With Red Imported Fire Ants, *Solenopsis invicta* (Hymenoptera: Formicidae)?

by

Lekhnath Kafle¹, Wen-Jer Wu¹, Robert K. Vander Meer², Yi-You Huang³
& Cheng-Jen Shih^{1*}

ABSTRACT

The preference of red imported fire ants, *Solenopsis invicta*, for microencapsulated (MC) pyriproxifen based corn grit baits (P-bait) was conducted in laboratory and field conditions. A positive correlation between the microencapsulation rate and water tolerance ability of P-bait was observed. A 20% increment of water tolerance ability of P-baits was observed when P-baits were microencapsulated with 5% poly- ϵ -caprolactone. The microencapsulated and wet P-baits were less preferred than normal P-baits by the fire ants either in laboratory or field conditions. Based upon these results, poly- ϵ -caprolactone coated fire ant baits cannot be considered as perfect fire ant baits.

Key words: *Solenopsis invicta*; P-bait; microencapsulation; preference; water tolerance

INTRODUCTION

Since its accidental introduction in the southern United States in 1930s, the red imported fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae), has been an invasive pest threatening agriculture, local biodiversity and public health (Williams *et al.* 2001). Morrison *et al.* (2004) had predicted that many areas around the world, including large portions of Europe, Asia, Africa, Australia, and numerous island nations, are at risk for *S. invicta* introduction. That prediction was proved by the recent invasion of this devastating invasive pest in Hong Kong and mainland China in 2004, Mexico in 2005 (Sánchez-Peña *et al.* 2005), Taiwan in 2003 (Chen *et al.* 2006) and Australia in 2001 (Henshaw *et al.* 2005). Several techniques are in practice

¹Department of Entomology, National Taiwan University, Taipei 106, Taiwan

²Agricultural Research Service, U.S. Department of Agriculture, Gainesville, Florida 32608, USA

³Department of Biomedical Engineering, National Taiwan University, Taipei 100, Taiwan

*corresponding author: Email: shihcj@ntu.edu.tw

around the infested areas either to control the fire ant population or to limit their further expansion. To control *S. invicta* in the larger areas, toxic bait products are reported as the most effective tool (Vogt *et al.* 2003). The fire ant baits are prepared with three major components, a toxicant, a vegetable oil phagostimulant, and a carrier (usually corn grits) for the phagostimulant/toxicant solution, that allows easy distribution of the bait. One of the major drawbacks of bait formulations is their propensity to degrade when wet. Consequently, it is generally recommended that baits not be applied to wet ground or grass, nor if rain is expected within 24 h. Therefore, there is a need for the development of a high humidity or water resistant fire ant bait that would make the success or failure of a fire ant bait less weather dependent. In addition, such a bait could play a critical role in the success of control programs in humid areas, including Taiwan, Hong Kong, and mainland China. Microencapsulation is a technique that could be applied to fire ant baits to make them more resistant to high humidity and water. This method allows a particle to be encapsulated inside a tiny polymer sphere, and can stabilize the core active ingredient against environmental degradation (Shukla & Sivaram 1999). Poly- ϵ -caprolactone is biodegradable and could be used as a microencapsulation coating material (Schubert & Hochwimmer 2001). The objectives of this study were i) evaluation of the water resistance of microencapsulated (MC) pyriproxifen based corn grit bait (P-bait) and, ii) determination of the effect of microencapsulation on the feeding activity of *S. invicta* workers under laboratory and field conditions.

MATERIALS AND METHODS

Microencapsulation of P-bait

Poly- ϵ -caprolactone (typical Mn = 80,000, Aldrich, St. Louis, MO, USA) was used as a coating agent. The 0.5, 1, 3 and 5 g of poly- ϵ -caprolactone each were dissolved in 200 ml acetone and P-bait (100 g) was dispersed in each poly- ϵ -caprolactone concentration as specified above to prepare a 0.5%, 1%, 3% and 5% (w/v %) microencapsulated P-baits, respectively and stirred for 1h. After that, mixture was kept in a laminar flow hood for 12 h to evaporate the acetone and dry for another 24 h. During the solvent removal, P-baits were coated with a thin film of poly- ϵ -caprolactone automatically.

Source of *S. invicta*

Solenopsis invicta polygyne colonies were collected from Hsinchu County (Taiwan), separated from soil by a water drip out method (Chen 2007), and reared under laboratory conditions as described by Kafle *et al.* (2008) at least one week before conducting experimental studies. Laboratory ants were starved two days prior to experiments to ensure that their mid-gut was empty and ants were hungry enough to search for food (Furman & Gold 2006).

Foraging area, nest preparation and bait placement

Foraging area, nest preparation and bait placement was adapted from Kafle *et al.* (2008). A circular artificial foraging area, 40 cm x 17 cm (dia. x height; Fig. 1a), was used for the study. The detailed arrangement of the artificial nest and bait placement was as described in Kafle *et al.* (2008). Baits prepared for evaluation (ca. 1 g) were kept on the plastic weighing plate and placed at the five equidistant locations at the inner wall of the circular foraging area (Fig. 1c).

Determination of water tolerance ability of MC and normal P-bait

To determine the water tolerance ability of MC P-baits and normal P-bait, 1 g of each bait was transferred into 50 ml tube with 15 ml DD-water for 1 h. After 1 h, water was drained for 15 min and weighted again. The weight

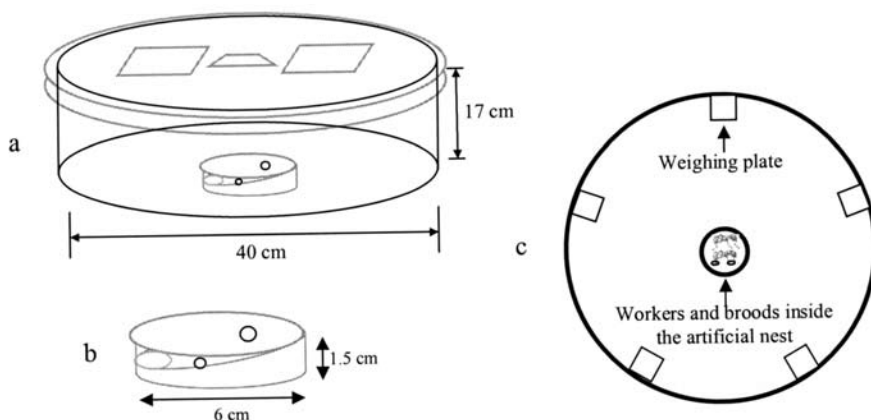


Fig. 1. Artificial foraging area (a), artificial nest (b) and baits placement design (c) (adapted from Kafle *et al.* 2008)

difference before and after water soaked was recorded. As control, 1 g of each bait was transferred into 50 ml tube without DD-water for 1 h. This study was replicated ten times.

Determination of fire ant's preference on wet MC and normal P-bait in the laboratory

To determine the fire ants' preference on water soaked (wet) or normal baits, two-days-starved fire ant workers (600) and brood (120) were kept in the previously described artificial nest and a circular artificial foraging area and 1 g of each bait was offered to the ants. The numbers of fire ants feeding on the baits were recorded every 30 min for 2.5 h (Kafle *et al.* 2008). This study was replicated four times. All experiments in the laboratory were conducted at $27 \pm 1^\circ\text{C}$ and $50 \pm 3\%$ RH with 14L10D condition.

MC P-bait's evaluation on field colonies

The field studies were conducted at the Longtan area of Taoyuan County (Taiwan). A total of eight active fire ant mounds were selected randomly; four colonies were used for normal and another four colonies were used in wet bait preference studies respectively. Baits (ca. 1 g) were kept in plastic weighing plates and placed ca. 1 m away from the center of the fire ant mounds. Numbers of fire ants feeding on the baits were recorded every 30 min for 5 h. Each fire ant mound was considered a replicate. Means were compared using SNK test (SAS 2007). During the study, the field temperature, RH and wind speed was $26.8 \pm 2^\circ\text{C}$, $64.87 \pm 4.14\%$, and 5.97 ± 0.74 km/h, respectively.

RESULTS

Water tolerance ability of MC and non-MC P-baits

A positive correlation between concentration of poly- ϵ -caprolactone and water tolerance ability of P-bait was observed. As concentration of poly- ϵ -caprolactone was increased from 0.5% to 5%, water tolerance ability of P-bait increased by 5% to 20%. When MC and normal P-baits were soaked in water for 1 h, P-bait gained weight significantly more than the microencapsulated baits evaluated ($P = 0.0013$, $F = 32.26$), however weight gained by the 0.5% and 1% or 1% and 3% MC P-baits were not significantly different (Fig. 2).

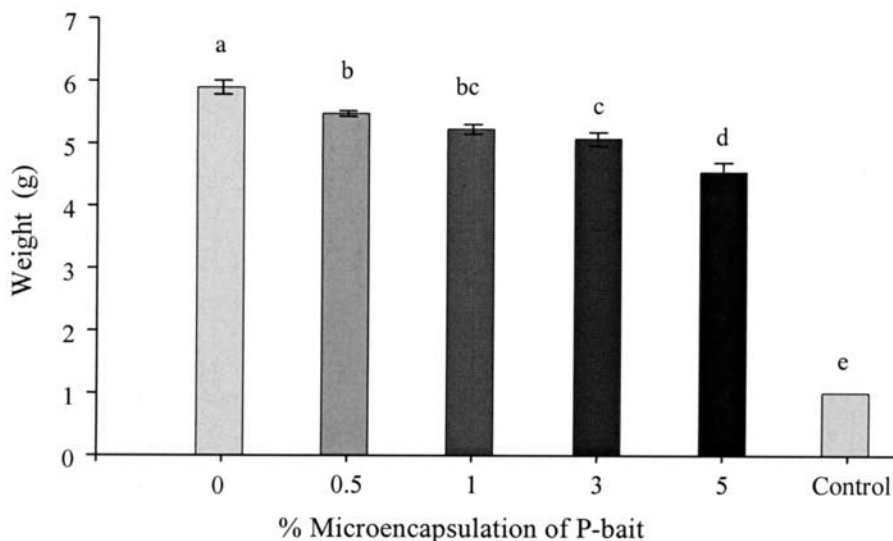


Fig. 2. Microencapsulated and normal P-bait gained weight after soaking in water for 1 h. Columns with different letters are significantly different ($P < 0.05$, SNK test).

Fire ant preference for MC-P-bait in the laboratory

When 0.5%, 1%, 3% and 5% MC and normal P-baits were offered to fire ants, significantly higher numbers of ants were feeding on the normal P-baits than all MC P-baits ($P = 0.0001$, $F = 26.62$). However, numbers of ants feeding on 0.5%, 1% and 3% MC P-baits or 1%, 3% and 5% MC P-baits were not significantly different (Table 1).

Similarly, when 0.5%, 1%, 3% and 5% MC and normal P-bait were soaked in the water for 1 h and offered to fire ants, the number of fire ants feeding on the normal P-baits and all MC P-baits were not significantly different ($P = 0.703$, $F = 0.55$) (Table 1).

When normal and wet P-baits were offered to the ants, a significantly higher number of ants fed on normal P-baits than wet P-baits ($P = 0.0013$, $F = 32.26$). Similarly, when 0.5% MC P-baits were compared to wet MC P-baits, significantly higher number of ants were fed on normal 0.5% MC P-baits than wet P-baits ($P = 0.03$, $F = 7.84$). However, the number of ants feeding on 1% MC P-baits normal vs. wet ($P = 0.3040$, $F = 1.26$), 3% MC P-bait normal vs. wet ($P = 0.3040$, $F = 1.26$), and 5% MC P-bait normal vs. wet ($P = 0.13$, $F = 0.7304$) were not significantly different (Table 1).

Table 1. Fire ants' preference on microencapsulated P-bait in laboratory conditions

Bait	No. of ants (mean \pm SE) ^a	
	Normal bait	Wet bait
P-bait	19.75 \pm 2.87 aA	2.75 \pm 0.85 aB
0.5% MC P-bait	5.75 \pm 0.48 bA	3.50 \pm 0.65 aB
1% MC P-bait	4.25 \pm 0.63 bcA	3.25 \pm 0.623 aA
3% MC P-bait	3.75 \pm 0.75 bcA	2.75 \pm 0.48 aA
5% MC P-bait	2.50 \pm 0.29 cA	2.25 \pm 0.63 aA

^aMeans within the same column (lower case) and same row (upper case) followed by the same letter are not significantly different ($P < 0.05$, SNK test).

MC-P-bait's evaluation on field colonies

When 0.5%, 1%, 3% and 5% MC and normal P-baits were offered to field colonies, a significantly higher number of ants fed on normal P-baits than any MC P-baits formulation ($P = 0.0001$, $F = 106.60$). However, number of ants feeding on the 0.5%, 1%, 3% or the 5% MC P-baits were not significantly different (Table 2).

Table 2. Fire ants' preference on microencapsulated P-bait in field conditions

Bait	No. of ants (mean \pm SE) ^a	
	Normal bait	Wet bait
P-bait	22.50 \pm 2.02 aA	0.75 \pm 0.48 aB
0.5% MC P-bait	0.50 \pm 0.29 bA	0.75 \pm 0.48 aA
1% MC P-bait	0.75 \pm 0.48 bA	0 aA
3% MC P-bait	0.25 \pm 0.25 bA	0 aA
5% MC P-bait	0.50 \pm 0.29 bA	0 aA

^aMeans within the same column (lower case) and same row (upper case) followed by the same letter are not significantly different ($P < 0.05$, SNK test).

When 0.5%, 1%, 3% and 5% MC and normal P-baits were soaked in the water for 1 h and offered to field colonies, the number of ants feeding on the normal P-baits and all MC P-baits were not significantly different ($P = 0.1735$, $F = 1.84$) (Table 2).

When normal and wet P-baits were offered to the field colonies, a significantly higher number of ants fed on normal P-baits than wet P-baits ($P = 0.0001$, $F = 109.70$). Furthermore, the number of ants feeding on 0.5% MC P-baits, normal and wet ($P = 0.20$, $F = 0.6704$), 1% MC normal and wet ($P = 0.1682$, $F = 2.45$), 3% MC normal and wet ($P = 0.3559$, $F = 1.00$) and 5% MC normal and wet ($P = 1340$, $F = 3.00$) were not significantly different (Table 2).

DISCUSSION

P-baits, after microencapsulation, could tolerate water better than normal P-baits. That was due to a micro layer around the outer surface of P-baits created during the microencapsulation process. A positive correlation between concentration of coating material and water tolerance ability of P-bait was observed. As the concentration of the coating agent (poly- ϵ -caprolactone) was increased from 0.5% to 5%, water tolerance ability of P-bait increased by 5% to 20%. However, the microencapsulated P-baits lost their fire ant's phagostimulant activity (Tables 1, 2). During the microencapsulation process, P-baits were exposed to air for 36 h. Flanders & Graham (2007) reported that if oil used in P-bait as a phagostimulant came in contact to air for a prolonged period, the oil becomes rancid and baits could not attract fire ants. This may explain why microencapsulated P-bait did not score more ants. Most fire ant bait products lose its ant phagostimulant activity once they encounter water (Nester 2007), which was observed during this study too. Both microencapsulated and normal P-bait did not stimulate more ants to feed under laboratory or field conditions, when those baits were soaked in water. Furthermore, the microencapsulation creates an outer layer that blocks not only water penetration but also blocks the release of fire ant phagostimulants from the baits. Therefore, microencapsulation of P-bait could create a barrier in the chemical communication between P-baits and ants. Furthermore, acetone probably dissolves the vegetable oil and toxicant, and then as the solvent evaporates, everything is redeposited, but not evenly throughout

the bait particles. It probably changes the amount of phagostimulant available for worker ant detection. Other authors (Shukla & Sivaram 1999; Tsuji 2001) also reported that the microencapsulation slows down the release of active ingredients. Based upon those experiences, we realized that changing the coating material or techniques for microencapsulation of P-baits might not be worthy for further work towards the development of a water or high humidity resistant fire ant bait. We think this because all coating materials or microencapsulation techniques finally create a micro layer around the P-bait that ultimately blocks the chemical communication channels between ants and baits. Although microencapsulated P-baits could resist water, fire ants were not stimulated to feed. Therefore, using the microencapsulation technique presented here and other similar methods with P-bait may not be a suitable alternative towards the development of a high humidity or water resistant fire ant bait. However, further investigation of different techniques to encapsulate water insoluble bait components is necessary, since there is a powerful need for a water resistant fire ant bait system. Application of an encapsulation system using a polymer that contains a true attractant - pheromone could be a better alternative to improve phagostimulant activity of MC P-bait. However there are no reports yet about such coating materials. In conclusion, based on the fire ants' preference and its water resistant properties of P-baits observed in this study, poly- ϵ -caprolactone treated fire ant baits did impart greater water resistance, but also inhibited worker feeding on the resulting bait formulations, thus future research will focus on different polymer formation methods.

ACKNOWLEDGMENTS

We would like to express our special thanks to the Laboratory of Extension Entomology and Science Education, Dept. of Entomology, National Taiwan University, Taiwan for facilities and financial supports. We also would like to thank Mr. Bao, Shi-Zhe for his cooperation during this study.

REFERENCES

- Chen, J. 2007. Advancement on techniques for the separation and maintenance of the red imported fire ant colonies. *Insect Sci.* 14: 1-4.

- Chen, J.S.C., C.H. Shenand, & H.J. Lee. 2006. Monogynous and polygynous red imported fire ants, *Solenopsis invicta* Buren (Hymenoptera: Formicidae), in Taiwan. *Environ. Entomol.* 35: 167-172.
- Flanders, K.L., & L. Graham. 2007. Getting the most out of your fire ant bait. Alabama Cooperative Extension System, ANR-1161.
- Furman, B.D., & R.E. Gold. 2006. Determination of most effective chemical form and concentration of indoxacarb, as well as the most appropriate grit size, for use in Advion. *Sociobiology* 48: 309-334.
- Henshaw, M.T., N. Kunzmann, C. Vanderwoude, M. Sanetraand, & R.H. Crozier. 2005. Population genetics and history of the introduced fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae), in Australia. *Aust. J. Entomol.* 44: 37-44.
- Kaffe, L., W.J. Wu, R.K. Vander Meer, & C.J. Shih. 2008. Simplified approaches to determine the attractant preference of *Solenopsis invicta* (Hymenoptera: Formicidae). *Appl. Entomol. Zool.* 43: 383-390.
- Morrison, L.W., S.D. Porter, E. Daniels, & M.D. Korzukhin. 2004. Potential global range expansion of the invasive fire ant, *Solenopsis invicta*. *Biol. Invas.* 6: 183-191.
- Nester, P.R. 2007. Red imported fire ant control around bodies of water. Texas Cooperative Extension, Fire Ant Plan Fact Sheet no. 021.
- Sánchez-Peña, S.R., R.J.W. Patrock, & L.A. Gilbert. 2005. The red imported fire ant is now in Mexico: documentation of its wide distribution along the Texas-Mexico border. *Entomol. News* 116: 363-366.
- SAS Institute. 2007. SAS user's guide: Statistics, version 8 ed. SAS Institute, Inc., Cary, NC.
- Schubert, U.S. & G. Hochwimmer. 2001. Biodegradable polymers with specific metal binding sites based on bipyridine-containing poly(lactic acid) and poly(ϵ -caprolactone): towards high-molecular weight polyesters. *Macromol. Rapid Commun.* 22: 274-280.
- Shukla, P.G., & S. Sivaram. 1999. Microencapsulation of the water-soluble pesticide monocrotophos by an oil in oil interfacial polyaddition method. *J. Microencapsulation* 16: 517-521.
- Tsuji, K. 2001. Microencapsulation of pesticides and their improved handling safety. *J. Microencapsulation* 18: 137-147.
- Vogt, J.T., W.A. Smith, R.A. Grantham, & R.E. Wright. 2003. Effects of temperature and season on foraging activity of red imported fire ants (Hymenoptera: Formicidae) in Oklahoma. *Environ. Entomol.* 32: 447-451.
- Williams, D.F., H.L. Collins, & D.H. Oi. 2001. The red imported fire ant (Hymenoptera: Formicidae): An historical perspective of treatment programs and the development of chemical baits for control. *Am. Entomol.* 47: 146-159.



