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Effectiveness of an Artisanal McPhail Trap for Mass Trapping Tephritid Fruit Flies in Haiti

Fractyl Mertilus

Louisiana State University and Agricultural and Mechanical College, fractyl007@yahoo.fr

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EFFECTIVENESS OF AN ARTISANAL MCPHAIL TRAP FOR MASS TRAPPING
TEPHRITID FRUIT FLIES IN HAITI

A Thesis

Submitted to the Graduate Faculty of the
Louisiana State University and
Agricultural and Mechanical College
in partial fulfillment of the
requirements for the degree of
Master of Science

in

The Department of Entomology

by
Fractyl Mertilus
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TABLE OF CONTENTS

ACKNOWLEDGEMENTS.....	ii
LIST OF TABLES.....	v
LIST OF FIGURES.....	vi
ABSTRACT.....	vii
CHAPTER 1. INTRODUCTION AND REVIEW OF LITERATURE.....	1
1.1 Introduction.....	1
1.2 Literature review.....	3
1.2.1 Indian fruit fly, Caribbean fruit fly, and their host plants.....	3
1.2.2 Anastrepha traps and lures.....	5
1.2.2.1 McPhail trap (MP).....	6
1.2.2.2 Multilure trap (M).....	7
1.2.3 Layout of trapping network.....	7
CHAPTER II. DEVELOPMENT, EVALUATION, AND COMPARISON OF THE ARTISANAL MCPHAIL TRAP WITH THE COMMERCIAL MCPHAIL TRAP.....	9
2.1 Introduction.....	9
2.2 Objective and Hypotheses.....	10
2.3 Materials and Methods.....	10
2.3.1 Study Site.....	10
2.3.2 Trap models description and lures.....	11
2.3.3 Experimental Design.....	12
2.3.4 Data collection and analysis.....	12
2.4 Results and discussion.....	13
2.4.1 Trap Model Effectiveness under Field Conditions, and Mass Trapping.....	13
CHAPTER III. DEVELOPMENT, EVALUATION, AND COMPARISON OF THE ARTISANAL MCPHAIL TRAP WITH THE COMMERCIAL MCPHAIL TRAP.....	18
3.1 Introduction.....	18
3.2 Objective and Hypotheses.....	18
3.3 Materials and Methods.....	19
3.3.1 Study Site.....	19
3.3.2. Experimental design.....	19
2.3.4 Data collection and analysis.....	19
3.4 Results and discussions.....	20
3.4.1 Performance of the clear artisanal trap model in mass trapping conditions.....	20
CHAPTER IV. DETERMINATION OF AN OPTIMAL DENSITY FOR THE ²³ ARTISANAL TRAP (AT2) UNDER MASS TRAPPING CONDITIONS.....	23
4.1 Introduction.....	23
4.2 Objective and Hypotheses.....	24
4.3 Materials and Methods.....	24

4.3.1 Study Site.....	24
4.3.2 Experimental design	25
4.3.3 Data collection and analysis	25
4.4 Results and Discussion.....	26
CHAPTER V. SUMMARY AND CONCLUSION	32
5.1 Summary	32
5.2 Conclusion.....	34
REFERENCES	35
APPENDIX A: TRAP MODELS EVALUATION AND COMPARISON, STATISTICAL ANALYSIS (<i>ANOVA</i>) (EXPERIMENT #1).....	39
APPENDIX B: BINOMIAL TEST FOR PROPORTIONS	422
APPENDIX C: BINOMIAL TEST FOR EQUAL PROPORTIONS.....	466
APPENDIX D: NONLINEAR REGRESSION.....	477
VITA.....	49

LIST OF TABLES

Table 2.1: Proportion of Fruit Fly Species Caught per Trap Model, and Comparison of the Cost-Effectiveness of the Trap Models.....	17
Table 4.1: Regression Analysis: Nonlinear Model Test (PROC NLIN, SAS (English 9.4), SAS/STAT 2006).....	27
Table 4.2: Regression Analysis: Parameter Estimates (PROC NLIN, SAS (English 9.4)).....	27
Table 4.3: Change in Capture Rates of Flies Compared to Increase in the Trapping Density.....	30

LIST OF FIGURES

Fig. 2.1: Trap models developed and used for the study: AT1, yellow bottom artisanal trap; AT2, clear artisanal trap; and MP, Commercial McPhail trap.....	11
Fig. 2.2: Mean (\pm SE) number of fruit flies caught per trap model. Bars labeled with identical letters were not significantly different after comparisons among trap models (<i>ANOVA</i> , Fisher's LSD $\alpha=0.05$).....	13
Fig. 2.3: Number of fruit flies caught per trap model. Bars labeled with identical letters were not significantly different after comparisons among trap models (Binomial test for equal proportion, Bonferroni's correction $\alpha=0.017$).....	14
Fig. 2.4: Average proportion of female and male fruit flies caught per trap model. Bars labeled with identical letters were not significantly different after comparisons among trap models(<i>ANOVA</i> , Fisher's LSD $\alpha =0.05$).....	15
Fig. 3.1: Total number of fruit flies caught in 24MP/ha and 36AT2/ha. Bars labeled with the same letters were not significantly different (Binomial test for equal proportion, $\alpha=0.05$).....	20
Fig. 3.2: Evolution of the number of flies caught in both trap densities throughout this experiment.....	21
Fig. 4.1: Squatter plot of the dependent variable (Y) with the independent variable (X): Y * X. The letters A are observations (number of fruit flies caught) for each level of trap density.....	26
Fig. 4.2: Curve Showing Predictive Relationship between Number of Fruit Flies Caught (Y: dependent variable) and Trap density (X: independent variable).....	30
Fig. 4.3: Fruit Flies Population Density in the Experimentation Site (Mango Orchard), during 2014. Ministry of Agriculture, Natural Resources, and Rural Development (MARNDR/DPV-PS) (National Program for Detection and Control of Fruit Flies). Unpublished Data.....	31

ABSTRACT

Three field experiments were conducted in a mango orchard in Croix-des-bouquets (Haïti) to develop an effective artisanal McPhail trap, less expensive than the commercial traps, for mass trapping *Anastrepha obliqua* (Macquart) and *Anastrepha suspensa* (Loew). A field trial conducted to evaluate the effectiveness of two newly made artisanal trap models (AT1 and AT2) with the commercial McPhail trap (MP) demonstrated that the artisanal traps yielded similar results in the average number of fruit flies caught (8.9 ± 2.6 , 13 ± 2.9 , and 16 ± 4.1 respectively). Moreover, the cost-efficacy ratio was a lot higher in the artisanal trap models (AT1: 0.42 \$ per flies caught, AT2: 0.28 \$ per flies, and MP: 0.69 \$ per flies), even if the total number of fruit flies was higher in the commercial trap (319 flies) compared to the others (AT1: 178 flies and AT2: 253 flies). Another field trial conducted in the same mango orchard compared a density of 24 McPhail traps per ha to 36 traps/ha using the most cost-effective artisanal trap, and revealed that they were not different in number of fruit flies caught (AT2: 236 flies and MP: 239 flies). In addition, the capture rate of *Anastrepha spp.* in both trap densities had a similar increasing trend line throughout the mango fruiting season. To determine an optimal trap density for the artisanal trap (AT2) under mass trapping conditions, a field experiment assessed six different trap densities (4, 8, 12, 16, 24, and 36 traps/ha), and suggested that a density of 25 traps/ha could protect the mango orchard from the growing phase to the maturation phase of mango fruits. However, analysis of fruit fly data available throughout the year suggested that trapping density should be increased during the ripening phase, when the *Anastrepha spp.* density reach their peak in this orchard. These findings indicated that cost-effective artisanal trap models can be developed to substitute the expensive commercial traps for implementation of fruit fly control programs with mass trapping methods.

CHAPTER 1. INTRODUCTION AND REVIEW OF LITERATURE

1.1 Introduction

Tephritid fruit flies represent one of the most economically important insects in the Tropical and Sub-Tropical regions. Besides their great impact on the international marketing of fresh fruits and vegetables, infestations of these insects have resulted in the implementation of area-wide or national control programs in order to comply with Sanitary and Phytosanitary Standard (SPS) measures (IAEA 2003, Aluja 1994). In 2007, losses caused by tephritids were estimated at over 4 million USD in Haiti, which represented 40% of the price of mango exports (Pierreval 2012). Consequently, six processing plants went out of business due to the increased costs related to the new processing and export standards (Weiner 2009). Since 2008, a nation-wide program has been implemented to detect and control fruit flies, and protect Haiti's mango as the first export crop (MARNDR 2009).

Haiti is among the world's twenty largest mango producers (FAO 2012), and the sixth largest mango exporter to the US market after Mexico, Peru, Ecuador, Brazil, and Guatemala, (Ward 2011). Moreover, the Central Bank of Haiti estimates Haiti's mango export at about \$10 million USD (BRH 2012, Pierreval 2012). Besides its economic importance, mango is an important source of Vitamin A, and mango trees constitute a major part of the vegetation cover in some areas of Haiti. Moreover, Haitian mango is well appreciated for its qualities, and for being organically grown. Even though smallholder associations do not have the financial and technical capability to obtain and renew certification for all of their mango fields, during the 2009 export season 2% of the total production was certified as "organic" (USAID 2010). The National Mango Forum organized by two USAID-funded programs (USAID-MarChE and USAID-WINNER) on April 20 and 21st 2010 in Port-au-Prince set a goal to help Haiti increase

its export from 2.5 to 5 million cases of USDA-certified mangoes by 2015 (USAID 2010). As a result, maintaining mango production areas pesticide-free throughout the country is a crucial asset for the mango industry.

Another weakness of the mango industry in Haiti is the lack of commercial orchards. In fact, Haitian mango production derives from individual mango trees dispersed throughout smallholders' farms. Given their number and the pricing of mango during the harvest season, these mango trees do not represent an important and permanent source of income to smallholders (Castañeda et al. 2011). Some farmers who have small or average orchards might be able to manage their own plantation by applying sanitation and fruit fly control methods. However, smallholders, on which mango production relies in Haiti, have not shown interest in investing their money in such activities. This situation makes it difficult for the Division of Plant Protection (DPV/PS) to technically and financially manage the National Program for Detection and Control of Fruit Flies (PNDCMF).

According to reports from the Division of Plant Protection of the Ministry of Agriculture (DPV/PS), the detection phase of that project revealed that the two tephritid fruit fly species were the Caribbean fruit fly, *Anastrepha suspensa*, Loew (Diptera: Tephritidae) and the Indian fruit fly, *Anastrepha obliqua*, Macquart (Diptera: Tephritidae) (MARNDR 2009). Moreover, the trapping network established to survey fruit fly densities in Haiti has yielded significant results, and contributed to a significant reduction of *Anastrepha spp.* density through time (MARNDR 2013). These data demonstrated that the environment is appropriate for mass trapping as a management method (Kogan and Jepson 2007). However, the density of McPhail traps recommended for mass trapping methods to control fruit flies (25-50 traps/ha) (Martinez-Ferrer et al. 2012) results in a financial cost that is too high for the fruit fly Haitian control program

(Malo and Zapien 1994, Burrack et al. 2008; Lasa et al. 2013). Thus, in order to be sustainable, and to reduce food safety risks, new trapping methods and trap devices should be designed to control fruit flies in a most cost-effective way.

This research had the following objectives: (1) to develop an artisanal McPhail trap less expensive than the commercial McPhail trap; (2) to evaluate and compare the effectiveness of the artisanal trap to the commercial McPhail for mass trapping in Haiti; (3) to determine an optimal density for the artisanal trap under mass trapping conditions.

1.2 Literature review

1.2.1 Indian fruit fly, Caribbean fruit fly, and their host plants

The Indian fruit fly and the Caribbean fruit fly are considered to be economically important in tropical and subtropical countries such as Mexico, Brazil, Costa Rica, and Haiti, where control programs have been established to limit their impact (Aluja et al. 1987, White and Elson-Harris 1992, AIEA 2003). These *Anastrepha spp.* are known to be polyphagous, with a wide host range. The adults require a balanced amount of carbohydrates, water, minerals, and protein to survive, develop and reproduce. The mated females oviposit in the pulp of mature or ripened fruit of suitable hosts. After completing three larval instars inside the fruit, the larvae leave the fruit to pupate in the soil, and finally emerge as adults. Optimal conditions of temperature, light, and moisture are critical for the completion of their life cycle (Christenson and Foote 1960, Bateman 1972, Aluja et al. 1994, Aluja and Piedra 2000). According to the quarantine rules in every fruit exporting country, the tolerance threshold for fruit flies infestation is zero in fruit for export. As a result, infestation by fruit flies makes fruits and vegetables lose their commercial values (Mitcham and Yahia 2010).

The adults of the genus *Anastrepha* are medium sized (1.5 to 6 mm), and have spots or stripes on their wings, which they slowly raise up and down when they are at rest. They have large eyes, and a combination of the colors yellow-orange, black, and brown on their body parts (Christenson and Foote 1960, Bateman 1972).

Of the *Anastrepha* species, *Anastrepha obliqua* (Macquart) and *A. suspensa* (Loew) were reported to be the most economically important in the Caribbean region (White and Elson-Harris 1992, Malavasi 2000). Known as the Indian fruit fly, the West Indies fly, fruit fly of the West Indies, or fly hockey, *A. obliqua* is distributed in Mexico, Central America, the Caribbean, and South America (White and Elson-Harris 1992, Malavasi 2000). It is a medium sized fruit fly, yellowish brown, with a central strip in the chest and two lateral widening strips before the suture of the scutellum. The reproductive activity of the adults reaches its maximum at the age of 4-6 weeks, and the females lay an average of 1376 eggs for an average longevity of 79 days (maximum 175 days) (Liedo et al. 1992, Aluja 1994). They mainly breed on fruit trees in the Anacardiaceae family such as mango (*Mangifera indica*, L.), hockey, plum (*Spondias spp.*), cashew (*Anacardium occidentale*, L.), but also attack alternate hosts such as guava (*Psidium guajava*), citrus (*citrus spp.*), coffee (*Coffea arabica*, L.), inga (*Inga spp.*), Surinam cherry (*Eugenia uniflora*), mamey (*Pouteria sapota*), granadilla (*Passiflora edulis*), sapote (or sapodilla) (*Achras zapota*), and rosa apple (*Syzigium jambos*) (Norrbon & Foote 1989).

On the other hand, *Anastrepha suspensa*, known as the Caribbean fruit fly, is distributed from Florida (USA) to Puerto Rico, and throughout the Caribbean islands which includes Cuba, Haiti, the Dominican Republic, Jamaica, and Bahamas (White and Elson-Harris 1992, Malavasi 2000). It is a small fruit fly, yellowish brown, that differs from the other *Anastrepha* species by a large dark spot (absent in some specimen from Jamaica, and small in *A. fraterculus*) at the

junction of the scutum and scutellum (Foote et al. 1993). It is an oligophagous fly whose main hosts are guava, cherry (*Eugenia spp.*), Suzigium (*Syzigium cumini*), and tropical almond (*Terminalia catapa*, L.) (Norrbon & Foote 1989), with a preference to the host plants in the Myrtaceae family (Whervin 1974). However, it has been collected also in mango, citrus, loquat (*Eriobotrya japonica*), avacado (*Persea americana*, L.), more than 36 other alternate hosts. Furthermore, an inventory of host plants throughout the areas of mango production in Haiti indicated several cultivated and wild host species for *A. obliqua* and *A. suspensa*, particularly: mango, yellow mombin (*Spondias mombin*, L.), red mombin (*Spondias purpurea*, L.) (Anacardiaceae), and guava (*Psidium guyava*, L.) (Myrtaceae) (MARNDR 2008).

1.2.2 Anastrepha traps and lures

Over the past few decades, much emphasis has been put on developing effective traps and lures for survey of *Anastrepha* species, while less progress has been made for control purposes (Heath et al. 1997, Epsky et al. 1995, Epsky et al. 1999, Lasa et al. 2013). Trapping devices vary in form, color, and size, but flies are captured using either a sticky material, a liquid solution, or materials soaked in insecticides. In addition, the nature of the attractants (pheromone or food-based attractant) is the most important element of the trap and has been designed to attract a specific species or a restricted group (IAEA 2003). The two types of traps commonly used for detection, monitoring and control of *Anastrepha* species throughout the areas of fruit production are the Multilure trap (Better World MFG Inc., Fresno.CA), the McPhail trap (McPhail 1934-1935), and some variants of these traps (IAEA 2003, Epsky et al. 1995, Vargas et al. 1997). The trap devices vary in shape, color, and size, depending on the manufacturer, but the attractants remain unchanged in each type of trap, Torula yeast/ borax and hydrolyzed protein

(NuLure) for the McPhail trap and a combination of ammonium acetate + putrescine (Biolure) for the Multilure trap (IAEA 2003).

1.2.2.1 McPhail trap (MP)

According to Steyskal (1977), the development of the conventional McPhail trap (McPhail 1934-1935) started in Europe at the end of the 19th century (by Dahl F.1896) as a household fly trap, and was improved from 1930 (Costantino, Italy) to 1935 (McPhail M. 1934-1935, Key West Florida). After further improvement by McPhail in 1944, the final bell-shaped version was adopted as the standard trap, and has been widely used for survey work on fruit flies around the world. It is an open bottom transparent glass container.

In 1971, Lopez, Steiner, and Holbrook (1971) used hydrolyzed protein Torula yeast/borax as bait for McPhail trap, and it is still in use. Indeed, an aqueous formulation of protein bait (NuLure) can be used, or Torula yeast/borax tablets (5 grams) are added to water to make a 250ml of liquid food bait. The pH of this solution, which attracts mostly female fruit flies, must remain stable at 8.5 for the hydrolyzed protein and 9.2 for the Torula yeast/borax to be attractive. After eight days, the solution becomes too acidic, and loses its attractiveness in the case of hydrolyzed protein. As a result servicing/re-baiting is conducted on a weekly basis before the pH of the solution drops (Epsky et al. 1993, IAEA 2003).

For better efficacy in capturing flies, better cost, and for convenience in managing the trapping networks, different variants of the McPhail trap were developed to replace the conventional version (glass container) such as Plastic McPhail IPS 235 (Great Lakes IPM, Inc., Vestaburg), Dome Trap (Agrisense BCS Ltd., Pontypridd, United Kingdom), Tepri-trap Ecological, Multilure trap device (Better World MFG Inc., Fresno.CA). Contrary to the conventional

McPhail trap, these variants offer different options to be used either as a wet trap, or as dry trap with synthetic lures (Lasa et al. 2014a).

1.2.2.2 Multilure trap (M)

The Multilure trap (Better World Manufacturing Inc., Fresno, CA, USA) is an open bottom trap consisting of a two-piece plastic cylinder. The top is clear and the bottom is yellow in order to be attractive to flies. The same container is used for the McPhail trap as well (described above). The trap is baited with a dry synthetic lure consisting of three separate small dispensers (ammonium acetate, putrescine, and trimethyl amine) (Biolure) attached inside the trap's wall or in the ceiling. Water is used in the trap as part of the retention system, with 10% propylene glycol to reduce its evaporation in hot climates, and to decrease the decomposition of captured flies. The three lures attract female Mediterranean fruit flies (*Ceratitis capitata*, Weidemann (Diptera: Tephritidae)), but the trimethyl amine must be removed in order to attract and capture *Anastrepha spp.* (Epsky et al. 1993, Health et al. 1993, Epsky et al. 1995, IAEA 2003).

1.2.3 Layout of trapping network

The layout of the trapping network is an important step of a trapping method. It depends on the intrinsic characteristics of the sample area, such as urban/rural, vegetation types, host plants, and dispersed fruit trees/orchard. Moreover, it includes two important elements, trap placement and trap density (IAEA 2003).

Traps should be placed 2-4 meters from the ground, in shady areas of primary or secondary fruit host trees. During the fruit maturation period, protein-baited traps should be placed on primary hosts, rather than secondary hosts, and on other potential fruit fly pathways (IAEA 2003).

Trapping density depends on the survey objectives (detection, suppression, eradication, or exclusion), and varies greatly with the characteristics of the area (production area, marginal area, urban area and point of entry) (IAEA 2003). Indeed, in the case of eradication (control), where mass trapping methods should be applied, the trap density should be 20-50 traps/km² (IAEA 2003) or 25-50 traps/ha (Martinez-Ferrer et al. 2012) to yield the best results.

For detection and monitoring, a trapping network has been in place in Haiti since 2008, consisting of a density of 2 Jackson traps/km² to specifically survey the Mediterranean fruit fly (*Ceratitidis capitata*, Weidemann), 2 Multilure traps and 0.5 McPhail trap per square kilometer (km²) for *Anastrepha spp.* (MARNDR 2009).

CHAPTER II. DEVELOPMENT, EVALUATION, AND COMPARISON OF THE ARTISANAL MCPHAIL TRAP WITH THE COMMERCIAL MCPHAIL TRAP

2.1 Introduction

Reduction of environmental impact caused by pesticide use, food safety concerns, and prevention of movement of invasive species into new areas are the most important objectives for the development of trapping methods to survey, detect, and control fruit flies (Diptera, Tephritidae). Many trap types have been developed and used for these purposes, depending on the goal and the fruit fly species (IAEA 2003).

Trap designs vary in dimension, color, and shape, but the core principles of a trap is based on two main factors: attractiveness and physical retention. The baited trap aims to attract the flies within a certain distance from the trap by releasing different volatile compounds (El Sayed et al. 2006). On the other hand, the trap device must offer certain physical characteristics (color, dimension, shape) that attract flies and prevent them from escaping after entering the trap. In sum, these factors are incorporated to take advantage of the fruit flies' behavior. Based on these principles, development of artisanal traps has been initiated with the goal of reducing the cost of trapping systems. Lasa et al. (2014b) tested two handmade trap models (a 500 ml blue polyethylene bottle and a 500 ml transparent colorless polyethylene bottle, both with three 10 mm diameter holes perforated at 2/3 above the base, baited with CeraTrap Lure) for control of *Anastrepha ludens* (Diptera, Tephritidae) in orange orchards in Mexico. These artisanal traps have proven to be more efficient under cage and field conditions in number of fruit flies trapped than two commercial traps (MS2 trap (Fitosanitaria S.A. de C.V., Texcoco, Mexico) and A&C trap (Mubaqui, Tamaulipas, Mexico), also baited with CeraTrap Lure) tested under the same conditions.

2.2 Objective and Hypotheses

The present experiment was conducted to (1) develop two artisanal McPhail trap models: AT1 and AT2, and (2) to evaluate their performance in comparison to the commercial McPhail (MP) trap under field conditions.

H_0 = The number of fruit flies caught in the three trap models: AT1, AT2, and MP are equal.

H_A = The number of fruit flies trapped is different, in at least one of the trap models.

2.3 Materials and Methods

2.3.1 Study Site

This experiment was conducted from December 31st 2014 to January 14th 2015, during the driest season (November-February) in a mango orchard (*Mangifera indica*, L., cv. ‘Tommy Atkins’) located in the municipality of Croix-des-Bouquets, (N 18°34’00.0” W 72° 13’45.0”W) in Ouest Département, Haïti. The site has an elevation of 90-95 m altitude, and less than 8% slope.

The orchard had ca. 238 trees per ha, spaced at 7m x 6m. It was considered to be a commercial orchard fifteen years ago, and now is characterized by its lack of sanitation practices. The mango trees were 10-15 meters high at the time of the study. Like all areas of mango production across the country where the fruit fly program has been implemented, this area has been surveyed since 2008, using Jackson, McPhail and Multilure traps which have reduced the fruit fly density to 2.54 flies/trap/day (FTD) before the period of this study (MARNDR 2014). Other host plants such as guava, yellow mombin, and red mombin are not abundant around the orchard. An area of 3 ha was delimited for this experiment.

2.3.2 Trap models description and lures

The artisanal trap models (AT1 and AT2), developed based on principles indicated below, and the conventional McPhail trap (MP) (Fig.2.1) were used in this study:

- i) **the yellow bottom artisanal trap:AT1** is made from a 0.59 liter clear recycled plastic soda bottle. To allow attractant diffusion and fly entrance to the trap, two 1cm diameter circular holes were symmetrically cut at two thirds (2/3) the height of the bottle. In addition, the base of the bottle was painted yellow, up to 1/5 of its height (Fig.2.1)
- ii) **the clear artisanal trap:AT2** is just the artisanal trap AT1 as described above, without the yellow base (Fig.2.1).
- iii) **the commercial McPhail trap: MP** (Model: AR933 McPhail Trap/ ISCA Technologies), with clear top and yellow opened bottom as described above (Fig.2.1), was used for the study.



Fig. 2.1: Trap models developed and used for the study: AT1, yellow bottom artisanal trap; AT2, clear artisanal trap; and MP, Commercial McPhail trap.

The attractant used in these trap models for the entire study was composed of two pellets (5 grams) of Torula Yeast/borax and 250ml of water. Torula yeast is a proteinaceous food that releases volatile compounds that are highly attractive to fruit flies. Protein sources are critical for adult fruit flies soon after emergence, for growth, ovaries development and other reproductive activities (Bateman 1978, IAEA 2003, Aluja and Rull 2009).

2.3.3 Experimental Design

Traps were labeled, then placed in the orchard in groups of three, including one model for each set. Traps were hung in mango trees at 3-4 meters height, in a triangular pattern at 15 meters apart. Two pellets of Torula yeast and 250ml of water were put in each trap, and 20 replicates of the set of three traps were randomly distributed throughout the mango orchard.

2.3.4 Data collection and analysis

Traps were sampled every 3-4 days, and each set of three traps was rotated clockwise (sampling/rotation) to minimize any effect of trap location. The liquid bait was replaced every second sample time. The insect specimens were placed in labeled vials with 75% alcohol, and returned to the laboratory for counting and identification under a binocular microscope of species and sex using a specific key. The Australian handbook for the identification of fruit flies (version 1.0; ed. Woods N) was used for this purpose (Plant Health Australia 2011).

Statistical analysis was performed by Analysis of variance (PROC General Linear Model, SAS Institute Inc. SAS/STAT 2006), followed by Fisher's LSD mean separation procedure ($\alpha = 0.05$) for significant ANOVAs, in order to compare the trap models on their average number of fruit flies (Appendix A). A binomial test for proportions ($\alpha = 0.05$) (Bonferroni's correction for 3 comparison, $\alpha = 0.017$) was performed to compare the trap models on their proportion of fruit flies trapped (Appendix B).

2.4 Results and discussion

2.4.1 Trap Model Effectiveness under Field Conditions, and Mass Trapping

During the 15 days when fruit flies were collected from the traps, significant differences were not observed in the average number of fruit flies captured among the yellow bottom artisanal trap (AT1) (8.9 ± 2.6), the clear artisanal trap (AT2) (13 ± 2.9), and the commercial McPhail trap (MP) (16 ± 4.1) ($F = 1.2$; $df = 2$; $P = 0.3084$) (Fig.2.2).

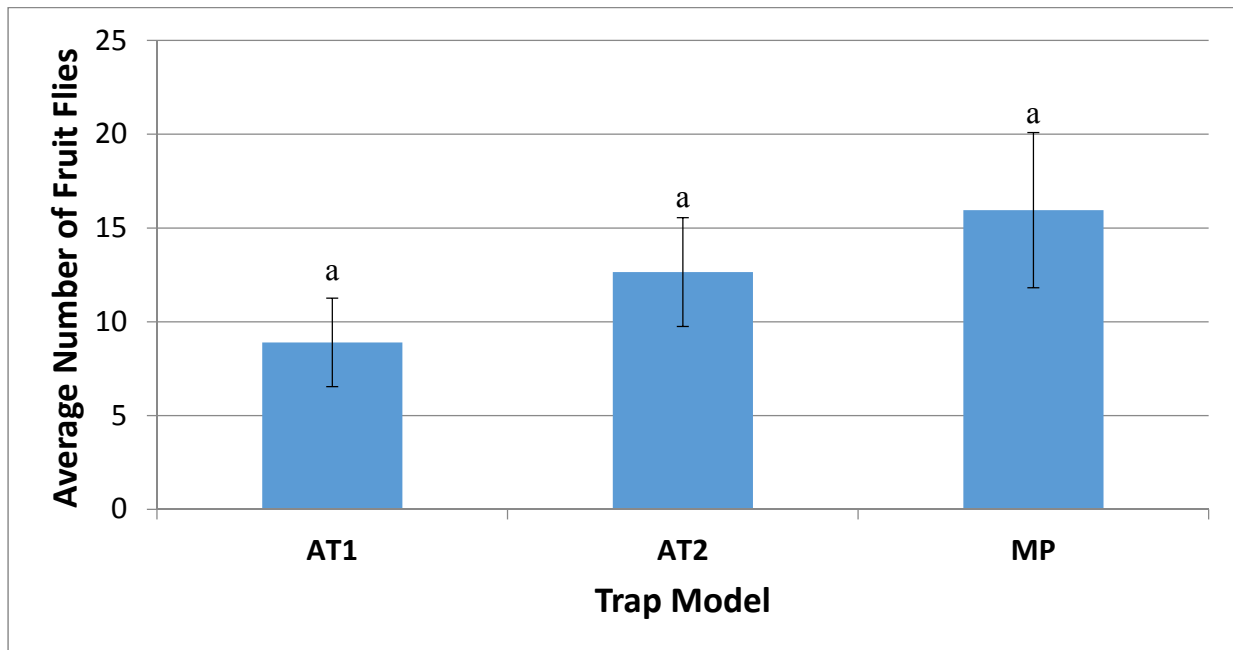


Fig. 2.2: Mean (\pm SE) number of fruit flies caught per trap model. Bars labeled with identical letters were not significantly different after comparisons among trap models (*ANOVA*, Fisher's LSD $\alpha=0.05$).

However, significant differences were observed between the three trap models in total number of fruit flies caught (Fig.2.3). The total number of fruit flies trapped in the clear artisanal trap (253 flies) was significantly higher than that in the yellow bottom artisanal trap (178 flies) ($Z = 3.56$; $P < 0.001$); the total number of fruit flies caught in the McPhail trap (319 flies) was significantly higher than that in the yellow bottom artisanal trap (178 flies) ($Z = 6.28$; $P < 0.001$), and the total

of fruit flies trapped in the McPhail trap (319 flies) was significantly greater than that in the clear artisanal (253 flies) ($Z = 2.72$; $P < 0.001$) (Fig. 3).

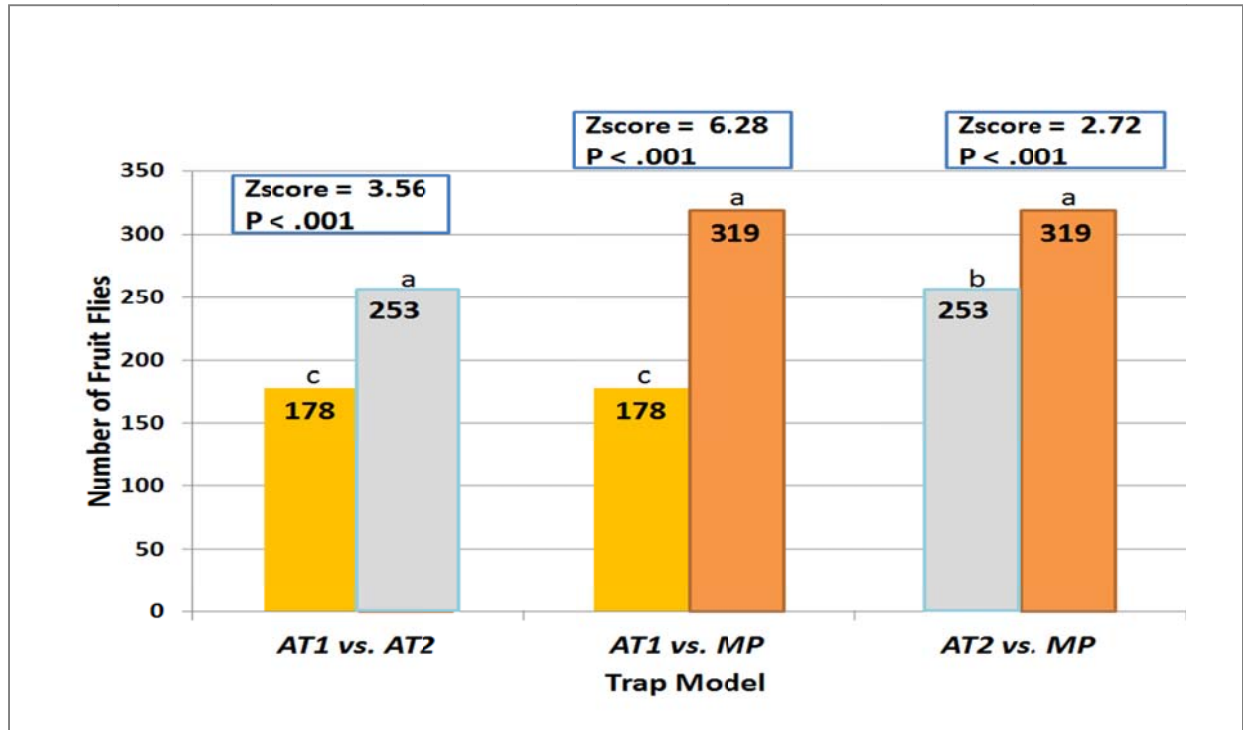


Fig. 2.3: Total number of fruit flies caught per trap model. Bars labeled with identical letters were not significantly different after comparisons among trap models (Binomial test for equal proportion, Bonferroni's correction $\alpha=0.017$).

Certain intrinsic characteristics of each trap model contribute to make the observed difference in number of fruit flies caught between the three trap models. First, the entrance hole in the McPhail trap (6cm) is six times larger than in the artisanal trap models (AT1 & AT2) (1cm), which might facilitate a better diffusion of the attractant and the entrance of the flies (F.M. Personal observation). Second, in addition to the chemical cues released from the food bait, the yellow bottom in the McPhail and the artisanal type AT1 is considered a visual cue that attracts flies when they come close to the traps (Cytrynowicz et al. 1982, Sivinski 1990, Aluja and Rull 2009). This coloration plays a key role in the McPhail trap by guiding the flies through the entrance hole, while it may have a negative effect on the performance of the yellow artisanal trap

by distracting flies from entering the holes, because the holes are not located in the yellow part of this trap. Furthermore, the volume of the McPhail trap is approximately three times larger than the artisanal trap models (591ml), which might improve the physical retention of the McPhail trap and prevents flies from escaping after entering (F.M. Personal observation). These factors contribute in whole or in part to improve trapping efficiency in the McPhail trap compared to the clear artisanal trap as well as to provide a higher number of flies in the clear artisanal trap compared to that caught in the yellow bottom artisanal trap.

Significant differences were observed between the proportion of females and males captured in the three trap models ($F = 10.91$; $df = 5$; $P < 0.001$). The percentage of females trapped in the yellow bottom artisanal trap (74%), the clear artisanal trap (63%), and the McPhail trap (63%) was significantly greater than that of males, which was respectively 26%, 37%, and 37% (Fig.2.4).

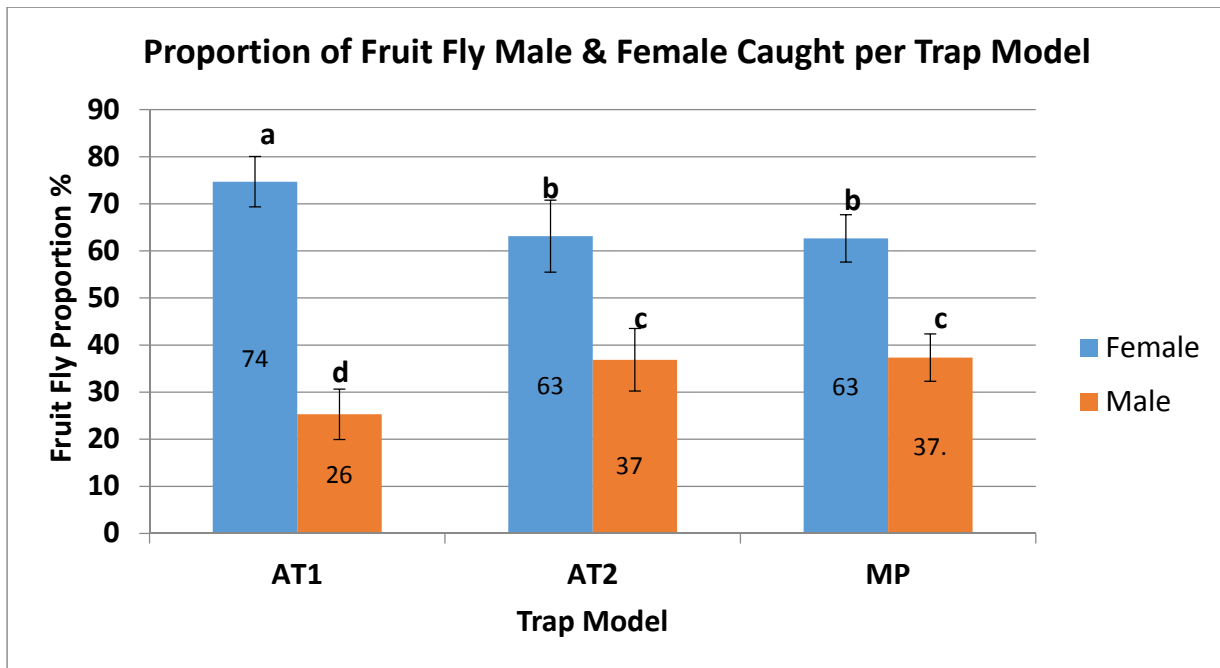


Fig. 2.4: Average proportion of female and male fruit flies caught per trap model. Bars labeled with identical letters were not significantly different after comparisons among trap models (ANOVA, Fisher's LSD $\alpha = 0.05$).

Further analysis demonstrated that the proportion of females caught in the yellow bottom artisanal trap was significantly greater than that of the clear artisanal trap ($Z = 2.54$; $P = 0.011$), and that of the McPhail trap ($Z = 2.66$; $P < 0.008$). However, significant differences were not observed in the proportion of females trapped in the clear artisanal trap and that of the McPhail trap ($Z = 0.099$; $P = 0.921$) (Fig.2.4). These observations have shown that the McPhail traps, commercial or artisanal models, are female-biased (Lasa et al. 2014a). Female fruit flies need much more protein than do males, because of requirements for oviposition. As a result, they are more attracted to the volatile compounds emitted by the proteinaceous bait (Torula yeast) (Bateman 1978, Aluja and Rull 2009).

Laboratory analysis of the insect specimens from the three trap models revealed that these traps caught many insect families of different orders, of which non-tephritid dipterans and wasps were predominant. Only the two fruit fly species formerly reported by the Ministry of Agriculture through the Fruit Fly Control Program were identified: the Indian fruit fly and the Caribbean fruit fly (MARNDR 2008). Nevertheless, the proportion of *A. obliqua* was significantly higher than that of *A. suspensa* in the three trap models (Table 2.1). In the yellow bottom artisanal, the clear artisanal, and the McPhail traps a proportion of 99%, $\approx 100\%$, and $\approx 100\%$ *A. obliqua* were respectively identified, while only 1%, $< 1\%$, and $< 1\%$ *A. suspensa* were respectively keyed out of these trap models (Table 2.1). These data confirm that mango is the main host for *A. obliqua* and alternate host for *A. suspensa* (Whervin 1974, Norrbon & Foote 1989). In addition, the population of *A. obliqua* might be much higher in this mango growing area of Haiti than that of *A. suspensa*, because studies have shown that the McPhail trap is effective against both fruit flies species (Burditt Jr 1982).

Table 2.1: Proportion of Fruit Fly Species Caught per Trap Model, and Comparison of the Cost-Effectiveness of the Trap Models

Trap model	Flies caught	% Fly per species		Price	Cost-effectiveness
	Total number	<i>A. obliqua</i>	<i>A. suspensa</i>	20traps(\$)	Dollars/Fly caught
AT1	178 c	98.87	1.23	75	0.42
AT2	253 b	99.6	0.4	70	0.28
MP	319 a	99.69	0.31	220	0.69

The number of flies followed by different letter are significantly different ($Z(AT1,AT2) = 3.56$, $P < 0.001$; $Z(AT1,MP) = 6.56$, $P < 0.001$; $Z(AT2,MP) = 3.56$; $P < 0.001$), Binomial for equal proportion ($\alpha = 0.017$).

A comparison between the prices of these three trap models has shown that the McPhail trap (MP) is a much more expensive than the artisanal trap models (AT1 and AT2). The total cost of the 20 McPhail traps used for the experiment (\$220 for 319 flies caught) is more than three times the total cost of 20 clear artisanal trap (AT2) (\$70 US for 253 flies caught), and approximately three times the cost of 20 yellow bottom artisanal (AT1) (\$75 US for 178 flies caught) (Table 2.1). As a result, an analysis of the cost/fly caught demonstrated that the clear artisanal trap (AT2) was the most cost-effective model at 0.28 dollars/fly caught, followed by the yellow artisanal trap model (AT1) at 0.42 dollars/fly caught, and the commercial McPhail trap (MP) that was the least cost-effective model tested at 0.69 dollars/fly caught (Table 2.1). The importance of inexpensive traps was highlighted by Lasa et al. (2014b) to ensure the best cost-benefit ratio possible with the mass trapping technique. In fact, cost, the most important factor in determining the Economic Injury Level (EIL), should be the first criterion to be taken into account while developing any management plan (Stern et al. 1959). If the cost of the management plan is too high, the crop yield and the market value of the product may not be sufficient enough to ensure a good profit. In this case, mass trapping with commercial McPhail traps would require a prohibitive number of traps per ha, making a fruit fly control program very expensive and financially impossible to manage. Based on its cost-effectiveness, the clear artisanal trap model (AT2) was retained for use in subsequent experiments (Chap. 3-4).

CHAPTER III. DEVELOPMENT, EVALUATION, AND COMPARISON OF THE ARTISANAL MCPHAIL TRAP WITH THE COMMERCIAL MCPHAIL TRAP

3.1 Introduction

The trap design and bait combination represent the most important factors that make a trap effective against fruit flies (Lasa et al. 2014a). However, using mass trapping as a pest control method, the number of traps per surface area and the trap distribution throughout the orchard are crucial in yielding satisfactory results (El Sayed et al. 2006). This technique consists in placing an optimal number of baited traps throughout an area, in order to reduce as much as possible the foraging adult fruit flies population in this area (Martinez-Ferrer 2010). Based on the results obtained from field trials in citrus groves in Spain, Martinez-Ferrer (2010) reported that a density of 25 traps/ha can be used as a stand-alone method to control the Mediterranean fruit fly (*Ceratitis capitata*, Wiedman), but when the population density increases during the fruiting period, the trap density should be adjusted for a successful control of the pest. Other research has shown that 20-25 traps/ha was the required density using a mass trapping technique for any eradication program (IAEA 2003).

Earlier research described in this thesis (Chapter II) demonstrated that the clear artisanal model (AT2) was less effective than the conventional McPhail trap (MP) in capturing *A. obliqua* and *A. suspensa* (Table 2.1). Therefore, the performance of this trap under mass trapping conditions needed to be evaluated, by testing an optimal density of McPhail trap and a high density of the artisanal trap.

3.2 Objective and Hypotheses

This experiment was conducted in order to compare the performance of a density of 36 clear artisanal traps per ha (36AT2/ha) (high density) to a density of 24 McPhail traps per ha (24MP/ha), considered as an optimal density.

H_0 = The number of fruit flies caught using 36AT2/ha and 24MP/ha are equal

H_A = The number of fruit flies caught using 36AT2/ha and 24MP/ha are not equal.

3.3 Materials and Methods

3.3.1 Study Site

This experiment, conducted from January 17th to March 7th 2015, started three days after the first one finished, in another part of the same orchard described above in the first experiment. In addition, it was conducted at the same period as the third experiment, in the northern side of the mango field, and at approximately 50 meters distance from the experimental field of the third experiment.

3.3.2. Experimental design

A total of six plots of 0.25 ha (50m x 50m) were scattered throughout the experimental field, and plots were located at 25-30 m from each other. Six MP traps and 9 AT2 individually labeled traps were placed on mango trees in a regular pattern, 2 x 3 and 3 x 3, respectively within each plot. All traps were baited with 2 tablets of Torula yeast, and 250ml of water added to each trap model: AT2 and MP (Fig. 2.1).

Three replicates of both treatments were assigned to plots in a completely randomized design.

2.3.3 Data collection and analysis

Sampling was performed on a weekly basis, and the liquid bait was replaced at the same time. The insect specimens were placed in labeled vials with 75% alcohol, and brought to the laboratory for counting and identification. The Australian handbook for the identification of fruit flies (version 1.0; ed. Woods N) was used for this purpose (Plant Health Australia, 2011).

Statistical analysis was performed using a binomial test for equal proportion ($\alpha = 0.05$) to compare the proportion number of fruit fly trapped in 36AT2/ha and 24MP/ha (Appendix C).

3.4 Results and discussions

3.4.1 Performance of the clear artisanal trap model in mass trapping conditions

After a seven-week period, 236 and 239 fruit flies were captured using 36AT2/ha and 24MP/ha, respectively (Fig.3.1). Analysis of the proportion of fruit flies caught using both trap densities failed to find differences in the total number of tephritid flies caught ($Z = 0.13$, $P=0.734$ ($\alpha=0.05$)) (Fig.3.1).

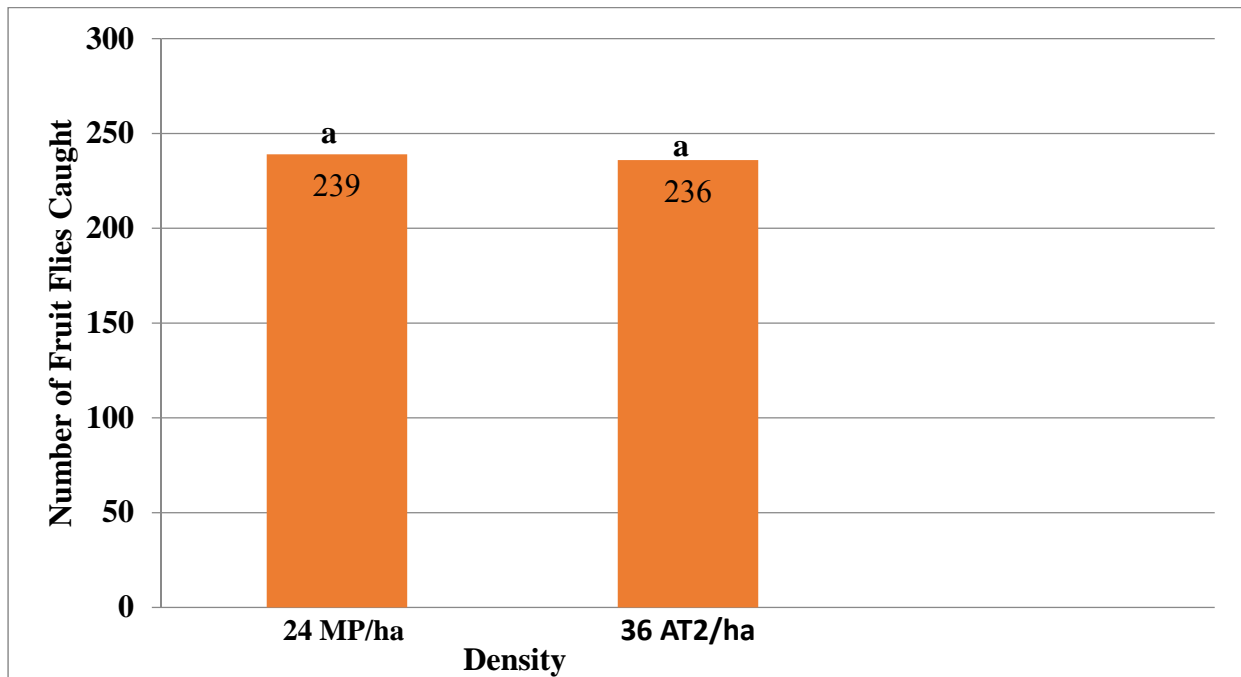


Fig. 3.1: Total number of fruit flies caught in 24MP/ha and 36AT2/ha. Bars labeled with the same letters were not significantly different (Binomial test for equal proportion, $\alpha=0.05$).

These results showed that the clear artisanal trap model (AT2) is effective in capturing tephritid flies in a mass trapping program. Moreover, in this experiment, a density of 36 traps per ha (36AT2/ha) was as effective as 24 McPhail traps per ha (24MP/ha). According to Martinez-Ferrer (2010) the trapping density must be increased from 20 traps per ha to 50 traps per ha during the fruiting period to ensure sufficient protection. In this case, the 24MP/ha and the 36AT2/ha might not be high enough to protect the mango orchard throughout the fruiting period.

When the traps were set in the orchard to start the experiment, mango fruits were just developing. By the end of the experiment (March 7th 2015), the mango fruits were maturing. Several studies demonstrated that fruit flies have a direct coevolution with their host plants during their life history, which enables them to delay emergence during periods of food scarcity and to emerge in mass during seasons of food abundance (Backer 1944, Nishida 1963, Bateman and Sonleitner 1967). The analysis of the number of fruit flies caught every week using both trap densities (24MP/ha and 36 AT2/ha) showed that the number fruit flies increased every week as mango fruits matured (Fig.3.2).

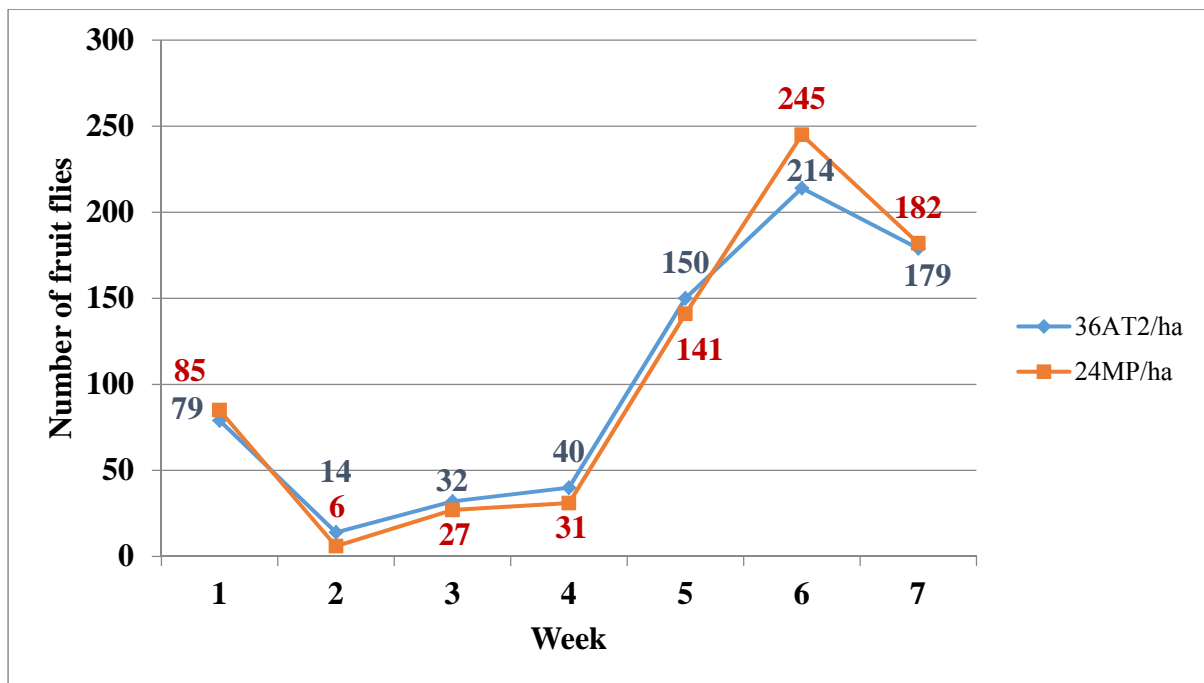


Fig. 3.2: Number of flies caught in both trap densities throughout this experiment.

During the first week of the experiment the fruit fly densities were low, with very limited food and water sources in the orchard, because it was the driest season of the year and the beginning of the mango production period. That likely explains why high numbers of flies were caught (85 flies in MP and 79 flies in AT2) during the first week of sampling and reduced to 6-

14 flies during the second week. Backer (1944) and Bateman (1967) reported that water is essential to fruit flies survival, growth, and reproduction, especially during the dry season. This observation showed that fruit flies water dependence is a good factor that can be exploited by incorporating chemical control strategies in the management plan during the dry seasons. Aerial applications of Spinosad in commercial citrus in Florida resulted in a reduction of 54-73% of the population of the Caribbean fruit fly (*Anastrepha suspensa*, Loew) (Burns et al. 2001). This organic pesticide (Dow Chemical) could be incorporated into a control program to protect Haitian organic mangoes for export, especially during long dry season of December-March. Due to time and logistic constraints, this study did not evaluate the amount of fruit damage by both species of fruit flies. A future study is needed to correlate the efficacy of trapping regarding fruit infestation by these species.

CHAPTER IV. DETERMINATION OF AN OPTIMAL DENSITY FOR THE ARTISANAL TRAP (AT2) UNDER MASS TRAPPING CONDITIONS

4.1 Introduction

Trapping has been used for decades for detection, monitoring, and eradication of fruit flies (IAEA 2003). Depending on the objectives pursued in this management method (detection, delimitation, monitoring, exclusion, suppression, or eradication), the characteristics of the area in question (production area, urban area, points of entry), and the types of trap used, the trapping density may vary greatly. Indeed, according to the International Atomic Energy Agency (2003), for detection and monitoring, whose goals are respectively to detect the presence of fruit fly species and to study the population dynamics within an area, the trapping density is relatively low (0.25- 5 traps/km²). However, when the purpose is to reach a low prevalence of fruit flies in an area (suppression) or to reach a fruit fly free area (eradication) the trapping density is respectively 10-20 traps/km² and 20-50 traps/km², because a high density of traps is needed to capture significant number of flies (IAEA 2003, Martinez-Ferrer et al. 2010). Navarro-Llopis et al. (2004) reported that mass trapping has been used for decades to control the Mediterranean fruit fly (*Ceratitidis capitata*, Wiedemann) with the conventional McPhail trap, and demonstrated that a density 50 traps per ha had good efficacy protecting citrus groves against the Mediterranean fruit fly (Navarro-Llopis et al. 2008). Moreover, besides other methods and combinations of techniques that were developed to control tephritid flies such as chemical control, insect sterile technique (IST), chemosterilant, and biological control with parasitoids, mass trapping has been the most promising technique against the Medfly, because of their non-negative impacts on the environment (Navarro-Llopis et al. 2008, Martinez-Ferrer et al. 2010). Because of the large number of traps required and their costs, the trend in research has been oriented over the last decade towards developing inexpensive traps and determining optimal trap

densities for mass trapping. Recent trials in Mexico, conducted to develop and to evaluate inexpensive handmade trap designs demonstrated that they were more efficient than two commercial traps: MS2® (Fitozoosanitaria S.A. de C.V., Texcoco, Mexico) and A&C Trap® (Mubarqui, Tamaulipas, Mexico) (Lasa et al. 2014b). On the other hand, trials conducted by Martinez-Ferrer et al. (2010) in citrus groves in Spain to optimize mass trapping density revealed that a density of 25 traps per ha (Maxitrap Model baited with Ferag. CC D TM® attractant) can be a good stand-alone control method against the Medfly, depending on the fruiting season. In addition, other factors such as attractant efficacy, host plant abundance, climate, fruit fly species, and trap efficacy were highlighted to have the most influence on trapping densities (IAEA 2003).

4.2 Objective and Hypotheses

The present experiment was conducted to determine the optimal mass trapping density by comparing the efficacy of six different trap densities (4, 8, 12, 16, 24, and 36 traps per ha) for the clear artisanal trap model (AT2) (Fig.2.1).

H_0 = The number of fruit flies caught in all six trap densities are equal

H_A = The number of fruit flies caught, at least in one of the trap densities, is not equal.

4.3 Materials and Methods

4.3.1 Study Site

This trial was conducted during the same period as the second experiment (from January 21st 2015 to March 3rd 2015), on the southern side of the same orchard described previously. This period coincides with the major season of mango production in this region under conditions of low altitude in Haiti.

4.3.2 Experimental design

Throughout the study area, a total of eighteen (18) plots of 0.25 ha (50m x 50m) were delimited, and plots were separated by 25-30 meters from each other. As a result, the number of traps was divided by four (4) in order to have six densities (treatments) of 1, 2, 3, 4, 6, and 9 clear artisanal traps (AT2)(Fig.2.1) for 0.25 ha.

Each trap was baited with 2 tablets of Torula Yeast and 250 ml of water. After replicating each trap density three times, each trap density was randomly assigned to a plot, in order to have a completely randomized design (CRD). Traps were individually coded based on their respective plot, and traps of a same plot were dispersed equidistantly on mango trees throughout the plot area.

4.3.3 Data collection and analysis

Sampling was conducted every seven days, and the food bait (Torula Yeast + water) was replaced at the same time. After labeling, the insect specimens were placed in vials with 75% alcohol, and brought to laboratory for counting and identification by using specific key. The Australian handbook for the identification of fruit flies (version 1.0) was used for the purpose (Plant Health Australia, 2011).

Statistical analysis was performed using SAS (English 9.4) (SAS/STAT 2006). Because of difference in the variances of the treatments, with Hartley's $F_{max}=3337.31$, the data were transformed using the Negative Exponential function: $Y = asymptote * (1 - Exp^{(curve * (X + Shift))})$. Gauss-Newton iterative Method was used to estimate the parameters: asymptote, curve, and shift. Nonlinear regression analysis (Proc NLIN) (Nonlinear Model – Negative Exponential) was performed to assess the relationship between the dependent variable (Y= number of fruit flies caught) and the dependent variable (X= trap density) (Appendix D). The initial parameter

estimates for the nonlinear regression were asymptote=50, curve=-0.40, and shift=-14 (Appendix D). The Gauss-Newton algorithm was used to estimate parameters for the model. This algorithm regresses residuals onto the partial derivatives of the model with respect to the parameters until the estimates converge (SAS/STAT 2006). Convergence criteria were met when the sum of squares was minimized (Appendix D).

4.4 Results and Discussion

The analysis of the scatter plot of the number of fruit flies (dependent variable) with the trap density (independent variable): plot of $Y * X$ shows some outliers at different levels of trap density ($D_4=0$, $D_8=71$, $D_{12}=4$, $D_{16}=31$, $D_{24}=38$, 44, and $D_{36}=12$ fruit flies caught) (Fig.4.1). These small numbers of fruit flies caught in high trap densities, so call outliers, resulted in undetectable variability throughout the mango orchard. The soil conditions in some area of the field must have altered adult fruit fly emergence (Bateman and Sonleitner 1967).

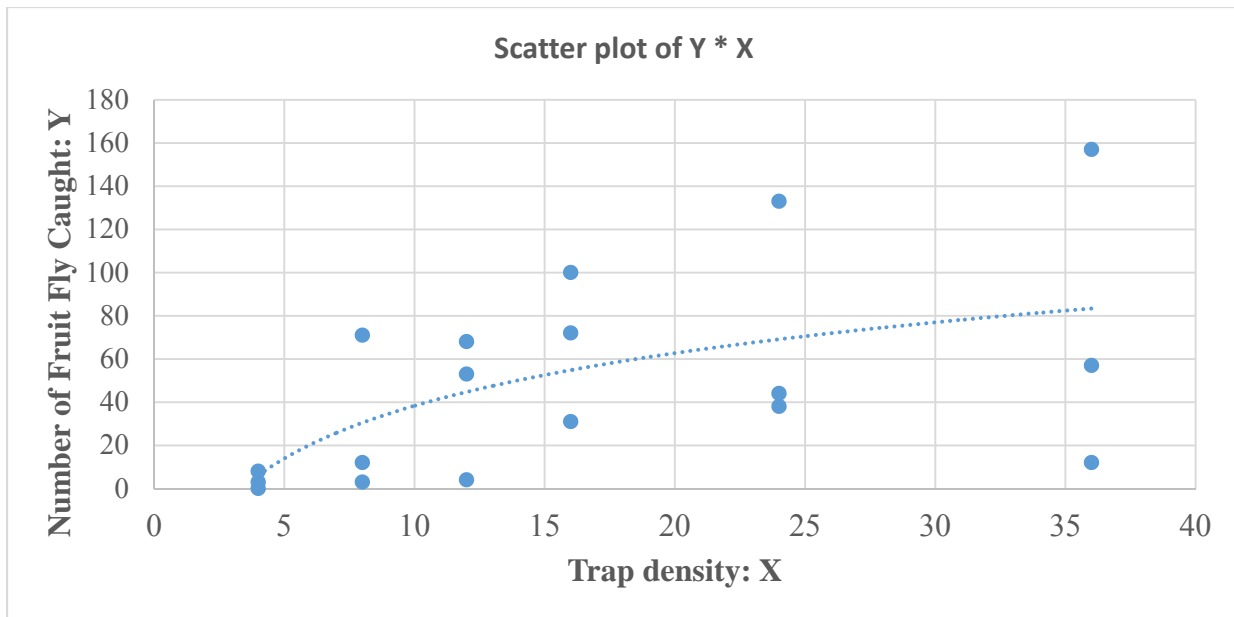


Fig. 4.1: Scatter plot of the dependent variable (Y) with the independent variable (X): $Y * X$. The letters A are observations (number of fruit flies caught) for each level of trap density.

Indeed, soil plowing used by small farmers who practice intercropping in parts of the mango grove might have reduced the amount of pupae in the soil, by exposing them to sunlight and to predators on the soil surface.

Even though the data yields a scatter plot with many outliers, data transformation with the Gauss-Newton iterative method and Negative Exponential function provided a significant model ($F=3.64$, $P=0.0514$, $\alpha = 0.1$) (Table 4.1).

Table 4.1: Regression Analysis: Nonlinear Model Test (PROC NLIN, SAS (English 9.4), SAS/STAT 2006)

Source	DF	Sum of Squares	Mean Square	F Value	Approx.(Pr > F)
Model	2	11868.2	5934.1	3.64	0.0514
Error	15	24439.6	1629.3		
Corrected Total	17	36307.8			

Table 4.2: Regression Analysis: Parameter Estimates (PROC NLIN, SAS (English 9.4))

Parameter	Estimate	Approx. Std Error	Approximate 95% Confidence Limits	
asymptote	79.6108	26.2964	23.5614	135.7
curve	-0.1119	0.1109	-0.3483	0.1244
Shift	-3.7450	2.6720	-9.4403	1.9503

Moreover, the parameter estimates (asymptote = 79.61 ± 26.29 , curve = -0.112 ± 0.11 , Shift = -3.7450 ± 2.67) (Table 4.2) had a good correlation that allowed the development of a predictive equation to assess the relation between the number of fruit flies caught (dependent variable) and the trapping density (independent variable): $Y = 79.61 * (1 - \text{Exp}^{-0.112 * (X - 3.745)})$.

Analysis of the deductive curve resulting from the equation demonstrated a closed relationship between the number of fruit flies caught (Y) and the trapping density (X) (Fig. 4.2). The rate of the curve (additional flies caught per additional traps) increased more than proportionally, from 4

traps/ha to 22 traps/ha, when the density was increased by 2 trap units (2 more traps yields > 2 fruit flies). And, this rate has a proportional increase (2 more traps yield 2 more fruit flies) around 24-25 traps/ha, to become less than proportional (2 more traps yield < 2 more flies) from 26 traps/ha to 36 (Table 4.3) (Fig.4.2). These data suggested that the trapping density should be set at around 25 traps/ha, because at 24-25 traps/ha each trap unit added yields 1 additional fly. Moreover, each trap unit added will catch less than 1 fly when the trapping density is greater than 25 traps/ha (Table 4.3).

This study demonstrated that the number of fruit flies caught increases as the trapping density increases. But, when there are too many traps per surface area, the fruit fly density during the period may not be high enough to justify a high trap density (Martinez-Ferrer et al. 2010), because it will be a waste of money. Indeed, according to Martinez-Ferrer et al. (2010), the mass trapping density must be adjusted with respect to the fruit flies population density, which is the key factor that triggers any management decision. His research conducted in citrus groves in Spain revealed that a density of 25 traps/ha was sufficient enough to protect the citrus groves, but this trapping density needed to be increased up to 50 traps/ha during the early-season when the fruit flies reaches its peak. Similarly, data collected from a McPhail trap that has been installed in the mango orchard (experimental site in Haiti) for monitoring purposes (MARNDR 2014) shows that the fruit flies population density reaches its peak in April, which coincides with the end of the mango season the trial is conducted (Fig. 4.3).

This experiment was conducted from January 21st to March 3rd 2015, period during which the rate of fruit fly capture was increasing exponentially (Fig. 3.2). At the last sampling of this field trial (March 3rd), the mango fruits were in their maturation phase (personal observation). Thus, the rate of capture, indirect estimation of the fruit fly density, would be expected to

increase until the end of the season in April. Therefore, the density of 25 traps/ha should be increased at the end of the mango season, specifically during the ripening phase, in order to reach a fruit fly low prevalence, because fruit fly tolerance threshold is very low in marketable fruit and must equal to zero to satisfy quarantine requirements (Mitcham and Yahia 2010).

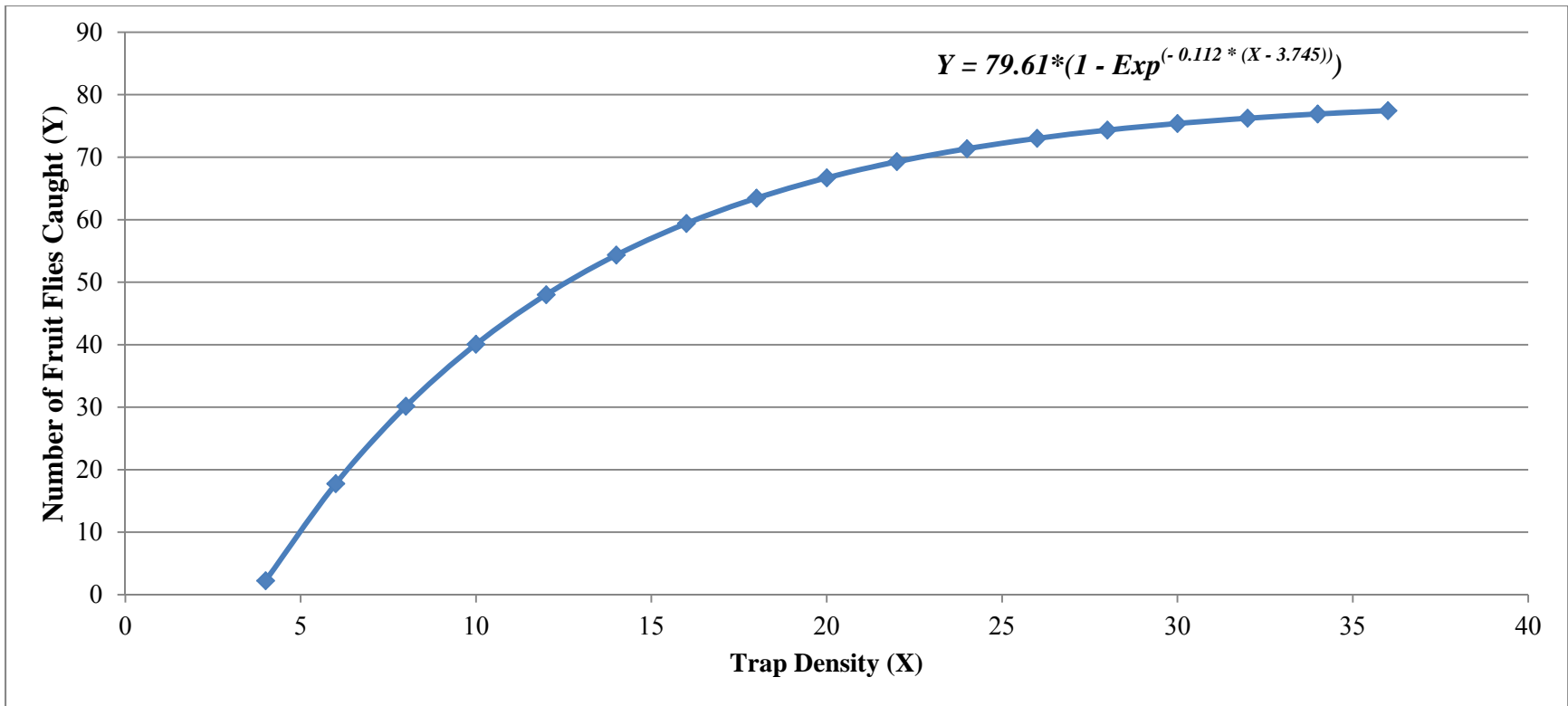


Fig. 4.2: Predicted Curve Showing Predictive Relationship between Number of Fruit Flies Caught (Y: dependent variable) and Trap density (X: independent variable)

Table 4.3: Predicted Change in Capture Rates of Flies Compared to Increase in the Trapping Density.

Trap Density: X	4	6	8	10	12	14	16	18	20	22	24	26	28	30	32	34	36
Number of Flies: Y	2.24	17.8	30.2	40.1	48	54.3	59.4	63.5	66.7	69.3	71.4	73	74.3	75.4	76.2	76.9	77.5
Capture Rate: $Y_n - Y_{n-1}$	--	15.5	12.4	9.9	7.9	6.3	5.1	4	3.2	2.6	2.1	1.6	1.3	1.1	0.84	0.68	0.54

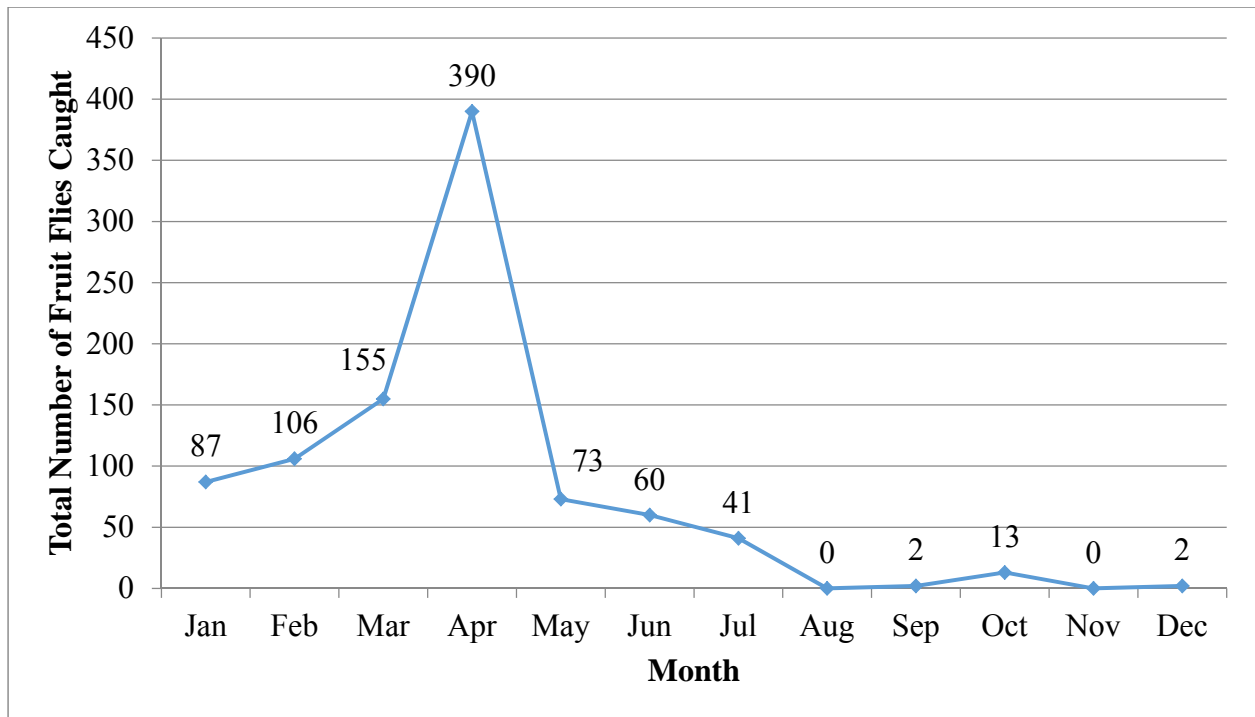


Fig. 4.3: Fruit Flies Population Density in the Experimentation Site (Mango Orchard), during 2014. Ministry of Agriculture, Natural Resources, and Rural Development (MARNDR/DPV-PS) (National Program for Detection and Control of Fruit Flies). Unpublished Data.

CHAPTER V. SUMMARY AND CONCLUSION

5.1 Summary

Tephritid fruit flies trigger both direct and indirect costs that cause them to be the major threat to fruit production and trade in the tropical and subtropical region (Aluja 1994, IAEA 2003). For instance, since 2007, the mango industry in Haiti has severely suffered the attack of the Indian fly (*Anastrepha obliqua*, Macquart) and the Caribbean fly (*Anastrepha suspensa*, Loew) that have caused losses of tons of mangoes, closure of two mango processing plants (Pierreval 2012), and cost of implementing a management program (MARNDR 2008). The management strategies that aim at controlling these pests include biological control, insect sterile technique (IST), chemical control, and mass trapping methods. However, due to environmental and health concerns, the trend has been directed towards the latter, which entails high implementation costs. As a result, this research was conducted to develop an effective artisanal trap model, less expensive than the commercial traps for mass trapping.

A field trial conducted during the mango season of December 2014-April 2015 in Haiti revealed that inexpensive artisanal traps can be highly effective in mass trapping fruit flies. Indeed, analysis of variances (ANOVA) and Binomial test for proportions indicated that two newly made artisanal trap models (AT1 & AT2) and a commercial McPhail trap (MP), baited with the same attractant and tested in the same conditions, yielded similar results in the average number of fruit flies caught (8.9 ± 2.6 , 13 ± 2.9 , and 16 ± 4.1 respectively). Even though the total number of fruit flies was higher in the commercial trap (319 flies) compared to the others (AT1: 178 flies and AT2: 253 flies), the cost-efficacy ratio was higher in the artisanal trap models (AT1: 0.42 \$ per flies caught, AT2: 0.28 \$ per flies, and MP: 0.69 \$ per flies). This study confirmed the presence of both fruit fly species: *A. obliqua*, Macquart and *A. suspensa*, Loew ,

already detected by the ministry of agriculture of Haiti (MARNDR 2008); with a significantly higher abundance of *A. obliqua* ($\approx 100\%$) compared to *A. suspensa* ($< 1\%$). Moreover, this study confirmed the female-biased characteristics of McPhail traps. These findings demonstrated that the budget of a control program using mass trapping can be significantly reduced utilizing cost-effective artisanal traps using a lower dose of attractant, compared to commercial traps with a high dose of attractant.

A seven-week field experiment was conducted in the same mango field as the previous study, to compare the efficacy of the commercial McPhail trap (MP) to the most cost-effective artisanal model (AT2) under mass trapping conditions. After seven weeks of data collection where a density of 36 AT2/ha was compared to a density of 24 MP/ha, binomial test for equal proportions indicated that the two trap densities were not different in their number of fruit flies caught (AT2: 236 flies and MP: 239 flies). In addition, this study indicated that the capture rate in both trap densities followed the same trend line during the mango season. This study demonstrated that the artisanal trap model (AT2) is effective for mass trapping.

Regression analysis performed on data collected from six different trap densities tested suggested that a density of 25 traps/ha was enough to protect the mango orchard during the growing phase to the maturation phase of the mango season. Even though the experiment concluded before the fruit ripening phase, data from the Ministry of Agriculture (MARNDR, 2013) showed that the fruit flies density reaches its peak at the end of the mango season (in April). As a result, the density of 25 artisanal traps/ha will need to be increased to protect the mango groves. These findings confirmed results from other research on trapping density (Martinez-Ferrer et al. 2012, IAEA 2003).

5.2 Conclusion

This research indicated that cost-effective artisanal trap models could be developed as a substitute for expensive commercial traps, to implement fruit fly control programs with mass trapping methods. In addition, the amount of attractant can be reduced for a significant improvement in control program cost/benefit. Therefore, due to the great variation in the fruit fly population density throughout different ecosystems and different seasons in Haiti, the trapping density should also be adjusted to provide sufficient protection to fruit groves (Matinez-Ferrer et al. 2012, IAEA 2003). Moreover, the integration of a chemical control method could yield promising results by using organic insecticides during dry seasons. Future research in this direction should be focused on developing and testing artisanal trap models similar to the AT2 Model, over a larger scale of time and space, to determine different trapping densities for artisanal trap models.

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APPENDIX A: TRAP MODELS EVALUATION AND COMPARISON, STATISTICAL ANALYSIS (ANOVA) (EXPERIMENT #1)

```

DM 'LOG; clear;output;clear;';
options ps=512 ls=105 nocenter nodate nonumber nolabel
      FORMCHAR="|----|+|----+=|-\<>*";
ODS listing;
ods graphics on;
ods html close;
ODS HTML style=minimal;
TITLE 'Completely Randomized Design';
TITLE 'Comparison between two Artisanal McPhail Traps(AT1 & AT2), and the
Conventional McPhail(MP)'
'(FF=Number of fruit fly)';
DATA;
INPUT trap$ rep FF;
Label FF = Number of Fruit Fly;
CARDS;
AT1  1  5
AT1  2  7
AT1  3 10
AT1  4 28
AT1  5 27
AT1  6 22
AT1  7 36
AT1  8  1
AT1  9  2
AT1 10 10
AT1 11  5
AT1 12  7
AT1 13  1
AT1 14  1
AT1 15  1
AT1 16  2
AT1 17  3
AT1 18  3
AT1 19  4
AT1 20  3
AT2  1  6
AT2  2  9
AT2  3 10
AT2  4 25
AT2  5 30
AT2  6 40
AT2  7 38
AT2  8  0
AT2  9  5
AT2 10  9
AT2 11  2
AT2 12  5
AT2 13  0
AT2 14  0
AT2 15  1
AT2 16  4
AT2 17 10
AT2 18 21

```

```

AT2 19 9
AT2 20 29
MP 1 5
MP 2 5
MP 3 22
MP 4 10
MP 5 25
MP 6 20
MP 7 78
MP 8 1
MP 9 4
MP 10 16
MP 11 6
MP 12 17
MP 13 5
MP 14 1
MP 15 3
MP 16 3
MP 17 12
MP 18 15
MP 19 23
MP 20 48
;

```

```

PROC PRINT;
proc means N mean var stderr;
  var FF;
  class trap;
run;
proc glm;
class trap;
model FF = trap;
means trap/lsd lines;
run;

```

Comparison between two Artisanal McPhail Traps (AT1 & AT2), and the Conventional McPhail (MP) (FF=Number of Fruit Fly)

The MEANS Procedure
Analysis Variable: FF

trap	N		Mean	Variance	Std Error
	Obs	N			
AT1	20	20	8.9000000	111.1473684	2.3574071
AT2	20	20	12.6500000	168.4500000	2.9021544
MP	20	20	15.9500000	342.0500000	4.1355169

Comparison between two Artisanal McPhail Traps (AT1 & AT2), and the Conventional McPhail (MP) (FF=Number of Fruit Fly)

The GLM Procedure

Class Level Information

Class	Levels	Values
trap	3	AT1 AT2 MP

Number of Observations Read	60
Number of Observations Used	60

Comparison between two Artisanal McPhail Traps (AT1 & AT2), and the Conventional McPhail (MP) (FF=Number of Fruit Fly)

The GLM Procedure
Dependent Variable: FF

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	2	497.70000	248.85000	1.20	0.3084
Error	57	11811.30000	207.21579		
Corrected Total	59	12309.00000			
R-Square	Coeff Var	Root MSE	FF Mean		
0.040434	115.1599	14.39499	12.50000		

Source	DF	Type I SS	Mean Square	F Value	Pr > F
trap	2	497.7000000	248.8500000	1.20	0.3084
Source	DF	Type III SS	Mean Square	F Value	Pr > F
trap	2	497.7000000	248.8500000	1.20	0.3084

Comparison between two Artisanal McPhail Traps (AT1 & AT2), and the Conventional McPhail (MP) (FF=Number of Fruit Fly)

The GLM Procedure
t Tests (LSD) for FF

NOTE: This test controls the Type I comparisonwise error rate, not the experimentwise error rate.

Alpha	0.05
Error Degrees of Freedom	57
Error Mean Square	207.2158
Critical Value of t	2.00247
Least Significant Difference	9.1154
Means with the same letter are not significantly different.	
t Grouping Mean	N trap
A	15.950 20 MP
A	12.650 20 AT2
A	8.900 20 AT1

APPENDIX B: BINOMIAL TEST FOR PROPORTIONS

BINOMIAL TEST FOR EQUAL PROPORTIONS

Comparing the total number of fruit flies caught in the trap models: AT1, AT2, and MP (Experiment # 1)

The hypothesis test that two numbers r_1 and r_2 have equal proportion of success $p = q = \frac{1}{2} = 0.5 = 50\%$. Thus they are not statistically different.

$$H_0: r_1 = r_2$$

$$H_a: r_1 \neq r_2$$

$Z_c = \frac{(r - np) / -0.5}{\sqrt{npq}}$, where r represents the numbers to compare (r_1 or r_2), n is the sum of these numbers, p and q are success for both numbers ($p = q = 1/2 = 50\%$).

Comparing AT1 vs AT2:

AT1 caught $r_1 = 178$ fruit flies and AT2 caught $r_2 = 253$ fruit flies.

so $n = 178 + 253 = 431$ and $np = 431 * 0.5 = 215.5$

$$Z_c = \frac{(178 - 215.5) / -0.5}{\sqrt{(431 * 0.5 * 0.5)}} = 3.56$$

with $\alpha = 0.05/3 = 0.017$ (Bonferroni's correction for 3 comparisons: AT1 vs MP, AT1 vs AT2, and AT2 vs MP)

$$Z_c = 3.56, \alpha = 0.017$$

Since we have a two-tailed test, the P-value is the probability that the z-score is less than -3.56 or greater than 3.56.

We use the Normal Distribution Calculator to find $P(z < -3.56)$ plus $P(z > 3.56)$.

Thus, the P-value $< 0.001 < 0.017$

So we reject H_0 , and conclude that the number of fruit flies caught in both trap models: AT1(178 flies) and AT2 (253) are significantly different.

Comparing AT1 vs MP:

AT1 caught $r_1 = 178$ fruit flies and MP caught $r_2 = 319$ fruit flies.

so $n = 178 + 319 = 497$ and $np = 497 * 0.5 = 248.5$

$$Z_c = \frac{178 - 248.5 / -0.5}{\sqrt{(497 * 0.5 * 0.5)}} = 6.28$$

$$Z_c = 6.28, \alpha = 0.017$$

We use the Normal Distribution Calculator to find $P(z < -6.28)$ plus $P(z > 6.28)$.

Thus, the P -value $\ll 0.001 < 0.017$

So we reject H_0 , and conclude that the number of fruit flies caught in both trap models: AT1(178 flies) and MP (319) are significantly different.

Comparing AT1 vs MP:

AT2 caught $r_1 = 253$ fruit flies and MP caught $r_2 = 319$ fruit flies.
so $n = 253 + 319 = 572$ and $np = 572 * 0.5 = 286$

$$Z_c = \frac{253 - 286 / -0.5}{\sqrt{(572 * 0.5 * 0.5)}} = 2.72$$

$$Z_c = 2.72, \alpha = 0.017$$

We use the Normal Distribution Calculator to find $P(z < -2.72)$ plus $P(z > 2.72)$.

Thus, the P -value = $0.0065 < 0.017$

So we reject H_0 , and conclude that the number of fruit flies caught in both trap models: AT2 (253 flies) and MP (319) are significantly different.

TEST FOR COMPARING TWO PROPORTIONS

Comparing the proportion of female fruit flies caught in the trap models: AT1, AT2, and MP (Experiment # 1)

The hypothesis test that the two binomial proportions are equal is

$$H_0: p_1 = p_2$$

$$H_a: p_1 \neq p_2$$

P_1 is the proportion of success in the event #1 (X_1, N_1)

P_2 is the proportion of success in the event #2 (X_2, N_2)

Test Statistic: $Z = \frac{P_1 - P_2}{\sqrt{P(1-P)(\frac{1}{N_1} + \frac{1}{N_2})}}$, with $\alpha = 0.05/3 = 0.017$ (Bonferoni's correction for 3

comparisons: AT1 vs MP, AT1 vs AT2, and AT2 vs MP)

where P is the proportion of successes for the combined sample and

$$P = \frac{X_1 + X_2}{N_1 + N_2}$$

Comparing AT1 vs MP:

AT1: 133 females out of 178 fruit flies (45 males), $P_1 = 133/178 = 0.744$

MP: 202 females and 117 males for a total of 319 fruit flies, $P_1 = 202/319 = 0.627$

$$P = (133 + 202) / (178 + 319) = 0.674$$

$$Z = (0.744 - 0.627) / \sqrt{(0.674)(1 - 0.674)\left(\frac{1}{178} + \frac{1}{319}\right)} = 2.665$$

$$Z = 2.2665, \alpha = 0.017$$

Since we have a two-tailed test, the P-value is the probability that the z-score is less than -2.2665 or greater than 2.2665.

We use the Normal Distribution Calculator to find $P(z < -2.2665)$ plus $P(z > 2.2665)$.

Thus, the P-value = 0.0008 < 0.017

So we reject H_0 , and conclude that the proportion of female flies in both trap models: AT1 and MP is significantly different.

Comparing AT1 vs AT2:

AT1: 133 females out of 178 fruit flies (45 males), $P_1 = 133/178 = 0.744$

AT2: 172 females and 81 males for a total of 253 fruit flies, $P_1 = 172/253 = 0.631$

$$P = (133 + 172) / (178 + 253) = 0.708$$

$$Z = (0.744 - 0.631) / \sqrt{(0.708)(1 - 0.708)\left(\frac{1}{178} + \frac{1}{253}\right)} = 2.54$$

$$Z = 2.54, \alpha = 0.017, P_{\text{-value}} = 0.011 < 0.017$$

So we reject H_0 , and conclude that the proportion of female flies in both trap models: AT1 and AT2 is significantly different

Comparing AT2 vs MP:

AT2: 172 females and 81 males for a total of 253 fruit flies, $P_1 = 172/253 = 0.631$

MP: 202 females and 117 males for a total of 319 fruit flies, $P_1 = 202/319 = 0.627$

$$P = (172+202)/(253+319) = 0.654$$

$$Z = (0.631 - 0.627) / \sqrt{(0.654)(1 - 0.654)\left(\frac{1}{253} + \frac{1}{319}\right)} = 0.099$$

$$Z = 0.099, \alpha = 0.017, P\text{-value} = 0.921 > 0.017$$

So we failed to reject H_0 , and conclude that the proportion of female fruit flies in both trap models: AT2 and MP is not significantly different

APPENDIX C: BINOMIAL TEST FOR EQUAL PROPORTIONS

Comparison of a density 24 MP/ha to a density of 36AT2/ha in total number of fruit flies caught (Experiment # 2)

The hypothesis test that two numbers r_1 and r_2 have equal proportion of success $p = q = \frac{1}{2} = 0.5 = 50\%$. Thus they are not statistically different.

$$H_0: r_1 = r_2$$

$$H_a: r_1 \neq r_2$$

$Z_c = \frac{(r - np) / -0.5}{\sqrt{(npq)}}$, where r represents the numbers to compare (r_1 or r_2), n is the sum of these numbers, p and q are success for both numbers ($p = q = 1/2 = 50\%$).

Comparing 24 MP/ha vs 36 AT2/ha:

36AT2/ha caught $r_1 = 236$ fruit flies and 24MP/ha caught $r_2 = 239$ fruit flies.
so $n = 236 + 239 = 475$ and $np = 475 * 0.5 = 237.5$

$$Z_c = \frac{(236 - 237.5) / -0.5}{\sqrt{(475 * 0.5 * 0.5)}} = 0.13, \text{ with } \alpha = 0.05$$

Since we have a two-tailed test, the P-value is the probability that the z-score is less than -0.13 or greater than 0.13. We use the Normal Distribution Calculator to find $P(z < -0.13)$ plus $P(z > 0.13)$.

Thus, the P-value $< 0.734 > 0.05$

So we failed to reject H_0 , and we conclude that the number of fruit flies caught in both trap densities: 24 MP/ha (239 flies) and 36 AT2/ha (236) are not significantly different.

APPENDIX D: NONLINEAR REGRESSION

Trap density (X) VS Number of Fruit flies caught (Y)

```
DM 'LOG; clear;output;clear;';
options ps=512 ls=105 nocenter nodate nonumber nolabel
      FORMCHAR="|----|+|----+=|-\<>*";
ODS listing;
ods graphics off;
ODS HTML style=minimal body='';

Title1 'Analysis for Fractyl Mertilus - Entomology';
TITLE2 'NonLinear Regression for Density (X) and Number of fruit fly
caught(Y) using the Artisanal McPhail Trap (AT2)';
```

```
DATA density;
INPUT X Y;
      LABEL X= 'Density' Y= 'Number';
CARDS;
4 0
4 8
4 3
8 12
8 3
8 71
12 4
12 53
12 68
16 31
16 72
16 100
24 44
24 133
24 38
36 157
36 12
36 57
;
proc nlin data=density;
  title3 'Nonlinear model - negative exponential';
  parms asymptote = 50 curve = -0.40 shift = -14;
  model Y = Asymptote * (1 - exp(curve * (X + Shift)));
run;
ods html close;
run;
quit;
```

```
Analysis for Fractyl Mertilus - Entomology
Linear Regression for Density (X) and Number of fruit fly(Y) caught using the
Artisanal McPhail Trap (AT2)
Nonlinear model - negative exponential
```

```
The NLIN Procedure
Dependent Variable Y
Method: Gauss-Newton
```

Iterative Phase				
Iter	asymptote	curve	shift	Sum of Squares
0	50.0000	-0.4000	-14.0000	22511025
1	70.4409	-0.3849	-10.9158	2780388
2	70.8460	-0.3471	-9.1546	418078
3	71.9620	-0.2734	-7.7822	80077.3
4	74.5261	-0.1778	-6.5876	31429.8
5	78.3047	-0.1212	-5.0731	25149.7
6	79.7452	-0.1119	-3.9115	24449.5
7	79.6085	-0.1120	-3.7469	24439.6
8	79.6108	-0.1119	-3.7450	24439.6

NOTE: Convergence criterion met.

Estimation Summary

Method	Gauss-Newton
Iterations	8
R	3.376E-6
PPC(curve)	0.000013
RPC(shift)	0.000493
Object	4.65E-8
Objective	24439.63
Observations Read	18
Observations Used	18
Observations Missing	0

Source	DF	Sum of Squares	Mean Square	F Value	Approx Pr > F
Model	2	11868.2	5934.1	3.64	0.0514
Error	15	24439.6	1629.3		
Corrected Total	17	36307.8			

Parameter	Estimate	Approx		
		Std Error	Approximate 95% Confidence Limits	
asymptote	79.6108	26.2964	23.5614	135.7
curve	-0.1119	0.1109	-0.3483	0.1244
shift	-3.7450	2.6720	-9.4403	1.9503

Approximate Correlation Matrix			
	asymptote	curve	shift
asymptote	1.0000000	0.8198238	0.2461422
curve	0.8198238	1.0000000	0.5291801
shift	0.2461422	0.5291801	1.0000000

VITA

The last born into an agrarian family in Bombardopolis, North West part of Haiti, Fractyl spent the early years of his life helping his family in agricultural activities while carrying out his academic duties. After finishing primary school in his home town, he moved to the capital city for his high school, and graduated in 2002. He received a Bachelor's degree in Agronomy from Université d'Etat d'Haïti (UEH) in 2008. His tremendous work during his undergraduate research on phytosanitary problems of cabbage developed his interest for Phytopathology and Entomology.

Fractyl worked at the division of plant protection (DPV/PS) of the Ministry of Agriculture of Haiti for 4 years and 5 months. He won a scholarship from USAID/USDA to pursue his Master's degree in the United States at Louisiana State University (LSU). In the context of these studies, he conducted his research project in Haiti from December 2014 to March 2015 under the guidance of Dr. Timothy Schowalter and Dr. Jorge Peña (from University of Florida), in order to start addressing some pest management problems his country is facing.

Fractyl's plan after his graduate studies is in perfect correlation with his belief that Haiti needs people with big heart, honesty, besides expertise to finally make it move forward. Indeed, he plans, as Agronomist and Entomologist, to bring meaningful contributions to agriculture in Haiti by staying active in both academic and extension level. Furthermore, he will continue involving in community development in his home town.