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Efficacy of kaolin, spinosad and malathion against *Ceratitis capitata* in *Citrus* orchards

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Abstract

Field experiments were conducted in 2005 on orange, *Citrus sinensis* (L.) Osbeck, var. Thomson in two different sites, Sbikha and Sidi Bouali, in Tunisia to assess the effectiveness of kaolin, spinosad and malathion against the Mediterranean fruit fly (medfly), *Ceratitis capitata* (Wiedemann) (Diptera Tephritidae). In both study sites, the materials were applied 3 times (kaolin at a concentration of 5 kg formulated product /100 l, spinosad at a concentration of 1 l formulated product /ha and malathion at a concentration of 200 ml formulated product /100 l). The efficacy of the different treatments was evaluated by assessing male captures in modified Steiner traps and fruit damage (punctured and dropped fruits).

The results indicated that (1) no significant differences among treatments in male captures were detected (2) fruit damage (no. punctured fruits and fallen fruits) was lower on kaolin-treated trees than on spinosad, malathion, and untreated trees. Kaolin successfully protected fruits from medfly infestations and provided long term control, from fruit development until harvest, whereas the insecticide, malathion, and the naturally derived insect control agent, spinosad, failed to protect fruits. Kaolin appears to be an important and helpful tool to reduce medfly fruit damage, and could be a valid alternative to intensive applications of insecticides, currently commonly used in *Citrus* orchards.

Key words: *Ceratitis capitata*, medfly, kaolin, spinosad, malathion, field trial, *Citrus*, Tunisia.

Introduction

The Mediterranean fruit fly (medfly), *Ceratitis capitata* (Wiedeman) (Diptera Tephritidae), is a serious economic pest infesting more than 350 species of fruits and vegetables throughout the world most of which are of high commercial value (Liquido *et al*., 1991). In Tunisia *C. capitata* is a multivoltine species, present all year round and essentially active from May to January on different host plants, such as apricot, peach, fig, plum, apple, pear, and *Citrus*. The number of generations per year varies with local temperatures and host plant.

Fruits attacked by *C. capitata* can be found in any season in the warm coastal regions. Economically, *Citrus* is the main host, especially early cultivars like Thomson and Washington navel, but also other *Citrus* including late cultivars (e. g. Valencia late), and late apricot and peach cultivars (ripening after the second week of May) can show heavy *C. capitata* infestation levels.

The control of the medfly in the main *Citrus* producing areas in Tunisia (North Eastern regions of the country) is centralized and carried out by the Ministry of Agriculture: malathion mixed with a hydrolyzed protein bait (food attractant) is applied in aerial and ground treatments. *C. capitata* control in other areas and on non-citrus crops, is conducted by the individual farmer with a wide range of different pesticides.

Although very effective when correctly applied, bait sprays are known to create ecological, toxicological and environmental problems (Troetschler, 1983). Malathion was found to be highly toxic to a number of beneficial insects, such as honey bees (Gary and Mussen, 1984) and parasitoids (Cohen *et al*., 1987; Daane *et al*., 1990), and therefore this active ingredient needs to be replaced with other, less harmful agrochemicals.

A possible alternative to malathion for medfly control is spinosad, an insecticide obtained from the naturally occurring actinomycete, *Saccharopolyspora spinosa* Mertz and Yao, (Dow Agrosciences, 2001). This active ingredient has been combined with a food attractant and feeding stimulant, formulated, and the formulated product is sold in Tunisia under the brand name Success Appât (Dow Agro sciences, Indianapolis, Indiana, USA). Recent studies show that spinosad is effective in controlling medfly infestations (Burns *et al*., 2001; Vargas *et al*., 2002). Furthermore spinosad was demonstrated safe to honeybee (Miles, 2003).

Kaolin (Surround® WP, Engelhard Corp. Iselin, NJ, USA) is also a potential alternative pest management product with improved safety and reduced environmental impact. Kaolin, is a white non-porous, chemically inert, non-swelling, low-abrasive, and fine grained aluminium-silicate mineral that easily disperses in water (Glenn and Puterka, 2005). Kaolin is used to protect plants from insect pests as well as from sunburn and heat stress (Glenn *et al*., 1999; Glenn and Puterka, 2005; Melgarejo *et al*., 2004; Wand *et al*., 2006). On the plants kaolin forms a particle film altering insect/pathogen behaviour on the plant. It can easily be removed from harvested commodities. On plants coated with hydrophobic particle films, repellence, ovipositional deterrence, and reduced survival of insects were observed (Glenn *et al*., 1999; Glenn and Puterka, 2005). The formulated product Surround WP was shown to be effective against different diseases and insects (Glenn *et al*., 1999; Knight *et al*., 2000; Saour and Makee, 2003; Mazor and Erez, 2004; Saour, 2005), in particular pear psylla, *Cacopsylla pyricola* (Foerster) and *C. pyri* (L.)

(Puterka *et al*., 2000; Pasqualini *et al*., 2002), olive fruit fly, *Bactrocera oleae* (Gmelin) (Saour and Makee 2003), pistachio psyllid, *Agonoscena targionii* (Lichtenstein) (Saour, 2005), and *C. capitata*, on nectarine, apple and persimmon (Mazor and Erez, 2004).

This study aimed at evaluating the efficacy of the kaolin-based product Surround WP and of the spinosad-based insecticide Success Appât against *C. capitata* on orange in two Tunisian study sites with different climatic conditions in comparison with the organo-phosphate insecticide malathion and an untreated control.

Materials and methods

Field trials were conducted in two study sites with Mediterranean climate, the mainland site Sbikha (at approximately 60 km from the sea, 35.40° N, 10.06° E, 60 m above sea level) and the coastal site Sidi Bouali (at approximately 5 km from the sea, 35.55° N, 10.33° E , 15 m above sea level).

Sbikha orchard

The Sbikha study orchard was located on a farm of 20 ha in size, at approximately 30 km from the town Kairouan in the centre of Tunisia.

The trial orchard (size: approximately 0.5 ha) cultivated with *Citrus*, *C. sinensis* (L.) Osbeck, var. Thomson, consisted of 19 rows with 23 trees per row (distance between rows: 4 m; distance between trees along row: 3 m; plant age: 28 years). The plot was surrounded to the north by a row of cypress trees bordering a local road, to the east by a pear and fig orchard, to the west by a pomegranate orchard, and to the south by a *Citrus* orchard (Clementine, Valencia late, and Orange).

To evaluate the efficacy of the different treatments, a randomized block design with 4 replicates of 4 trees per treatment was used. Blocks and plots were arranged in one area of the study orchard along 8 adjacent rows using 8 trees per row. The following treatments were compared: 1. Kaolin (formulated product -f.p-. Surround WP; concentration a.i. 95%; applied rate: 5 kg f.p. /100 l); 2. spinosad (f.p. Success Appât; concentration a.i. 24%; applied rate: 1 l f.p. /ha; 3. malathion (f.p. Fyfanon 50 EC from Cheminova Agro A/S, Denmark; concentration a.i.: 50%; applied rate: 200 ml f.p. /l00 l); 4. untreated control. The first treatment was applied on September 28, 2005; at that date the mean weekly male captures in the traps were 49 (see below), and fruits were still developing. Two additional applications were carried out on October 19, and November 10, 2005. Products were applied with a manually operated knapsack sprayer with 16 l of capacity (Model H-103 Himatic Agro, India) calibrated to deliver 2.0 l of liquid suspension per tree.

The remaining area of the study orchard was treated with the grower's standard control strategy (applications of malathion on September 3, 19, 29, October 15, 30, and on November 10 and 24, 2005).

Temperature data were collected from a meteorological station located at about 26 kilometres from the study orchard.

C. capitata flight activity

To monitor the flight activity of *C. capitata* males, after the first treatment (September 28), 16 modified Steiner traps (Steiner, 1952), produced locally and each containing a cotton wick (3.5 cm in length by 1 cm in diameter) baited with a 2-3 ml solution of trimedlure (90%) and malathion (10%) (v:v) and refilled every four to five weeks, were used: in each plot, one trap was hung on the first of the four trees (4 monitoring traps per treatment) at a height of 1.2 - 1.5 m. The distance between traps varied from 6 m along blocks to 8 m between blocks. The number of males captured per trap was recorded weekly.

Fruit damage

Due to the relatively low number of fruit per tree, we decided to inspect all 4 trees of each plot for fruit damage. To avoid biasing of data due to old fruit damage, prior to applying the first treatment, all trees were surveyed for damaged fruits, and attacked fruits (punctured and dropped fruits) were removed. Additional assessments for damaged fruits were conducted once a week throughout the study period. Punctured fruits were recorded and marked, and dropped fruits were recorded and removed. At the end of the study period, the total number of punctured and dropped fruits per tree was calculated.

Sidi Bouali orchard

The Sidi Bouali study orchard was located on a farm of approximately 6 ha in size, cultivated mostly with different *Citrus* and pomegranate cultivars. The study orchard was surrounded to the north by prickly pear, to the east by pomegranate, to the south by orange (*C. sinensis* (L.) Osbeck) var. Maltaise and Clementine (*C. clementina* Hort. ex Tanaka), and to the west by different *Citrus* fruits (orange var. Valencia late, lemon, and sweet orange).

The study orchard was approximately 0.5 ha in size, and consisted of 15 rows of 24-year-old *Citrus* trees, *C. sinensis* (L.) Osbeck var. Thomson, with 8 trees per row. The study orchard was divided into 4 plots, different in size: one plot of two rows was sprayed with kaolin (f.p. Surround WP; applied rate: same as above), an adjacent additional plot of two rows was treated with spinosad (f.p. Success Appât; applied rate: same as above), one plot of 10.5 rows was treated with malathion (f.p. Fyfanon 50 EC; applied rate: same as above), while 4 trees of the last row were used as untreated control plot.

Treatments (kaolin, spinosad, and malathion) were applied for the first time on October 14, when the average weekly number of males captured per trap was 180.5, and repeated on October 29, and November 17. Due to the high infestation level, two additional sprays were applied to the spinosad- and the malathion-treated plot on November 8 and 21. All sprays were applied with the same knapsack sprayer used in Sbikha, but the sprayer was calibrated to deliver 3 l of solution per tree.

Before the beginning of the trial, the entire study orchard was treated with malathion by the grower on September 8, 22, and on October 8.

Throughout the study period, weather data were recorded by an automatic meteorological station (National Institute of Rural Engineering, Water and Forest, INGREF) situated at about 10 km from the study orchard.

C. capitata flight activity

To monitor *C. capitata* flight activity, one modified Steiner trap baited with trimedlure-malathion (for details see paragraph of Sbikha orchard) was hung on a tree in the centre of the first row of the plot, and checked weekly for *C. capitata* male captures.

Fruit damage

Within each plot, one tree was selected and checked weekly for punctured fruits. The 4 selected trees bore approximately the same number of fruits. At every check, new punctured fruits were marked with indelible ink; dropped fruits were recorded, collected, and removed. In order to assess fruit damage during ripening, at the end of the study period, the total number of punctured and dropped fruits per tree was determined.

To assess for fruit damage at harvest, on December 1, three days before harvest, 50 fruits per treatment were collected from at least 5 different trees and from the four trees in the control plot, brought to the laboratory, and examined for medfly damage (sting marks).

Statistical analysis

In the Sbikha orchard, the effects of blocks and treatments on the total male captures and fruit damage were analyzed using a two-way ANOVA: blocks and treatments were used as independent variables and "total number of male capture", "total number of punctured fruits", and "total number of dropped fruits" as dependent variables. All analyses were done using Minitab 13.1 Software (Minitab. Inc., PA, USA).

In the Sidi Bouali orchard, since only one large plot per treatment was used and plots were different in size, the total number of male captures and fruits damage during development (number of punctured and dropped fruits) were reported but were not statistically analyzed.

Results

Sbikha orchard

C. capitata flight activity

The flight activity of *C. capitata* males was low in early October, increased after October 19, peaking at November 16, and remained relatively stable after November 23 till the end of the experiment. In all treatments, the trap captures were highest during the week from 10 to 16 November, period when fruits were ripening (figure 1). The combination blocks /treatments show no significant difference in males captures between blocks nor between treatments (two way ANOVA: Blocks, $F_{3,15} = 2.4$, $P = 0.135$. Treatments, $F_{3,15} = 1.27$, $P = 3.341$). However, weekly male captures trap were lowest in the kaolin-treated plots, intermediate in the malathion- and spinosad-treated plots, and highest in the untreated control plots (figure 1).

Fruit damage

Differences among blocks in both the total number of punctured and dropped fruits were not significant (two-way ANOVA: $F_{3,15} = 0.74$, $P = 0.555$ for punctured fruits; $F_{3,15} = 2.02$, $P = 0.182$ for dropped fruits), showing that the *C. capitata* population was distributed uniformly within the study area. Differences among treatments in the total number of punctured and dropped fruits, instead, were significant (Two-way ANOVA: $F_{3,15} = 4.13$, $P = 0.043$ for punctured fruits; $F_{3,15} = 5.54$, $P = 0.020$ for dropped fruits). The average numbers of punctured and dropped fruits over the study period are given by figures 2 and 3.

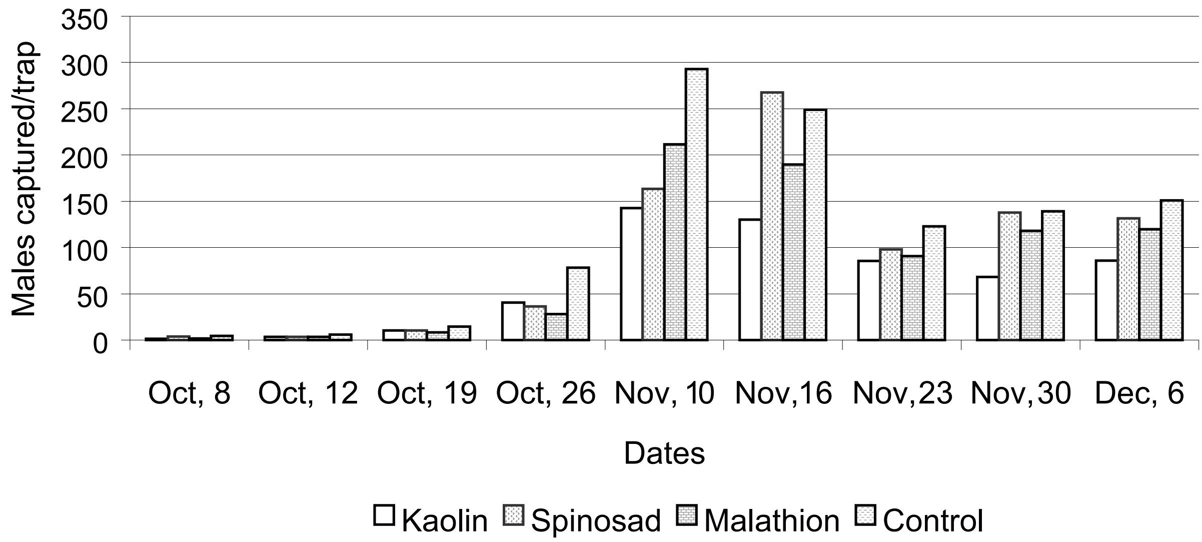

Figure1. Mean number of *C. capitata* males captured weekly per trap in the modified Steiner traps baited with trimedlure-malathion in the Sbikha orchard (four traps per treatment).

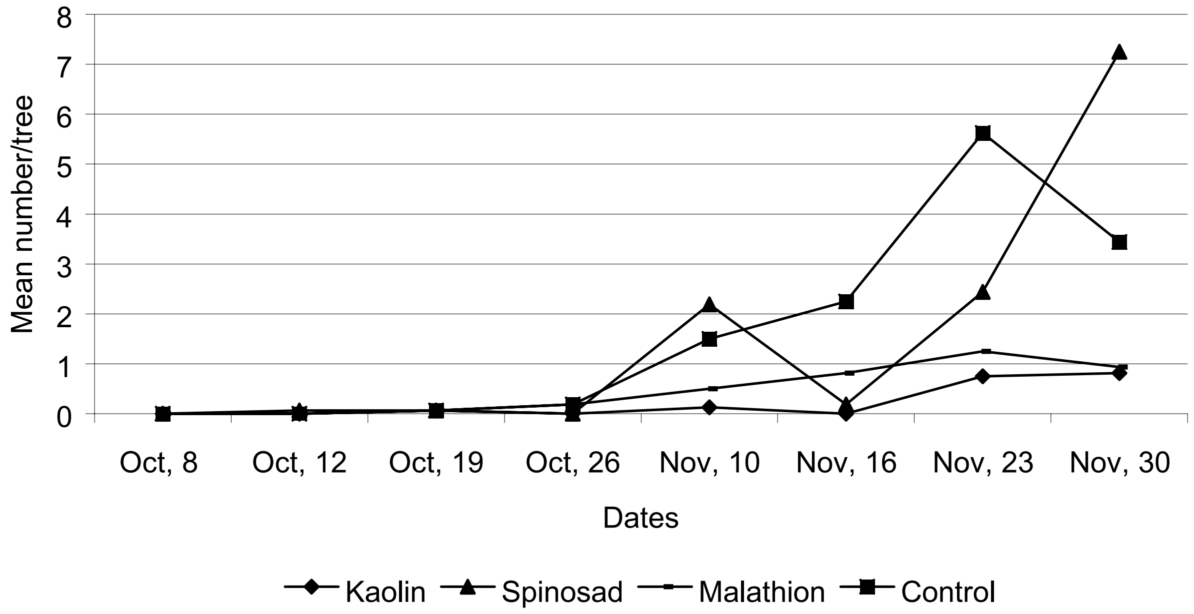

Figure 2. Mean number of punctured fruits per tree in the different treatments in the Sbikha orchard.

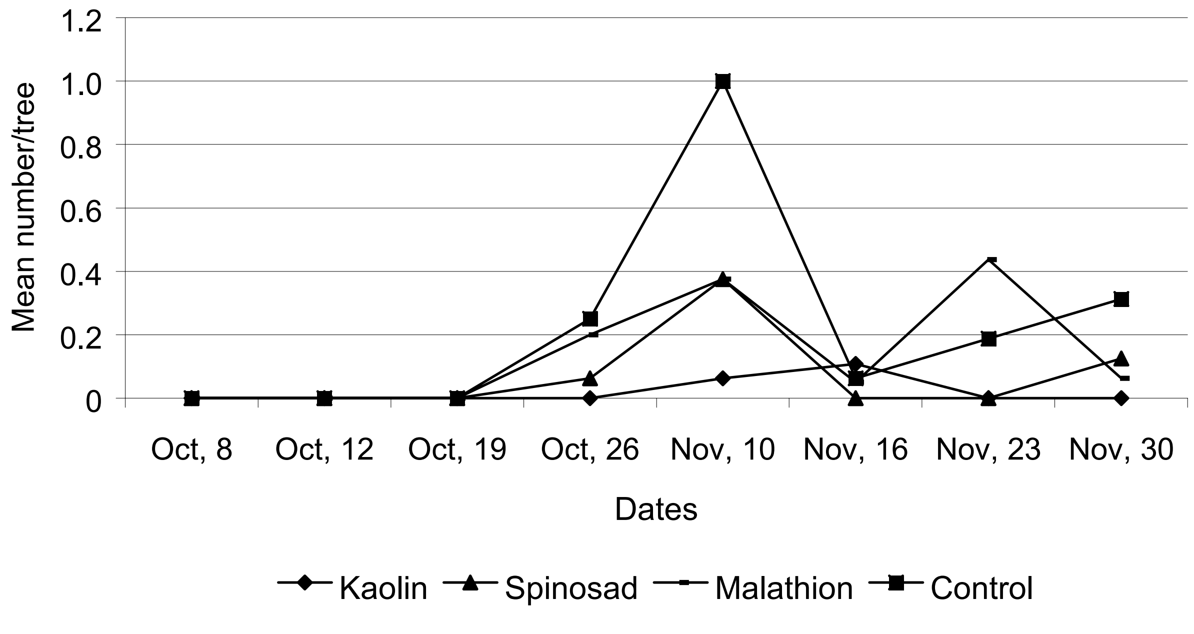

Figure 3. Mean number of dropped fruits per tree in the different treatments in Sbikha orchard.

Sidi Bouali orchard

C. capitata flight activity

The flight activity of *C. capitata* males was important during the study period peaking at the end of October – beginning of November (figure 4). The average weekly male capture recorded was 340.14 in kaolin-treated plot, 457.85 in spinosad-treated plot, 429.14 in malathion-treated plot and 661.85 in the untreated plot.

Fruit damage

Fruit damage during fruit ripening:

The number of punctured fruits during ripening is given by the figure 5. For kaolin treated plot only 7 fruits per tree were attacked throughout the study period specifically after November 1, (they were 2, 1, 2 and 2 fruits respectively on Nov.11, Nov. 17, Nov. 24 and Dec. 1, 2005). However, the total numbers of punctured fruit in the other treatments plots were high with 101,

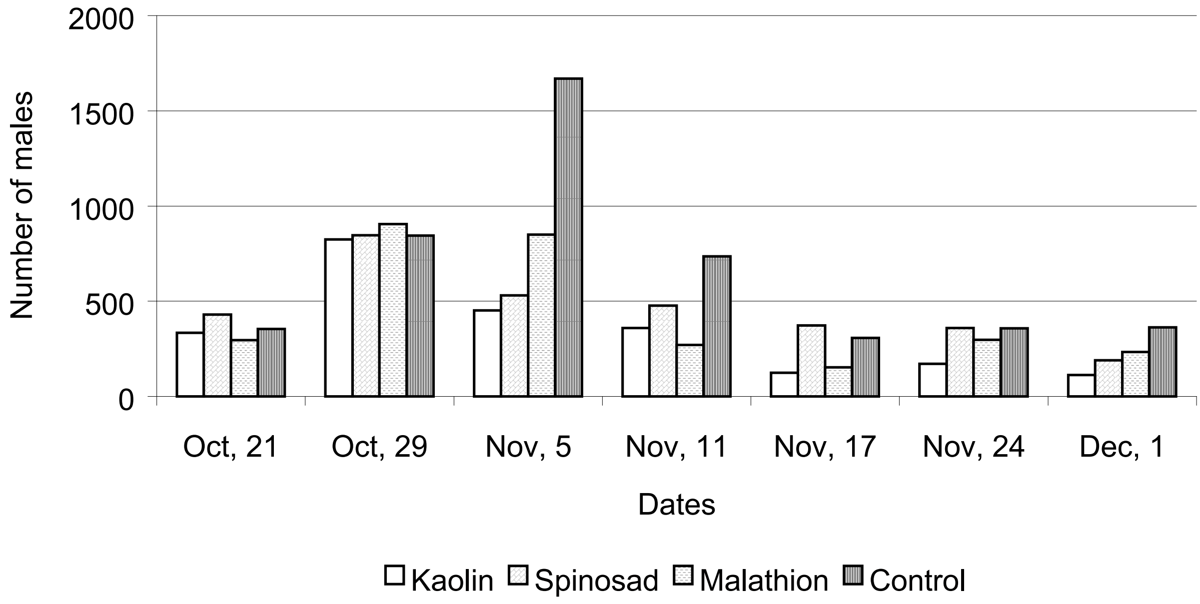

Figure 4. Number of males captured weekly in the different treatments in the modified Steiner trap baited with trimedlure-malathion in Sidi Bouali.

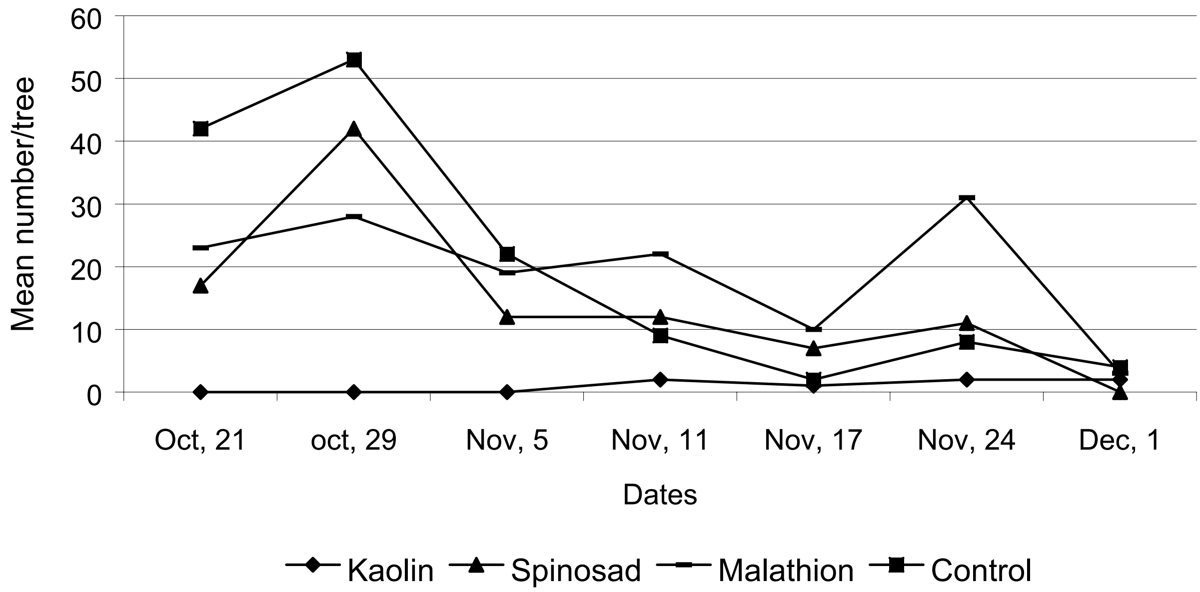

Figure 5. Mean number of punctured fruits per tree in Sidi Bouali orchard.

136, and 140 respectively for spinosad, malathion and control plots.

Regarding the fallen fruits the kaolin treated plot shows the lowest damage with only a total of 6 dropped fruit during the study period in comparison with 69, 56, and 98 respectively for spinosad, malathion and control plots (figure 6).

Fruit damage at harvest:

The results on fruit damage at harvest reflect those on fruit damage during fruit ripening: in the Sidi Bouali orchard, 3 days before harvest, in the kaolin-treated plot, only 2 out of 50 randomly selected fruits were punctured, while respectively 45, 38, and 47 fruits out of 50 showed *C. capitata* punctures in the plot treated with spinosad, malathion and in the untreated control plot.

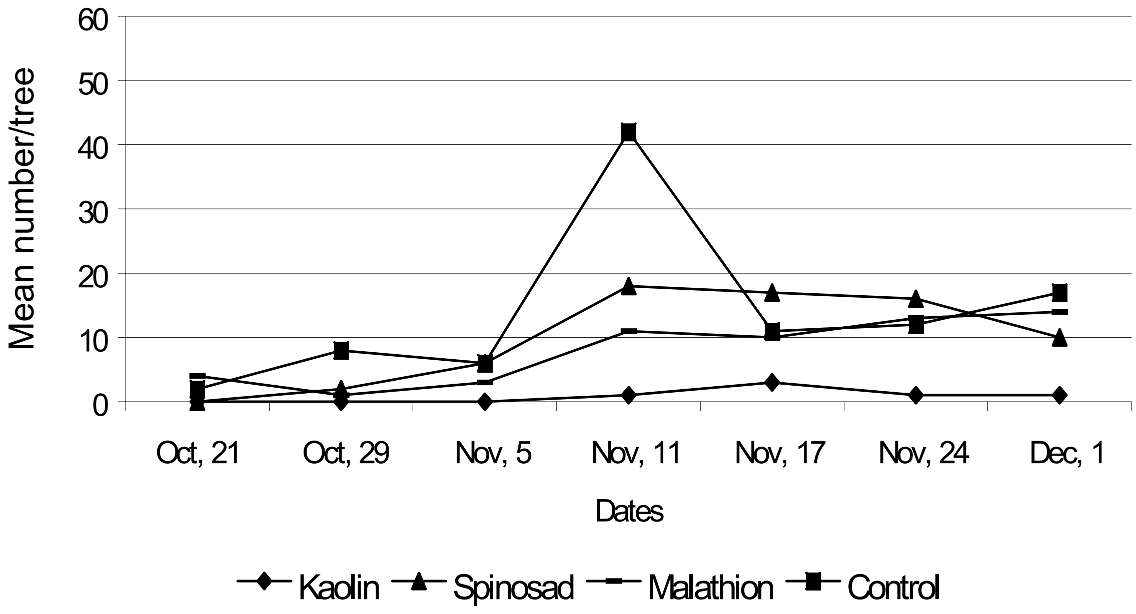

Figure 6. Mean number of dropped fruits per tree in Sidi Bouali orchard.

Discussion

This study aimed at verifying whether the organic insecticides kaolin and spinosad could be used instead of conventional agrochemicals for *C. capitata* control in *Citrus* orchards. In one trial, kaolin was more efficient than spinosad and the insecticide malathion. Formulated kaolin, forming a particle film on plants, thus seems to be an important and helpful tool for the control of *C. capitata* in *Citrus* groves. In Israel, Mazor and Erez (2004) conducted both laboratory and field tests with formulated kaolin (Surround WP), and obtained an almost complete protection on nectarine, apple, and persimmon against infestations of the medfly. Kaolin also showed high efficacy in controlling an another Tephritid fly, the olive fruit fly, *B. oleae*, in olive groves in Syria (Saour and Makee, 2003): fruit damage on kaolin-treated olive trees was significantly lower than on untreated control trees. Kaolin provided season-long insect control (> 14 weeks) outperforming the insecticide Dimethoate.

As other species of the family Tephritidae, also adults of *C. capitata* are known for their flying foraging behaviour between trees and adjacent orchards (Prokopy and Roitberg, 1989). In our trials, experimental plots were side by side without buffer zone permitting free adult *C. capitata* movement and consequently a uniform fruit damage and relatively similar adult captures. This is true for spinosad-treated plots, malathion –treated plots and un-sprayed plots but not in kaolin treated plots where both damaged fruits and males captures were less important suggesting a relatively good protection of kaolin treated trees.

Many species of phytophagous arthropods use visual cues to locate their hosts (Owens and Prokopy, 1986). Tephritid fruit flies are attracted to colors and shapes reminiscent of host fruit and foliage. Katsoyannos (1989) tested seven colours in attracting *C. capitata* in *Citrus* orchards and found that the yellow colour was very attractive to fertilized females, whereas blue and white were the least attractive colours. Also Economopoulos (2002) stated that the preferred colour of *C. capitata* females is yellow, especially fluorescent yellow.

Little research has been published on male *C. capitata* preference for visual cue in traps baited with trimedlure probably because trimedlure is a such potent attractant (Nakagawa *et al*., 1971). Epsky *et al*., (1996) tested different colored inserts in Jackson traps baited with trimedlure and found that traps containing either yellow or orange inserts capture more *C. capitata* males than those with standard inserts.

The trees, leaves and fruits treated with kaolin and thus coated with a white particle film could influence the landing of females (Saour and Makee, 2003).

In our trials, we did not find significant differences among treatments in the number of males captured in modified Steiner traps, probably due to (1) the high adult population level (figure 1, figure 4) and (2) the powerful attraction of males by the parapheromone trimedlure over relatively long distances (Cunningham, 1989). Furthermore, the modified Steiner traps were yellow-orange in colour, and thus easy to locate for *C. capitata* adults.

Saour and Makee (2003) suggested that the bright white colour of kaolin-sprayed olive trees may disrupt the orientation of the olive fruit fly, *B. oleae*, within the olive grove. However, the Authors did not support their hypothesis with data on female captures in MacPhail traps baited with bicarbonate solution.

The use of the effective female trapping system consisting of McPhail traps baited with three synergistically acting food attractants (ammonium acetate, putrescine and trimethylamine) (Epsky *et al*., 1995; Heath *et al*.,

1997) would probably have enabled us to collect detailed data on female captures.

In the Sbikha orchard, the slight decrease in the average weekly male captures with 94.88 and 85.69 recorded respectively in the plots treated with spinosad and malathion compared to untreated plots (117.55) could be due to mortality of males caused by the insecticide applications. However, the number of captured males was always lowest in the kaolin treated plots (mean of 63.11 weekly males captured).

Puterka *et al*. (2000) had successfully used formulated kaolin against the pear psylla *C. pyricola*, and the authors identified six mechanisms or mode of action: repellence, ovipositional deterrence, reduced feeding efficacy, impeded grasping of the host, host camouflage, and direct mortality. The efficacy of kaolin against *C. capitata* may be attributed to repellence, and/or ovipositional deterrence, because we found males in monitoring traps, and we observed adults flying in the canopy. Furthermore, in the kaolin-treated plots, fruits showed female punctures only on their untreated parts (usually the spray did not reach the bottom part of the fruits).

In both trials, spinosad seems to show little efficacy with regard to fruit damage. The efficacy of the spinosad-based product against *C. capitata* was very low. Our results do therefore not confirm the positive results obtained by other researchers with spinosad, where in both field and laboratory tests spinosad-based treatments provided good control of *C. capitata* and Caribbean fruit fly, *Anastrepha ludens* (Leow) (Burns *et al*., 2001; King and Hennessey, 1996; Vargas *et al*., 2002; Mangan *et al*., 2006). The poor efficacy of spinosad as well as of the chemical standard malathion may be due to (1) the unusually high *C. capitata* populations present during the study period. Temperature is the most important factor affecting *C. capitata* life cycle (Vargas *et al*., 1997, Israely *et al*., 2004). The population build-up in our study orchards was probably due to the high temperatures that occurred during summer and autumn (figures 7 and 8), permitting a rapid development of generations. (2) Also, malathion and spinosad, volatiles molecules could have lost persistence on the crop by rapid degradation owing to excessive temperatures and intense sunlight, while kaolin consisting of particles remained on the trees.

Dust particles are known to be disruptive for natural enemies (Debach, 1979) and resurgence of pest species populations that are regulated by natural enemies may occur in particle-film treated trees. The likelihood of reduced beneficial insect activity should not be ignored. Knight *et al*., (2000) described increased levels of the western tentiform leafminer *Phyllonorycter elmaella* Doganlar and Mutuura, as well as significantly lower parasitism of this pest on kaolin (M96-018)-treated apple trees compared with untreated trees. We did not evaluate the efficacy of natural parasitoids and predators which is generally low in *Citrus* orchards due to heavy insecticide applications yearly conducted against the medfly and other important pests such as *Phyllocnistis citrella* Stainton (citru*s* leafminer), aphids and mealy bugs.

The results of our study suggest that three applications of kaolin are sufficient for the control of *C. capitata* in *Citrus* orchards, outperforming the insecticides malathion and spinosad. However, the efficacy of kaolin may be reduced when rainy conditions follow the kaolin sprays. In fact, Mazor and Erez (2004) indicated that products such as kaolin are best suited to dry regions because they may be washed off by heavy rainfalls.

In conclusion, kaolin seems to be a promising and helpful tool for the control of medfly populations in *Citrus* orchards. However, to optimize its efficacy, further studies on the number, the concentration, and the timing of the applications (early in the season before egg-laying) are needed.

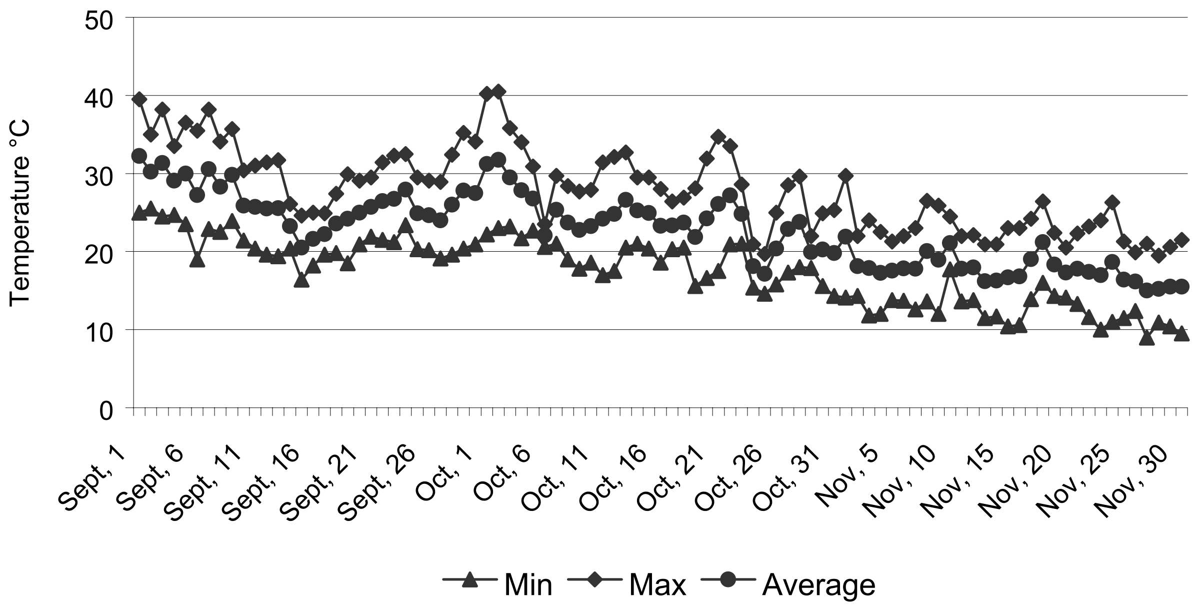

Figure 7. Temperature value (minimum, maximum and average) during the trial September-November 2005, in Sbikha orchard (data from meteorological station located at about 26 km from the study site).

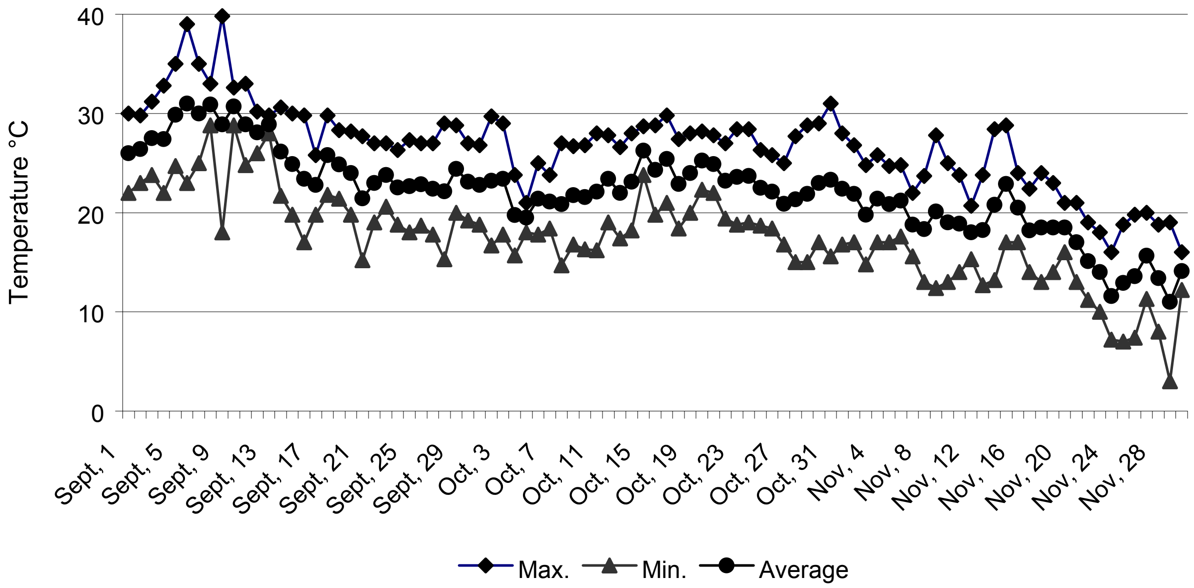

Figure 8. Temperature value (minimum, maximum and average) during the trial September-November 2005, in Sidi Bouali orchard (from meteorological station located at about 10 km).

Acknowledgements

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