Response of *Tuta absoluta* Meyrick (Lepidoptera: Gelechiidae) to different pheromone emission levels in greenhouse tomato crops

Sandra Vacas, Jesús López, Jaime Primo and Vicente Navarro-Llopis

Centro de Ecología Química Agrícola – Instituto Agroforestal del Mediterráneo (CEQA-IAM), Universidad Politécnica de Valencia. Camino de Vera s/n, edificio 6C, 5ª planta. 46022 Valencia (Spain).

ABSTRACT

The response of *Tuta absoluta* (Lepidoptera: Gelechiidae) to different emission rates of its pheromone, (3*E,8Z,11Z*)-tetradecatrienyl acetate, was evaluated in two greenhouse trials with traps baited with mesoporous dispensers. For this purpose, weekly moth trap catches were correlated with increasing pheromone emission levels by multiple regression analysis. Pheromone release profiles of the dispensers were obtained by residual pheromone extraction and gas chromatography quantification. In the first trial carried out in summer 2010, effect of pheromone emission was significant as catches increased linearly with pheromone release rates up to the highest studied level of 46.8 μg/d. A new trial was carried out in spring 2011 to evaluate the effect of the emission factor when pheromone release rates were higher. Results demonstrated that trap catches and pheromone emission fitted to a quadratic model, with maximum catches obtained with a release level of 150.3 μg/d of (3*E,8Z,11Z*)-tetradecatrienyl acetate. This emission value should provide enhanced attraction of *T. absoluta* and improve mass trapping, attract-and-kill or monitoring techniques under greenhouse conditions in the Mediterranean area.

RESUMEN

En el presente trabajo se evalúa la respuesta de *Tuta absoluta* (Lepidoptera: Gelechiidae) frente a diferentes niveles de emisión de su feromona, acetato de (3E,8Z,11Z)-tetradecatrienilo, en dos ensayos de invernadero usando trampas cargadas con emisores mesoporosos. Para ello, los datos de polillas capturadas cada semana se correlacionaron con niveles crecientes de emisión de feromona mediante un análisis de regresión múltiple. La cinética de emisión de los emisores se estudió por extracción de la feromona residual y cuantificación posterior por cromatografía de gases. En el primer ensayo realizado en el verano de 2010 se obtuvo que el efecto de la emisión sobre las capturas era significativo y que la respuesta de *T. absoluta* aumentaba linealmente con la emisión de feromona hasta el nivel máximo estudiado de 46.8 μg/d. En la primavera de 2011 se realizó un nuevo ensayo para comprobar la respuesta frente a niveles de emisión más altos. Los resultados demostraron que la relación emisión-capturas se ajustaba a un modelo cuadrático, indicando la existencia de un máximo relativo de capturas correspondiente con un nivel de emisión de 150.3 μg/d de acetato de (3E,8Z,11Z)-tetradecatrienilo. Este valor podría emplearse para promover la atracción de *T. absoluta* y mejorar las técnicas de seguimiento de poblaciones y de atracción y muerte en condiciones de cultivo de invernadero en el área Mediterránea. Se discute el alcance de este tipo de estudios.

KEYWORDS: tomato leaf miner; monitoring; mass trapping; attract and kill; mesoporous dispenser; release rate

Tuta absoluta Meyrick (Lepidoptera: Gelechiidae), or tomato leaf miner (TLM), is an invasive pest considered an important threat for tomato production. Native to South America, it has been involved in the invasion and rapid colonization of the full length of the Mediterranean and South-Atlantic coasts of the Iberian Peninsula and the rest of European and North African Mediterranean Basin countries (Desneux et al. 2010). The exceptional speed with which it spreads suggests that *T. absoluta* will invade important exporting countries by 2016, such as USA and China (Desneux et al. 2011). For these reasons, control of *T. absoluta* has become a key issue for both outdoor and greenhouse crops. Controlling this pest entails repeatedly applying chemicals to affect the larvae when they are outside of galleries, which has led to pesticide resistance (Siqueira et al. 2000, 2001; Lietti et al. 2005). These insecticides could also affect natural enemies, thus making the consolidation of biological control systems very difficult. Thus, alternative means of suppressing TLM populations are needed and new IPM programmes could include other cultural, biotechnological and biological methods, such as application of entomopathogenic fungi or nematodes (Rodríguez et al. 2006, Batalla-Carrera et al. 2010), treatments with *Bacillus thuringiensis* Berliner (Giustolin et al. 2001, Theoduloz et al. 2003, Niedmann and Meza-Basso 2006, González-Cabrera et al. 2011), use of new biological control agents for *T. absoluta* (Urbaneja et al. 2009), or their combinations (Mollá et al. 2011), as well as techniques based on pheromones.

It has been demonstrated that virgin TLM females release a sex pheromone that strongly attracts males (Quiroz 1978), which was later characterized as (3*E*, 8*Z*, 11*Z*)-tetradecatrienyl acetate (Attygalle et al. 1995, 1996). This component represents about 90% of the volatile material found in the sex gland of calling females. Nevertheless, a minor component (~10%) was identified as (3*E*, 8*Z*)-tetradecadienyl acetate (Griepink et al. 1996, Svatos et al. 1996). These findings enabled detection and monitoring of *T. absoluta* populations (Guedes et al. 1996, Benvenga et al. 2007, Salas 2007) and the development of pheromone dispensers for the purpose of testing attract-and-kill (Michereff et al. 2000a) or the mating disruption technique (Michereff et al. 2000b, Vacas et al. 2011b).

Many companies have developed pheromone dispensers to detect and monitor *T. absoluta* populations. Most of them are rubber septa, a commonly used pheromone dispenser. In most cases however, their performance is not optimized. A dispenser with an appropriate pheromone release rate is required to not only achieve good efficacy, but to expand use of pheromones in pest control systems. To improve the control methods based on pheromones as attractants (monitoring, mass trapping, or 'attract-and-kill'), the key factor is to know the optimum emission level because release rates strongly affect the attractiveness of the lure, and catches may decrease below and above this level (Jacobson and Beroza 1964, Anshelevich et al. 1994, Zhang and Amalin 2005). Although there have been a few reports of *T. absoluta*'s responses to different pheromone loads of dispensers (Ferrara et al. 2001, Chermiti and Abbes 2012), emission rates have not been assessed, thus optimal release rates were not proposed. Generally, producers tend to increase pheromone load of dispensers to obtain maximum efficacy and longevity. However, pheromone cost is one of the main drawbacks to its implementation in *T. absoluta* management. Thus, knowledge and optimization of emission rates and pheromone release profiles would be preferred, rather than simply increasing dispensers' loads.

The main aim of our study was to determine an optimum pheromone emission rate to help control *T. absoluta* in greenhouse trials. For this purpose, the number of moths caught each week in white Delta traps with different release rates of (3*E*,*8Z*,*11Z*)-tetradecatrienyl acetate using mesoporous pheromone dispensers were compared in two different years.

Methods and Materials

Pheromone Dispensers and Traps. Pheromone dispensers were formulated based on the technology of inorganic molecular sieves developed by Corma et al. (1999, 2000). The dispenser matrix is sepiolite (Tolsa SA, Madrid, Spain), a natural clay mineral with a high adsorptivity for organic molecules. The formulation procedure involves the impregnation of sepiolite with the corresponding amount of pheromone in dichloromethane solution, together with different additives to give consistency and protect the dispenser against humidity. The impregnated material is then compressed in a cylindrical mold by means of a hydraulic press. This manufacturing process has been licensed to Ecologia y Protección Agrícola S.L. (Valencia, Spain) who has manufactured the dispensers for these trials.

(3*E*,*8Z*,*11Z*)- tetradecatrienyl acetate (TDTA hereafter) was employed as the sex pheromone at a 90% isomeric purity, synthetized by Ecología y Protección Agrícola S.L. The minor component of the pheromone, (3*E*, 8*Z*)-tetradecadienyl acetate (TDDA), was not included in the study as Michereff et al. (2000a) reported that the addition of this secondary component does not improve the attraction of TDTA.

The mesoporous dispenser employed in 2010 (referred to as TU1 hereafter) contained a 1 mg pheromone load. It was a cylindrical tablet of 9 mm in diameter and 3.5 mm in height. New mesoporous dispensers (denoted as MD hereafter) were prepared for the trial carried out in 2011: MD1, MD5 and MD25, with initial pheromone loads of 1 mg, 5 mg and 25 mg of pheromone, respectively. They were all cylindrical tablets: MD1, 9 mm in diameter and 3.5 mm in height; MD5, 9 mm in diameter and 7 mm in height; MD25, 13 mm in diameter and 7 mm in height. In both trials, dispensers were placed in white Delta traps, with 19x40 cm sticky bases (Biagro, Valencia, Spain).

Greenhouse Trials. The relationship between pheromone emission level and number of moths captured was studied in two trials; one in 2010 and the other in 2011, inside two greenhouses growing tomatoes (*Solanum lycopersicum* L.) over rock wool hydroponic substrate, which were owned by Anecoop S. Coop. (Valencia, Spain). Greenhouse dimensions and trap distribution are shown in Fig.1. Hourly temperature and relative humidity were recorded by means of a Hobo® Data Logger (Onset, Cape Cod, MA, USA). Data obtained are depicted in Fig. 2. The ventilation system of the greenhouses is controlled by zenithal windows, programmed to open when the temperature exceeds 25ºC. Air recirculators ensure uniform climate conditions inside the greenhouse.

The preliminary study was carried out in 2010 in a 4000 m^2 9x6 mesh (threads/cm^2) greenhouse with four blocks of four traps. The distance between blocks was around 20 m, and the intertrap distance was 15 m. The traps on each block were placed randomly in a grid and were baited with different pheromone doses. They are referred to hereafter as TU1, 2TU1, 3TU1 and 4TU1 (baited with 1, 2, 3 or 4 TU1 dispensers, respectively). Traps were hung on 8 July 2010 and the number of moths caught was counted weekly over six weeks.

A second trial was carried out in 2011 in a 4000 m^2 plastic greenhouse with four blocks of five traps with the same aforementioned distances and arrangement. The traps on each block were baited with different pheromone dispensers: MD1 (1 mg pheromone dispenser), MD5 (5 mg dispenser) and MD25 (25 mg dispenser). Thus, emission levels will be referred as to MD1 (one MD1 dispenser), MD5 (one MD5 dispenser), 2MD5 (two MD5 dispensers), MD25 (one MD25 dispenser) and 2MD25 (two MD25 dispensers). Traps were hung on 15 March 2011 and captures were revised weekly over six weeks.

The traps in both trials were hung at 1 m above the ground and their position inside each block was rotated clockwise every week. None of these dispensers was replaced during the trials.

Pheromone Release Profiles. In parallel with the greenhouse trials, additional dispensers were simultaneously aged in a 4000 m^2 9x6 mesh greenhouse in 2010 and inside a plastic greenhouse in 2011, located 100 m away from the respective trial greenhouses and having the same aforementioned cropping conditions. The residual TDTA content was extracted at different ageing intervals. Three dispensers per ageing time were extracted by solvent extraction at 40ºC for 2 h, with magnetic agitation and dichloromethane as the solvent.

The TDTA content was measured by gas chromatography with a flame ionization detector (GC/FID) using a Clarus®500 gas chromatograph (PerkinElmer Inc., Wellesley, USA). Extracts were analysed, and quantification was done using *n*-dodecane as an internal standard. Each extract was injected in triplicate on a ZB-5 (30 m × 0.25 mm × 0.25 mm) column (Phenomenex Inc., Torrance, CA), maintained at 120ºC for 2 min and then raised by 20ºC/min to 260ºC, to be then maintained for 3 min. The carrier gas was helium at 1.5 ml/min.

Statistical Analysis. The quantified residual pheromone loads, *P* (μg) for each dispenser were fitted by polynomial regression with independent variable *t* (number of ageing days). The first derivative of the resulting equations provided an estimation of the emission rates for each trapping period (t_i) [i.e., d(TDTA)⁄dt ($t = t_i$)]. For example, the 2MD5 traps inspected on 29 March 2011 corresponded to the traps baited with two MD5 dispensers collecting moths during the period of 7-14 days (i.e., t = 7 to t = 14). Thus, the pheromone emission rate was estimated by applying t = 10.5 (this being the midpoint of the 7-14 day period) to the respective derived equation (MD5 release profile), and the resulting value was multiplied by two, as two MD5 dispensers were used in this trap. The release rate was assumed to be constant throughout each time interval.

The Box-Cox power transformation (λ) was employed to normalize trap catch data prior to analysis of variance (ANOVA). The equation employed to correlate the estimated release rates and trap captures was obtained following the same methodology used in previous works (Vacas et al. 2009, Vacas et al. 2011a, Navarro-Llopis et al. 2011), which is now described. A multifactor ANOVA followed by Fisher's LSD test ($P \leq 0.05$) was applied to study the effects of three factors on trap catch: week (time), block (position of the block inside the greenhouse) and emission level. Once significance of the emission factor was confirmed, we proceeded with analysis of the variability in trap catch data due to time and position of the blocks. For this purpose, a two-way ANOVA was performed with catch data only with factors week and block. The residuals of this ANOVA did not account for variance due to the two factors week and block, and still provided evidence for variance due to the emission level factor. Thus, these residuals were employed to perform multiple regression analysis in order to evaluate the linear and quadratic effects of the emission factor over trap catches and to obtain the equation relating trap catch and emission level. Statistical analyses were performed using the Statgraphics Centurion XVI package (StatPoint Technologies, Warrenton, VA, USA).

Results

Pheromone Release Profiles. The release profile of the mesoporous dispenser (TU1) employed in the preliminary study is depicted in Fig. 3. Multiple linear regression demonstrated that the quadratic effect was not statistically significant ($P = 0.49$) and that the residual load of TDTA fitted a linear model ($P = 0.01$; Eq. 1, $R^2 = 0.90$). The independent variable was the number of days since the dispensers were installed in the greenhouse (*t* (days)). Thus, it was assumed that the residual pheromone load decreased at a constant rate throughout the study period, which is given by the slope of the linear model and is equal to 11.71 μg/d.

The release profiles of the three mesoporous dispensers employed in 2011 are also provided in Fig. 3. The quadratic effect was statistically significant for the MD1 dispenser ($P < 0.001$); thus, TDTA (μg) emission was not constant, but fitted the quadratic model ($R^2 = 0.92$) given by Eq. 2. A quadratic equation was also obtained for the MD5 release profile, resulting in $R^2 = 0.84$ (Eq. 3), while the MD25 dispensers mean release rate was assumed constant and equal to 99.95 μg/d, according to the linear fitting given by Eq. 4 ($R^2 = 0.81$, significance of the quadratic term P = 0.14). The slope of the lines based on equations 2 and 3 was not constant (Fig. 3), implying that the daily emission rate of these pheromone dispensers decreased over time. The first derivatives of Eq. 2 and 3 allowed the estimation of the emission rates for each trapping period (t_i). All the estimated emission values are indicated in Table 1.

Greenhouse Trials. The weekly average number of catches (MTW) obtained with the different traps in the 2010 trial are depicted in Fig. 4. The power-transformed ($\lambda = 0.36$) catches were analyzed by a multifactor ANOVA using three factors: week, block and emission. None of the possible interactions between factors were statistically significant (week × block: $F = 1.00$; df = 12,36; $P = 0.47$, week × emission: $F = 1.45$; df = 12,36; $P = 0.19$, block × emission: $F = 0.44$.; df = 9,36; $P = 0.90$). The week factor was significant ($F = 6.93$; df = 4,69; $P < 0.001$), according to the increasing trend of the registered captures. The block factor also had a significant effect ($F = 4.15$; df = 3,69; $P = 0.01$), which could be explained by the pest's natural clumped distribution. As expected, the emission factor effect was also statistically significant ($F = 9.31$; df = 3,69; $P < 0.001$): the captures obtained with the traps baited with one TU1 dispenser were significantly lower than those traps with 4TU1, suggesting that attractant power increased with the emission level.

Considering that the estimated mean release rate for TU1 was 11.7 μg/d, the emission factor could be considered a quantitative variable, providing the following relationship in terms of the traps baited for the test: 1TU1 = 11.7 μg/d, 2TU1 = 23.4 μg/d, 3TU1 = 35.1 μg/d, and 4TU1 = 46.8 μg/d. The multiple regression analysis performed with these emission rates and the residues of the two-way ANOVA (week and block factors) shows that the quadratic effect of emission was not statistically significant ($P = 0.99$). This indicates the absence of a relative maximum of catches corresponding to an optimum emission level and confirming the linearity of the trend observed in Fig. 5 ($P < 0.001$).

A new trial was carried out in 2011 to evaluate the effect of the emission factor when emission levels were higher. The population dynamics in this greenhouse are provided in Fig. 6, and indicate that the lowest mean captures were obtained in those traps baited with one MD1. Following the same statistical procedure as above, the effect of the factors week, block and emission was first evaluated by a multifactor ANOVA with the power-transformed ($\lambda = 0.23$) MTW data. None of the possible interactions between factors were statistically significant (week × block: $F = 0.86$; df = 15,60; $P = 0.61$, week × emission: $F = 1.05$; df = 20,60; $P = 0.42$, block × emission: $F = 0.82$; df = 12,60; $P = 0.63$). The effects of block and week factors were significant (block: $F = 4.20$; df = 3,119; $P = 0.008$; week: $F = 110.18$; df = 5,119; $P < 0.001$). The significance of the emission level effect ($F = 42.72$; df = 4,119; $P < 0.001$) confirmed the influence of pheromone emission on attractant power.

As described in the previous section, MD25 emission was constant and emission levels of traps baited with this dispenser took the following values: MD25 = 99.95 μg/d and 2MD25 = 199.89 μg/d. Release profiles of MD1 and MD5 followed polynomial models, and their release rates for each trapping period were calculated according to their derived equations (Table 1). All the estimated release rates were employed in a subsequent multiple regression analysis with the residues saved from the two-way ANOVA performed with the week and block factors. The quadratic effect evaluated in the regression analysis was significant ($P < 0.001$), which highlights the existence of a relative maximum of captures corresponding to a particular emission value. The regression gave the relationship represented by the Eq. 5, which is depicted in Fig. 7. To obtain the emission value corresponding to the maximum catches, the first derivative of the fitted model (Eq. 5) was equated to zero, resulting in em = 150.3 μg/d.

Discussion

Sex pheromone-mediated systems are now viable tools to control *T. absoluta*. Currently, use of pheromone-baited traps for monitoring purposes is a common practice, although efforts are being made to develop direct control methods, such as mating disruption and attract-and-kill techniques. Vacas et al. (2011b) demonstrated the efficacy of mating disruption by using mesoporous dispensers inside high-containment greenhouses; however, the application of this technique has constraints. In contrast, many pheromone dispensers have been developed for attraction purposes, but very little information is available about use of mass trapping or attract-and-kill systems (Hassan and Al-Zaidi 2010). Most of the dispensers available are rubber septa, commonly characterized by irregular release kinetics, high emission rates during the first week of exposure and rapid loss of efficacy. In addition, this emission is highly temperature-dependent (McDonough et al. 1989). For these reasons, the performance of rubber septa dispensers is not always optimized, which may lead to irregular captures and provide a mistaken estimation of pest populations.

There are many examples in the literature of studies comparing catches and pheromone doses for Lepidopteran pests with diverse results. Kehat et al. (1994) found growing catches of codling moth (*Cydia pomonella* (L.)) males with increasing pheromone doses of up to 100 μg; yet rubber septa loaded with 5000 μg were significantly less attractive than 100 μg or 1000 μg dispensers. A similar response was obtained for rice leaffolder moth, *Cnaphalocrocis medicinalis* (Guenée) (Kawazu et al. 2004) and *Mocis latipes* (Guenée) (Landolt and Heath 1989). Vacas et al. (2009) found less catches of *Chilo suppressalis* (Walker) both below and above an optimal release rate of 34 μg/d. However, other response types, i.e., asymptotic, were exhibited by other Lepidoptera species, as found for pine processionary moth (*Thaumetopoea pytiocampa* Denis and Schiffermüller), giving increasing doses of its pheromone up to 20 mg, with 95% of the maximum catch obtained with dispensers loaded with 10 mg (Jactel et al. 2006). Other lepidopterans have shown this asymptotic pattern, such as some species of the genus Geometridae, Pyralidae or Noctuidae (Evenden et al. 1995, Knutson et al. 1998, Rao and Subbaratnam 1998).

Response of *T. absoluta* to increasing pheromone doses was first shown by Ferrara and co-workers (2001), who obtained an increasing number of moths caught in field trials with increasing doses of TDTA, ranging from 1 μg to 100 μg. More recently, Chermiti and Abbes (2012) reported significant differences between number of catches obtained in traps baited with 800 μg TDTA and those with 500 μg dispensers, in crops with high population levels (> 30 MTW). These works, like others mentioned above, discuss insect responses based on the dispensers' initial pheromone loads. Nonetheless, this does not provide information on the actual release of pheromone given that daily emission rates and, therefore the amount of airborne pheromone depend on weather conditions, dispenser type or formulation. In fact, the present work employed two dispensers loaded with 1 mg of TDTA, TU1 and MD1, which showed different release patterns, even though they had the same matrix and load. Although release profiles of these dispensers were studied in different periods (TU1 in summer months and MD1 in spring), their different release pattern could be due to slight differences in the manufacturing process as temperature does not explain why release rate of MD1 decreases while temperature increases. In fact, results reported by Dominguez-Ruiz et al. (2008) demonstrated that performance of mesoporous dispensers is independent of temperature in the range 20-40ºC.

Very few studies have determined the optimal release rate of attractants (de Groot and DeBarr 1998, Cross et al. 2006, Vacas et al. 2009, Vacas et al. 2011a, Navarro-Llopis et al. 2011, Ryall et al. 2012). In the present work, mesoporous dispensers were employed as tools to obtain different tested pheromone doses. In the first trial carried out in 2010, a linear relationship was found for *T. absoluta*'s response to increasing release rates, ranging from 11.71 to 46.84 μg/d. According to this result, higher pheromone doses (a maximum of ca. 200 μg/d) were tested in the second trial (2011) to verify the existence of an optimum release value, or whether the trend becomes asymptotic at higher release rates. The model obtained by the multiple regression analysis shows the existence of a relative maximum of the captures corresponding to a release rate of *ca.* 150.3 μg/d. Thus, emission rates above and below this value offer lower catch efficacy in Mediterranean greenhouse conditions. It must be taken into account the limitations of the obtained value because the study has been conducted in a particular region and with Delta traps; thus, this result must be validated for other regions, seasons and types of traps. Air flow throughout the greenhouse may also affect results as pheromone could be washed away. Therefore, ventilation system is another factor that affects the estimation of optimum release values.

Research on this topic is essential to develop effective formulations for attraction purposes because commercial dispensers could be designed in accordance with these values for better pheromone use. Optimum release rates for attraction could also be useful to help develop mating disruption formulations. According to the exhibited response, the release rates that are higher than optimum emission values could tend to create proper pheromone environments to disrupt the chemical communication of insects in accordance with the mechanism involved in mating disruption.

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Tables

Table 1. Estimated pheromone emission rates for traps baited with dispensers MD1 and MD5 in trial 2011

Day period[1]	Date[2]	Trap code	Emission (µg/day)	procedure[3]
0-7	22/03/2011	MD1	9.45	$d(2)/dt_{t=3.5}$
		MD5	48.50	$d(3)/dt_{t=3.5}$
		2MD5	96.99	$2 \cdot d(3)/dt_{t=3.5}$
7-14	29/03/2011	MD1	8.38	$d(2)/dt_{t=10.5}$
		MD5	39.91	$d(3)/dt_{t=10.5}$
		2MD5	79.83	$2 \cdot d(3)/dt_{t=10.5}$
14-21	05/04/2011	MD1	7.32	$d(2)/dt_{t=17.5}$
		MD5	31.33	$d(3)/dt_{t=17.5}$
		2MD5	62.66	$2 \cdot d(3)/dt_{t=17.5}$
21-28	12/04/2011	MD1	6.25	$d(2)/dt_{t=24.5}$
		MD5	22.75	$d(3)/dt_{t=24.5}$
		2MD5	45.50	$2 \cdot d(3)/dt_{t=24.5}$
28-35	19/04/2011	MD1	5.18	$d(2)/dt_{t=31.5}$
		MD5	14.17	$d(3)/dt_{t=31.5}$
		2MD5	28.33	$2 \cdot d(3)/dt_{t=31.5}$
35-42	26/4/2011	MD1	4.12	$d(2)/dt_{t=38.5}$
		MD5	5.58	$d(3)/dt_{t=31.5}$
		2MD5	11.17	$2 \cdot d(3)/dt_{t=31.5}$

[1] Day 0 corresponds to 15 March 2011 at which all traps were installed.
[2] Date at which traps were inspected for counting.
[3] Procedure used to calculate emission values: applying t=i (i=midpoint of the period) to the respective derived equation indicated within parentheses.

Figure Legends

Figure 1 Sketch of trap layout and greenhouse dimensions (m) in trial 2010 (A) and trial 2011 (B).

Figure 2 Temperature profiles recorded in trial greenhouses 2010 (A) and 2011 (B). Mean (Tm), minimum (Tmin) and maximum (Tmax) temperature profiles are depicted.

Figure 3 Release profiles of (3*E*,8*Z*,11*Z*)-tetradecatrienyl acetate (TDTA) from the mesoporous dispensers employed: TU1 (trial 2010), and MD1, MD5 and MD25 (trial 2011). Fitted models (eqs. 1-4) describe the mean pheromone content (TDTA) of the dispenser vs. time (t = number of days in greenhouse). Three replicates were extracted per ageing time.

Figure 4 Mean ± SE number of moths caught per trap and week (MTW) for each of the four types of pheromone-baited trap tested in trial 2010. Moths were captured in white Delta traps and pheromone dispensers were not replaced throughout the study.

Figure 5 Means and 95% LSD intervals of MTW (males per trap and week) data corresponding to factor emission throughout trial 2010. The line represents the model that best fits the mean values.

Figure 6 Mean ± SE number of moths caught per trap and week (MTW) for each of the five types of pheromone-baited trap tested in trial 2011. Moths were captured in white Delta traps and pheromone dispensers were not replaced throughout the study.

Figure 7 Scatter plot and fitted regression model (eq. 5), for trial 2011 data, of residuals vs. emission rates (*em*). The dependent variable is the residuals from the ANOVA performed with factors week and block.