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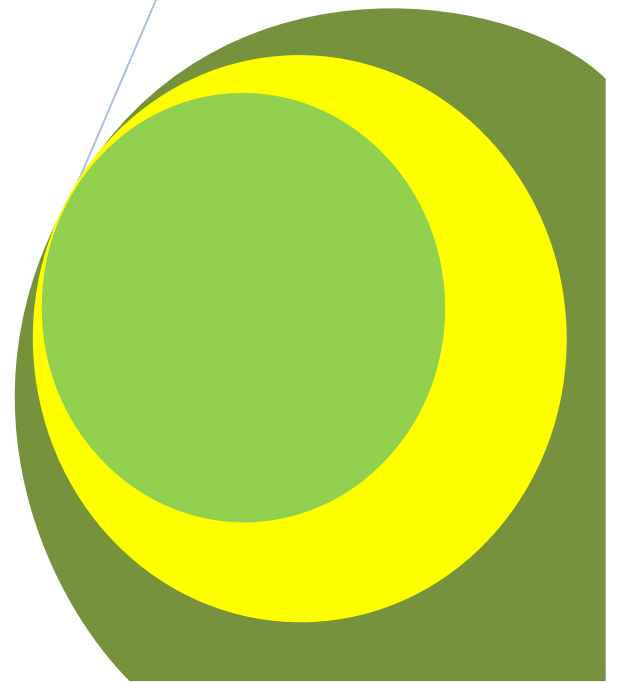
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Is Mass Trapping Technique useful for the Control of the Tomato Leafminer, *Tuta absoluta* (Lepidoptera: Gelechiidae)?

By

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Research Article

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ABSTRACT

The effectiveness of the mass trapping technique for the control of the leafminer, *Tuta absoluta* was evaluated in an open field tomato in 2011 and under greenhouse conditions in 2012 in Tunisia. A field of un-staked tomato was used in Kalaâ Kebira region. The trial was set up in an area of about 12000 m² in a randomized block design with four replications at three water trap densities (D1 = 20 pheromone traps per ha; D2 = 40 pheromone traps per ha and D3 = 80 pheromone traps per ha). Traps were inspected approximately at weekly interval; leaves and fruits were sampled and examined for insect infestation. Results indicate that the mean number of *T. absoluta* eggs, larvae and mines per leaflet do not statistically vary between the three tested densities. The percentage of fruit infestation by *T. absoluta* larvae related to sampling dates were respectively 17.5 %, 18.75 %, 18.33 % and 33.75 % for D1 and 15 %, 20 %, 16.25 % and 23.75 % for D2 and 11.25 %, 22.5 %, 18.75 % and 20% for D3. Over all, there is no clear difference in fruit infestation regarding the three densities suggesting the possibility of adult migration from nearby tomatoes.

The technique was evaluated in two plastic greenhouses planted with tomato located in Saheline region in comparison with another greenhouse sprayed chemically. High trap densities (12 per greenhouse) were used. Tomato leaves and fruits were sampled and checked for *T. absoluta* larval infestation. Results suggested that there was no significant difference between mass trapping technique and chemical control strategy. In average, the percentages of fruit infestation were respectively 16.66 %, 23.80 % and 44.44% in the first greenhouse; 18.75 %, 6.66 % and 35 % for the second greenhouse and 14.28 %, 15.38 % and 41.66 % for the control greenhouse managed chemically.

Lessons learnt are that mass trapping strategy demonstrate the need to apply this technique over an isolated field, in the whole area or under greenhouse conditions to minimize the influence of adult migration.

Keywords: *Tuta absoluta*, tomato, mass trapping, pheromone, trap densities, greenhouse, Tunisia.

1. INTRODUCTION

The tomato leafminer, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) is now considered to be one of the most damaging pests of tomato production, both in open fields and under greenhouse conditions. The species originates from South America and has been introduced in Europe and in the Mediterranean basin countries between 2006 and 2010 (Desneux et al., 2010). In the area of origin of the insect, the primary management strategy is chemical control (Siqueira et al., 2000) showing low to moderate efficacy mainly because of the endophytic behavior of the larvae and to the resistance to insecticides observed in many populations (Lietti et al., 2005; Silva et al., 2011). In Tunisia, the most widely insecticide used to control the insect is the neurotoxin obtained from a soil actinomycete, spinosad (Tracer) which is effective (Braham et al., 2012). Nevertheless, recently, a South American population of *T. absoluta* has been found to be resistant to spinosad (Reyes et al., 2011).

Reducing the quantity of insecticides applied on crops and in the environment has been the major objective that drives research for the implementation of other control strategies. The use of pheromone traps for mass trapping is an insect control method that has been sufficiently studied. The concept of mass trapping is using species-specific synthetic chemical lures such as sex and aggregation pheromone and food/host attractant, to attract insects in a trap where they would be confined and die. Mass trapping using color-baited traps is one of the old approaches to direct control for suppression and eradication of insect population (Steiner, 1952).

Virgin female tomato leafminer releases a sex pheromone that strongly attracts males (Quiroz, 1978). This pheromone was identified by Attygalle et al. (2006) as (3E, 8Z, 11Z)-3,8,11-tetradecatrien-1-yl acetate. Though there are some studies of mating disruption (Michereff et al., 2000; Vacas et al., 2011; Cocco et al., 2013) and mass trapping in Italy (Cocco et al., 2002) and Egypt (Taha et al., 2013), to our knowledge, no attempts have been made to control *T. absoluta* with mass trapping in Tunisia especially under greenhouse conditions.

The main objective of the present study was to assess whether the male mass trapping results in a decrease of *T. absoluta* male abundance and a subsequent reduction in damage in tomatoes. This approach has been proposed by El-Sayed et al. (2006).

2. MATERIAL AND METHOD

Trials were undertaken in two cities located in the central part of Tunisia; Kalâa Kebira and Saheline.

2.1. Open field tomato study

The mass trapping technique was studied over a 12000 m² in an open field un-staked tomato in the Kalaa Kebira region (Sousse governorate, 35°54' North, 10°25' East) comparing three trap densities; 20 traps per ha, 40 traps per ha and 80 traps per ha. Tomato plants (cv Justar) were purchased from a local nursery in polystyrene trays at about 15 cm height and transplanted in rows on 16 and 17 March 2011. Drip irrigation and fertigation were used. Weeds were handily controlled; harvest took place between 24 June and 13 July 2011.

The trial tomato field was divided into four equal plots of 60 x 50 m separated by 30 m to avoid trap effect. Every plot consisted of three sub-plots of 50 x 20 m (1000 m²) in which one of the three trap densities was allocated. Traps were set up in three plots within each bloc. Each block contained 60 rows of tomato (0.8 m between rows and 0.4 m in the row) for a density of about 30,000 plants per ha. The distance between the traps was minimum 7 m (80 traps/ ha) (Fig. 1). The plot was surrounded by an almond- olive orchard in the East, in the South - by a local road, in the West - by a field of artichoke and in the North - by a tomato field.

Pan traps were made of plastic, red in color (height from bottom to top = 14 cm, diameter = 35 cm) purchased from a local store. Two small holes were made on the top in which a wire was introduced housing the pheromone dispenser in a punctured small plastic tube (7 cm length and 3 cm diameter) to provide shade for the lure. Pheromone capsules were purchased from Atlas Agro (Atlas Agro, 2013) produced in January 2011, (Switzerland) and sold in Tunisia by the Company Agrichimie in June 2011 and held in the refrigerator at 0°C in the laboratory. Approximately 7 L of irrigation water was added to the trap and renewed every week. Traps were setup on April 29, 2011 and inspected regularly usually at week interval from 6 May 2011 to 13 July 2011. Moths caught were carefully removed from the trap using a piece of wood and water was renewed. Pheromone capsules were changed at four weeks interval (IPS, 2012).

Sampling of leaves and fruits

To evaluate trap densities, 10 tomato leaves per plot per density were randomly picked during the period from 13 May to 13 July 2011. Samples were put in plastic bags and stored in the refrigerator for further assessment. Leaves were examined under stereomicroscope for *T. absoluta* eggs, larvae, pupae and mines. In addition, 10 fruits were collected per trap density per plot (= 40 fruits per density) on June 24, July 1, 8 and 13, 2011 and checked for *T. absoluta* larval damage (entry holes). Fruits attacked by Noctuidae larvae (large entry holes) were not included in the calculation of infestation percentage.

2.2. Greenhouse trial

Three greenhouses were used in the current study, two for mass trapping experiment and one conducted as a control using insecticides. Each greenhouse measured 64 m in length and 8 m wide located in Saheline region (Monastir Governorate, 35°42' North, 10°42' East). Four double rows of tomato (cv Amel), planted on 17 November 2011 spaced 0.75 m apart. The distance between each double row was 1 m. In the row, tomato plants were spaced at 0.45 m. During the period of the study, the two greenhouses used for mass trapping received no insecticide sprays. However four sprays were done for control greenhouse (table I). Two fungicide sprays were made on 21 January 2012 and on 12 March, 2012 (Kocide at 150 g of formulated product/ 100 L water and Curvax at 300 g of formulated product/ 100 liters water) to control the late blight, *Phytophthora infestans* for all plots. Tomato fruits were harvested on 11 May, 17 May and 21 May 2012. Thirty fruits were picked at each date and inspected for the presence of *T. absoluta* larval entry holes.

For mass trapping, pheromone traps were set up in April and May 2012 (Table 1). In greenhouse 1, before the beginning of the trial, one water trap was put approximately in the center for the monitoring of the insect on 8 March, 2012 and checked twice a week to make a decision when to begin mass trapping trials. The trap was removed on 6 April 2012.

Water traps as described above and Delta traps were used. Delta traps were commercial traps with sticky inserts. They were white in color and each one had a sticky surface of about 420 cm² with two main delta shaped entrance approximately 300 cm² each. The pheromone dispenser was placed on the sticky surface approximately in the middle. A total of four water traps and 8 delta traps at two heights (4 traps at 0.4 m, and 4 traps at 1.5 m above the ground) were setup in April and May (table I). The distance between traps varied from 6

and 9 m (Fig. 2). A third greenhouse with the same tomato variety was chemically sprayed and used as a positive control. It was not possible to allocate a control greenhouse (without sprays) due to the high value of this crop. The details of the three greenhouses are given in table I.

Table I: Greenhouse parameters, trapping and treatment data on tomato leafminer

	Greenhouse 1	Greenhouse 2	Control Greenhouse
Monitoring trap	1 water trap (8 March 2012)	No	No
Traps set up	9 April 2012 (12 traps)	17 May 2012 (12 traps)	No
Insecticide spays	- One spray of Chlorpyrifos -ethyl (Pyrical 480 at 100 ml per 100 l water) one day after transplanting to control Noctuid larvae	- One spray of Chlorpyrifos -ethyl (Pyrical 480 at 100 ml per 100 l water) one day after transplanting to control Noctuid larvae - 1 spray before the set up of mass trapping with spinosad, (Tracer at 50 ml per 100 l) water on 23 April 2012	- One spray of Chlorpyrifos -ethyl (Pyrical 480 at 100 ml per 100 l water) one day after transplanting to control Noctuid larvae - 4 sprays on 23 April 2012, on 4 May, on 17 May and on 28 May 2012) with Indoxacarb (avaunt at 50 ml per 100 l water) Triflumuron (Alystin at 50 ml per 100 l water), emamectin benzoate (Proclaim at 30 ml per 100 l water) and spinosad (Tracer at 50 ml per 100 l water)

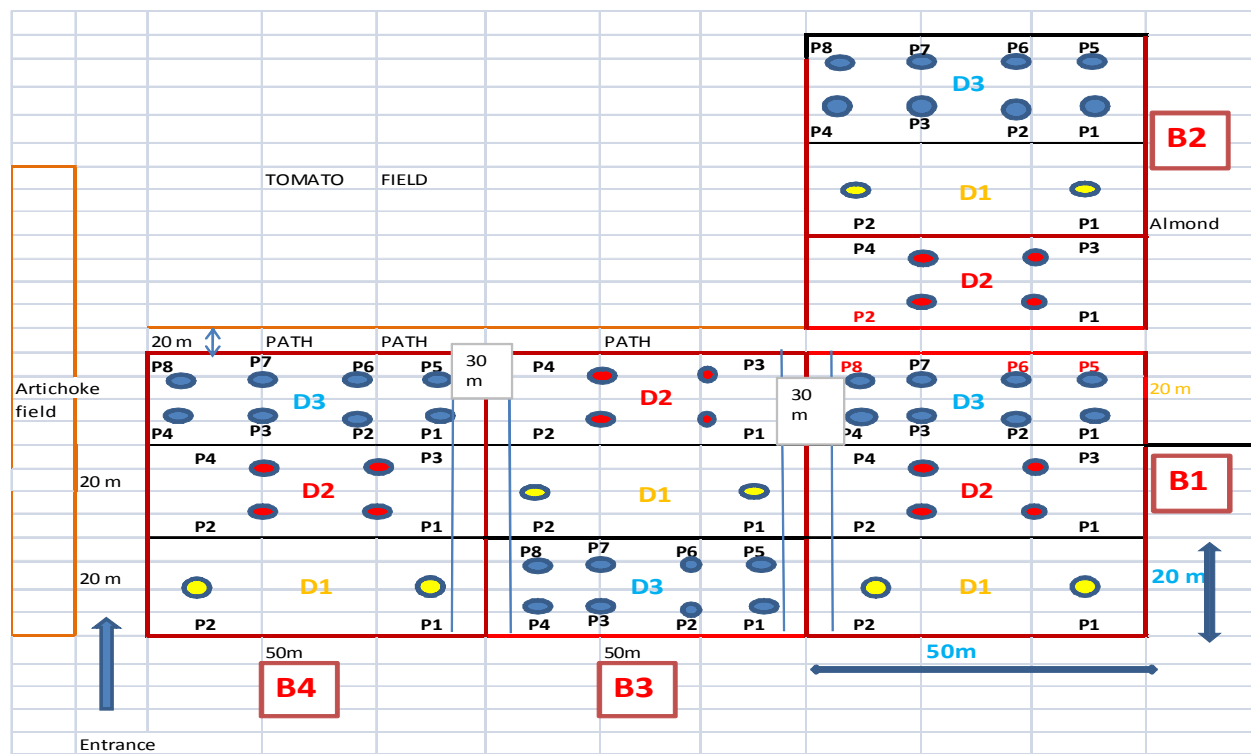


Fig.1: Schematic representation of placement of the traps used for *T. absoluta* in an open field tomato.

B: denotes block number
 P: denotes trap

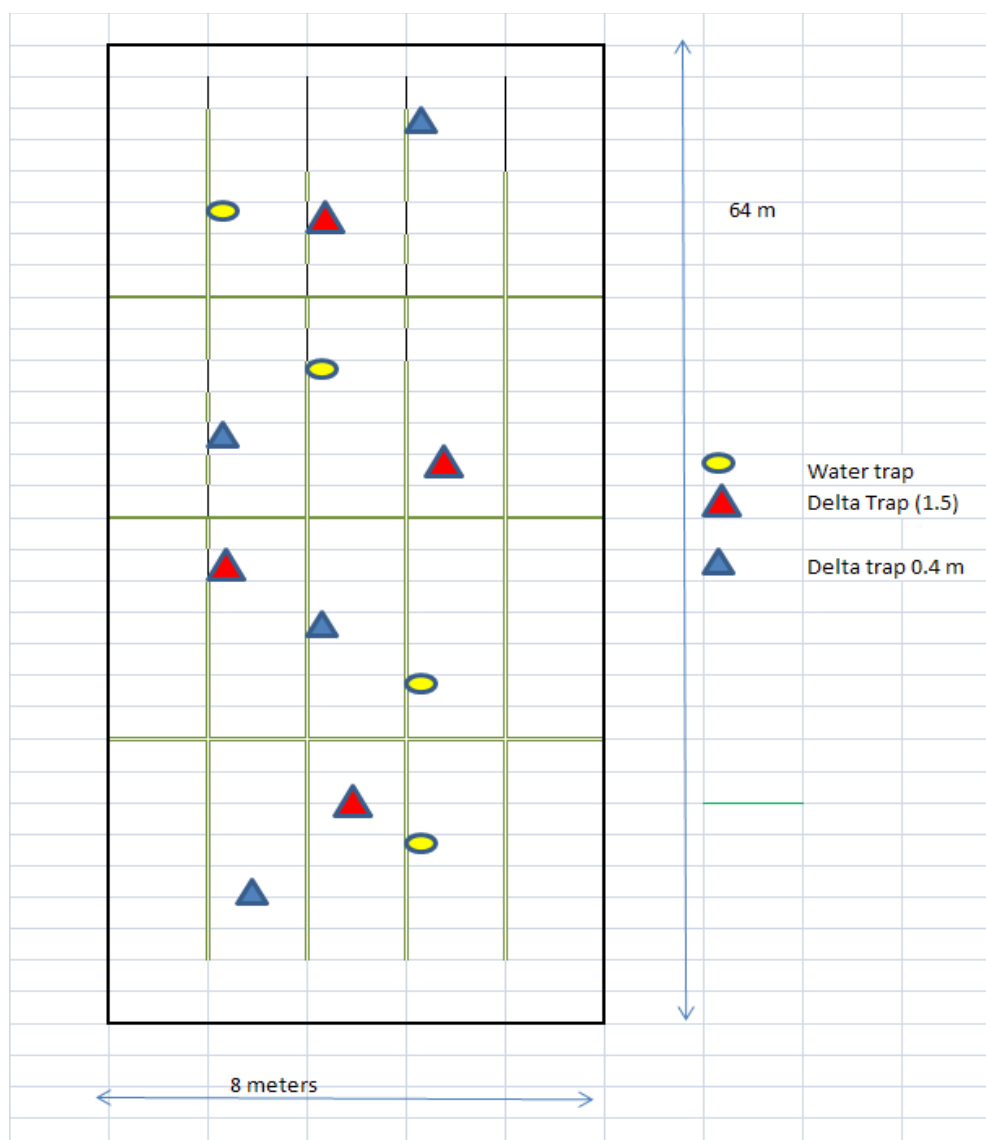


Fig.2: Lay out of the greenhouse used for *T. absoluta* male mass-trapping.

2.3. Data analysis

Trap counts were transformed to $\log(x+1)$ to reduce heterogeneity of variances, then data were submitted to a one-way analysis of variance (ANOVA) using the software SPSS 17.0 (2008). The dependant variable was the number of trapped moths per trap and the independent variable was trap densities. In addition Pearson's correlation analysis was used to correlate total moth capture per week according to trap densities.

The densities of eggs, larvae, empty mines per leaf relating to trap densities or between, treatments were subjected to one way ANOVA after data transformation to meet the normality assumption. For all analysis, treatments were compared using Fisher's protected least significant difference (LSD at $P < 0.05$).

3. RESULTS

3.1. Open field tomato

3.1.1. Male *T. absoluta* flight pattern Dynamics of population density of male *T. absoluta*

The average number of male captured per trap per inspection date varied between 13 and 52. The flight activity of the male moths seems to be moderate to high during the study period (from May to July) without real distinction between generations (Fig. 3).

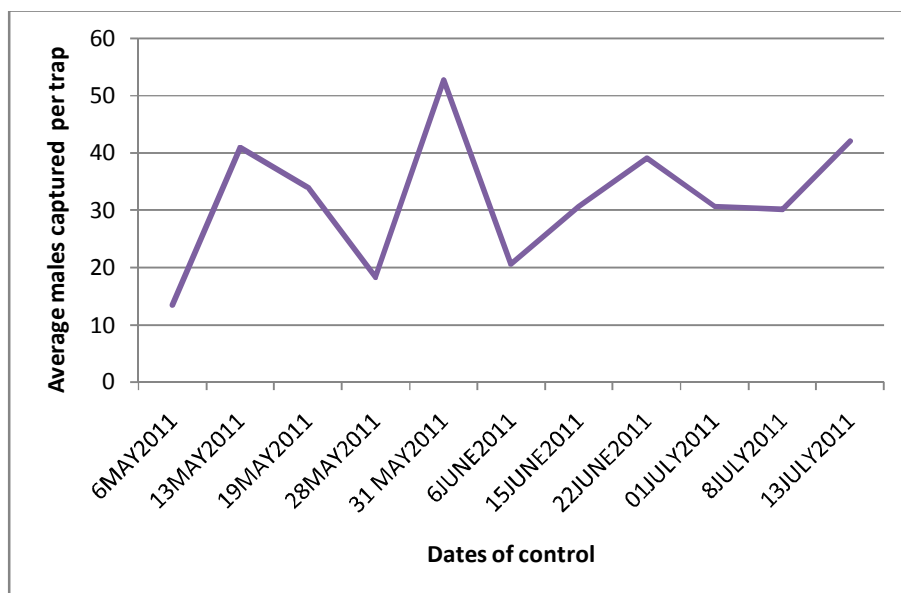


Fig.3: Male flight activity of *T. absoluta* recorded in different trap densities (open field tomato, 2011).

3.1.2. Average number of captured males

The mean numbers of captured males per inspection date were 31.27 ± 16.84 for D1; 31.51 ± 12.59 for D2 and 33.26 ± 14.20 for D3. Generally, there is no significant difference between trap densities regarding the mean number of males captured per water trap (One- way analysis of variance; $F = 2.97$; $P = 0.060$, $df = 2, 53$; Fig 4.). Indeed, for each inspection date, there are no significant differences between trap densities ($P > 0.05$; table II).

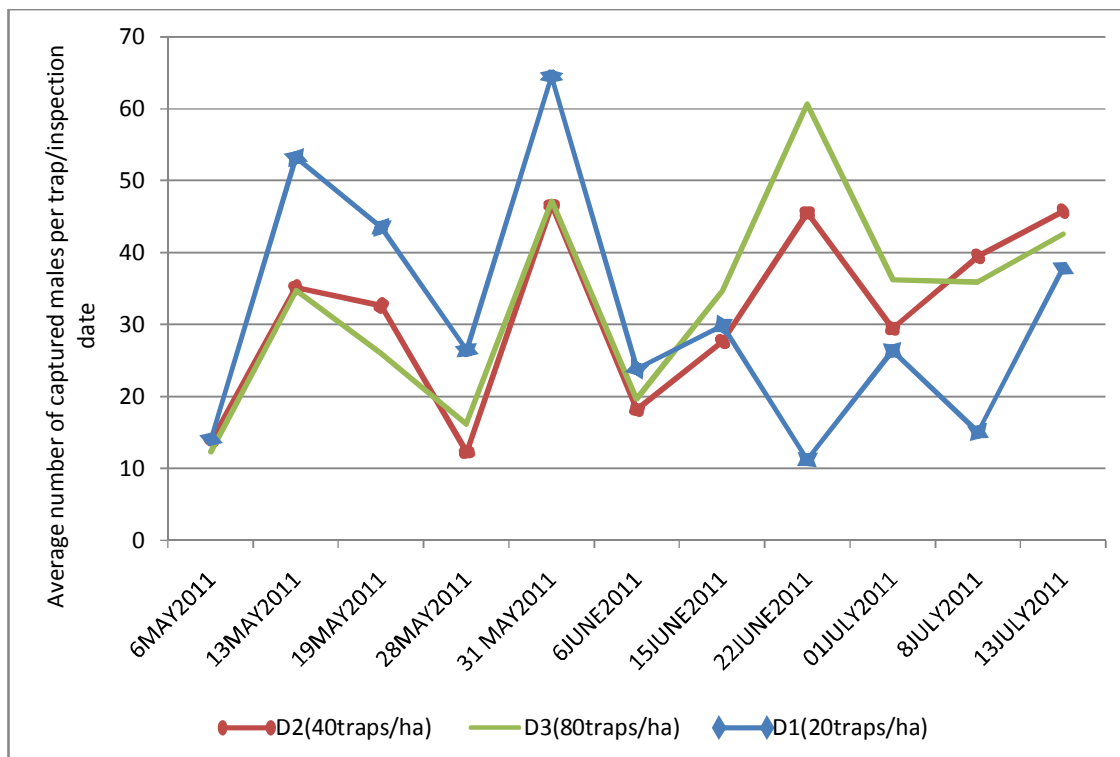


Fig.4: Average number of captured males per trap /inspection date.

Table II: Average number of captured *T. absoluta* males according to inspection dates

Trap densities	Mean number of males per trap										
	6 May 2011	13May 2011	19May 2011	28May 2011	31May 2011	6June	15June	22June	1July	8July	13July
D1 (20 traps/ha)	14.13 ±11.82	53.25 ±63.51	43.50± 45.10	26.50± 26.77	64.50± 48.5	23.88 ±18.38	29.9± 21.8	11.2±2 1.2	26.3± 47.9	15.1 ±15.6	39.4 ±25.3
D2 (40 traps/ha)	13.88 ±10.06	35.13 ±39.26	32.69± 19.28	12.25± 6.33	46.63± 27.04	18.19 ±10.70	27.3± 16.1	45.5±6 0.8	29.4± 40.9	39.5 ±35.5	45.8 ±35.8
D3 (80 traps/ha)	12.31 ±15.50	34.69 ±27.59	26.06± 30.62	16.56± 11.53	47.16± 32.27	19.7± 31.3	34.5± 29.3	60.7±9 6.2	36.2± 46.6	35.9 ±26.8	42.6 ±26.4
Statistical test	F= 0.081 P= 0.92; df 2, 53	F= 0.954 P= 0.392 ; df 2, 53	F= 1.11 P= 0.335; df 2, 53	F= 2.97 P= 0.06; df 2, 53	F= 0.93 P= 0.4; df 2, 53	F= 0.13 P= 0.87; df 2, 53	F= 0.42 P= 0.65; df 2, 53	F= 1.22 P= 0.30; df 2, 53	F= 0.21 P= 0.8; df 2, 53	F= 2.13 P= 0.12 ; df 2, 53	F= 0.11 P= 0.89 ; df 2, 53

It seems that at low population density (from 6 May to 15 June 2011), the number of captured males per trap is higher for D1 compared with D2 and D3. But at moderate to high population density, more moths were captured in D2 and D3 (Fig. 4).

3.1.3. Total number of captured males

The total number of captured males at different trap densities varies significantly; more moths were captured in plots with high trap density (Table III, Fig. 5). When comparing trap densities, there are significant differences (D1 versus D2, ANOVA $F = 11.92$ $df = 1, 20$; $P = 0.003$), D2 versus D3 $df (1, 20)$ $F = 13.97$; $P = 0.001$); D1 versus D3 $df (1, 20)$ $F = 32.93$ $P = 0.001$). Pearson's correlation values show positive correlation between trap densities (Table III).

Table III: Total number of captured male moths in relation to trap densities and Pearson's correlation coefficients

Densities	Total number of captured males per inspection date ± SD*	Pearson's correlation coefficients
D1	252 ± 134.80	D1- D2; $R = 0.611$; $P = 0.003$
D2	504 ± 201.50	D2- D3 $R = 0.641$; $P = 0.01$
D3	1065 ± 454.40	D1- D3; $R = 0.786$; $P = 0.000$

*for a total of 8 traps for D1, 16 traps for D2 and 32 traps for D3

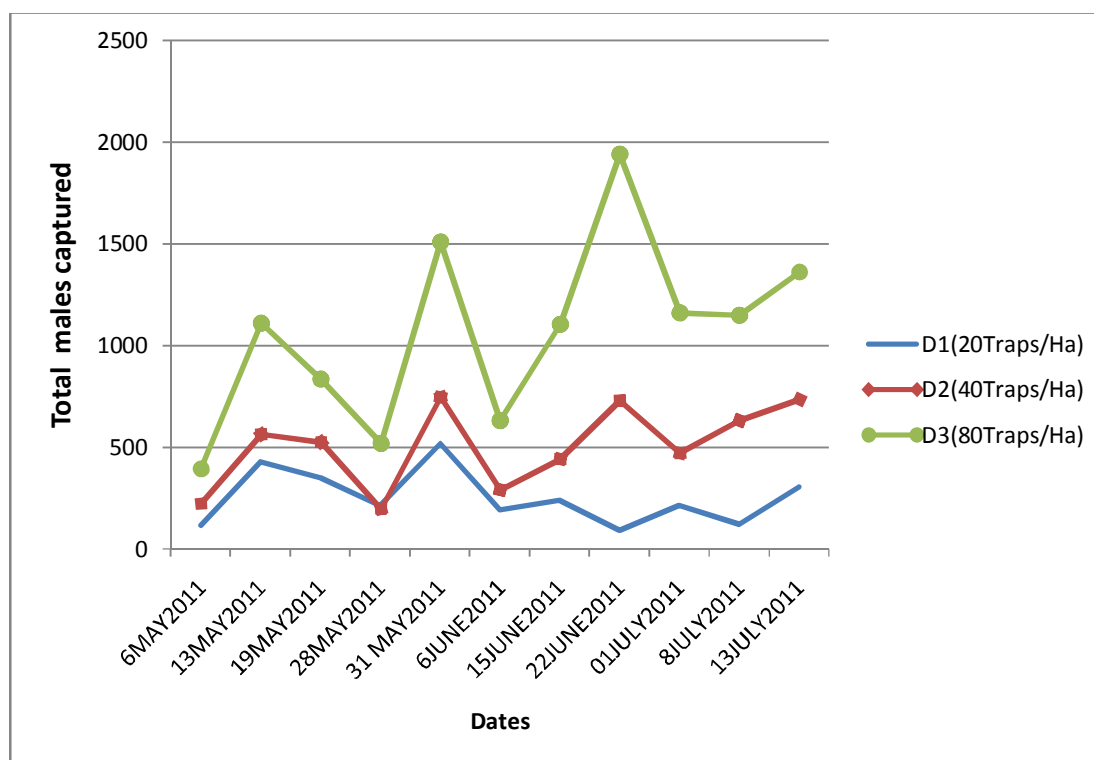


Fig. 5: Total number of *T. absoluta* moths captured in water traps at different trap densities (for a total of 8 traps for D1, 16 traps for D2 and 32 traps for D3).

3.1.4. Leaf infestation

On the whole, when we consider weekly sampling as the repeated factor, the densities of eggs, larvae and *T. absoluta* mines do not statistically vary between the three tested densities (Eggs: $F_{2, 27} = 0.42$; $P = 0.65$. Larvae: $F_{2, 27} = 0.02$; $P = 0.97$. Empty mines: $F_{2, 27} = 0.15$; $P = 0.86$) (Table IV). A further more detailed data for each sampling date (tables V.a to V.i) show no significant differences between trap densities except the sampling on 13 May (old larvae table V.a) and 15 June 2011 for empty mines (table V.e).

Table IV: Mean number of eggs, larvae, empty mines and fruit infestation percentages at different trap densities

Trap densities	Eggs SD (Min-Max)	Larvae SD (Min-Max)	Empty mines* SD	Fruit infestation SD (%) ⁽¹⁾
D1(20Traps/ha)	0.050.02 (0.02-0.1)	0.07+0.09 (0-0.3)	0.13+0.09 (0-0.27)	0.21+0.07 (0.17-0.33)
D2(40Traps/ha)	0.07+0.04 (0-0.12)	0.07+0.07 (0-0.2)	0.16+0.14 (0-0.5)	0.18+0.03 (0.15-0.23)
D3(80 Traps/ha)	0.06+0.03 (0.02-0.12)	0.08+0.08 (0-0.24)	0.14+0.11 (0-0.32)	0.17+ 0.04 (0.11-0.22)
Statistical analysis	$F = 0.427$; $df = 2, 27$; $P = 0.657$	$F = 0.026$; $df = 2, 27$; $P = 0.975$	$F = 0.152$; $df = 2, 27$; $P = 0.86$	$F = 0.49$ $df = 2, 27$; $P = 0.626$

⁽¹⁾ Mean of four sampling dates (on 24 June, 1 July, 8 July and 13 July 2011)

* mines are either empty or with larvae which are counted as alive larvae

Table V.a: Mean number of alive eggs, larvae of *T. absoluta* and empty mines per tomato leaflet at different trap densities

13 May 2011					
Trap Densities	Eggs \pm SD	Small larvae \pm SD	Old larvae \pm SD*	Total(eggs-larvae) \pm SD	Empty mines \pm SD
D1 (20traps/ha)	0.03 \pm 0.15	0.05 \pm 0.31	0b	0.08 \pm 0.35	0.20 \pm 0.64
D2(40 traps/ha)	0	0.08 \pm 0.26	0b	0.08 \pm 0.26	0.48 \pm 1.0
D3(80 traps/ha)	0.03 \pm 0.22	0.03 \pm 0.15	0.08 \pm 0.26a	0.16 \pm 0.36	0.33 \pm 0.82
Statistical analysis	F=1.07; P= 0.36 df (2,117)	F=0.38; P= 0.68 df (2,117)	F=3.16; P= 0.046 df (2,117)	F=1.07; P= 0.36 df (2,117)	F=0.69; P= 0.5; df (2,117)

*Denotes Significant difference at P<0.05)

Table V.b: Mean number of alive eggs, larvae of *T. absoluta* and empty mines per tomato leaflet at different trap densities

19 May 2011					
	Eggs \pm SD	Small larvae \pm SD	Old larvae \pm SD	Total(eggs-larvae) \pm SD	Empty mines \pm SD
D1 (20traps/ha)	0.10 \pm 0.30	0.05 \pm 0.22	0.03 \pm 0.15	0.20 \pm 0.51	0.38 \pm 1.17
D2(40 traps/ha)	0.13 \pm 0.46	0.10 \pm 0.37	0.05 \pm 0.22	0.28 \pm 0.6	0.10 \pm 0.37
D3(80 traps/ha)	0.13 \pm 0.40	0.23 \pm 0.82	0.03 \pm 0.15	0.38 \pm 1	0.28 \pm 0.84
Statistical analysis	F=0.05; P= 0.94 df (2,117)	F=1.1; P= 0.33 df (2,117)	F=0.25; P= 0.77 df (2,117)	F=0.56; P= 0.56 df (2,117)	F=1.04; P= 0.35 df (2,117)

Table V.c: Mean number of alive eggs, larvae of *T. absoluta* and empty mines per tomato leaflet at different trap densities

28 May 2011					
	Eggs \pm SD	Small larvae \pm SD	Old larvae \pm SD	Total(eggs-larvae) \pm SD	Empty mines \pm SD
D1 (20traps/ha)	0.08 \pm 0.26	0.08 \pm 0.26	0.03 \pm 0.15	0.18 \pm 0.44	0.18 \pm 0.50
D2(40 traps/ha)	0.05 \pm 0.22	0.15 \pm 0.24	0	0.20 \pm 0.51	0.30 \pm 1
D3(80 traps/ha)	0.05 \pm 0.22	0.05 \pm 0.31	0.03 \pm 0.15	0.13 \pm 0.40	0.20 \pm 0.60
Statistical analysis	F=1.14; P= 0.86 df (2,117)	F=0.92; P= 0.40 df (2,117)	F=0.5; P= 0.60 df (2,117)	F=0.27; P= 0.75 df (2,117)	F=0.29; P= 0.74 df (2,117)

Table V.d: Mean number of alive eggs, larvae of *T. absoluta* and empty mines per tomato leaflet at different trap densities

6 June 2011					
	Eggs \pm SD	Small larvae \pm SD	Old larvae \pm SD	Total(eggs-larvae) \pm SD	Empty mines \pm SD
D1 (20traps/ha)	0.05 \pm 0.22	0.03 \pm 0.15	0	0.08 \pm 0.44	0
D2(40 traps/ha)	0.05 \pm 0.22	0.03 \pm 0.11	0	0.08 \pm 0.26	0
D3(80 traps/ha)	0.08 \pm 0.26	0	0.03 \pm 0.15	0.1 \pm 0.37	0.3 \pm 0.15
Statistical analysis	F=0.11; P= 0.89 df (2,117)	F=0.5; P= 0.60 df (2,117)	F=1; P= 0.37 df (2,117)	F=0.08; P= 0.91 df (2,117)	F=1; P= 0.37 df (2,117)

Table V.e: Mean number of alive eggs, larvae of *T. absoluta* and empty mines per tomato leaflet at different trap densities

15 June 2011					
	Eggs ±SD	Small larvae ±SD	Old larvae ±SD	Total(eggs- larvae) ±SD	Empty mines ±SD*
D1 (20traps/ha)	0.03±0.15	0	0.03±0.15	0.05±0.22	0.13±0.33a
D2(40 traps/ha)	0.05±0.22	0	0	0.05±0.22	0
D3(80 traps/ha)	0.08±0.26	0	0.03±0.15	0.1±0.30	0.3±0.15a
Statistical analysis	F=0.59; P= 0.59 df (2,117)		F=0.5; P= 0.60 df (2,117)	F=0.52; P= 0.52 df (2,117)	F=3.8; P= 0.025 df (2,117)

Table V.f: Mean number of alive eggs, larvae of *T. absoluta* and empty mines per tomato leaflet at different trap densities

22 June 2011					
	Eggs ±SD	Small larvae ±SD	Old larvae ±SD	Total(eggs- larvae) ±SD	Empty mines ±SD
D1 (20traps/ha)	0.05±0.22	0	0	0.05±0.22	0.08±0.26
D2(40 traps/ha)	0.10±0.30	0	0	0.10±0.30	0.05±0.22
D3(80 traps/ha)	0.3±0.14	0	0.3±0.15	0.05±0.22	0.08±0.26
Statistical analysis	F=1; P= 0.35 df (2,117)		F=1; P= 0.37 df (2,117)	F=0.52; P= 0.59 df (2,117)	F=0.13; P= 0.87 df (2,117)

Table V.g: Mean number of alive eggs, larvae of *T. absoluta* and empty mines per tomato leaflet at different trap densities

1 July 2011					
	Eggs ±SD	Small larvae ±SD	Old larvae ±SD	Total(eggs- larvae) ±SD	Empty mines ±SD
D1 (20traps/ha)	0.05±0.22	0.03±0.15	0	0.08±0.26	0.10±0.30
D2(40 traps/ha)	0.05±0.22	0	0	0.05±0.22	0.15±0.36
D3(80 traps/ha)	0.05±0.22	0	0	0.05±0.22	0.08±0.26
Statistical analysis	F=0; P= 1 df (2,117)	F=1; P=0.37 df (2,117)		F=0.14; P= 0.86 df (2,117)	F=0.59; P= 0.55 df (2,117)

Table V.h: Mean number of alive eggs, larvae of *T. absoluta* and empty mines per tomato leaflet in relation to trap densities

8 July 2011					
	Eggs ±SD	Small larvae ±SD	Old larvae ±SD	Total(eggs- larvae) ±SD	Empty mines ±SD
D1 (20traps/ha)	0.08±0.26	0	0	0.08±0.26	0.25±0.49
D2(40 traps/ha)	0.10±0.30	0	0	0.10±0.22	0.15±0.42
D3(80 traps/ha)	0.03±0.15	0	0	0.03±0.22	0.08±0.26
Statistical analysis	F=0.92; P= 0.39 df (2,117)			F=0.59; P= 0.55 df (2,117)	F=1.86; P= 0.16 df (2,117)

Table V.i: Mean number of alive eggs, larvae of *T. absoluta* and empty mines per tomato leaflet at different trap densities

13 July 2011					
	Eggs ±SD	Small larvae ±SD	Old larvae ±SD	Total(eggs- larvae) ±SD	Empty mines ±SD
D1 (20traps/ha)	0.1±0.30	0.23±0.42	0.08±0.26	0.40±0.59	0.28±0.55
D2(40 traps/ha)	0.05±0.22	0.20±0.40	0	0.26±0.43	0.20±0.40
D3(80 traps/ha)	0.08±0.22	0.20±0.40	0	0.28±0.50	0.25±0.43
Statistical analysis	F=0.35; P= 0.70 df (2,117)	F=0.05; P=0.95 df (2,117)	F=3.16; P=0.046 df (2,117)	F=0.97; P= 0.30 df (2,117)	F=0.26; P= 0.76 df (2,117)

3.1.5. Fruit infestation

On average, the number of infested fruits did not vary in relation to trap densities ($F = 0.49$ $df(2,9)$; $P = 0.62$). However, D3 seems to be the least infested (Fig 6.).

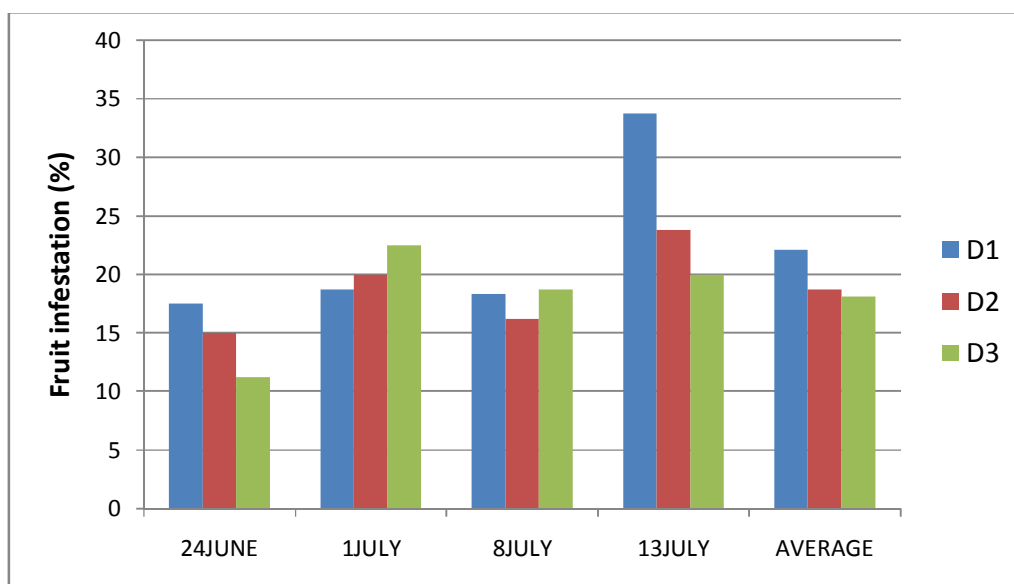


Fig. 6: Mean number of fruits infested by *T. absoluta* larvae.

3.2. Greenhouse experiments

3.2.1. Male capture

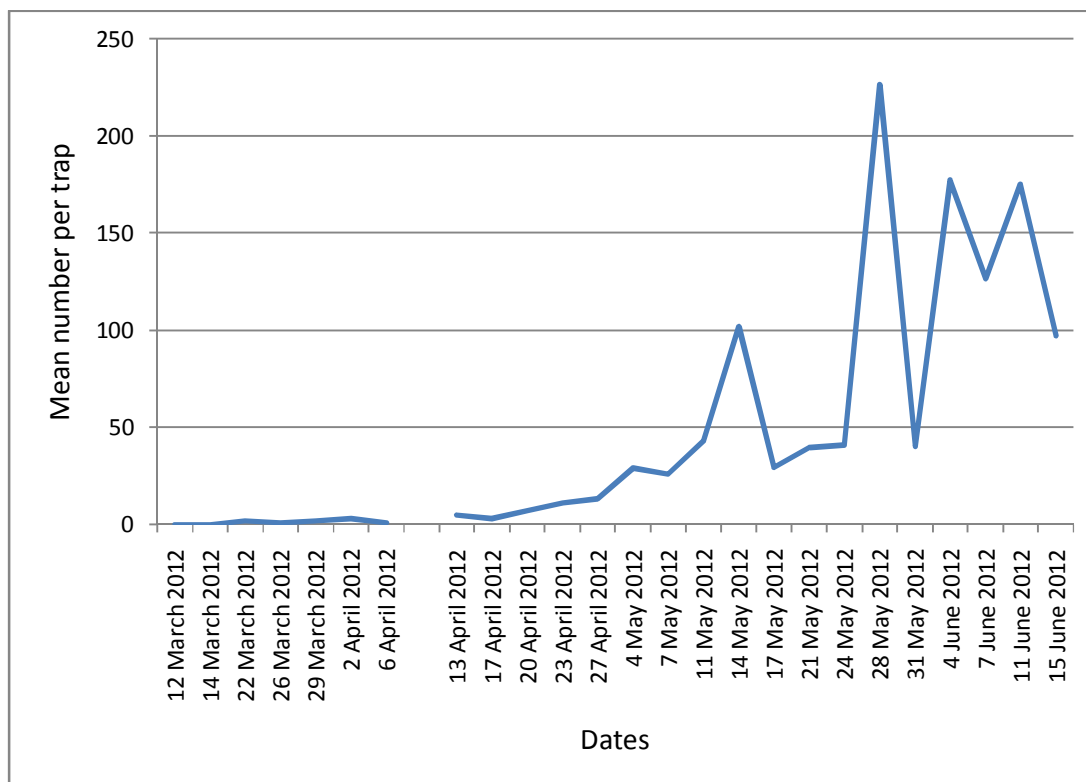


Fig. 7: Number of moth captured in water trap before the beginning of the experiment in greenhouse 1.

In greenhouse 1, the first 2 adults were captured on 22 March 2012 in monitoring water trap. Then the number of trapped males varied from 1 to 3 per trap per inspection date until 6 April 2012 (Fig 7.). The population dynamic of the adult demonstrated that the flight activity begins from the second decade of April until late June in correlation with the increasing of temperature and tomato fruiting period. The two kinds of traps (delta and water) functioned well and captured large number of males (Fig. 8).

For the greenhouse 2, traps used for mass trapping experiments were set relatively late (on 17 May 2012). Insect flight was concentrated in May and June (Fig. 9).

For the greenhouse 2, traps used for mass trapping experiments were set relatively late (on 17 May 2012). Insect flight was concentrated in May and June (Fig. 9).

For the two greenhouses, both trap kind (delta and water traps) functioned very well, there is no real distinction between them regarding the number of trapped moths (Fig 10).

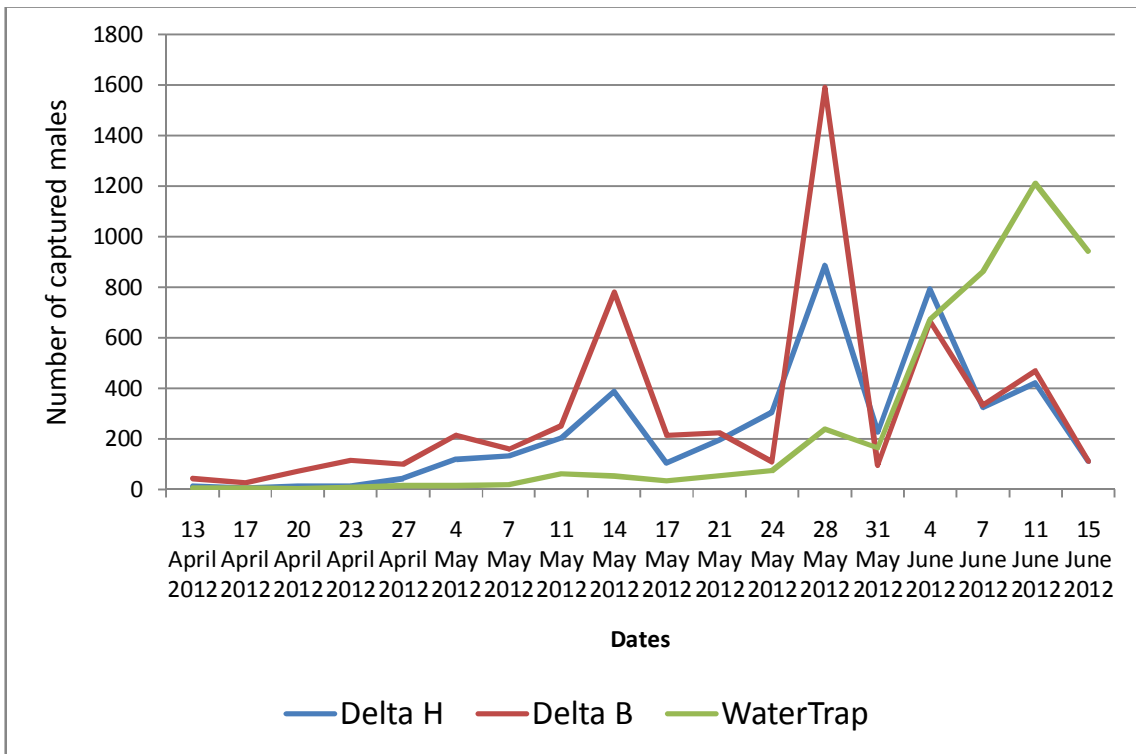


Fig. 8: Total number of captured males in tomato Greenhouse 1.

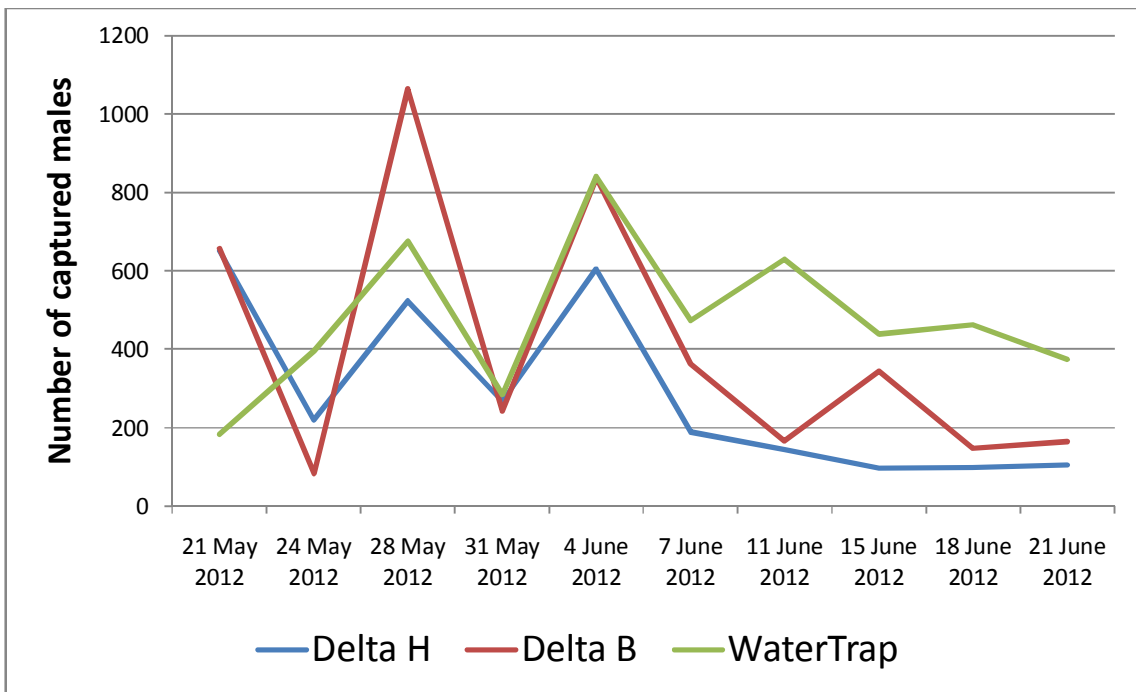


Fig. 9: Total number of captured males (greenhouse 2).

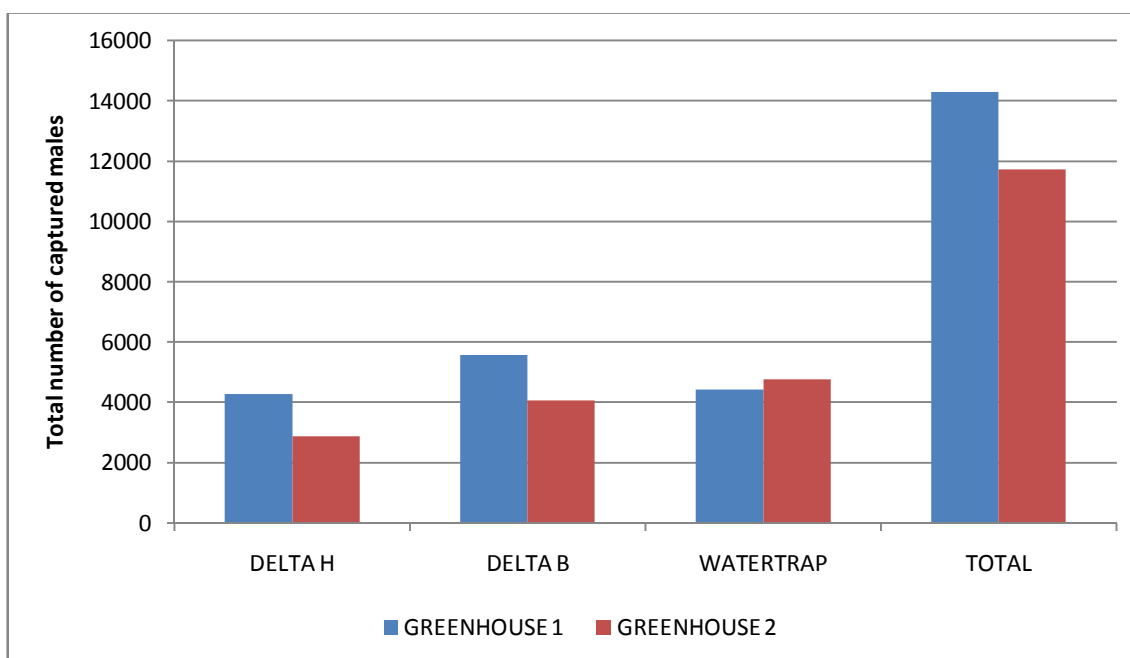


Fig. 10: Total number of male captured in the two greenhouses used for mass trapping experiments.

3.2.2. Leaf infestation

For the tree sampling dates, there are no significant differences in relation to the densities of eggs, larvae and empty mines except for larvae on 21 May 2012 (tables VI.a, VI.b and VI.c).

Table VI.a: Mean number of eggs, larvae and empty mines per tomato leaflet (sampling on 17 May 2012)

17 May 2012			
Greenhouses	Eggs \pm SD (Min-Max) ⁽¹⁾	Larvae \pm SD (Min-Max) ⁽¹⁾	Empty mines ⁽¹⁾ \pm SD
Greenhouse 1	0.07 \pm 0.25(0-1)	0.07 \pm 0.25(0-1)	0.63 \pm 0.86(0-3)
Greenhouse 2	0.07 \pm 0.25(0-1)	0.17 \pm 0.46(0-2)	0.43 \pm 0.67(0-2)
Control Greenhouse	0.06 \pm 0.24(0-1)	0.13 \pm 0.34(0-1)	0.77 \pm 0.58(0-2)
Statistical analysis	F= 0.8; df = 2, 87; P= 0.97	F= 2; df = 2, 87; P= 0.131	F= 2.87; df = 2, 87; P= 0.58

Table VI.b: Mean number of eggs, larvae and empty mines per tomato leaflet (sampling of 21 May 2012)

21 May 2012			
Greenhouses	Eggs \pm SD (Min-Max) ⁽¹⁾	Larvae \pm SD (Min-Max) ⁽¹⁾	Empty mines ⁽¹⁾ \pm SD
Greenhouse 1	0.07 \pm 0.25(0-1)	0.27 \pm 0.45(0-1)	0.50 \pm 0.82(0-3)
Greenhouse 2	0.07 \pm 0.18(0-1)	0.07 \pm 0.25(0-1)	0.60 \pm 0.77(0-3)
Control Greenhouse	0	0.07 \pm 0.25(0-1)	0.20 \pm 0.61(0-3)
Statistical analysis	F= 1.02; df = 2, 87; P= 0.36	F= 3.62; df = 2, 87; P= 0.031	F= 2.38; df = 2, 87; P= 0.09

Table VI.c: Mean number of eggs, larvae and empty mines per tomato leaflet (sampling of 28 May 2012)

28 May 2012			
Greenhouses	Eggs \pm SD (Min-Max) ⁽¹⁾	Larvae \pm SD (Min-Max) ⁽¹⁾	Empty mines ⁽¹⁾ \pm SD
Greenhouse 1	0	0.17 \pm 0.46(0-2)	0.77 \pm 0.62(0-2)
Greenhouse 2	0.03 \pm 0.18(0-1)	0.13 \pm 0.34(0-1)	0.53 \pm 0.81(0-3)
Control Greenhouse	0.03 \pm 0.18(0-1)	0.07 \pm 0.25(0-1)	0.33 \pm 0.60(0-2)
Statistical analysis	F = 0.50; df = 2, 87; P = 0.60	F = 0.58; df = 2, 87; P = 0.55	F = 2.98; df = 2, 87; P = 0.06

3.2.2. Fruit infestation

There is no significant difference between mass trapping technique and chemical control alternative. (F = 0.219 df(2,6) P = 0.80) In average, the percentage of fruit infestations were respectively 16.66 %; 23.80 % and 44.44% in the first greenhouse 18.75 %; 6.66 % and 35 % for the second greenhouse and 14.28 %; 15.38 % and 41.66 % for the control greenhouse managed chemically (Fig 11).

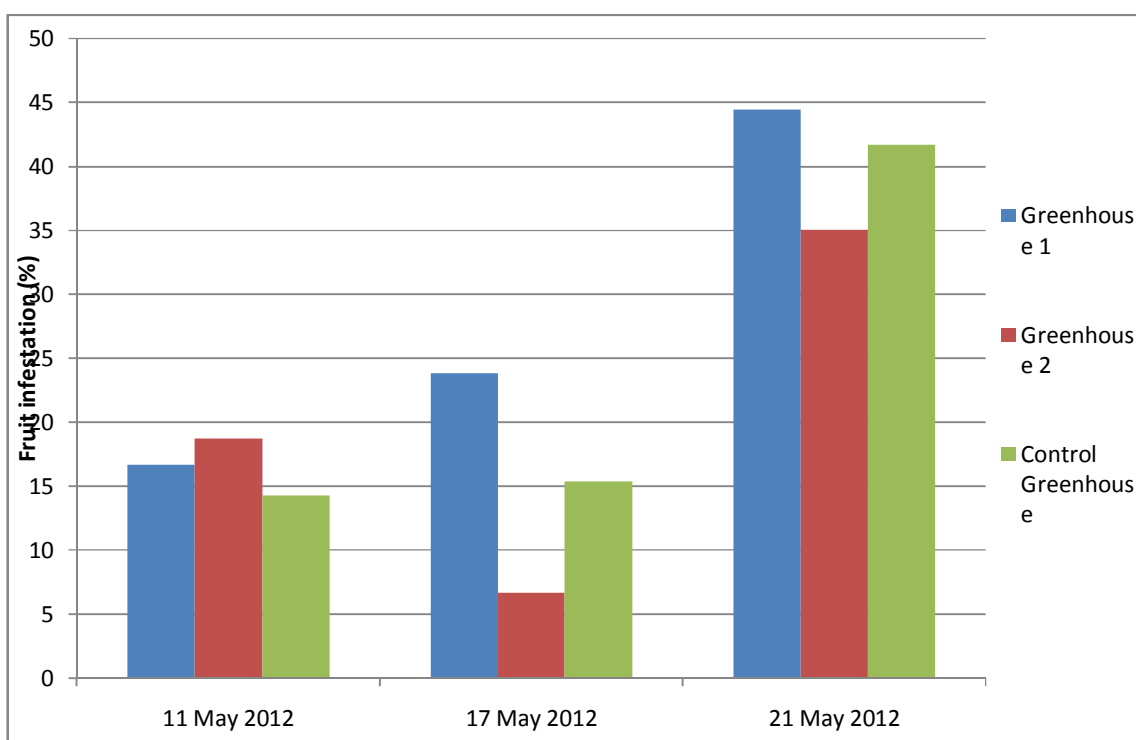


Fig. 11: Percentage of fruit infestation (greenhouse trials).

4. DISCUSSION

The main objective of the present study was to assess whether male mass trapping results in a decrease of *T. absoluta* male abundance and a subsequent reduction in tomato leaf and fruit damages in two main tomatoe cropping systems in Tunisia; open field un-staked and greenhouse tomatoes. Our study was motivated by the increasing use of pheromone lures of *T. absoluta* for monitoring and mass trapping in the world; according to Witzgall *et al.* (2010), the estimated number of *T. absoluta* pheromone lures is 2 millions used each year.

Mass-trapping using attractants is a method of pest control experimented for several insects. El-Sayed *et al.* (2006) cited more than 100 studies in the literature. The technique of mass trapping with pheromone had been widely used for the control of different insect species (Howse *et al.*, 1998). However, unlike Coleopteran and Dipteran species, only a few examples of successful application of pheromone baited mass trapping for Lepidopteran species had been reported; for example, Sternlicht and Tamin (1990) reported that male mass

trapping of *Prays citri* was more effective than insecticide control; the most effective treatment being 120 traps per ha. Also, Mafra-Neto and Habib (1996) used mass trapping technique to control the pink bollworm, *Pectinophora gossypiella* (Saunders) (Lepidoptera: Gelechiidae) populations in cotton fields in Brazil. Oil traps containing lures with a high dose of pheromone (0.2 g per ha), installed at a density of 20 traps per ha soon after the occurrence of the first cotton bolls, suppressed pink bollworm populations below economic injury levels using Delta traps. Other authors suggested the need to associate mass trapping to other techniques to achieve good control (Khan et al., 2005). For example, Raman (1988) reported that although mass trapping of the potato tuber moth *Phthorimaea operculella* is successful in potato field and at storage, it should be supplemented by other means of control, especially pre-harvest control measures.

In tomato open field, the mean number of males captured per inspection date did not vary between trap densities. However the total number varied suggesting that the increase in the number of trap did increase male capture. The inter-trap distances can affect the trap capture due to competition among traps that are placed at short distance (Bacca et al., 2006). The interference between traps in this study was not clearly noticed. If interference works, the increase in trap density would decrease trap capture. Also, it is possible that *T. absoluta* females can be captured in water traps due to the proximity of traps; until now, there is no report on female trapping in such traps or females captured in such traps are ignored because of the specific nature of the pheromone and the difficulty to distinguish male from female in the field. High trap density meant high concentration of pheromone plumes suggesting a possible effect of mating disruption not only in plots with high trap densities but also in the other plots.

For greenhouse trails, on the whole, there is no significant difference in fruit infestation between tomato greenhouses used for mass trapping and control (chemically sprayed greenhouse). However, economically mass trapping is advantageous since a single chemical spray costs between 10 and 15 US \$ per tomato greenhouse. For mass trapping, one trap costs about 1 US\$ (can be reutilized for several seasons) and pheromone capsules are sometimes free of charge (or purchased for about 0.6 US \$ each). Other advantages are related to the shortage of workers in agriculture, the increasing cost of working force and no chemical residue in fruits. So, the introduction of mass trapping as part of the integrated control program would be improved because the technique is environmentally friendly, efficient, non-poisonous and non-hazardous to natural enemy populations.

Successful examples of mass trapping to control Lepidopterous pests have targeted isolated low-density populations (Madsen and Carty, 1979). It may be important to implement mass trapping of *T. absoluta* at the beginning of the flight activity when populations are low.

For Lepidoptera insects, pheromone traps capture adults and often only males, thus trapping information is to be used as a predictive manner to quantify damage caused by the next generation of larvae. With female-produced sex pheromone only males are caught. Since male insects typically mate more than once, a high proportion of the male population must be removed to produce an effect.

The relative efficiency of pheromone traps depends on factors such as proper placement of traps (McNeil, 1991). The success of mass trapping technique using water basin traps depends on the isolation of the site in order to reduce the effect of immigration of adults particularly of gravid females from adjacent fields.

Even if large numbers of male individuals can be caught by coupling pheromone releasers with use of insect trapping devices, the success of pheromone-based control strategies is usually low. One of the hypotheses of this failure could be that the insect used asexual or parthenogenetic reproduction.

Under open field conditions, the mass trapping technique is a labor-intensive technique needing between 5 and 8 minutes for maintenance of a single trap (adding water, removing insects, putting pheromone capsule during the season). Nevertheless, this method provides a good alternative to conventional insecticide application eliminating insecticide residues in fruits and preserving natural enemy populations.

The mass trapping technique of Lepidoptera species is based on an important biological trait: the insect must breed through sexual reproduction.

Although, large numbers of male individuals were caught in pheromone traps (more than 14000 males in the greenhouse 1, almost 14000 in greenhouse 2 and 20027 in open field), leaf infestation and particularly fruit infestation are relatively high (Fig. 6 and 11). There is no clear relationship between trap capture and leaf/fruit infestations. Two hypotheses may explain this (1) the insect uses asexual or parthenogenetic reproduction (Caparros et al., 2012) and the unfertilized females can lay viable eggs; (2) the non-isolation of tomato field permitting insect flying. In fact, the success of the mass trapping technique depends strongly on the isolation of the experimental plot. The isolation of the area reduces the effect of immigration of adults particularly of fertilized females from adjacent fields. In our studies, the treated and untreated plots were small and not isolated from each other; thus, migration of moths was possible and fertilized females could be introduced in from outside of the treated areas to lay eggs. The problem of insect migration from untreated to treated plots was also discussed by other researchers (Ioriatti and Angeli, 2002; Mazomenos et al., 2002). This probably could be overcome by increasing treatment area to cover the whole area of insect occurrence and/or by a preceding decrease of population density (by insecticide sprays) to a level appropriate for pheromone application.

The conclusion made is that mass trapping strategy demonstrates the need to apply this technique over an isolated field, in the whole area or under greenhouse conditions to minimize the effect of adult migration. More studies will be needed to evaluate the effectiveness of the mass trapping technique.

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