

Comparative efficacy of mass trapping and attract-and-kill technique in the control of medfly (*Ceratitis capitata*, Wiedemann) in Central Moroccan peach orchards

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SUMMARY

To develop eco-friendly alternative control strategies for medfly, mass trapping trials were conducted in the central Morocco during the crop season 2016. Two control methods, mass trapping and the attract-and-kill technique were compared for the control of medfly on two peach varieties ('Rome Star' and 'Ryan Sun') in Sefou district. For mass trapping, 62 traps/1.23 ha of MagnetTMMed type baited with ammonium acetate, trimethylamine, putrescine and 0.01 g of deltamethrin were installed on both varieties. In plots testing the attract-and-kill technique, treatments with malathion in mixture with protein hydrolyzate were applied to straw tufts attached to branches of the same varieties whenever 1 fly was caught on a Trimedlure trap installed at the center of each plot. The results showed that the number of flies captured by mass trapping reached 508 and 489 adults on 'Rome Star' and 'Ryan Sun', respectively. The average number varied from 1 to 3 adults/trap/day, depending on the date of capture and the variety, and females accounted for 62-100% of total caught flies. In plots protected by the attract-and-kill technique, and taking into account the threshold adopted by the farm, 11 and 15 treatments were carried out respectively on the 'Ryan Sun' and 'Rome Star' varieties. Overall, infestation rates in plots did not exceed 0.3% before or at harvest with mass trapping versus 0.9% with the attract-and-kill technique. On fruit dropped on the ground, the infestation rate did not exceed 4% in mass trapping, compared to 11.5% in the chemically treated plots. Mass trapping was therefore proved to be an effective and eco-friendly tool for managing medfly on peach fruit.

Keywords: *Ceratitis capitata*; Peach; Mass trapping; Attract-and-kill; Morocco

INTRODUCTION

The medfly, *Ceratitis capitata*, is considered to be one of the most important horticultural pests worldwide due to its high invasive and adaptive capacities (Fimiani, 1989; Carey, 1991; Malacrida et al., 2007). It is a polyphagous species, and more than 250 host plants are exploited by the fly (Christenson & Foote, 1960; Liquido et al., 1990; Tremblay, 1994). Damage caused by the medfly severely affects yields, which is a real problem for fruit exports to fruit fly-free countries (White & Elson-Harris, 1992). Damage caused by this pest can be enormous without appropriate control measures, and reported losses reach 30%-68% and 71% on citrus and khaki, respectively (Primo Millo, 2004; Khalaf et al., 2011). On stone fruits, medfly can lead to a total destruction of production (Thomas et al., 2001). Chemical control of this pest remains very widespread in orchards despite its side effects on human health and the environment. In addition, resistance of this pest to commonly used insecticides, especially malathion, has already been reported (Magaña et al., 2007). Faced with this serious problem, several research studies have emphasized a need for developing alternative control methods relating to the use of insecticides. Some of these methods have proven to be practical, effective and less hazardous to the environment and human health. This is the case, for example, with chemosterilisation (Mazih et al., 2008; Navarro-Llopis et al., 2010), kaolin use (Lo Verde et al., 2011; D'Aquino et al., 2011), and mass trapping (Epsky et al., 1999; Broughton & De Lima, 2002; Navarro-Llopis et al., 2008; Martinez-Ferrer et al., 2012; García-Mari & Alonso Muñoz, 2004; Hafsi et al., 2015; Mediouni Ben Jemâa et al., 2010; Cabrita & Ribeiro, 2006; Eltazi et al., 2008; Boulahia-Kheder et al., 2015). Other methods have been developed for post-harvest elimination of medfly infestation. Such disinfestation methods include gamma radiation (Moy et al., 1983) and fumigation with methyl-bromide (Tebbetts et al., 1983). In Morocco, the fight

against *C. capitata* has evolved through several stages, ranging from generalized chemical control to bait spraying, which consists of spraying straw tufts with a mixture of an insecticide and a food bait, and to mass trapping control (Benziane et al., 2003). Concerning mass trapping control of *C. capitata*, this technique has expanded rapidly due to improving attractiveness and selectivity, and areas protected by this method of control are constantly expanding (Heath et al., 1997; Epsky et al., 1999; Miranda et al., 2001; Alemany et al., 2004). Therefore, the main objectives of our work were to evaluate the effectiveness of mass trapping by the MagnetTMMed trap as a stand-alone tool for combating medfly versus the attract-and-kill technique on two seasonal peach varieties in the region of Sefrou, Morocco. To the best of our knowledge, this work is the first investigation of mass trapping of medfly with MagnetTMMed traps on rosaceous fruit trees in Morocco.

MATERIAL AND METHODS

Experimental orchards

In this work, we determined trap attractiveness based on medfly gender and compared the counts to data from plots representing the attract-and-kill technique. In order to compare the effectiveness of each control method, infestation rates generated by medflies were determined on trees at the harvesting period and on fruits discarded in the orchard during harvest. These percentage rates were then compared to the acceptable level of crop loss adopted by the farm, which is around 1% of harvested fruit. The trials were conducted in two plots of peach trees, *Prunus persica* (L.) (var. 'Rome Star' and 'Ryan Sun'), located in the Sefrou region in Central Morocco during the crop season 2016. The orchards were 11 years old with a density of 666 trees/ha. The characteristics of the experimental plots are listed in Table 1.

Table 1: Characteristics of experimental design in peach plots in Sefrou region during 2016 growing season

Varieties	Trials	Geographical coordinates	Plot size (in ha)	Number of traps/ha		Starting dates of harvest
				Magnet Med	Trimedlure	
Rome Star	MT*	33°53'44.7"N	1.23	50	1	July 31
	A K	4°40'49.5"W	1.23	0	1	
Ryan Sun	PM	33°53'35.8"N	1.23	50	1	August 30
	A K	4°40'59.3"W	1.23	0	1	

*MT: Mass trapping; AK: Attract-and-kill

Assessments

In order to evaluate the effectiveness of mass trapping of medfly, plots of each peach variety were compared with those protected by the attract-and-kill technique adopted by the farm. The latter method of control consists of spraying a mixture of malathion insecticide and food attractant on tufts of straw (167 units/ha) previously installed on trees, once the threshold of 1 fly per trap is exceeded. Details of the experimental design in peach plots are given in Table 1.

Mass trapping by MagnetTM Med

The MagnetTM Med trap (Suterra-USA) is composed of 6.19 g of ammonium acetate, 2.81 g of trimethylamine, 0.05 g of putrescine (1,4-diaminobutane), and 0.01 g of deltamethrin. Its persistence activity is around 6 months and it is marketed in Morocco by the company Koppert. The recommended number of units per ha is 50. The traps were installed 9 and 18 days before harvest on the varieties ‘Rome Star’ and ‘RyanSun’, respectively. Each trap was set on a branch on the southwest side of each tree at 1.5 m height from the ground. The traps were homogeneously dispersed in the plots, each trap covering an area of 200 m². To count the visiting adults of *C. capitata* per trap, the faces of traps were coated with a glue. All traps were checked once or twice a week. The captured adults from each trap were separated based on their gender and detached from the glued trap face.

Monitoring trap for medfly populations

To monitor the medfly flight dynamics, “Maghreb-Med” pheromone traps containing each a capsule of Trimedlure (2 g) with dichlorvos were installed at the center of each experimental plot. This trap was also used as an indicator for insecticide treatments in plots protected by the attract-and-kill technique. The traps were suspended from branches on the southwest side of trees, in the shade, and at 1.5 m height from the ground. To maintain their maximum effectiveness, the capsules were changed every 4 weeks. Pheromone traps were also checked 3 to 4 times a week. The flies captured by each trap were counted based on their gender (male or female) in all experimental plots.

Attract-and-kill technique

Tufts of straw were suspended from tree branches in the test plots, away from fruits, one unit per 4 trees. The tufts were used for the application of insecticide solution baited with protein hydrolyzate. The chemical treatment was carried out wherever the threshold of one fly per

pheromone trap was exceeded. Such threshold was adopted for medfly control in stone fruit orchards at the farm level. Chemical treatments consisted of a mixture of food attractant (Blouz 30% protein hydrolyzate) and malathion (500g/l) as an insecticide. The doses applied were 0.2 l of malathion and 1.5 kg of protein hydrolysate diluted in 100 l of water; the volume of the mixture applied to straw tufts was 100 l/ha. Chemical treatment was conducted using a MATABI type lever-operated knapsack sprayer with 16 l capacity at a pressure of 2.5 bars.

Assessment of infestation rates

The rate of damage caused by the medfly was evaluated by calculating fruit infestation rates, corresponding to the ratio between the number of insect-infested fruits and total number of fruits examined, multiplied by 100. The pre-harvest medfly damage control was carried out once in 4 days by examining 20 fruits/tree out of a total of 50 trees randomly selected. At the harvest of the ‘Rome Star’ peach variety, 12335 fruits in total were checked in harvesting boxes on 6 different dates regardless of the technique/method used to control *C. capitata* in the orchards. On the ‘Ryan Sun’ variety, 350 fruits were checked at each harvest time. At the same time, 6 checks of non-compliant fruit discarded in the orchard were carried out at harvest by examining 100-200 fruits/control, i.e. a total of 1000 fruits per variety.

Temperature monitoring

Knowing that the numerical fluctuations of pests depend on thermal conditions (e.g. Nyamukondiwa et al., 2013), temperatures were recorded using a thermohygrograph placed in the experimental orchard throughout the study period.

Data analysis

The numbers of medfly adults captured by MagnetTMMed traps were compared by an analysis of variance in the statistical software Statistica ver. 7 and Microsoft Excel 2010 using the assessment dates as a factor. The means were separated by Tukey’s HSD multiple-range test at $P \leq 0.05$ when the samples had the same sizes or by Bonferroni’s multiple-range test at $P \leq 0.05$ when the samples had different sizes. Catches in the surveillance sexual traps representing mass trapping and attract-and-kill technique were compared by using Student’s t-test at 5%. Statistical analyses were done on raw data for catches or transformed into arcsin SQRT (%) in the proportion cases (Sokal & Rohlf, 1995).

RESULTS

Mass trapping by Magnet™Med

The flight dynamics of *C. capitata* monitored by Magnet™Med traps on peach trees in the Sefrou region (Morocco) during the study period is presented with

temperature conditions in Figure 1. The number of captured adults varied depending on peach variety, date of trapping, and environmental conditions such as temperature.

On the ‘Rome Star’ variety, 508 adults were recorded on traps about one week before the harvest started. Catches recorded on August 14 and 18 are statistically comparable, but significantly higher than those recorded

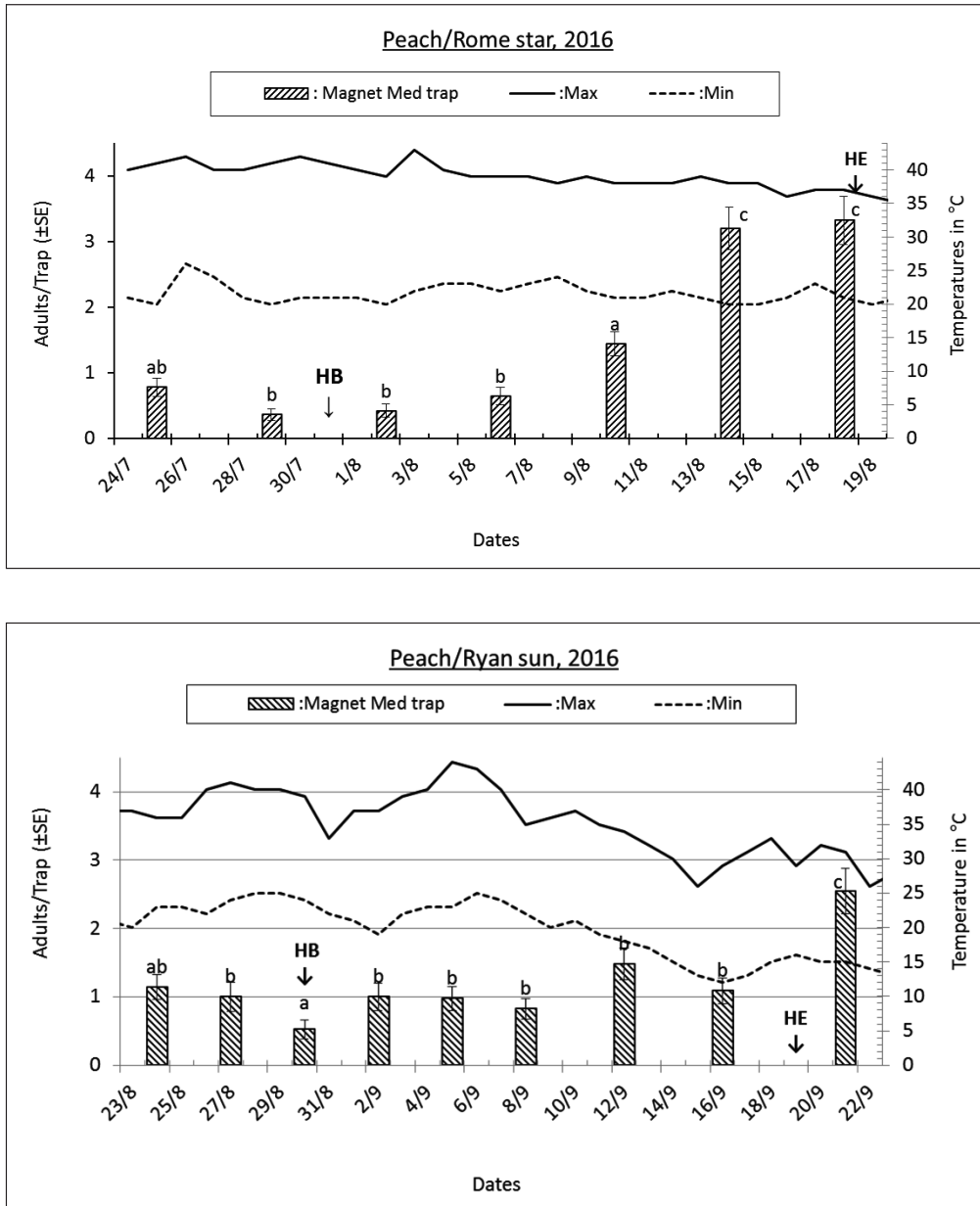


Figure 1: Flight dynamics of *Ceratitis capitata* adults captured by Magnet™Med traps in peach orchard in Sefrou region, Morocco, 2016 [Histograms bearing the same letter are not statistically different (ANOVA, followed by Tukey’s HSD multiple-range test at $P \leq 0.05$)]; HB: Harvest beginning; HE: harvest end; number of traps used = 50/ha)

on the other dates ($F_{6, 343}=34.94, P \leq 0.05$). During this period, temperature conditions were favorable for medfly activity (Figure 1).

On the ‘Ryan-Sun’ variety, catches reached a total of 489 flies over the assessment period. Their densities depended on the date of catching ($F_{8, 405}=7.51, P \leq 0.05$). The highest number of trapped adults occurred at the beginning of the

second half of August. The temperatures read were also suitable for *C. capitata* on that variety, despite a marked temperature decrease in September (Figure 1).

On both varieties, insect flight continued until the end of harvest. With respect to the overall number of adults caught throughout the trapping period, there was no significant difference between the two varieties ($F_{1,95}=0.25, P=0.62$).

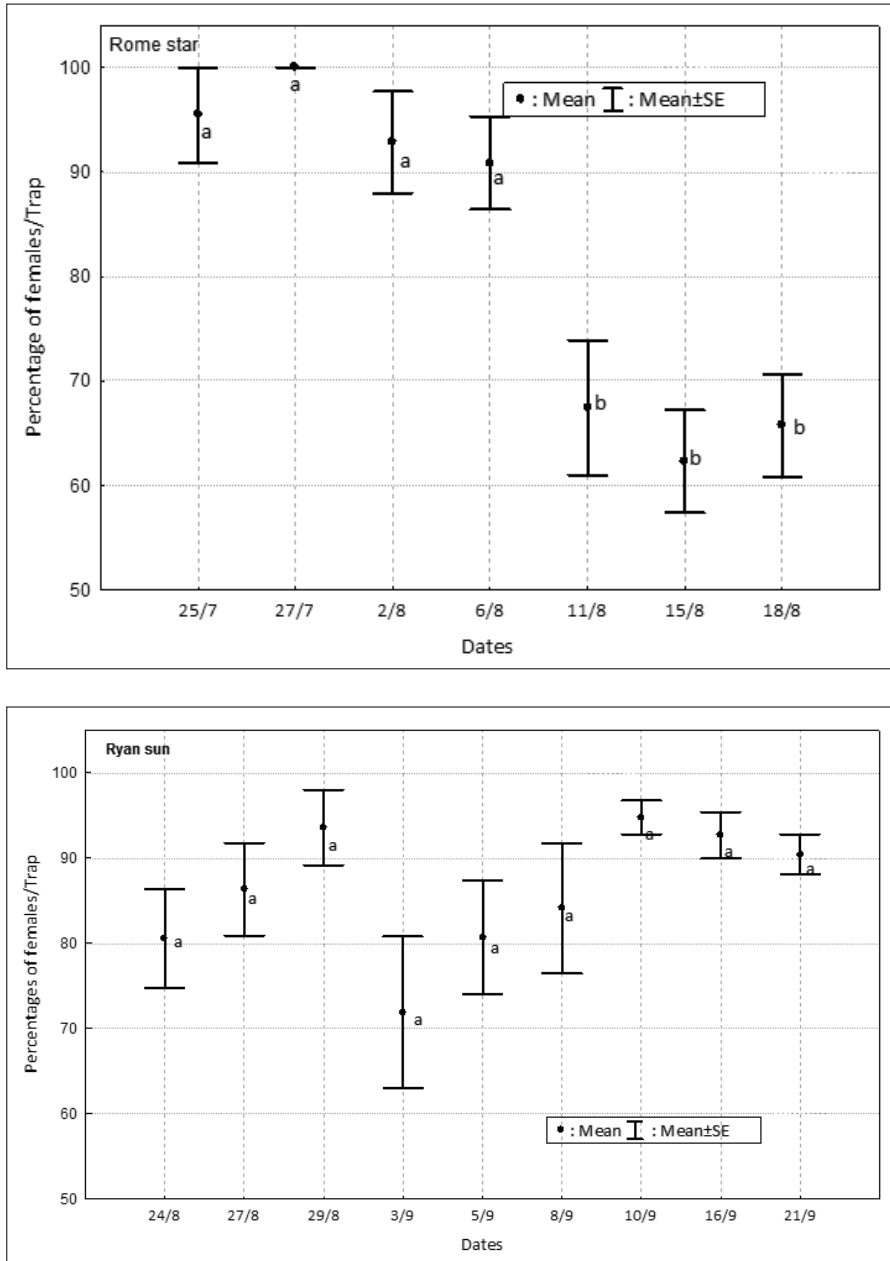


Figure 2: Percentage of *Ceratitis capitata* females caught in Magnet™ Med traps on peach trees in Sefrou region, Morocco, 2016 (Means bearing the same letter do not differ statistically, ANOVA followed by Bonferroni’s multiple-range test at $P \leq 0.05$)

Gender ratio

The gender of flies caught in Magnet™ Med traps revealed a relative importance of females compared to males throughout the study period. Considering the traps, however, by capturing at least one adult, the average percentages of captured females far exceed 60% of the trapped fly population, regardless of peach variety (Figure 2). On ‘Rome Star’, the proportion of caught females varied significantly over time ($F_{6,194}=7.90, P=0.05$), and the highest percentages were recorded in late July and early August. On ‘Ryan Sun’, the pattern of females percentage is statistically comparable to that of ‘Rome Star’ ($F_{8,211}=1.83, P=0.05$) (Figure 2). However, high variability which was observed between the dates of trapping is noteworthy.

The coefficients of variation are very high, ranging from 0 to 59% for ‘Rome Star’, and from 11 to 55% for ‘Ryan Sun’.

Catches by monitoring trap and treatments

Regular monitoring of *C. capitata* flights in plot trials was carried out using the Trimedlure trap; the catching timeline is shown in Figure 3. Adult males accounted for approximately 95% and 100% of the adults captured on the ‘Rome Star’ and ‘Ryan Sun’ peach varieties, respectively. The numbers of *C. capitata* captured by Trimedlure in plots protected by mass trapping or by attract-and-kill control were the same for ‘Rome Star’ ($t_{12}=0.139, P=0.05$) and for ‘Ryan Sun’ ($t_{56}=-1.066, P=0.05$). Medfly infestation of the two plots was therefore of the same intensity, and

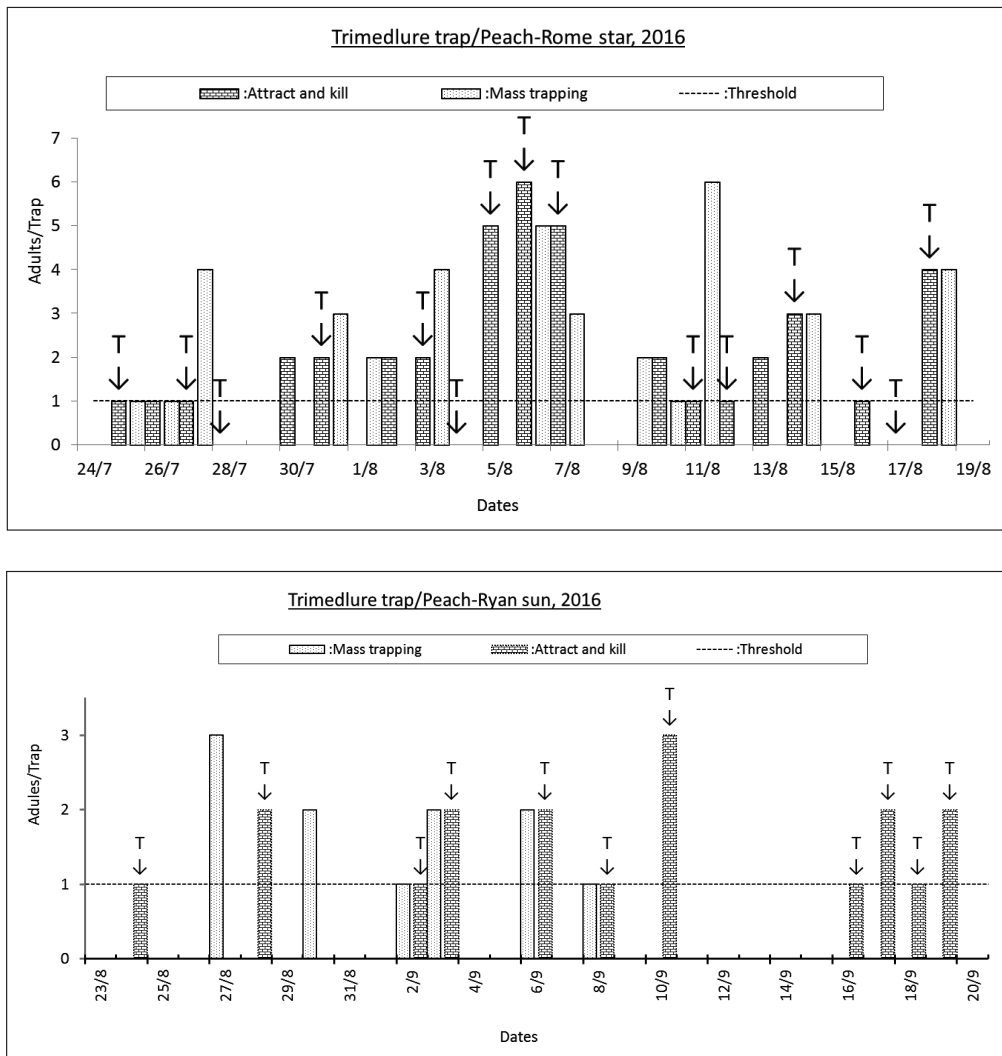


Figure 3: Dynamics of *Ceratitis capitata* adults caught in Trimedlure traps in peach orchard in Sefrou region, Morocco, 2016 (T = treatment of straw tufts)

the level of fly populations was homogeneous in the plots separately undergoing the two management methods.

In terms of harmfulness, the number of adults caught by the Trimedlure trap reached or exceeded the threshold adopted by the farm, but no treatment was undertaken in the plots protected by mass trapping. On the other hand, 15 and 11 treatments, using straw tufts, were carried out in the attract-and-kill controlled plots of the 'Rome Star' and 'Ryan Sun' varieties, respectively (Figure 3). Over the trial period, 'Rome Star' appeared to be more exposed to medfly attacks than 'Ryan Sun'.

Infestation rates

Pre-harvest

Medfly infestation rates of the 'Rome Star' variety of peach were very low, remaining 0% or less than 1% of fruits collected from trees throughout the trial period regardless of control methods. Infestation occurred during the last week of harvest (Table 2). In the plot protected by the attract-and-kill technique, infestation rates were statistically higher than those recorded in the plot protected by mass trapping [$(t_{1998} = (6.32, 4.54 \text{ and } 6.60 \text{ for August } 4, 13, \text{ and } 17 \text{ respectively in } 2016 \text{ growing season}), P = 0.05]$. During the pre-harvest period, damage generated by *C. capitata* was therefore perfectly countered both by the attract-and-kill technique and mass trapping as control methods.

On the 'Ryan Sun' variety, and throughout the study period (from 8/30 to 2016/9/19), there were no fruits on trees that showed signs of fly damage, and all sampled peaches (1000 fruit) were free of pest infestation regardless of the sampling period.

Table 2: Fruit infestation rate caused by *Ceratitis capitata* on trees of 'Rome Star' variety in Sefrou region, Morocco, 2016 (N = 1000 fruits examined by date and control method)

Assessment dates	Mass trapping	Attract-and-kill
	Infestation rates ± SE (%)	Infestation rates ± SE (%)
July 23	0.0 ± 0	0.0 ± 0
July 27	0.0 ± 0	0.0 ± 0
July 31	0.0 ± 0	0.0 ± 0
August 4	0.0 ± 0	0.1 ± 0.02
August 8	0.0 ± 0	0.0 ± 0
August 13	0.1 ± 0.02	0.3 ± 0.04
August 17	0.3 ± 0.05	0.9 ± 0.08
Mean ± SE (%)	0.001* ± 0.0003	0.002 ± 0.001

*: Infestation rates are statistically different (Student's test at 5%).
SE: Standard error

At harvest

The infestation rates of fruits at each harvest of 'Rome Star' were less than 1% of all fruits examined, and they ranged from 0 to 0.88%, depending on control method and harvest date. Infestation occurred between 2016/8/5 and 2016/8/18 (Table 3). The overall infestation rate observed in the plot controlled by the attract-and-kill technique was statistically higher than the rate in the mass trapping plot ($t_{12334} = 15.25, P = 0.05$) but infestation rates remained below the threshold tolerated by the farm (1%) regardless of control method.

No fruit infested with medfly was found after harvests of the 'Ryan Sun' variety, regardless of control methods, throughout the trial period from 2016/8/30 to 2016/9/19.

Table 3: Fruit infestation rate caused by *Ceratitis capitata* at the harvest of 'Rome Star' variety in Sefrou region, Morocco, 2016 (N = 6245 fruits examined at the rate of 560-1330 fruits/date in mass trapping control, and N = 6091 at the rate of 607-1262 fruits/date under attract-and-kill control)

Assessment dates	Mass trapping	Attract-and-kill
	Infestation rates (%)	Infestation rates (%)
July 31	0.0	0.0
August 1	0.0	0.0
August 5	0.0	0.09
August 8	0.0	0.26
August 16	0.08	0.88
August 18	0.23	0.87
Mean ± SE (%)	0.0001* ± 0.0001	0.0033 ± 0.0007

*: Infestation rates are statistically different (Student's test at $P \leq 0.05$).

Non-compliant fruits discarded in orchard

Regarding 'Rome Star', the infestation rates of fruits discarded at each harvest date varied from 0 to 11.5% of all sampled fruits, depending on control method and sampling date (Table 4). Overall, infestation rate in the attract-and-kill plot was statistically higher than in mass trapping plot ($t_{1998} = 2.19, P = 0.05$).

Regarding the 'Ryan Sun' variety, 1/350 and 2/350 sampled fruits were infested with *C. capitata* in the mass trapping plot. Fruits infested in the plot protected by mass traps were picked on October 3 and 6 of the growing season 2016. In the attract-and-kill plot, no fruit infested by the fly was observed throughout the trial period, i.e. from 2016/8/30 to 2016/9/19.

Table 4: Infestation rates of discarded fruits caused by *Ceratitis capitata*, collected in each harvesting period on 'Rome Star' variety in Sefrou region, Morocco, 2016 (N = 1000 fruits examined at 100 to 200 fruits by date and by control method)

Assessment date	Mass trapping	Attract-and-kill
	Infestation rates (%)	Infestation rates (%)
July 31	0.0	0.0
August 1	0.0	2.0
August 5	1.0	10.0
August 8	2.0	7.5
August 16	3.5	11.5
August 18	4.0	7.5
Mean \pm SE (%)	0.381* \pm 0.05	1.383 \pm 0.16

*: For each method of control, infestation rates are statistically different (Student's test at $P \leq 0.05$).

DISCUSSION

The results of this study highlighted the effectiveness of mass trapping of *C. capitata* by MagnetTMMed traps on two studied peach varieties in Morocco. The average infestation rate did not exceed 0.3% before and at harvest versus 0.9% in the plot protected by attract-and-kill technique. Among the discarded fruits, the rate of those damaged by *C. capitata* reached 11.5% in the plot controlled by attract-and-kill, while damage rate did not exceed 4% of all examined fruits protected by mass trapping. Our results confirmed the effectiveness of mass trapping against medflies, as reported previously for several other crops (Navarro-Llopis et al., 2008; Martínez-Ferrer et al., 2012; García Mari & Alonso Muñoz, 2004; Hafsi et al., 2015; Mediouni Ben Jemâa et al., 2010; Cabrita & Ribeiro, 2006; Eltazi et al., 2008; Boulahia-Kheder et al., 2015; Rahman & Broughton, 2016). It is also important to note that no chemical treatment was applied in the mass trapping plot, whereas 11 and 15 treatments, localized on straw tufts, were carried out in the plots controlled with the attract-and-kill technique on 'Ryan Sun' and 'Rome Star' varieties, respectively. In this respect, using the attract-and-kill technique against medfly on citrus, Benziane et al., (2003) carried out 7 sprays with an infestation rate equivalent to the generalized chemical control. Similarly, other previous studies also showed that mass trapping alone or accompanied by a limited number of treatments gave satisfactory control of *C. capitata* on citrus (Eltazi et al., 2008; Martínez-Ferrer et al., 2012; Boulahia-Kheder et al., 2015). Our results also

demonstrate that MagnetTMMed catches more females than males throughout the trapping period, suggesting that females are more responsible for the damage than males by their feeding and oviposition on fruits. This might be due to the trap's chemical composition, which is more attractive to medfly females. Previous studies had reported similar results with different types of traps baited with the same compounds as those used in our work (Midgarden et al., 2004; Heath et al., 2004; Broughton & De Lima, 2002; Katsoyannos & Papadopoulou, 2004; Papadopoulou et al., 2001; Epsky et al., 1999; Navarro-Llopis et al., 2008; Alonso-Muñoz & García Mari, 2007; Gazit et al., 1998; Katsoyannos et al., 1999; Alfawwer et al., 2009). In addition, the traps baited with three food attractants that were used in our work also catch more female flies than Trimedlure trap (Midgarden et al., 2004; Papadopoulou et al., 2001; Epsky et al., 1999; Katsoyannos et al., 1999). According to Broughton and De Lima (2002), Midgarden et al., (2004) and Pezhman et al., (2011), Trimedlure traps are specific to males. They are also highly recommended for monitoring *C. capitata* (e.g. Alfawwer et al., 2009; Pezhman et al., 2011; Başpınar et al., 2013). In view of the results obtained in this work, mass trapping is a clean, environment-friendly and very effective tool for controlling medfly in peach orchards. The density of *C. capitata* populations was regulated by mass trapping alone. This technique has the advantage of continuously regulating medfly populations throughout the time required for fruit protection. It can therefore be a part of an integrated management program against the medfly. However, for use on a large scale, it would be very appropriate to assess its impact on natural enemies, as it was done with Ceratrap traps (Pezhman, 2016). In addition, given the long persistence of MagnetTMMed traps (6 months), they can be moved from one variety to another as needed over periods in which fruit orchards are susceptible to pests. Thus, growers can protect at least 4 varieties with staggered maturity, which would significantly reduce the cost of this technique. In addition, the MagnetTMMed trap is allowed in organic farming (EU Directive EU 2092/91).

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DISCLOSURE STATEMENT

No potential conflict of interest was reported by the authors.

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Komparativna efikasnost masovnog izlovljavanja i tehnike “privuci i ubij” u suzbijanju voćne muve (*Ceratitis capitata*, Wiedemann) u voćnjacima breskve u centralnom Maroku

REZIME

U svrhu razvoja ekološki prihvatljivih, alternativnih strategija suzbijanja voćne muve sprovedeni su ogledi sa masovnim izlovljavanjem u centralnom Maroku tokom vegetacione sezone 2016. Na dve sorte breskve ('Rome Star' i 'Ryan Sun') u voćnjacima u okrugu Sefou upoređeni su efekti primene dve metode, suzbijanje masovnim izlovljavanjem i tehnikom “privuci i ubij”. Za masovno izlovljavanje, na obema sortama korišćeno je 62 klopki tipa MagnetTMMed/1.23 ha sa mamcima od amonijum-acetata, trimetilamina, putrescina i 0.01g deltametrina. Na parcelama na kojima je testirana tehnika “privuci i ubij”, mešavina malationa i protein hidrolaze nanošena je na gomilice slame tamo gde je bar jedna muva uhvaćena pomoću klopke Trimedlure postavljene u sredini svake parcele obeju sorti. Rezultati pokazuju da je broj izlovljenih muva kod masovnog izlovljavanja bio 508 adulta na sorti 'Rome Star' i 489 na 'Ryan Sun'. Prosečan broj se kretao od 1 do 3 adulta/klopki/danu, u zavisnosti od dana izlovljavanja i sorte, a odnos ženki je bio 62-100% u odnosu na ukupan broj muva. Na parcelama na kojima je ispitivana tehnika “privuci i ubij”, a uzimajući u obzir prag koji je odredila sama farma, na sortama 'Ryan Sun' i 'Rome Star' obavljeno je 11, odnosno 15 tretmana. Ukupna infestacija na parcelama za masovno izlovljavanje nije prelazila 0.3% pre ili nakon berbe, odnosno 0.9% kod tehnike “privuci i ubij”. Na opalim voćkama, infestacija nije prelazila 4% u varijanti masovnog izlovljavanja, odnosno 11.5% na hemijski tretiranim parcelama. Masovno izlovljavanje se pokazalo kao efikasan i ekološki povoljan način zaštite breskve od voćne muve.

Cljučne reči: *Ceratitis capitata*; Breskva; Masovno izlovljavanje; Privuci i ubij; Maroko