

Monitoring oak processionary moth *Thaumetopoea processionea* L. using pheromone traps: the influence of pheromone lure source, trap design and height above the ground on capture rates

David T. Williams*, N. Straw*, M. Townsend†, A. S. Wilkinson‡ and A. Mullins‡

*Forest Research, Centre for Forestry and Climate Change, Alice Holt Lodge, Farnham, Surrey GU10 4LH, U.K., †Gristwood & Toms, Harris Lane, Shenley, Herts WD7 9EG, U.K., and ‡University of Derby, Department of Biology and Forensics, Kedleston Road, Derby DE22 1GB, U.K.

- Abstract**
- 1 A field trial conducted in the summer of 2011 evaluated three key parameters that might be influential for determining the number of adult male oak processionary moths (OPM) *Thaumetopoea processionea* (L.) caught in pheromone traps. Two types of traps (Delta and funnel; Oecos, U.K.) containing one of three different commercially available pheromone lures for OPM were placed out in the lower (3–5 m), mid (5–10 m) and upper (10–15 m) canopy of 72 individual oak trees in Richmond Park, in London, U.K.
 - 2 The traps were placed out for 8 weeks covering the main flight period of OPM, and significantly more male moths were captured in traps positioned in the upper canopy (76.6%) compared with either mid-canopy (18.6%) or lower canopy (4.8%) positions. Funnel traps caught significantly more male OPM than Delta traps, catching almost six times as many moths over the trapping period.
 - 3 Traps containing one of the commercially available pheromone lures did not catch any moths, whereas traps with the other two lures caught similar numbers of moths. Chemical analysis revealed considerable differences between the three pheromone lures used in the trial in terms of the initial starting concentration of the primary component (Z,Z)-11,13-hexadecadienyl acetate and its dissipation over a 28-day period.
 - 4 The results obtained in the present study indicate some of the main factors that need to be taken into account when using pheromone traps to monitor OPM populations and also contribute to the establishment of a standardized monitoring system for this recently established insect pest.

Keywords Lepidoptera, monitoring, pheromones, *Quercus* spp., survey, Thaumetopoeidae, traps.

Introduction

The oak processionary moth (OPM) *Thaumetopoea processionea* (L.) is widely distributed across western, central, and southern Europe, where it is considered as a serious defoliator of oak *Quercus* spp. in urban and amenity areas and also in woodlands and forests (EFSA, 2009). In recent decades, the distribution of OPM in north-west Europe has expanded, potentially in response to changes in regional climate patterns (van Oudenhoven *et al.*, 2008; Groenen & Meurisse, 2011; Moraal

& Jagers op Akkerhuis, 2011). Human-mediated transportation of insect pests as a result of increasing global trade, however, facilitates range expansion at a considerably faster rate than natural dispersal and spread via changes in climate in an insect's native area. Human-mediated dispersal has been observed to be responsible for the long-distance jumps in range expansion of many non-indigenous forest insect pests and pathogens (Liebhold *et al.*, 1995; Liebhold & Tobin, 2008), including pine processionary moth *T. pityocampa* (L.) in France (Robinet *et al.*, 2012). In 2006, OPM larvae were discovered on oak trees at two separate locations in the west of London (Townsend, 2008, 2009). These breeding populations were the first to be found in the U.K. and appear to have originated through the importation

Correspondence: David T. Williams. Tel.: +44 (0) 1420 22255; fax: +44 (0) 01420 23653; e-mail: david.t.williams@forestry.gov.uk

and planting of semi-mature Cypress oak trees *Quercus robur* f. *fastigiata* from continental Europe, on which undetected eggs of OPM were present (Townsend, 2008, 2009).

High populations of OPM have occurred in Belgium, Germany, Italy, and the Netherlands in recent years, which have resulted in serious defoliation of oak trees (Stigter *et al.*, 1997; Roversi, 2008; EFSA, 2009; Wagenhoff & Veit, 2011). In combination with other biotic factors (e.g. other Lepidopteran defoliators, buprestid beetles, and fungal pathogens) and abiotic factors (e.g. drought), OPM is likely to be a significant factor contributing to oak decline in general and, in some cases, to tree mortality (Thomas *et al.*, 2002; EFSA, 2009). The larvae of OPM also pose a serious risk to human and animal health. The third- to sixth-instar larvae possess defensive urticating hairs (setae) containing a soluble protein (thaumetopoein) that can cause severe allergic skin reactions in humans and animals (Lamy & Novak, 1987; Lamy, 1990; Jans & Franssen, 2008). Typical reactions to exposure to the hairs include pruritus, dermatitis, conjunctivitis, respiratory problems (e.g. pharyngitis or asthma), and, in rare cases, an anaphylactic reaction (Maier *et al.*, 2003; Gottschling & Meyer, 2006; Gottschling *et al.*, 2007).

The public health issues associated with OPM are a particular concern in west London, which is a heavily urbanized and densely populated area, and, combined with the threat to plant health, had prompted a major control programme to eradicate the pest from the U.K. Subsequently, because the pest population has continued to increase and expand its range, the aim of the control strategy has changed from eradication to containment within the current outbreak area and slowing the rate of spread. The implementation of this control strategy and managing the pest outbreak is crucially dependent on survey and monitoring, aiming to identify areas of infestation and to assess the effectiveness of treatments (insecticide spraying or manual removal of larval nests). Monitoring to date has involved indecisive ground surveys to locate infested trees, as well as the placement of pheromone traps baited with the commercially available sex pheromone of OPM in a grid across the west London area. The results obtained from this network of pheromone traps, however, have been disappointing, with relatively few moths being caught even in areas where the ground surveys indicated that there were appreciable numbers of OPM larval nests (Forestry Commission, unpublished data; Table 1).

Pheromone trapping for monitoring populations and dispersal of Lepidopteran pests is a well established technique and has been exploited for use with numerous non-indigenous invasive species worldwide (Sharov *et al.*, 2002; Myers & Hosking, 2002; Suckling *et al.*, 2005; Tobin *et al.*, 2007). It is particularly effective for detecting insect pests at low population densities, although trap captures are influenced by a wide range of factors. Some of the key factors include trap design and colour, the height of trap in or above the canopy, and the effectiveness of the pheromone lure at attracting males not only in terms of the blend of components, but also with respect to the dosage (release rate) and longevity of the lure. Additionally, climatic factors such as temperature, humidity, light intensity, wind direction, and speed can also influence the number of insects caught (McNeil, 1991). The deployment of the pheromone

Table 1 The number of adult male *Thaumetopoea processionea* caught in the Forestry Commission network of pheromone traps in west London compared with the number of *Thaumetopoea processionea* larval nests observed by ground surveys

	2007	2008	2009	2010	2011
Number of traps	84	84	136	136	164
Trap type	Delta	Delta	Funnel	Funnel	Funnel
Total number of male OPM caught	0 ^a	16	28	51	18
Number of traps with one or more adult male	—	12	14	14	10
Number of OPM larval nests ^b	708	508	2450	2071	4410

^aZero catch probably as a result of an ineffective pheromone lure.

^bData from Parks and Townsend (2011) and Forestry Commission (unpublished data).

Traps were placed at 2–3 m above the ground in oak trees, for the whole of the moth flight period. OPM, oak processionary moth.

traps in London may not have been optimal for several of these factors, which may explain their relative ineffectiveness. Previous studies have shown that the formulation of the synthetic pheromone components of the OPM lure has a large influence on trap catches (Breuer *et al.*, 2003a, 2003b; Quero *et al.*, 2003; Gries *et al.*, 2004), and trap type and the height at which the trap is positioned in the tree canopy are also important (Breuer *et al.*, 2003a, 2003b). It remains unclear to what extent, however, these factors might have influenced the number of adult male OPM caught in the U.K.

The present study aimed to refine and optimize the trap and lure parameters for capturing male OPM in pheromone traps and to establish a more effective monitoring system. Three key factors were investigated: (i) the design of the pheromone trap; (ii) the height of the trap above the ground; and (iii) the OPM lures supplied by three different manufacturers. In addition, chemical analyses were carried out to determine the initial concentration and longevity of the main active component of the OPM pheromone lure supplied by the different manufacturers. Previous studies have suggested that the key active component of the OPM pheromone (*Z,Z*-11,13-hexadecadienyl acetate) may breakdown rapidly into less effective isomers at high temperatures or through ultraviolet (UV) exposure during the summer months (Breuer *et al.*, 2003a). It is important to determine how quickly this active pheromone component either volatilizes or isomerizes into the inactive isomers under field conditions because this determines if and when it might be necessary to replace the lures in the pheromone traps during the flight period of the moth.

Materials and methods

Study site and experimental design

The experimental trial was carried out in Richmond Park in west London (National Grid Reference: TQ 201 730), which is

the largest of the Royal Parks in London, covering an area of almost 1000 ha. The park has over 100 000 trees of which approximately 40 000 are oak trees, mainly *Quercus robur* (L.), although there are some *Quercus rubra* (L.), *Quercus cerris* (L.), and other oak species planted in various places as ornamentals. Many of the oaks are over 400 years old and several are over 750 years old. The park was selected for the experimental trial because it was located within the main OPM outbreak area, and a large number of oak trees in the park and in the surrounding area were infested with OPM in 2010 (Parks & Townsend, 2011).

Oak trees selected for the trial were at least 15 m in height and were accessible from a path, track or road. A total of 72 trees were selected (67 *Q. robur*, two *Q. rubra*, two *Quercus palustris*, and one *Q. cerris*) with a diameter at breast height (1.3 m above the ground) ranging from 44 to 187 cm. Each tree was located at least 50 m away from any of the other selected trees to minimize the effects of trap-poaching from one pheromone trap to another. The trees were assigned an experimental number, and treatments (lure supplier \times trap type \times canopy position) were allocated to each tree using random numbers.

OPM pheromone lures were obtained from Edialux (Belgium) (Lure 1), ISCA Technologies Inc. (Riverside, California) (Lure 2), and Pherobank (The Netherlands) (Lure 3). Each of these lures was tested in two types of commercially available pheromone trap: cardboard Delta traps and standard green funnel traps (both supplied by Oecos, U.K.), and at three different heights within the tree canopy: lower canopy (below 5 m), middle canopy (between 5 and 10 m) or upper canopy (10–15 m). Each combination of lure, trap type and canopy position was replicated four times, giving a total of 72 traps. A single trap was placed in each tree.

The positioning of traps in the middle and upper canopy was achieved by either using a mobile elevated work platform or by climbing the tree. These traps were attached to polyethylene throw lines, which were passed through 60-cm Lyon Nylon slings (Dmm, Wales, UK) that were wrapped around appropriate branches in the canopy. Traps were then pulled up into the correct position just below the sling, and the other end of the polyethylene throw line was tied off on a suitable lower branch at approximately 3 m. The line could then be reached, and the trap raised and lowered, from a step-ladder. Traps positioned in the lower canopy were attached directly to suitable branches using a step-ladder. Once the traps were set in position within the tree canopy, their actual height above the ground was recorded using a tape measure.

All traps were established and primed with the selected pheromone lure on the 18/19 July 2011. Disposable gloves were used and replaced when handling each individual pheromone lure to prevent cross-contamination of lures and traps. Approximately 250 mL of saline solution was added to the funnel traps, and the sticky boards were inserted into the base of the Delta traps. The traps were subsequently checked every 2 weeks until mid September for moth captures, which covered the main OPM flight period. On each collection date, moths were removed from the traps and the saline solution and the sticky boards were replaced. In addition, on the second collection date (4 weeks into the trial), the pheromone lures

were replaced with a fresh lure from the same supplier. Moths caught in the funnel traps were placed in numbered plastic containers and taken back to the laboratory to confirm their identification. Similarly, a transparent plastic sheet was placed over the adhesive surface of the sticky boards from the Delta traps and these were taken back to the laboratory to confirm the identification of any moths captured. All of the moths caught within each trap on each collection date were subsequently recorded.

Chemical analysis of lures

The chemical analysis of the pheromone lures was performed using a gas chromatograph-mass spectrometer (GC-MS), and was conducted in parallel with the main pheromone trapping trial in Richmond Park using lures from the same batch. The three commercially available lures consisted of either natural rubber septas (Lures 1 and 3) or synthetic rubber septas (Lure 2) coated with the synthetic pheromone lure components. Eight lures from each of the three suppliers were placed in labelled Delta traps, in accordance with the manufacturer's instructions, and these 24 traps were placed at a height of 3–4 m in the mid-canopy of suitable trees on 20 July 2011 (day 0). On the same day, two lures from each of the suppliers were removed from their packaging and were analyzed to determine the initial amounts of (Z,Z)-11,13-hexadecadienyl acetate on the rubber septas (day 0). Every 7 days thereafter, two of the Delta traps for each lure were selected randomly, removed from the tree, and the pheromone lures assessed for the (Z,Z)-11,13-hexadecadienyl acetate component. This key pheromone component was therefore assessed over a 28-day period on five separate dates (days 0, 7, 14, 21, and 28). In addition, for each individual lure, the GC-MS analysis was performed three times.

Pheromone lure components were extracted from the rubber septas using 1 mL of *n*-pentane solvent containing a 1% *n*-decane v/v internal standard. For each lure, on each date, the lure was placed in a clean 10-mL glass beaker, and 1 mL of *n*-pentane containing *n*-decane at 1% v/v was added. The solvent was rinsed over the outer surface of the lure and within the recess of the rubber septa for a period of exactly 1 min, such that all surface absorbed material was taken up by the solvent. The solvent was then pipetted into a sealed brown glass scintillation vial fitted with a screw top lid and polytetrafluoroethylene internal septa. This sample was placed immediately into a dark refrigerator at 2–8 °C until the analysis was performed (within 24–48 h of extraction).

A commercially available standard for the active compound (Z,Z)-11,13-hexadecadienyl acetate (93.9% stereochemically pure) was obtained from International Pheromone Systems Ltd (U.K.) as a 1 g/50 mL standard in diethyl ether (SEDQ, Spain). This standard was diluted to provide a range of external reference samples (containing 1% *n*-decane internal standard as a reference) of between 0.85 and 25.7 mg/mL. These samples were injected into the GC-MS as 1 μ L aliquots under the same separation conditions as the pheromone lure samples to produce a calibration between the (Z,Z)-11,13-hexadecadienyl acetate and the internal standard. Subsequently, the amounts of (Z,Z)-11,13-hexadecadienyl acetate represented by the peak

areas from the GC-MS analysis of the pheromone lure samples were estimated by comparison with the internal standard. The ratio between these two components was used to estimate the amount of the (Z,Z)-11,13-hexadecadienyl acetate on each of the rubber septa, using the calibration analysis.

The chemical analysis of the pheromone lures was performed on a QP2010S fitted with an AOC-20i auto injection system and running GC Lab Solution software, version 2.50 (Shimadzu, Japan). The instrument was fitted with a Zebron ZB1 column (internal diameter 30 m \times 0.25 μ m; film thickness 0.25 μ m) (Phenomenex, Torrance, California), and operated using a helium carrier gas at a column flow rate of 1 mL/min. With regard to GC separation parameters, 1- μ L aliquots of the pheromone lure samples were injected in a splitless injection mode at 250 °C within the injection port. The GC oven was set at a starting temperature of 50 °C, which was held for 4 min before rising at a rate of 5 °C/min to a final temperature of 280 °C. This final temperature of 280 °C was then held for a total of 10 min, and the total run time was 60 min. There was a solvent delay time set of 2.5 min before the mass spectrometer was turned on. The mass spectrometer source and interface temperatures were 200 and 280 °C, respectively, and the detector was set to detect m/z values from 25 up to 1000. The mass spectrometer was set to scan with a scan speed of 1000 u/s, with event time set at 0.5 s.

Statistical analysis

Moth captures were not normally distributed and, despite employing a variety of transformations (square root, cube root, and log) and testing for normality using the Shapiro–Wilk test, it was not possible to normalize the data. Consequently, the data were analyzed using the Mann–Whitney and Kruskal–Wallis tests, with Dunn’s test being used to identify significant differences between individual variables (Zar, 1999). Significant differences in trap detection ability (i.e. whether traps captured at least one moth) were determined using Fisher’s exact test. The relationship between trap capture and individual trap height above the ground was analyzed using stepwise polynomial regression, with the number of adult males caught per trap $\log(x + 1)$ transformed before the analysis. Polynomial regression was also used to describe the calibration between the external reference samples and internal standard in the GC-MS analysis. All analyses were performed using GENSTAT, 13th edition (Payne *et al.*, 2010).

Results

Field trial

The total number of male OPM caught over time varied considerably, with the majority of moths (68.9%) being caught in the 2-week period from 2–16 August 2011 (Table 2). This indicated that the main flight period for male OPM occurred within a relatively narrow time period in 2011.

Traps primed with either Lure 1 or Lure 3 captured similar numbers of adult male moths (combining trap type and canopy position of trap), with 189 and 187 moths caught, respectively;

Table 2 Total numbers of adult male *Thaumetopoea processionea* caught in the pheromone traps in the experimental trial in Richmond Park in 2011

Dates	Number of moths captured	Percentage of total
19 July to 2 August	27	7.2
2 August to 16 August ^a	259	68.9
16 August to 30 August	79	21.0
30 August to 13 September	11	2.9
Total	376	100

^aPheromone lures replaced on 16 August.

Totals for all traps, canopy positions, and lures combined.

Table 3 Total numbers of adult male *Thaumetopoea processionea* captured, and the number of traps that caught at least one male *Thaumetopoea processionea*, for Lures 1 and 3 at each canopy position

Trap position in canopy	Total number of moths captured		Number of traps with/without a moth		Percentage of traps capturing a moth
	Lure 1	Lure 3	Lure 1	Lure 3	
Lower (3–5 m)	4	14	1/7	5/3	37.5
Middle (5–10 m)	38	32	5/3	8/0	81.3
Upper (10–15 m)	147	141	7/1	8/0	93.8
Total	189	187	13/11	21/3	70.8

hence, no significant difference was observed in total moth trap captures between these two lures (Mann–Whitney, $U = 240.0$, $P = 0.320$; Table 3). By contrast, Lure 2 proved to be ineffective and failed to capture a single moth over the 8-week trapping period. Not unexpectedly, there was a significant difference between lures in their ability to capture adult moths [Kruskal–Wallis (adjusted for ties), $H = 29.97$, $P < 0.001$]. Because Lure 2 failed to capture any moths, it was not included in any of the subsequent analyses.

The capture of adult male OPM by Lures 1 and 3 was very similar in relation to trap type and trap height above the ground. There was no significant difference in the number of moths caught in Delta and funnel traps by Lures 1 and 3 (Delta traps: 30 and 26 moths, respectively; Mann–Whitney, $U = 51.5$, $P = 0.223$; funnel traps: 159 and 161 moths, respectively; Mann–Whitney, $U = 66.5$, $P = 0.765$). Similarly, there was no significant difference in the number of moths caught at the different canopy positions between Lures 1 and 3 (Mann–Whitney, lower canopy, $U = 16.5$, $P = 0.082$; middle canopy, $U = 23.0$, $P = 0.361$; upper canopy, $U = 26.0$, $P = 0.552$; Table 3).

There was a significantly higher number of moths caught in the funnel traps than in the Delta traps (320 and 56 moths, respectively) over the entire trapping period when combining the moth catches with both Lures 1 and 3 and at all canopy positions (Mann–Whitney, $U = 152.5$, $P = 0.004$; Fig. 1). There was no significant difference in moth catches between trap types with respect to Lure 1 data alone (Mann–Whitney, $U = 42.0$, $P = 0.070$); however, moth catches with Lure 3 did differ significantly between trap types, with funnel

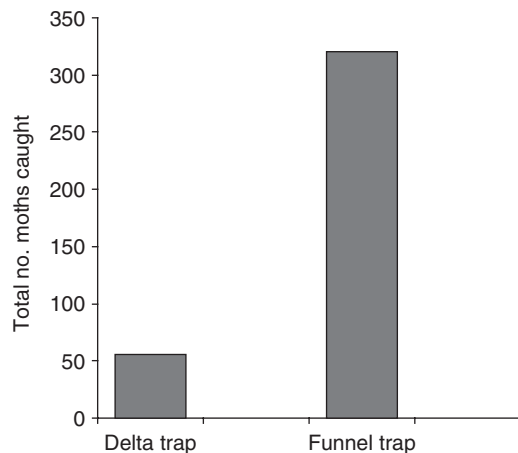


Figure 1 Total number of adult male *Thaumetopoea processionea* caught in Delta traps and funnel traps over the entire 8-week trapping period. Totals for all canopy positions and lure types combined for each trap type.

traps catching significantly more moths than Delta traps (Mann–Whitney, $U = 32.5$, $P = 0.019$).

The positioning of pheromone traps in the tree canopy was a highly influential factor in capturing male OPM. Traps in the upper canopy caught 76.6% of the total moths captured, 18.6% of moths were caught in the middle canopy, and only 4.8% were caught in traps in the lower canopy (Fig. 2). The number of moths caught at the three canopy positions differed significantly when catches for Lures 1 and 3 were combined [Kruskal–Wallis test (adjusted for ties); $H = 18.20$, $P < 0.001$; Fig. 2]. Further analysis indicated that there was a significant difference in trap catches between the upper and lower canopy positions (Dunn test, $Q = 4.266$, $P < 0.001$; Fig. 2) but not between lower and middle, or middle and upper canopy positions.

The relationship between trap height above the ground and the number of adult male OPM caught is shown in Fig. 3. A quadratic expression was the best fit to the data, indicating the higher the trap was placed in the canopy, the greater the chance of capturing a moth, without any evidence that trap catches reached an asymptotic maximum above a certain height (Fig. 3). There was no relationship between tree size (diameter at breast height) and the number of moths captured ($R^2 < 0.1$, $P < 0.05$ for linear, quadratic, cubic, logarithmic, and exponential regression models).

The ability of a trap to catch at least a single moth is perhaps a more useful indicator of trap efficiency for monitoring purposes, at least when trying to detect new outbreaks and in determining distances of spread. In this respect, funnel traps were more effective than Delta traps, with 19 out of 24 funnel traps (72.2%) catching at least one moth compared with 15 out of 24 delta traps (62.5%), although the difference was not significant (Fisher's exact test, $P = 0.341$). By contrast, Lures 1 and 3 differed significantly in their ability to catch a single moth. Thirteen out of 24 traps with Lure 1 (54.2%) caught at least one moth, whereas 21 out of 24 traps with Lure 3 (87.5%) caught at least a single moth (Fisher's exact test, $P = 0.024$; Table 3). Thus, traps containing Lure 3 appeared to

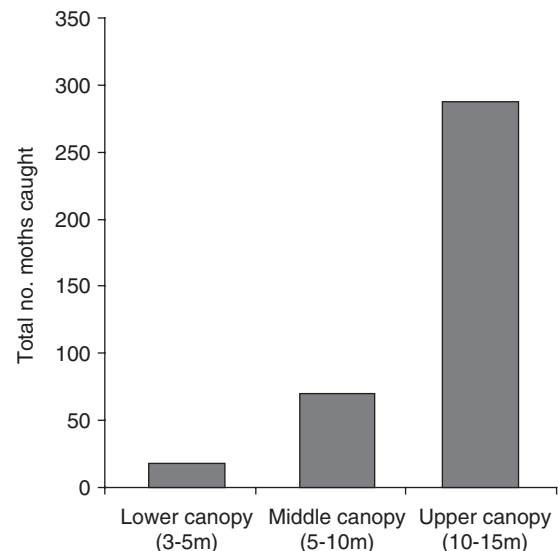


Figure 2 Total number of adult male *Thaumetopoea processionea* captured in lower, middle, and upper canopy positions. Totals for all trap types and lure types combined for each canopy position.

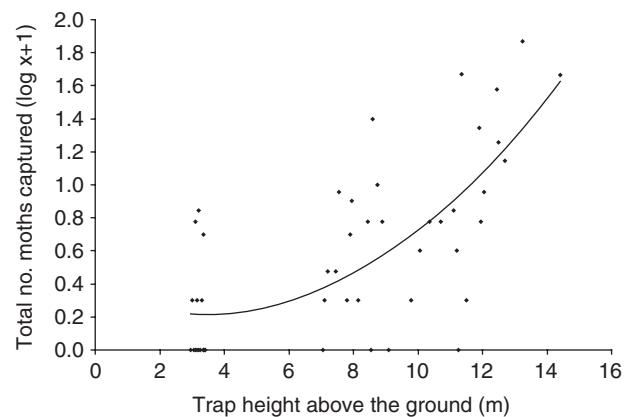


Figure 3 Relationship between the total number of adult male *Thaumetopoea processionea* captured and trap height above the ground (m). Regression line: $y = 0.012x^2 - 0.077x + 0.344$, $R^2 = 0.50$, $P < 0.001$, $n = 48$.

have been more consistent in catching moths, even though both lures caught a similar total number of moths.

Traps placed in the middle or upper canopy were equally likely to catch at least one moth, despite traps in the upper canopy catching many more moths overall (Fisher's exact test, $P = 0.60$; Table 3). Traps positioned in the lower canopy however, were significantly less likely to capture at least one moth than either traps in the middle canopy (Fisher's exact test, $P = 0.029$) or the higher canopy (Fisher's exact test, $P = 0.002$; Table 3).

Chemical analysis of lures

The calibration between the external reference samples of (Z,Z)-11,13-hexadecadienyl acetate and the internal standard

was described best by a quadratic regression ($y = 0.0153x^2 + 0.211x$, $R^2 = 0.99$, $P < 0.001$). GC-MS analysis of the pheromone lures revealed that Lure 2, despite not catching any adult moths, had 5–15 times the quantity of (Z,Z)-11,13-hexadecadienyl acetate compared with Lures 1 and 3 (Table 4). The decline in the amount of the (Z,Z)-11,13-hexadecadienyl acetate component over 28 days was variable but was least for Lure 2 (3.4%), moderate for Lure 3 (12.7%) and relatively rapid for Lure 1 (65.1%) (Table 4).

Discussion

The results obtained in the present study show that pheromone traps caught significantly higher numbers of adult OPM when placed in the upper part of the tree canopy of oak trees (10–15 m); that funnel traps caught significantly more adult moths than Delta traps; and that there were major differences in the numbers of moths caught in traps baited with pheromone lures obtained from different commercial suppliers. The introduction of OPM into the U.K. has necessitated the development of an efficient monitoring system to determine range expansion and population increase, and the findings of the present study clearly demonstrate that identifying the key variables that influence moth captures in pheromone traps is an essential part of the process when developing a successful monitoring programme.

The accidental introduction of exotic insect pests into new locations and their subsequent establishment and spread into new areas usually initiates the development of integrated pest management strategies that aim to eradicate, control or manage the exotic invader. An estimate of rates of spread and changes in the distribution range from the initial point of infestation of the non-indigenous insect pest is an essential part of the pest management process. To enable the detection, and to monitor rates of spread, particularly of non-indigenous Lepidopteran pests, pheromone traps have typically been used with a considerable degree of success (Myers & Hosking, 2002; Sharov *et al.*, 2002; Suckling *et al.*, 2005; Tobin *et al.*, 2007). One of the key factors that often influences trap capture rates is the positioning of the trap in the tree canopy (Cardé & Elkinton, 1984). Studies conducted by Breuer *et al.* (2003a, 2003b) in Germany have also demonstrated that higher numbers of adult male OPM were caught in pheromone traps when they were positioned at 10–15 m in the upper canopy of oak trees. Similarly, in their chemical analysis study of the lure components, Quero *et al.* (2003) note that traps were more efficient at capturing male OPM when placed at this height in the upper canopy, and were less efficient when placed at 2 or 6–8 m. Studies on pine processionary moth (PPM), a close relative of OPM, have also shown that higher numbers of adult males are captured when pheromone traps are placed in the crowns of pine trees (Einhorn *et al.*, 1983; Jactel *et al.*, 2006). For many other Lepidoptera species as well, pheromone trap catches are greater when the traps are placed in the upper part of the tree canopy (Sower & Daterman, 1977; Hanula *et al.*, 1984; Liebhold & Volney, 1984; Bhardwaj & Chander, 1992; Barrett, 1995; Suckling *et al.*, 2005; Kovanci *et al.*, 2006). The present study indicated that male OPM captures in traps did not reach a maximum, and continued to rise with an increasing height

positioning of the pheromone trap in the canopy. Although the present study only positioned traps at three heights within the canopy, traps positioned above the tree canopy might catch more moths, as observed with other Lepidopteran species (Aliniaze, 1983; Pasqualini & Natale, 1999).

The reasons why adult male OPM are caught mainly in the upper parts of the tree canopy remain unknown, although it may be a reflection of the males preferring to fly around the higher parts of the tree canopy because this is where the females tend to congregate and lay their eggs (Dissescu & Ceianu, 1968; Stigter *et al.* 1997; Breuer *et al.*, 2003a). Alternatively, it may relate to the pheromone being dispersed more effectively at greater heights (Breuer *et al.*, 2003a) or the pheromone components combining synergistically with crown volatiles to augment the attraction effect, as suggested for PPM (Jactel *et al.*, 2006).

Trap design also had a significant effect on the numbers of moths caught in the present study, with almost six times as many adult moths caught in the funnel traps compared with the Delta traps. This difference was not attributable to any trap saturation effect because the Delta traps tended to capture only single moths or small numbers of individuals (the maximum catch in a Delta trap was 12), and the sticky pads in the Delta traps were changed every fortnight, regardless of whether they had or had not captured an adult moth. Other studies have also shown a significant effect of trap design on capture efficiency. Breuer *et al.* (2003a) demonstrated that Pherocon traps were more efficient than Delta traps at capturing male OPM moths, yet Wagenhoff and Veit (2011) utilized Delta traps successfully in a 5-year OPM monitoring programme in south-west Germany. Similarly, both Gries *et al.* (2004) and Quero *et al.* (2003) utilized Delta traps to test the efficacy of various formulations of the OPM synthetic pheromone lure, and studies on PPM have shown that funnel traps are not as effective as either Delta traps or sticky traps (Jactel *et al.*, 2006; Athanassiou *et al.*, 2007).

Various aspects of trap design can be influential in determining the numbers of moths captured, including the colour, size, and shape of the trap; the size of the openings; and the structure of the pheromone plume emanating from the trap, along with other factors (Cardé & Elkinton, 1984; Muirhead-Thomson, 1991; Howse *et al.*, 1998). Funnel traps are assumed to generate a more consistent pheromone plume compared with Delta traps, which produce different pheromone plume forms depending on the direction of the wind (Howse *et al.*, 1998). Although the Delta traps did not catch as many moths in the present study, their ability to catch at least one male OPM was not significantly different from the funnel traps and was only marginally lower. Hence, even at relatively low overall OPM population densities, Delta traps may be sufficiently effective for detecting the presence or absence of the moth, and increases in range and colonization of new areas.

A future objective of the pheromone trap monitoring system is to link pheromone trap captures with local population size, especially the number of larvae likely to be found in the surrounding area of the subsequent year. This relationship would provide an early warning of serious infestations and whether control measures might need to be implemented. In this respect, because they catch higher numbers of adult OPM,

Table 4 Initial quantity (mg) (mean \pm SE) and decline over 28 days of the active component (Z,Z)-11,13-hexadecadienyl acetate for three commercially available *Thaumetopoea processionea* pheromone lures

Lure	Time from start of trial (days)					Percentage loss
	0	7	14	21	28	
1	3.16 \pm 0.24	1.45 \pm 0.22	1.10 \pm 0.08	1.03 \pm 0.03	1.10 \pm 0.02	65.1
2	17.3 \pm 2.7	16.6 \pm 5.1	16.6 \pm 12.2	17.5 \pm 0.4	16.7 \pm 0.2	3.4
3	1.12 \pm 0.08	1.16 \pm 0.12	1.06 \pm 0.14	0.95 \pm 0.00	0.97 \pm 0.05	12.7

funnel traps are likely to provide a more robust relationship with local population density and more readily identifiable threshold trap catches for initiating subsequent surveys and management action.

The effectiveness of the synthetic pheromone lure at attracting male moths is fundamental to an efficient monitoring system. Synthetic sex pheromones are now used worldwide in detection and monitoring systems for a vast number of insect pests, particularly Lepidopteran pests, and are essential for determining whether specific insects are present in an area, as well as their seasonal flight periods (Witzgall *et al.*, 2010). The present study demonstrated that the synthetic OPM pheromone lures from Edialux (Lure 1) and Pherobank (Lure 3) were very effective at attracting adult male OPM, although the lures formulated by ISCA Technologies Inc. (Lure 2) were ineffective at catching moths. Breuer *et al.* (2003a) and Quero *et al.* (2003) identified (Z,Z)-11,13-hexadecadienyl acetate as the key active component of the OPM pheromone lure and found it to be effective in attracting adult male OPM. However, Gries *et al.* (2004) demonstrated that (Z,Z)-11,13-hexadecadienyl acetate was not effective on its own as a lure but required the second component (Z,E)-11,13,15-hexadecatrienyl acetate in a 1 : 1 ratio to be effective in attracting the male moths. Communication with the manufacturers of the lures used in the present study indicated that this second component was present in Lure 3, was probably present in Lure 1, and was not present in Lure 2, tending to support the findings of Gries *et al.* (2004) in that a second component is required in the pheromone lure for OPM to be fully effective. Clearly, the correct mix of pheromone components is essential, and even subtle differences in lure composition can alter their effectiveness (Howse *et al.*, 1998; El-Sayed *et al.*, 2006). It is rare for a single chemical constituent to be the only component of a sex pheromone; it is generally more common for a blend of pheromone components to be required to elicit the full behavioural response from the male moth (Muirhead-Thomson, 1991). The (Z,Z)-11,13-hexadecadienyl acetate component of the OPM lure may be important for the initial attraction of the male moth, perhaps operating over a long-range, eliciting a behavioural response in the moth to fly towards the source of the pheromone. The (Z,E)-11,13,15-hexadecatrienyl acetate component of the pheromone lure, however, may be required by the male moth as it approaches the lure, and perhaps operates over a short-range, and elicits the behavioural response in the male to land close to the pheromone source. This clearly needs investigating further for OPM, although this type of behaviour in response to a blend of pheromone components has been observed in other Lepidopteran pests (Baker & Cardé, 1979).

Our chemical analysis showed that Lure 2, despite not catching any moths, had higher quantities of (Z,Z)-11,13-hexadecadienyl acetate than either Lures 1 or 3. Very high concentrations of sex pheromones can have an inhibitory or repellent effect on attracting male moths to traps compared with low or moderate concentrations (Baker & Cardé, 1979; Cardé & Elkinton, 1984; El-Sayed *et al.*, 2006). However, this is unlikely to have occurred in the present study because Breuer *et al.* (2003a) did not find any inhibitory effects on male OPM capture at high dosages of 15 mg, which is a dosage comparable to that found on Lure 2. Jactel *et al.* (2006) also demonstrated that there was no repellence effect to male PPM capture in increasing the pheromone dosage of lures, and a similar observation has been reported with pheromone lures for gypsy moth *Lymantria dispar* (L.) (Cardé *et al.*, 1977; Plimmer *et al.*, 1977). However, in both of these latter cases, plateauing of trap catch was either observed or speculated to occur at high dosages.

Another reason for Lure 2 being ineffective may relate to the rubber septas on which the OPM sex pheromone chemicals are coated. Lure 2 was a synthetic rubber septa, whereas Lures 1 and 3 were natural rubber septas, and there may have been differences in the release rates of the active pheromone component from the two types of rubber. Pheromone volatilization from the synthetic rubber septa of Lure 2 appears to have been much slower (only 3.4%) over 28 days than from Lures 1 and 3 (65.1% and 12.7%, respectively). In addition, varying amounts of anti-oxidants, stabilizers and UV protecting agents are incorporated into synthetic pheromone lures to protect the pheromone components from temperature, UV light, and oxidation, which could otherwise lead to excessive degradation rates or increased rates of isomerization (Howse *et al.*, 1998; Heuskin *et al.*, 2011). The varying quantities of these additional materials might influence the relative rate of release and breakdown of the pheromone components and the subsequent attractiveness of the lures. The rates of loss of the key pheromone component from the lures in the present study were variable and this may have been attributable to differences in these other chemical constituents. Lures 1 and 3 caught similar numbers of male OPM; however, Lure 3 catches were less variable and a higher proportion of the traps with this lure (87.5%) caught at least one moth compared with Lure 1 (54.2%). Therefore, Lure 3 was better at detecting the presence or absence of OPM, which is perhaps related to the key active pheromone components being less variable and more stable.

It is clear from the present study that the positioning of the pheromone trap in the tree canopy, the design of the trap,

and the source of the pheromone lure together influence the number of adult male OPM caught in pheromone traps, and all of these factors need to be taken into account when designing a standardized monitoring programme for this significant insect pest. The results of the present study clearly demonstrate that pheromone trapping would be suitable for monitoring OPM populations not only within the core outbreak area of the pest in west London, but also within the expansion range of the insect. Further research should aim to investigate the relationship between pheromone trap catches and local population densities, in terms of observed larval nests, so that appropriate control measures can be initiated and timed as efficiently as possible whenever threshold trap captures are attained.

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