

HIDROGENIÔNICO (pH) OF THE ATTRACTANT, TRAP DENSITY AND CONTROL THRESHOLD FOR *Ceratitis capitata* (DIPTERA: TEPHRITIDAE) ON HAMLIN ORANGES IN SÃO PAULO CENTRAL REGION, BRAZIL¹

PAULO EDUARDO BRANCO PAIVA² & JOSÉ ROBERTO POSTALI PARRA²

ABSTRACT – This study evaluated the effect of initial pH values of 4.5, 6.5 and 8.5 of the attractant (protein bait) Milhocina® and borax (sodium borate) in the field, on the capture of fruit flies in McPhail traps, using 1, 2, 4 and 8 traps per hectare, in order to estimate control thresholds in a Hamlin orange grove in the central region of the state of São Paulo. The most abundant fruit fly species was *Ceratitis capitata*, comprising almost 99% of the fruit flies captured, of which 80% were females. The largest captures of *C. capitata* were found in traps baited with Milhocina® and borax at pH 8.5. Captures per trap for the four densities were similar, indicating that the population can be estimated with one trap per hectare in areas with high populations. It was found positive relationships between captures of *C. capitata* and the number of Hamlin oranges damaged, 2 and 3 weeks after capture. It was obtained equations that correlate captures and damage levels which can be used to estimate control thresholds. The average loss caused in Hamlin orange fruits by *C. capitata* was 2.5 tons per hectare or 7.5% of production.

Index terms: true fruit flies, protein bait, damage, IPM, Citros.

POTENCIAL HIDROGENIÔNICO (pH) DO ATRATIVO, DENSIDADE DE ARMADILHAS E NÍVEL DE CONTROLE PARA *Ceratitis capitata* (DIPTERA: TEPHRITIDAE) EM LARANJA HAMLIN NA REGIÃO CENTRAL DE SÃO PAULO, BRASIL

RESUMO – Esta pesquisa teve como objetivos: avaliar o efeito do pH inicial, 4.5; 6.5 e 8.5, do atrativo proteico Milhocina® e bórax (tetraborato de sódio) na captura de moscas-das-frutas em armadilhas McPhail; estudar densidades de armadilhas, 1; 2; 4 e 8 por hectare, para estimar níveis de controle em laranja cv. Hamlin, na região central de São Paulo. A espécie predominante, com 99% das moscas-das-frutas capturadas, foi *Ceratitis capitata*, sendo 80% de fêmeas. As maiores capturas de *C. capitata* ocorreram nas armadilhas com Milhocina® e bórax em pH 8.5. As capturas, nas 4 densidades, foram semelhantes, indicando que a população pode ser estimada com uma armadilha por hectare em áreas de altas populações. Houve relações positivas entre capturas de *C. capitata* e o número de frutos danificados, 2 e 3 semanas após a captura. Assim, foram obtidas equações que relacionam a captura e o dano, possibilitando estimar níveis de controle desse inseto. As perdas médias causadas por *C. capitata* em laranja cv. Hamlin chegaram a 2,5 toneladas de frutos por hectare ou 7,5% da produção.

Termos para indexação: moscas-das-frutas, dano, atrativo proteico, MIP, Cítrros.

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²Universidade de São Paulo, Escola Superior de Agricultura Luiz de Queiroz, Departamento de Entomologia e Acarologia, Avenida Pádua Dias, 11, CEP 13418-900 Piracicaba, SP, Brazil. E-mails: paulopaiva@usp.br, jrpparra@usp.br

INTRODUCTION

The Mediterranean fruit fly, *Ceratitidis capitata* (Wiedemann) (Diptera: Tephritidae), is one of the main fruit tree pests in the world. This species was recorded in Brazil at the end of the 19th Century and is presently found in most Brazilian states (ZUCCHI, 2001). It is estimated that one box (40.8 kg) of oranges per tree may be lost, in early maturing varieties; due to fruit flies (RAGA, 2005). Besides losses caused by the fall and subsequent rotting of fruits, there are sanitary restrictions to the export of fresh fruit to countries where these insects do not occur. Apart from *C. capitata*, citrus fruits in Brazil are also attacked by *Anastrepha fraterculus* (Wiedemann), *Anastrepha obliqua* (Macquart) (Diptera: Tephritidae) and flies of the Lonchaeidae family (MALAVASI; MORGANTE, 1980; SOUZA FILHO et al., 2000). In São Paulo, large populations of *C. capitata* develops in mature coffee fruits during the autumn-winter period, and these insects may move to citrus and other groves, where they can cause serious damage (SOUZA FILHO et al., 2000; ZUCCHI, 2001; MONTES et al., 2011).

Economically important fruit flies have been monitored using traps with food attractants (GARCIA; CORSEUIL, 1998; NASCIMENTO et al., 2000; SCOZ et al., 2006) or other components, such as trimedlure for *C. capitata* (GROUT et al., 2011). McPhail traps with hydrolyzed proteins, or similar products, have been the most used, despite being considered expensive, difficult to use and inefficient (ALUJA, 1994), because they are able to capture different tephritid species. Characteristics of the attractant suspension, such as attractant concentration and quality, pH, addition of preservatives, production of ammonia and fermentation time, have influenced the capture efficiency of fruit flies (BATEMAN; MORTON, 1981; MAZOR et al., 1987; EPSKY et al., 1994; DUYCK et al., 2004; HEATH et al., 2009).

Pest population monitoring and utilization of control thresholds have become major components in integrated pest management programs (NORRIS et al., 2003). However, the best trap density for monitoring these insects in citrus is unknown and may depend on the fruit fly population. Although there are some indications on control thresholds for fruit flies (NASCIMENTO et al., 2000), little research has been done on the relationship between insect trap captures and the damage caused (AGUNLOYE, 1987; UCHÔA-FERNANDES et al., 2003; GROUT et al., 2011). The objectives of this research were to evaluate the effect of pH of the attractant suspension and also estimate the control threshold for *C. capitata*

at four trap densities, based on captures and fruit damage in Hamlin variety (sweet orange).

MATERIAL AND METHODS

The experiment was conducted in a Hamlin grove (*C. sinensis*), an early maturing variety, in Itaju, central region of the state of São Paulo, Brazil, from June to August, 2002. The attractant used was a corn steep water, with 25% protein content, Milhocina® (Corn Products, Mogi-Guaçu, Brasil), which is an acid hydrolyzed corn protein product. Sodium tetraborate decahydrate (borax) was added to this at 5% (weight/volume) to preserve the attractant and the captured insects (LOPEZ; HERNANDEZ BECERRIL, 1967). The pH of the mixture was adjusted to 8.5 with a solution of sodium hydroxide. A portable pH meter (Checkmite pH-10, Corning, NY) was used for verification. McPhail traps, Mata-Sete® model (Isca Tec, Ijuí, Brazil), 13.5 cm tall, with a yellow 17 cm diameter base and a transparent upper portion, were used.

The attractant was diluted in water to 5% (volume/volume), and 250 ml of the attractant suspension were placed in each trap. The traps were installed inside the orange trees in the shade at an approximate height of 1.5 m from the soil. Any branches beneath the trap that could stop or make the entrance of the insects more difficult were removed. The captured insects were separated by pouring the liquid trap contents onto a sieve, where they were retained. The sieve was then placed in a recipient with water and the insects were identified when they floated. The fruit flies were counted and *C. capitata* distinguished by sex, based on the male antennae and female ovipositor (ZUCCHI, 2001). *Anastrepha* species were identified by M. F. Souza Filho and Lonchaeidae by P. Strikis.

Effect of pH of the attractant on *C. capitata* capture. The experiment was conducted in a sweet orange grove (*C. sinensis*), with an early maturing Hamlin variety, in Itaju, central region of the state of São Paulo, Brazil, in June, 2002. The grove occupied an area of 10 hectares, with 5-year old trees, without fruits, and was harvested a week before the experiment began. The trees, spaced 7.5 by 3 m, were 2 to 2.5 m high. The experimental design was completely random with four attractants (treatments), (1) Milhocina® in pH 4.5, (2) Milhocina®+borax in pH 4.5, (3) Milhocina®+borax in pH 6.5 and (4) Milhocina®+borax in pH 8.5, with six repetitions. The attractant was diluted to 5% and placed in the traps. These traps were randomly

distributed, maintaining 30 m distance between them. Evaluations were made 2, 4, 6 and 8 days after installation, by counting captured fruit flies and measuring the pH. The traps were again randomized after each evaluation.

Effect of trap density on *C. capitata* capture and the relationship with damaged Hamlin orange fruits. The experiment was conducted in a Hamlin grove (*C. sinensis*), in Itaju, state of São Paulo, Brazil, between June and August, 2002, when there were large *C. capitata* populations, probably originating from coffee (*Coffea arabica* L.). The experimental area consisted of two neighboring 14 hectare plots, with 5-year old trees, 2 to 2.5 m high, within a continuous 700 ha area of citrus. Fruits were yellowish, approximately 9 months old (originating from flowering in October 2001), and were mature at the beginning of the experiment and ready for harvesting. The inter-tree spacing was 7.5 by 3 m, with a density of 444 trees per hectare.

The treatments were one, two, four and eight traps per hectare, with 5 repetitions. The experimental design was completely random, with plots of 100 by 100 meters. The traps were distributed equidistantly from each other inside the plot, with at least 25 m between them, one trap per tree. The traps were installed in the morning and the fruit fly evaluation was realized in the following morning, 24 hours later, since a preliminary evaluation of 7 days of captures had observed insect decomposition due to the excess number of insects collected. The trap contents were sieved and the insects identified, with fruit flies being separated by sex. The traps were kept on the trees without attractant suspension until the next evaluation, one week later.

Damage was evaluated by tagging nine trees at random within each plot and counting the number of damaged fruits by fruit fly, on the tree and on the ground. At the beginning of the experiment, all damaged fruits were removed from the tagged trees. At each evaluation, the damaged fruits were removed from the tagging trees, so they would not be recounted at the next evaluation.

Linear regressions, considering fruit damage as dependent variable, and the capture, one, two and three weeks later, was calculated at the four trap densities. The treatments were compared by analyses of variances (F test). All statistical analyses were conducted on SAS (SAS Institute, 2000).

RESULTS AND DISCUSSION

Effect of pH of attractant on *C. capitata* capture. Only *C. capitata* was captured in this experiment. The initial pH of the attractant suspension of Milhocina®+borax influenced the capture of *C. capitata* flies in a Hamlin orange grove with no fruits ($F = 77.26$, d.f. = 2; 70, $P < 0.01$) (Figure 1).

The mean number of *C. capitata* captured in two days in the most efficient suspension (pH 8.5) was 42.6 flies per trap, six times higher than that observed for the Milhocina®+borax suspension of pH 4.5, that is, 7.1 flies per trap. Similar results were observed by Bateman and Morton (1981) for *Bractrocera tryoni* (Froggatt) and the attractant NBC, where the mean capture was nine times higher for pH 8.5 than for the standard attractant with a pH of 4.8. With the NuLure attractant, the highest captures of female *C. capitata* also occurred at a pH of 8.5 (EPSKY et al., 1994).

There was no difference between captures in traps with an attraction suspension pH of 4.5, with and without borax ($F = 1.78$, d.f. = 1; 46, $P = 0.19$). Thus, the addition of sodium tetraborate decahydrate (borax) to the 5% attractant concentrate did not influence the capture of *C. capitata* with an acid pH of 4.5. The addition of 1, 2 and 3 % borax to the protein attractant PIB-7 (pH of 8.5 to 9) reduced the capture of *Anastrepha ludens* (Loew) (LOPEZ; HERNANDEZ; BECERRIL, 1967). Substances which inhibit microorganism development can reduce ammonia production (BATEMAN; MORTON, 1981), and, consequently, reduce the capture of tephritids.

The addition of 1% borax to the protein attractant PIB-7 allowed the stabilization of the suspension pH to 8.7 for up to 7 days, suggesting that attractant decomposition was inhibited (LOPEZ; HERNANDEZ BECERRIL, 1967). Epsky et al. (1994) verified that the number of female *Anastrepha suspensa* (Loew) fell with the addition of 3, 5 and 10% borax, and there was a stabilization of the solution pH at these concentrations. The present study used 5% borax in the attractant, which diluted to 5% in water resulted in a final borax concentration of 0.25% and this concentration was unable to avoid pH variations in the attractant suspensions and affect *C. capitata* capture, at pH of 4.5.

Effect of trap density on *C. capitata* capture in Hamlin variety oranges. A total of 51,532 fruit flies were collected, of which 50,956 were *C. capitata*, 304 tephritids of the genera *Anastrepha* (*A. obliqua*, *Anastrepha serpentina*

(Wiedemann) and *Anastrepha sororcula* Zucchi) and 272 were loncheids of the genera *Neosilba* [(*Neosilba zadolicha* McAlpine and Steyskal, *Neosilba pendula* (Bezzi) and *Neosilba* sp.)]. The mean percentage of females in the captures was 92% for *Neosilba*, 80% for *C. capitata* and 58% for *Anastrepha*.

In general, *C. capitata* captures per trap were unaffected by trap density (Figure 2). Thus, independent of the trap density of one, two, four or eight traps per hectare, the number of *C. capitata* captured per trap did not differ statistically between the different trap densities for the six evaluations.

Although there were no differences between *C. capitata* captures for each evaluation, when the captures for all the evaluations were considered, there was a significant difference for the total number of flies captured ($F=3.71$, d.f. = 3; 446, $P=0.012$) and females ($F=4.22$, d.f. = 3; 446, $P<0.01$), but not for males ($F=1.64$, d.f. = 3; 446, $P=0.18$). Considering the six evaluations, the highest individual trap captures were observed for the smallest trap density studied, one trap per hectare.

Relationship between *C. capitata* capture and damage in Hamlin oranges. Since the predominant species was *C. capitata*, corresponding to almost 99% of fruit flies captured, linear regressions were estimated between this insect capture and damage to ripe oranges. There was a positive correlation between *C. capitata* capture and the damage caused by these insects in the second and third week after capture (Table 1, Figure 3). There was no correlation between *C. capitata* captures and the damage in the same week to capture and in the first week.

Contrary to the result obtained in the present study, Agunloye (1987) concluded there was no relationship between the number of *C. capitata* captured in traps (with honey and sugar) and the number of damaged oranges, in plots of 4 trees. Uchôa-Fernandes et al. (2003) also observed no correlation ($p<0.01$) between *Neosilba* captured in traps containing protein attractant in orange groves and the number of larvae and adults of these flies in attacked fruits. Agunloye (1987) used small plots and Uchôa-Fernandes et al. (2003) found a positive correlation of *Neosilba* sp. captured in traps and adults emerged in oranges ($p=0.036$).

Since it is the female fruit flies which damage the fruits by ovipositing, the correlation between female trap captures and damaged fruits seemed to be more probable than the capture of both males and females. However, although females were the majority in the captures (80%), there was also a

correlation with the capture of males and females together. Therefore, separating insects by sex when monitoring *C. capitata* is unnecessary.

As the efficacy of fruit fly monitoring with food attractants (NASCIMENTO; CARVALHO, 2000) and capture have been considered low (ALUJA, 1994), trap captures would not represent the population of the monitoring area. Since there was a correlation between *C. capitata* capture and damage caused to Hamlin fruits, there should also have a correlation between the resident population of the area and the damage. Therefore, it may be inferred from the results obtained that a relationship may exist between the resident population of *C. capitata* and trap captures.

Using the linear equation obtained (Figure 3), for a capture of 50 *C. capitata* per trap per day, Hamlin oranges damaged is estimated in 230 kg/hectare. However, before a general recommendation can be made, control threshold should be tested and validated in different groves. At high population levels, the first capture may already be higher than this control threshold. However, if the females are in the pre-oviposition stage there will be enough time to begin control and avoid damage.

Damage caused by *C. capitata* in Hamlin oranges. With an accumulated mean damage of 306.7 fruits on 9 trees over 6 weeks (density of 444 orange trees per hectare and fruit weight of 165 g), a mean loss was estimated in 2,495 Kg of fruits per hectare. Losses within plots varied between 1,354 and 4,458 Kg of fruits per hectare (Figure 4).

Mean losses of 2.5 tons of fruits per hectare were estimated or 7.5% of the total production. This value is lower than that estimated by Raga (2005), and occurred under these experimental conditions due to three factors: a high *C. capitata* population, an advanced stage of fruit maturation, about two months after the harvest time, and no chemical control of the insect. However, in large areas of oranges, harvesting delays are common and the level of losses observed in the present study may occur.

TABLE 1 - Values F and P for linear regressions between mean number of *Ceratitits capitata* captured per McPhail trap and the mean number of damaged fruits in 9 trees at the same week to capture (week 0), one (week 1), two and three weeks (week 3) after capture.

capture x damage	F	P	d.f. ¹	linear equation	r ² coefficient
damage week 0	1.85	0.18	1;118	-	-
damage week 1	0.03	0.84	1;98	-	-
damage week 2	7.96	<0.01	1;78	Y = 0.12X + 38.14	0.10
damage week 3	49.91	<0.01	1;58	Y = 0.26X + 25.98	0.35

¹: degrees of freedom for regression and residue.

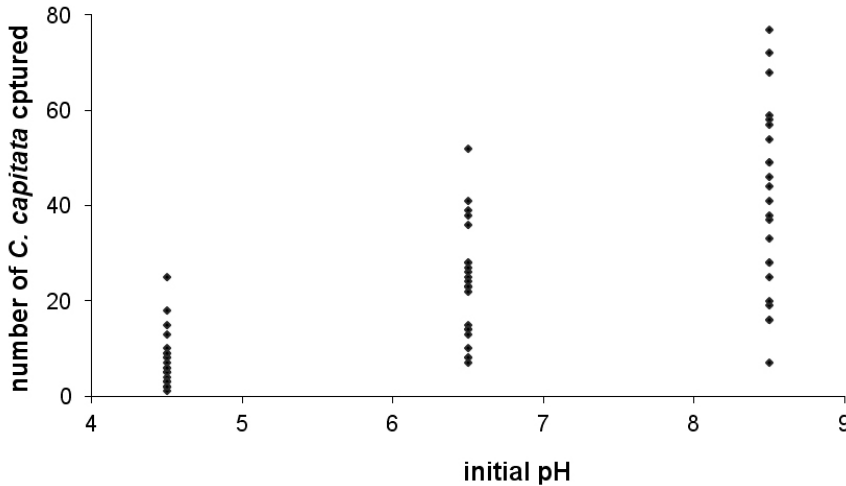


FIGURE 1- Effect of initial pH of attractant suspension of Milhocina® and borax on *Ceratitits capitata* capture, in a Hamlin orange grove without fruits. Itaju / SP.

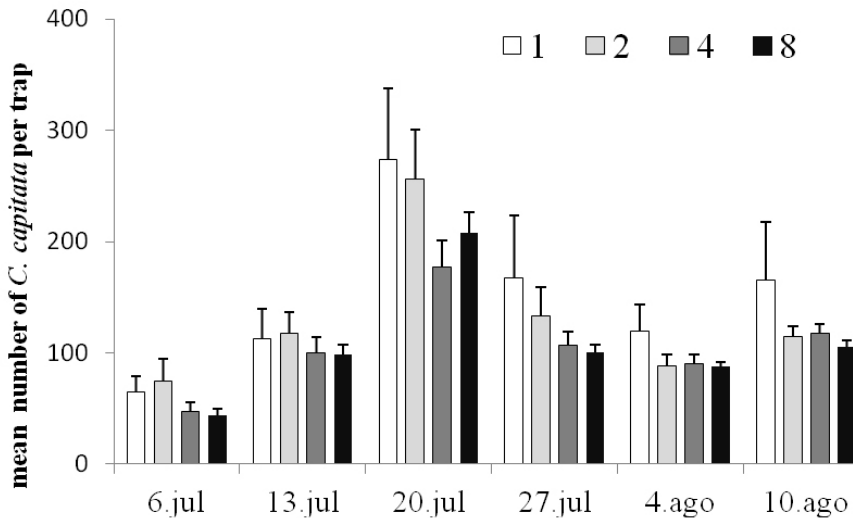


FIGURE 2- Mean number of *Ceratitits capitata* captured (standard error of mean) in one day, in a Hamlin orange grove with fruits, at 1, 2, 4 and 8 traps per hectare. Itaju / SP.

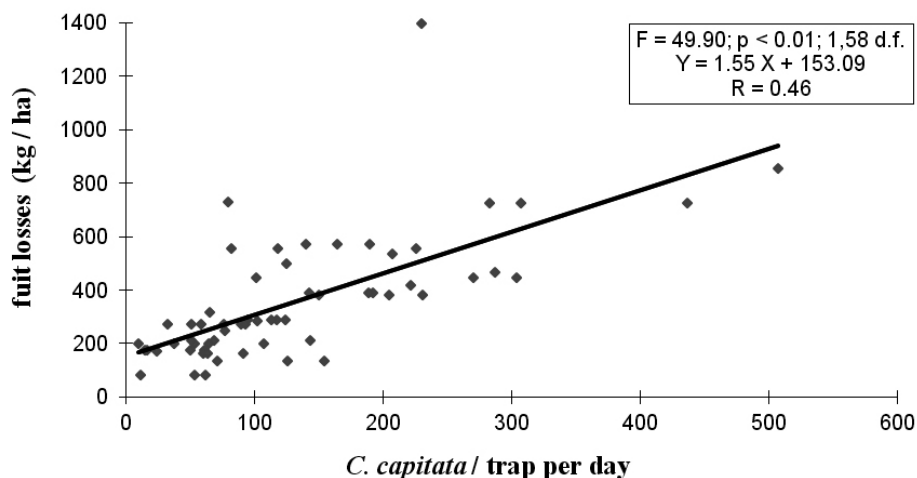


FIGURE 3 - Positive correlation between mean numbers of *Ceratitis capitata* captured per day and the orange fruit losses 3 weeks later. Itaju / SP, 2002.

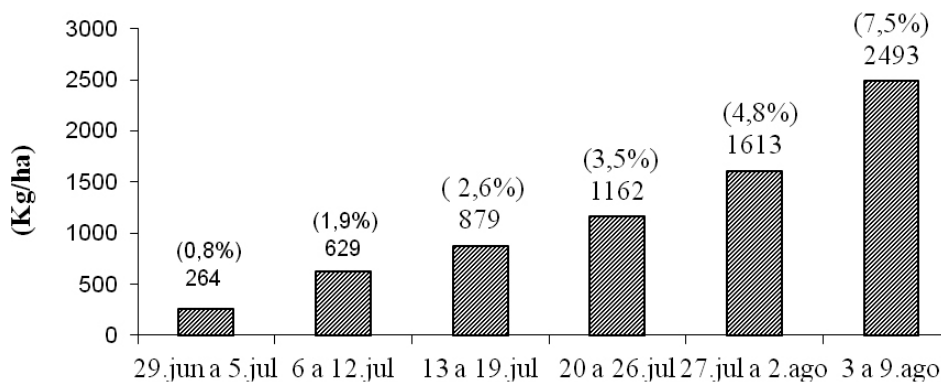


FIGURE 4- Accumulated mean damage caused by *Ceratitis capitata* to mature fruits of Hamlin oranges (percentage of losses in relation to total harvested). Itaju / SP.

CONCLUSIONS

1- The highest captures of *Ceratitis capitata* in McPhail traps occur with Milhocina® and borax adjusted to a pH of 8.5.

2- The mean number of *C. capitata* individuals captured is similar for the different trap densities, and this insect can be estimated at a trap density of 1 per hectare in areas with high insect populations.

3- There are positive correlations between the number of captured *C. capitata* and the number of damaged Hamlin oranges, two and three weeks after, allowing the estimation of control thresholds.

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