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chain waxes, low boiling point semi-VOCs, and other lipid components can also be identified by the same method, which can be adopted to be an automated high-throughput method for insect classification, surveillance and quarantine purposes.

**Keywords:** direct immersion solid phase microextraction (DI-SPME), fatty acids & sterol lipids, biomarker, stored grain insect, insect morphology and identification.

# **Webbing Clothes Moth, Tineola bisselliella (Hummel) Sex Pheromone Transfer from Monitoring Lures to Textiles**

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#### **Abstract**

The use of synthesized sex pheromone lures for the purpose of monitoring populations of webbing clothes moth, *Tineola bisselliella* (Hummel) in museum storage environments is typical in many museums. Questions about whether the pheromone incorporated in the dispensing lures could possibly transfer over to textiles that are in close proximity to the lures have been posed by museum conservators. Although some textiles may be naturally attractive to clothes moths, the concerns are that the textiles themselves may become even more attractive to insects due to exposure to the pheromone and that this could ultimately cause further damage to the collections. The focus of this study was to determine the degree to which textiles that have been exposed to pheromone lures absorb the pheromone and become attractive themselves. Based on the results of this study, the textiles observed here have little to no additional attraction to insect pests after focused exposure to synthetic pheromone lures over a two-week period.

**Keywords:** Webbing clothes moth, Tineola, sex pheromone, textile, monitoring.

### **1. Introduction**

The webbing clothes moth, *Tineola bisselliella*, is a cosmopolitan pest that carries economic importance due to damage caused by their larvae feeding on objects that incorporate wool, feather, hair and hide (Krüger-Carstensen and Plarre 2011). Textiles that incorporate cotton, silk, linen, paper and synthetic fibers can also be damaged by *T. bisselliella* if these items have been soiled with urine, sweat, beer, milk, soft drinks, tomato juice or other substances that contain nutritional needs for the moths (Sloderbeck 2004).

Being one of the most common pests in museums in many parts of the world, this species of moth has caused severe damage to cultural heritage objects (Querner 2014). The use of synthetically produced sex pheromone monitoring lures specifically for *T. bisselliella* for the purpose of early detection and locating sources of infestation has become commonplace in some museum institutions to prevent this damage. The use of a pheromone lure within a sticky trap increases the rate of capture twenty-fold over a sticky trap with no lure (Cox et al. 1996) and is a key factor in determining increases in population density and economic thresholds (Plarre 2013).

Concern over the practice of pheromone monitoring was raised by a prominent museum conservation scientist and author who believed that the pheromone incorporated in the dispensing lures would transfer over to museum objects (Florian 1997). Following up on this, this same author made a statement in an online museum conservation listserv that suggested that the volatile fatsoluble pheromone can be adsorbed by materials of artifacts and thus make the artifacts themselves attractive to insect pests (Florian 2011). This posting suggests that even after monitoring lures are removed, the museum collections would continue to attract and draw-in damaging museum pests. The question that this study aims to answer is if pheromone transfer between the sex pheromone lures and a variety of textiles found in museum storage environments is occurring and if these pheromones are making the textiles themselves attractive to pests.

### **2. Materials and Methods**

### 2.1 Exposing the Pheromone to the Textiles

In order to answer the question of whether textiles exposed to sex pheromone monitoring lures become more attractive to the insect pests themselves, it was first necessary to establish a means of exposure for the textiles so the theory could be tested. Pheromone plumes emanating from monitoring lures are typically carried by air currents out to the surrounding areas where they attract the insects back to the lure (Murlis et al., 1992). In order to ensure exposure of the textile to the pheromone in this study, a constant air current generated by electric fans was blown across the lures towards the textile at an air speed of  $40 \pm 1.5$  meters/min for a 2-week period in controlled temperatures between 21.1° C and 22.7° C and within a relative humidity between 40% - 50%. This exposure system was set up using eight 30.48 cm long sections of 10.16 cm diameter corrugated polyethylene field drainage tile as a conduit for the air flow (Figure 1). The fans were placed 40.6 cm away from the corrugated field drainage tiles and were directed to blow air through the open center of the tiles. The airflow was calculated using a hand-held anemometer (#DCFM8906, General Tools & Instruments, Secaucus, NJ, USA). The pheromone lures used in this study were standard, commercially available webbing clothes moth Bullet® lures (Insects Limited, Westfield, IN USA). The lures used in the study had been manufactured within the previous month of the study, were frozen to ensure freshness and were then taken fresh from the package. These lures incorporate a pheromone dose of 4.5 micrograms per lure. This dose can be considered on the high end of commercially available pheromone lures for webbing clothes moth (Van Ryckeghem, 2014). The lures were suspended on the inside of the drainage tile using a flexible metal wire positioned at the opposite end from the fan. A screen mesh was placed over the open end of the drainage tile on this same side. This screen was set in place for the purpose of creating a physical barrier between the lure and the textiles being exposed, while still allowing air to flow freely across the lure and onto the textile. No direct physical contact between the pheromone lures and the textiles was made in any of our studies. The mesh screens were standard fiberglass insect screening with a 7 X 6 mesh count per cm and the fabric was 0.3 mm thick. The close-range exposure between the lures and the textiles was performed using only the screen mesh between them at a distance of 0.3 mm. A single set a data points was retrieved at the greater distance of 152 mm between the lure and modern synthetic pile carpet to give data that represents a distance that is more commonly found in a museum setting.

The five textiles that were chosen to be exposed in this study were selected as being textiles commonly found in museum storage settings. These textiles include:

- Antique Wool Pile Carpet (mid to late 19th century)
- Modern Synthetic Pile Carpet (late 20th century)
- Modern Synthetic Plain Weave (early 21st century)
- Antique Wool Plain Weave (mid to late 19th century)
- Antique Wool Flannel (late 19th century)

Relatively larger 30 cm<sup>2</sup> sheets of the various textiles were cut into smaller 50 mm X 50 mm squares for use in the exposure study. The textile squares were secured to the screen mesh using metal paper clips and were placed directly on the opposite side of the screen from the lures to ensure exposure to the pheromone. The textiles were handled only while the technician was wearing latex gloves to prevent any exchange of pheromone from person to textile. After an exposure period of two weeks to allow the sex pheromone to blow directly across the lures onto the textiles, the textiles were immediately taken and placed into 10.16 cm X 15.24 cm, 4 mil Metalized PET (Polyethylene terephthalate) Zipper Pouches. The zipper pouches were then sealed and placed into a standard upright freezer (-20°C) until they were used in the insect portion of the study. The PET is considered a barrier film for oxygen (Frounchi and Dourbash 2009). Since pheromones are larger molecules than oxygen, the PET pouches can also be considered a barrier for the pheromone that will retain any pheromone absorbed onto the textile. Freezing the samples also slows molecular movement (Debenedetti and Stillinger 2001) and thus should slow any loss of pheromone out of the textile pouches and into the environment prior to use in the study.

After pheromone exposure of the textiles was performed, the second portion of this study was to determine if the adult clothes moths prefer pheromone-exposed textiles over non-exposed textiles of the same material. This determination was made with a choice test that included 4 different options for the adult moths to choose. The four options are:

1. Sticky trap containing a 50 mm X 50 mm square of textile that has been exposed to the pheromone and placed into the center of the base of the trap.

2. Sticky trap containing a 50 mm X 50 mm square of the same textile as above that has not been exposed to the pheromone and placed into the center of the base of the trap.

3. Sticky trap containing a pheromone Bullet lure specific for webbing clothes moths placed into the center of the base of the trap as a positive control.

4. Sticky trap with no textile or attractant inside, used as a control.

The pheromone lure option and the empty trap option were added as controls to the choice test to give comparative trap capture numbers: source moths are known to be attracted to (pheromone lure) and source that should have no attraction (empty trap).

The test arena that was used in the choice test was a 2.7 m X 4.0 m space that included a desk and storage cabinets (Fig. 2). This setup gave the moths plenty of hiding spaces other than the traps, if they preferred to not go to a trap at all. Moth colony jars, active with adult *T. bisselliella*, were opened on a platform 0.74 meters above the floor and at a distance of 2.06 meters from the wall where the traps were placed. The traps that incorporated a textile square on the inside for this study were prepared by taking a textile from the freezer and placing it into the center of a sticky trap. These traps were made of milk-carton stock, wax-coated cardboard with the interior base of the trap coated with a 1 – 2 mm layer of sticky adhesive (Flat Trap adhesive trap, Insects Limited, Westfield, IN, USA). The dimensions of the traps were 20.32 cm X 10.16 cm X 3.81 cm. All four traps were set on the floor and spaced at a distance of 53.34 cm apart from each other. These locations are marked 1 through 4 in Fig. 2.

Webbing clothes moth colonies were reared in 2-Quart (1.89 liter) screw-top canisters with 5 pin holes on the upper side of the canister. The pin holes were made to allow air exchange in and out of the canister. The diet consisted of a mixture of chicken feather meal with 1% brewer's yeast by weight. The colony jars were opened and placed on their side to aid in the release the adult moths.

After one week of allowing the moths to move out of the colony jar and enter the test arena, the individual trap captures were counted and recorded and the traps were rotated to a new trap location in the arena. Also, after one week, the existing colony jar of live moths was removed and a new colony jar with freshly emerged adult moths was opened in its place. A total of 4 repetitions, totaling 4 weeks of release for each textile, were performed. Each trap in the study would spend one week's time at each location of the four without duplication of location during that 4-week period. A total of six individual 4-week trials were run in this study. Five of those studies involved each of the different textiles exposed at the short 0.3 mm distance to the lure. The sixth trial was a single trial of the Modern Synthetic Carpet exposed to pheromone from the greater distance of 152 mm.

### **3. Results**

Totals of captured moths after 4 weeks varied from 107 to 323 based on the number of adult moths in the colony jars at the time of release and the attraction of the traps. Throughout the six trials, a total of 913 *T. bisselliella* were captured in the different traps. It is estimated that a total of > 2000 moths were released through these studies. The results of the capture numbers for each individual textile, as well as the controls and pheromone lures can be seen in Fig. 3 – 7.

A statistical analysis was prepared using the Kruskal-Wallis *H* test (Microsoft Excel 2013). The Kruskal-Wallis test is a rank-based non-parametric method for one-way analysis of variance test that compares the samples even though they may have different sample sizes. The results of the Kruskal-Wallis test can be found in Table 1.



**Fig. 1** Image of pheromone exposure to textiles with anemometer



**Fig. 2** Diagram of clothes moth test arena



**Fig. 3** - Choice test trap capture results for antique wool pile carpet exposed to pheromone at a distance of 0.3 mm.



**Fig. 5** Choice test trap capture results for modern synthetic plain weave exposed to pheromone at a distance of 0.3 mm.



**Fig. 4** Choice test trap capture results for modern synthetic pile carpet exposed to pheromone at a distance of 0.3 mm.



**Fig. 6** Choice test trap capture results for antique wool plain weave exposed to pheromone at a distance of 0.3 mm.



**Fig. 7** Choice test trap capture results for antique wool flannel exposed to pheromone at a distance of 0.3 mm. **Tab. 1** Results of non-parametric Kruskal-Wallis tests comparing trap capture rates of combined textile types.



## **4. Discussion**

The procedures used to expose the textiles to the pheromones in this study represent what could be considered a worst-case-scenario for pheromone exposure in a real-world setting.

A lure with a relatively high dose of pheromone was used in this study. This is not always the case in a museum storage environment as pheromones with low dosages are commonly used. Also, pheromone traps are typically not placed in direct contact with museum objects as this could be detrimental to the object if it were to get stuck in the glue. Pheromone traps for pest insects and particularly traps for *T. bisselliella* are usually placed in open areas along the wall or on the floor so they can be inspected easily (Trematerra and Fontana, 1996). Even a pheromone trap that is in direct contact with a museum object is going to have the pheromone lure at a minimum of 1 cm away from the object due to the distance from the paper outside of the trap to the adhesive pad within where the lure rests. It may even be as high as 15 mm away depending on where the lure sits in the trap. These distances are considerably higher than the 0.3 mm that we used in this study.

Air currents are the mechanisms that translocate quantities of the sex pheromone from the lure into the air or onto a textile. In this study, a constant air flow was blown across the lure at a rate of approximately 40 meters/min  $\pm$  1.5 meters/min for a full 2-week period. This type of constant air flow across a lure and onto a textile is usually not seen in a museum storage setting unless the textile is placed directly between a return air vent and the pheromone lure or if the pheromone lure is placed directly in front of an air supply vent and the textile is positioned directly in the air path of that vent.

Although this study does represent a worst-case scenario for textile exposure to pheromone, this type of exposure theoretically could occur in a museum setting. Because of this potential, the questions of concern for this type of exposure need to be considered valid. Correlations between this study and other similar studies could not be done since other studies regarding the pheromone transfer from monitoring lures to textiles were not found in the available research.

There was a wide range of materials incorporated into the textiles that were studied here. Antique natural fibers were used in three of the five samples; wool pile carpet, flannel and antique wool plain weave. Also incorporated in some of these samples were synthetic materials that contained no 12th International Working Conference on Stored Product Protection (IWCSPP) in Berlin, Germany, October 7-11, 2018

natural fibers at all. These were the new pile carpeting and modern synthetic plain weave. *Tineola bisselliella* larvae feed on a wide variety of dried material of animal origin (Griswold, 1944). This fact should theoretically make the woolen textiles more attractive than the synthetic textiles, at least to the female moths looking to lay eggs. When we look at the results from this study however, we find that only the antique wool carpet and the modern synthetic flat weave had apparently higher moth attraction than the control trap. No clear affinity for natural fibers over synthetic fibers could be found. It is possible that many of the females in the study were left unmated due to a large capture of the male moths in the traps. If this were the case, the unmated females were not looking for potential food sources to lay their eggs, so we did not see an affinity for the natural fibers. The addition of human sweat, urine or food stains to natural fibers can make these materials more attractive to *T. bisselliella* (Klass, 2010). A possible explanation for the low attraction is that the samples we used containing natural fibers did not contain any of these additional attractants.

#### **5. Conclusions**

The textiles in this study, whether exposed to pheromone or not, did not have greater captures than control traps (Table 1).

Given these results, it is unnecessary for museum staff to be overly concerned that they are making their textile collection objects more attractive to *T. bisselliella* if they are using pheromone monitoring traps within their collections storage. This study suggests that *T. bisselliella* monitoring traps are an effective, non-detrimental tool. The informational value gathered through use of the pheromone traps used to mitigate damage to collection textiles and objects, far outweighs any negative possibilities that the textiles themselves will attract pests into storage areas.

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