Livestock manure as an alternative attractant for fruit flies (Diptera: Tephritidae) in guava tree¹

Rosenya Michely Cintra Filgueiras², Francisco Roberto de Azevedo³, Raul Azevedo³, Ricardo Braga de Farias³, Cristiane Ramos Coutinho²

ABSTRACT

Fruit flies are typically managed using hydrolyzed protein, which is difficult for family farmers to obtain. This study aimed at assessing the efficiency of livestock manure for monitoring and/or controlling this pest in guava tree orchards. The first experiment tested the efficiency of guava juice and manure from cattle, sheep, pig, horse and chicken as attractants for fruit flies. Once the best bait had been established, a second experiment was conducted using guava juice and chicken manure extract at concentrations of 10 %, 30 %, 50 %, 70 % and 100 %. A third assay analyzed guava juice and chicken manure extract (10 %) at three attractant aging periods (3, 7 and 14 days after trap installation). The cost-effectiveness ratio between guava juice and extract was also analyzed. It was concluded that fruit flies prefer the chicken manure extract (10 %), with greater capture observed three days after trap installation, which can replace the guava juice in the agroecological management of fruit flies in guava trees in family farms, since it is low cost and efficient.

KEY-WORDS: *Psidium guajava* L.; fruit flies; natural pest control.

RESUMO

Esterco de animais domésticos como atrativo alternativo para moscas-das-frutas (Diptera: Tephritidae) em goiabeira

As moscas-das-frutas são manejadas com proteína hidrolisada, mas, para agricultores familiares, esse produto é de difícil acesso. Objetivou-se avaliar a eficiência de estercos de animais domésticos no controle e/ou monitoramento dessa praga em pomares de goiaba. No primeiro ensaio, avaliou-se a eficiência do suco de goiaba e de estercos bovino, ovino, suíno, equino e de galinha, como atrativos para a mosca-das-frutas. Após definição do melhor atrativo, instalou-se o segundo ensaio, utilizando-se suco de goiaba e extrato de esterco de galinha nas concentrações de 10 %, 30 %, 50 %, 70 % e 100 %. No terceiro ensaio, avaliou-se suco de goiaba e esterco de galinha a 10 %, em três períodos de envelhecimento do atrativo (3, 7 e 14 dias após a instalação das armadilhas). A relação custo/beneficio do suco de goiaba e do extrato também foi avaliada. Verificou-se que as moscas-das-frutas têm preferência pelo extrato de esterco de galinha a 10 %, com maior captura observada aos três dias após a instalação, o qual pode substituir o suco de goiaba no manejo agroecológico de moscas-das-frutas em goiabeiras, na agricultura familiar, pois é de baixo custo e eficaz.

PALAVRAS-CHAVE: *Psidium guajava* L.; moscas frugívoras; controle natural de pragas.

INTRODUCTION

Brazil is the third largest fruit producer in the world, with an annual average of 40 million metric tons (Anuário... 2010). According to Veloso et al. (2012), farmers who obtain high quality fruits, free of pests, diseases and physiological disorders, can find new markets.

Guava (*Psidium guajava* L.) is one of the fruits most affected by fruit flies in Brazil. *Ceratitis capitata* Wiedemann and *Anastrepha* spp. (Diptera: Tephritidae) are a significant problem for fruit

farming, not only due to the direct damage they cause in the field, especially in certain regions, but also because of the quarantine barriers imposed by countries that import fresh fruits (Lima et al. 2012).

Fruit fly populations are managed using toxic baits and insecticide sprays, primarily organophosphates and pyrethroids, to control eggs/larvae inside fruits and adult flies outside them (Carvalho 2004). Although this technique is considered efficient for controlling insect pests, insecticides, particularly organophosphates, are highly toxic, negatively affecting both natural enemies

E-mails: roberto.azevedo@ufca.edu.br, raulbiologo@gmail.com, ribrafa@live.com.

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Universidade Federal do Ceará, Departamento de Fitossanidade, Fortaleza, CE, Brazil.

E-mails: rosenya michelycf@hotmail.com, cris.ramos@yahoo.com.br.

^{3.} Universidade Federal do Ceará, Laboratório de Entomologia Agrícola, Crato, CE, Brazil.

and pollinator populations. These insecticides can remain in the environment for long periods of time, causing health and ecological concerns (Nondillo et al. 2007).

Integrated pest management programs in fruit farming have encouraged the use of different methods to control fruit flies. Among them is the use of attractants, aiming at reducing pest population density (Alves 2010).

Given the quarantine-based significance of fruit flies in commercial orchards, the trapping devices and baits used must be effective and capable of detecting the presence of these tephritids. As such, effective monitoring depends on the quality of the attractant (food or sexual) and its distribution in strategic points throughout the orchard. Effective capture of adults is important for monitoring populations and planning proper orchard management (Carvalho 2005).

Monitoring fruit flies is vital and generally achieved using a McPhail trap baited with hydrolyzed protein (Monteiro et al. 2007). However, for family farmers, this technology is difficult to obtain, due to its high cost and low availability. As such, alternative and less expensive attractants are being sought for monitoring systems, including fruit juices, urea, molasses, urine and chicken manure. Ammonia is also used to monitor fruit fly populations. Kendra et al. (2005) found that ammonia was efficient in capturing fruit flies, being especially attractive to females.

The present study aimed at assessing the efficiency of livestock manure, in comparison with guava juice, as an alternative for controlling and/or monitoring fruit flies (*Anastrepha* spp. and *Ceratitis capitata*).

MATERIAL AND METHODS

Studies were conducted in a family farm with a ten years old Paluma guava (*Psidium guajava* L.) orchard, in Barbalha (7°17'18"S, 39°20'57"W and altitude of 459.2 m), Ceará State, Brazil, from august 8th to October 31st, 2013.

Guava trees were planted using 6 m x 5 m spacing, totaling an effective area of 0.5 ha. Standard cultivation practices were adopted, including fertilization, weeding and irrigation. However, chemical insecticides were not applied to control pests. Surrounding the experimental area were the following orchards: papaya (*Carica papaya L.*), passion fruit (*Passiflora edulis Sims f. flavicarpa*), individual red

mombin trees (*Spondias purpurea* L.), ambarella (*Spondias dulcis* L.) and mangoes (*Mangifera indica* L.). The study was conducted during the fruiting and ripening stages.

The first experiment was performed from August 8th to 15th, using a plastic McPhail trap with a yellow base as the experimental unit, distributed in a randomized blocks design, with six treatments and five replications, totaling 30 experimental units. The traps were installed in the shade, in alternate rows, approximately 10 m apart and at an average height of 2.0 m, in trees bearing ripe fruits ready for harvest.

The treatments used were manure from cattle, sheep, pig, horse and laying hen. The control treatment contained 25 % of natural guava juice + 10 % of granulated sugar. Each type of manure was collected in a 20 L plastic bucket one day prior to its use and diluted in distilled water at a ratio of 3:1, to obtain a smooth liquid extract. Next, the liquid was homogenized, stored in 2 L transparent PET bottles and taken to the field. Approximately 400 mL of these extracts were placed into each trap.

The baited traps remained in the field for seven days. Insects were then collected and counted using a fine-mesh plastic sieve and a fine brush. Captured insects were placed in 100 mL plastic containers filled with 70 % of ethanol and labeled based on the respective treatments. Next, the material was taken to the Agricultural Entomology Laboratory of the Universidade Federal do Ceará, in Crato (Ceará State, Brazil), for screening and counting. The traps were removed from the orchard once the material had been collected. The aim of this first phase was to assess which manure achieved the best result in terms of attracting *Anastrepha* spp. and *C. capitata* adults.

The second experiment was carried out from September 4th to 11th, using a plastic McPhail trap with a yellow base as the experimental unit. Traps were distributed in a randomized blocks design, with six treatments and five replications, totaling 30 experimental units. The same methodology applied to the previous phase was used for treatments and fly collection. Treatments were 25 % of natural guava juice + 10 % of granulated sugar as control and laying hen manure at concentrations of 10 %, 30 %, 50 %, 70 % and 100 %.

The third experiment (September 16th to October 31st) evaluated two treatments (guava juice and 10% chicken manure), under three aging periods for bait (3, 7 and 14 days), with 10 replications.

Twenty experimental units were evaluated in each assessment, using the same treatment application and collection methods as the previous phase.

The number of adult fruit flies captured over seven days in McPhail traps baited with natural guava juice and livestock manure diluted (3:1) in distilled water and also the average number of fruit flies captured every seven days in McPhail traps baited with different concentrations of laving hen manure were submitted to statistical normality, using a Shapiro-Wilk's test. Non-normal data were converted into $\sqrt{(x+0.5)}$ and submitted to analysis of variance, and means were compared by the Tukey's test, at 5 %. All tests were carried out using the PAST 3.0 software. Data on the average number of collected fruit flies sampled by McPhail traps baited with three aging periods with 10 % of chicken manure extract and 25 % of guava juice plus 10 % of sugar were submitted to regression analysis, using the Excel software (Microsoft, Washington, USA).

Cost-effectiveness of the best treatments, expressed in monetary terms, was evaluated. The price of guava pulp and its application in the control of fruit flies was analyzed and contrasted with 10 % of laying hen manure extracted from an orchard owned by a family farmer in Cariri (Ceará State, Brazil).

RESULTS AND DISCUSSION

The laying hen manure extract resulted in the highest number of captured flies, significantly differing from the other extracts used. However, the number of captured flies was significantly different and twice as high for the guava juice control, when compared to chicken manure (Table 1).

Prokopy et al. (1992) compared chicken manure to commercial attractants in laboratory and found that the manure attracted significantly more *C. capitata*. However, similarly to our results, chicken manure was significantly less attractive than guava juice.

Epsky et al. (1997) compared a crude chicken manure extract with a methanolic chicken manure extract in laboratory and observed that chicken droppings captured more *A. suspensa* Loew (Diptera: Tephritidae) than the methanol extract. The authors attributed this difference to a possible decline in the microbial activity of methanol, thereby reducing the release of ammonia by the extract. Poultry manure attracts fruit flies because of the release of volatile compounds, including ammonia, by bacteria fermentation (Robacker et al. 2000). Protein baits, agricultural supplements such as synthetic fertilizers, animal waste and any decomposing material produce and release ammonia during putrefication. The course and speed of decomposition is the result of the interaction between biotic (microbial activity) and abiotic factors (temperature, precipitation and wind) (Sommer & Hutchings 2001).

Rull & Prokopy (2000) tested chicken manure and odoriferous ethyl hexanoate bait for capturing *Rhagoletis pomonella* Walsh (Diptera: Tephritidae). They found that chicken manure did not affect the capture of males, but considerably increased the capture rate of juvenile females. These results suggest that chicken manure can be used as an alternative method to control fruit flies, increasing the capture rate of females.

Piñero et al. (2003) compared the efficiency of chicken manure, human urine and hydrolyzed protein in the capture of fruit flies in a mango (Mangifera indica L.) crop. They observed more Anastrepha obliqua Macquart, A. serpentina Wiedemann, A. alveata Stone, A. chiclayae Greene, A. ludens Loew and A. zuelaniae Stone (Diptera: Tephritidae) individuals in the traps containing manure. However, this treatment was less effective in attracting A. obliqua and A. serpentina. One of the hypotheses raised by the authors is that the difference in pH among manure, protein and urine explains the variation in capture numbers. Robacker et al. (2000) compared different types of duck manure and found

Table 1. Average number of adult fruit flies ± SE (*Anastrepha* spp. + *Ceratitis capitata*) captured over seven days in McPhail traps baited with 25 % of guava juice + 10 % of granulated sugar and livestock manure diluted (3:1) in distilled water.

Treatment	Average number of flies ¹	
Guava juice	7.5 ± 14.2 a	
Chicken manure	$3.6 \pm 2.1 \text{ b}$	
Horse manure	2.3 ± 3.8 c	
Cattle manure	$1.6 \pm 2.2 \text{ c}$	
Pig manure	$1.6 \pm 0.2 \text{ c}$	
Sheep manure	$1.1 \pm 0.1c$	
CV (%)	42.19	

¹Means followed by the same letter do not differ significantly according to the Tukey's test at 5 %. Data were transformed with $\sqrt{(x + 0.5)}$.

that more alkaline pH tended to attract more *A. ludens* individuals.

By testing five different concentratios of hen manure, we observed that the dilution at 10 % captured more adult flies, with no significant difference from the control (Figure 1). Since chicken manure is quite viscous, only the 10 % concentration formed a liquid mixture (extract) capable of attracting flies. Capture rates declined as the concentration increased, due to the fast water evaporation and amount of manure in the traps. Aluja & Piñero (2004) tested liquid baits containing different concentrations of ammonia and found that high levels tended to be less attractive. The release of ammonina is generally regulated by pH.

In behaviorial assays with *A. suspense* under laboratory conditions, Kendra et al. (2005) observed that ammonia is released primarily during the decomposition of certain food proteins, naturally attracting flies. The authors also found that ammonia became less appealing at higher concentrations, potentially repelling flies.

Aluja & Piñero (2004) compared different concentrations of human urine and hydrolyzed corn (*Zea mays L.*) protein in guava trees and found no difference in the number of individuals captured for *A. fraterculus* Wiedemann (Diptera: Tephritidae), *A. ludens* and *A. striata Schiner* (Diptera: Tephritidae). The authors suggested that these species responded well to the nitrogen stimulus present in the urine, as occurs with laying hen manure. In this study, higher concentrations of manure extracts dried more easily due to the

45 $41.2 \pm 7.7 \text{ a}$ Mean number of flies captured 40 35 30 25 $19.6 \pm 5.9 \,\mathrm{h}$ 20 15 10 $3.0 \pm 0.7 c$ 5 1.8 ± 0.4 c 2.2 ± 0.6 c $1.0 \pm 0 c$ 0 Juice 10% 30% 50% 70% 100% Guava juice and concentrations of chicken manure

high temperatures in the semi-arid Cariri region, hampering the release of ammonia, which in turn reduced the attraction of adult fruit flies.

Guava juice attracted significantly more adult flies than chicken manure at the third and seventh days after bait installation. For both guava juice and chicken manure, bait effectiveness declined linearly. Two weeks after installation, both baits (guava juice and chicken manure) were ineffective and therefore not significantly different (Figure 2).

Contrasting the results exhibited in Figures 1 and 2, the decreasing trend of guava juice and chicken manure attractiveness with time could be explained by changes in physical and chemical composition, associated with water loss and ammonia volatilization (Sommer & Hutchings 2001, Aluja & Piñero 2004). This decline occurred because the attractants dried out after being exposed to temperatures reaching an average of 30 °C. Water loss concentrates the ammonia, and high concentrations of ammonia repels flies (Kendra et al. 2005).

Nascimento et al. (2000) recommended that traps should be inspected up to seven days after installment. After this point, the attractant solution becomes less effective at attracting flies, therefore capturing fewer insects. This occurred in the present study, because capture rates fell as the exposed attractants (juice and manure) aged (Figure 2).

According to Azevedo et al. (2012), guava juice is easy to obtain, inexpensive and can be used as a replacement for hydrolyzed corn protein to capture fruit flies. This hydrolyzed protein is used by most farmers to monitor and/or control these pest in



Figure 1. Average number of fruit flies \pm SE (*Anastrepha* spp. + *Ceratitis capitata*) captured over seven days in McPhail traps baited with guava juice or laying hen manure extract at different concentrations. Means followed by the same letter do not differ significantly according to the Tukey's test at 5 %.

Figure 2. Linear regression of the average number of adult fruit flies (*Anastrepha* spp. + *Ceratitis capitata*) captured over seven days in McPhail traps baited with 10 % of laying hen manure and 25 % of guava juice + 10 % of granulated sugar submitted to three aging periods.

Product	Unit value	Amount L ⁻¹	Volume of attractant 0.5 ha ⁻¹	Cost of application per week
	US\$		L	US\$
Hydrolyzed protein	6.78 (500 mL plastic bucket)	50 mL	12	8.14
Guava juice	0.27 (100 g)	200 g	12	6.48
Chicken manure	0.03 (100 g)	200 g	12	0.72

Table 2. Amount and application costs for 0.5 ha of a guava orchard using guava juice.

commercial fruit orchards. However, the high cost makes it difficult for family farmers to afford and find it, since only two companies in São Paulo sell the product. Currently, 500 mL of the hydrolyzed protein costs US\$ 6.78, while chicken manure costs only US\$ 0.72 for each 12 L (Table 2).

Laying hen manure was the most efficient attractant for capturing fruit flies, when compared to manure from other animals. The Cariri region, as well as many other regions in Brazil, is home to a high number of poultry farms, distributed across several municipalities, meaning that small-scale farmers have a variety of options to obtain this manure source to attract fruit flies in their guava orchards. In the guava orchard under study, 30 traps were baited with 400 mL of attractant. With this baiting system, the weekly cost of using guava juice was US\$ 6.48 (Table 2). Considering a period of four weeks, farmers would spend almost US\$ 26.00 per month, against only US\$ 2.88 per month, using the chicken manure extract.

Since guava juice captured more fruit flies than laying hen manure, we believe that the same efficiency can be achieved for the latter, by increasing the number of traps in the area. This would be more beneficial, from an economic perspective, because of the chicken manure low cost. Therefore, this manure constitutes a good option for family farmers to monitor and control fruit flies populations.

CONCLUSIONS

- 1. Adult fruit flies show a preference for the 10 % laying hen manure extract.
- 2. Most fruit flies were captured three days after the traps were installed, with numbers declining significantly thereafter.
- 3. Chicken manure extract can be used in guava orchards by small farmers to replace guava juice, because it is cost-effective.

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