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Review Article

Sex-Pheromone-Mediated Mating Disruption Technology for the Oriental Fruit Moth, *Grapholita molesta* (Busck) (Lepidoptera: Tortricidae): Overview and Prospects

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A great deal of progress has been made over the last three decades in research on pheromone-mediated mating disruption technology for the oriental fruit moth, *Grapholita molesta* (Busck). Pheromones can interrupt normal orientation, and the most likely mechanism of pheromone disruption, competitive-attraction (false-plume following), invokes competition between point sources of pheromone formulation and females for males. This technology, performed by broadcasting pheromones into orchards to disrupt mate finding, has been successfully implemented in oriental fruit moth control. Reservoir-style dispensers made of polyethylene tubes, which release pheromone throughout the full growing season, are the current industry standard. Although reasonably effective, they require labor-intensive hand application. Recently, a new formulation, paraffin wax, which maximizes competition between point sources of synthetic pheromone and feral females for males, was shown to have high disruption performance. As this formulation is highly effective, inexpensive, and easy to produce, further study and development are advisable. Increased understanding of the principles of mating disruption will aid in the design of more effective dispensers. Continued research is needed to meet grower concerns with regard to risk, efficacy, and cost and to identify other semiochemicals that can be applied to this delivery system. Greater knowledge of the integration of different biological control methods is therefore essential.

1. Introduction

The oriental fruit moth (OFM), *Grapholita molesta* (Busck) (Lepidoptera: Tortricidae), is a key pest of stone and pome fruit in most fruit-growing areas of China, with the exception of Tibet [1–6]. Until recently, this pest has been primarily controlled by use of one or more broad-spectrum insecticides [7]. Issues associated with the widespread use of insecticides, including insecticide resistance, toxicity to natural enemies, worker safety, and food residue, have provided an impetus for research and development regarding alternative control technologies. The application of pheromone-mediated mating disruption technology has resulted in excellent control of this pest and could be an alternative to conventional insecticide use [2, 8]. This review presents the principle of pheromone-mediated mating disruption, summarizes the typical application of this technology, introduces its pheromone release device (pheromone dispenser), and discusses future directions for research and development.

2. Principles of Pheromone-Mediated Mating Disruption

Pheromone-mediated mating disruption controls insect species mainly by using chemicals involved in its own communication system [9]. Although it is not necessary to understand the mechanism underlying mating disruption to verify its efficacy, analysis of probable modes of action is useful to determine the reasons behind success and failure of various formulations [2].

Sex pheromones are specific chemicals released by females into the air to attract conspecific males for mating. Males follow the sex pheromone upwind to locate and mate with the female [9]. However, if the air in an orchard is filled with synthetic sex pheromone, males would encounter high-dose artificial point sources of pheromone and low-dose calling females' point sources of pheromone during their upwind zigzag flight. As males preferentially orient toward artificial point sources of pheromone than pheromone plumes from calling females, they would be unable to accurately locate calling females and this would greatly modify moth flight tracks. Therefore, synthetic sex pheromone successfully prevents males from finding and mating with calling females. This sex-pheromone-mediated mating disruption technology can protect orchards from pests, including the OFM, and ultimately achieve long-term reduction of the pest population [2, 9]. Many studies have indicated that competitive attraction; that is, false plume following, may be a leading mechanism underlying disruption of OFM mating by synthetic sex pheromones [10–13] and the most feasible principle of pheromone disruption [2, 14–16] including sensory adaptation or central nervous system habituation on males, while sensory imbalance affects the female plume and the way in which it is perceived by males. When encountering a high concentration of formulated pheromone above that produced by calling females, males may show an increase in response or complete abolition of responsiveness to subsequent pheromone released by females, because of adaptation of peripheral receptors on the antennae or habituation at the central processing level. In lepidopterous insects, pheromones are generally comprised of more than a single component with a very narrow range of ratios. An imbalance in sensory input may be produced and male response to the natural ratio of the components may be decreased by the disproportionality of blends or lack of partial components. In addition, reduction of female copulation propensity may be a secondary mechanism of mating disruption that affects the mating behavior of females in addition to that of males [17]. As an indicator of high competition, the high levels of sex pheromone perceived by females are unsuitable for reproductive success, thus restricting their receptiveness to copulation and causing the loss of female receptivity to mating. Mating of females may be adversely affected following sex pheromone autoexposure due to abnormal behavioral activity, which is thought to be because the preexposed females may be unable to sense the aphrodisiac pheromone of conspecific males and/or antennal sensitivity, which is interpreted as the requirement for male aphrodisiac, which plays a role in courtship, for the occurrence of adaptation of antennal responses, following sex pheromone preexposure.

Attention should be paid to the interplay between these principles. With regard to male response threshold raised by encountering a high-dose, artificial source of pheromone, subsequent male responses minimize the odds of detecting females that emit relatively low levels of pheromone, and in turn the enhanced competition effect of the two point sources of pheromone may encourage males to visit a high-dose point-source formulation than to locate and mate with a female. Taken together, these principles are helpful to optimize the effect of mating disruption technology by guiding the behavioral peculiarities of the insect to be managed, its spatial distribution, the type of formulation employed, and the rate of formulation application [16, 18].

3. Application of Sex-Pheromone-Mediated Mating Disruption

The sex pheromone of OFM was determined to be a mixture of four components [19]: (*Z*)-8-dodecenyl acetate (*Z*8-12:Ac), (*E*)-8-dodecenyl acetate (*E*8-12:Ac), (*Z*)-8-dodecen-1-ol (*Z*8-12:OH), and dodecanol (12:OH). As a disruptant, only the (*Z*)- and (*E*)-isomers of the acetate were used in early work performed in Australia [20] and USA [21, 22], indicating that OFM was highly susceptible to communication disruption. Later studies [19, 23] indicated that the addition of *Z*8-12:OH and 12:OH reduced the amount of pheromone required for disruption and for accurate location of hosts over short distances, respectively. Field results in a number of countries [2, 8, 9, 20, 24–34] indicated that this technology was capable of truly managing pesticide-resistant populations throughout the whole growing season and was therefore fully equivalent or superior to conventional pesticides. Therefore, sex-pheromone-mediated mating disruption technology could provide complete crop protection [2, 35].

4. Pheromone Dispenser

Based on the above discussion, use of a good pheromone dispenser plays a key role in achieving high-performance mating disruption. First, an ideal pheromone dispenser should remain effective for a prolonged period, not waste active ingredients, be inexpensive to produce, be easy to use in the field, and be nontoxic [36]. Furthermore, pheromone dispensers should be amenable to use at varying densities and deployment dates according to pest pressure. In addition, it should achieve the availability of a controlled release device to encourage growers to adopt mating disruption technology [37–41].

4.1. Common Pheromone Dispensers. If competitive attraction is the foremost mechanism underlying mating disruption, as suggested by recent studies [10–12], various pheromone dispensers would be desirable as shown in Table 1. The three most popular types are illustrated here, that is, hollow fiber dispensers, polyethylene tube dispensers, and sprayable formulations of microscopic capsules.

In the 1970s, Cardé et al. (1977) deployed 1700 hand-applied hollow fiber pheromone dispensers per ha according to this behavioral modification tactic and achieved successful control of OFM, thus indicating that this is a promising alternative to broad-spectrum neurotoxins [21, 42].

In the 1990s, hand-applied polyethylene tube dispensers, such as Isomate-M, M 100, and M Rosso (Pacific Biocontrol Co., Litchfield Park, AZ) became available for commercial use for disruption of OFM [25, 27, 43–47]. These dispensers were filled with 75–250 mg of OFM pheromone and applied by hand at 500–1000 units/ha (corresponding to 1–4 dispensers/tree), and pheromone release per dispenser varied between ca. 600–1000-fold as much as that produced by a calling female. Accumulating evidence showed that

Table 1: Use of the pheromone-mediated mating disruption technique for oriental fruit moth in various countries [2, 8, 9, 20, 24–34].

Country	Type of dispensers	Density (/ha)	Efficacy and extension
France	Polyethylene tube	1000	The two kinds of dispensers were the most effective and simple to use.
	Polyethylene bulb	500	
US	Polyethylene tube	1000	The pheromone-treated peach and nectarine orchards in California and Virginia extended from 600 ha in 1987 to 4000 ha in 1990.
	Microcapsule	—	Male OFM captures were reduced by 77–98% in 35 d, and the formulated pheromone significantly disrupted male orientation.
Japan	—	—	The effect of mating disruption was greater than that of trapping.
Korea	—	—	The effectiveness of the disruption technique was enhanced through sequential suppression year after year at the same site.
Australia	400 mL microcentrifuge tube	—	In this extensive 3-year trial, use of pheromone treatment increased from 25 to 40 ha and confirmed that conventional insecticides could be replaced by mating disruption from the viewpoint of cost and efficacy. These successful tests encouraged use of mating disruption in over tens of thousands of hectares in the peach-growing districts in New South Wales and Victoria.
	Polyethylene capillary	500	Male OFM capture was reduced by 77–98%.
		1000	OFM males preferred untreated orchards adjacent to pheromone-treated orchards.
	Polyethylene tube	—	Pheromone treatment reduced the capture of moths in pheromone-baited traps by an average of 98%, suggesting a high level of disruption.
	—	—	Application of mating disruption barriers on pears during two consecutive seasons provided sufficient control of OFM on peaches, and this mating disruption barrier treatment was able to reduce the number of OFM caught in all experimental peach blocks. Extending the mating disruption treatment area for 54–60 m into the neighboring pear block significantly reduced the edge damage in mating disruption-treated peaches in the first season and almost eliminated OFM damage in the second season.
	—	—	Mating disruption was a viable alternative to conventional insecticides.
China	Rubber septa	—	In Liaoning province, Eastern China, the percentage of infested fruits in the pheromone-treated orchards was reduced to 50% and 72% compared with the insecticides-treated orchards in 1981 and 1982, respectively; this technology was extended on a large scale.
		1050	In pear orchards of Shanxi province, Northern China, male OFM orientation was disrupted by 97.43% and the percentage of infested fruits was reduced by 74.72% in 2009.
		750	In pear orchards of Shanxi province, Northern China, male orientation was disrupted by 81.83% and the percentage of infested fruits was reduced by 56.43% in 2009.
		3000	In peach orchards of Shanxi province, Western China, after 37 d, under no insecticide, male orientation was disrupted by 93.46% and the percentage of infested fruits was reduced by 73.72% in 2007.
	Twist tie	374	In peach orchards of Shanxi province, Western China, after 37 d, under no insecticide, male orientation was disrupted by 97.19% and the percentage of infested fruits was reduced by 81.61% in 2007.

the relatively high densities of pheromone release sites at common overall release rates of pheromone per ha could achieve superior disruption for most moths [12, 13, 23, 48–53]. However, because of the appreciable costs of purchase and labor for hand application, polyethylene tube dispensers have not been widely adopted in some production systems and in many developing countries [53].

Early in the 21st century, sprayable formulations of microscopic capsules that release pheromone for prolonged periods were developed and shown to be effective against OFM when properly applied [4, 46, 54]. They were generally applied with standard air-blast sprayers. Not only they were considered a cost-saving alternative to hand-applied dispensers, but also they can be tank-mixed and coapplied with other orchard management chemicals [4, 54]. However, they maintained effectiveness for only 2–4 weeks and required more frequent applications than hand-applied reservoir dispensers [4, 47, 54]. Other drawbacks also included wash-off of microcapsules by heavy rain and degradation of active ingredients by UV irradiation [55, 56].

In addition, due to their ease of production and constant release rate, rubber septa dispensers are common and effective means of controlling OFM in China. Special lures for mating disruption are directly suspended in the upper third of the tree canopy at 200–400 units/ha without traps. However, the cost associated with this method is ca. US$180/ha per year, so they are unsuitable for common orchards in China. In addition, the rubber septa age rapidly, the duration of pheromone release is short, and hand application is expensive [57, 58]. Although they are mainly used for baiting monitoring traps, many research groups spent considerable time and effort to develop more reliable and efficient methods of using rubber septa [32–34, 57, 58].

4.2. New-Style Pheromone Dispenser: Paraffin Wax. Dispensers in which insect sex pheromones are mixed at the required concentrations into paraffin wax emulsions have been used in USA for almost 16 years [59, 60]. Two typical wax-paraffin dispensers, that is, Confuse-OFM and SPLAT-OFM, are described below.

Confuse-OFM resembles white, liquid glue and is applied using squirting devices, such as forestry paint marking guns and plastic squirt bottles [53, 61]. Use of this type of dispenser was shown to inhibit capture of male moths in pheromone traps and shoot damage as effective as Hercon (Hercon Environmental, Emigsville, PA) and Consep (Consep, Inc., now Suterra LLC, Bend, OR) hand-applied pheromone dispensers [59]. The University of California at Davis patented this emulsion (U.S. Patent 6,001,346) [62] and it was later commercially developed by Gowan Co. (Yuma, AZ) [53].

SPLAT-OFM consists of microcrystalline wax emulsified in water and so it can be pumped from a storage reservoir and sprayed onto the crop, and it shows long-lasting adhesion of dispensed particles on plant surfaces [63]. Stelinski et al. (2006) reported that mechanical application of SPLAT-OFM could save time and labor for mating disruption of OFM in apple orchards [64]. In 2003, ISCA Technologies, Inc. (Riverside, CA), patented this wax emulsion, and extensive testing of this technology was later performed along with adaptation for a variety of pests and crop systems (ISCA Technologies, Riverside, CA). ISCA Technology's Specialized Pheromone & Lure Application Technology (SPLAT) has been granted a federal registration for OFM control by the U.S. Environmental Protection Agency, and it is now commercially available as SPLAT OFM 30 M-1 [53].

4.2.1. Efficacy. Optimization of mating disruption requires that the density and size of droplets, pheromone release rate, and duration are appropriate for the biology of the targeted pest [13, 14]. Researchers can not only easily manipulate the size, density, and distribution of wax droplets, but also flexibly investigate how moths could be actually disrupted [65].

In 2005, male OFM orientation was shown to be disrupted more effectively by deploying ~8,000 0.1-mL drops of SPLAT-OFM per ha (each containing ca. 1% of the total pheromone active ingredient of a standard Isomate dispenser) compared with the label rate of 500 Isomate-M Rosso dispensers per ha, probably because sufficient point sources of pheromone were provided for optimal disruption of OFM with the typical deployment density of pheromone twist ties [51]. In 2006, male OFM orientation was shown to be disrupted by 98% relative to untreated control plots during the whole season using SPLAT-OFM, and either increasing the size of wax drops above the average volume of 0.04 mL achieved by the initial applicator prototype or reformulating the wax to allow for a higher initial pheromone loading concentration for longer release over time, especially in hot temperatures, could maintain efficacy and improve longevity [64]. In 2007, two applications of Confuse-OFM were shown to be as effective against OFM as one application of Isomate-M 100. A new emulsified wax formulation, Wax Dollops, was developed in 2007 with a release rate exceeding a 5 mg/ha/h threshold and duration of action that is twice as long as Confuse-OFM. One application of 3 mL dollops (ca. 590 dollops per ha) provided season-long (ca. 15 weeks) control, which was equivalent to the effects of Isomate-M 100 and Confuse-OFM applied as described above [53].

4.2.2. Advantages. Paraffin wax dispensers are inexpensive and easy to produce. Paraffin wax consists mostly of water and wax, which is a byproduct of petroleum refining, and it is therefore readily available and inexpensive [63, 64]. Wax emulsions can easily be increased proportionally and manufactured on a large scale with minimal labor [53]. Therefore, commercial production of wax emulsions should be cheaper than other currently available hand-applied formulations.

Paraffin wax is a viscous homogenate that hardens on crop foliage or branches once applied and therefore can act as a long-lasting discrete source of pheromone emission. Delwiche et al. (1998) reported that one of the initial formulations (30% paraffin wax emulsified in water, vitamin E, soy oil, and antioxidant) was as effective as Shin-Etsu, Isomate-M 100 polyethylene-tube dispensers for 75 days in the field [61]. Subsequently, one application of a more viscous version of the above-described paraffin wax dispenser provided the same level of season-long disruption of OFM as Isomate-M 100 dispensers and could be hand-applied once in less time than Isomate-M 100 dispensers [50].

Paraffin wax dispensers are rapidly applied mechanically, and there is a cost-saving advantage to mechanical application [51–53, 60, 65]. For example, SPLAT can be easily applied with a machine forming numerous discrete point sources per area of crop [64]. A single operator can treat a hectare of crop with the current mechanized applicator in ca. 20 min, which is approximately 3.4-fold faster than hand-application of Isomate-M Rosso dispensers by three people [50]. Therefore, SPLAT-OFM currently represents an economical alternative to hand-applied reservoir dispensers for high-performance mating disruption of OFM.

In addition, the flowable, adhesive, and dispersible emulsified wax can be applied with a wide range of deposit sizes and spatial distributions [53]. Furthermore, paraffin wax dispensers contribute to effective disruption of communication for other moth species [13, 23, 48–52] and are not phytotoxic, so they do not damage foliage and/or mark fruit [66]. In addition, insecticides have been incorporated into emulsified wax to produce effective attracticide formulations (ISCA Technologies).

4.2.3. Drawbacks. Pheromones are costly—for season-long control of OFM, the cost of Confuse-OFM was three times that of Isomate-M 100 (148 g AI/ha vs 57 g AI/ha) [53], and 160 g/ha of pheromone of SPLAT-OFM exceeded the 125 g AI/ha label rate of the Isomate-M Rosso reservoir dispenser [63].

In contrast, hand-application of dispensers is both time-consuming and expensive. Season-long control of OFM requires two applications of Confuse-OFM, and its application is laborious because this liquid formulation requires care and time for application to the tree bark [53]. In addition, even though SPLAT-OFM is applied mechanically, machine applicators are not affordable for individual growers, so the initial investment in the applicator for application of SPLAT should be provided by the manufacturer and/or distributor [64].

5. Future Prospects

As a major fruit pest [1], OFM is a long-standing target for the development of mating disruption programs [19, 21, 67, 68]. Therefore, accumulating evidence of the reliable and economic applications of pheromone-mediated mating disruption will lead to more widespread adoption of this technology.

First, it is necessary to determine the actual costs of OFM pheromone dispensers as well as the relations between the total costs for one or two applications of pheromone dispensers (materials and labor) in comparison with three insecticide and/or miticide applications (materials and labor) [2].

Second, laboratory assays are required to predict the effects of various types of dispensers on OFM behavior in the field, because the development of mating disruption technology still relies on repeated field trials and therefore remains both costly and slow [69].

Finally, sex-pheromone-mediated mating disruption is currently specific for male OFM, and we should develop other semiochemical-based methods, such as plant volatiles [70, 71], directly targeting females as the most important complement to mating disruption. In addition, more emphasis should be placed on the integration of different biological control methods, such as use of microbial pesticides [72], to reinforce the effects of behavior-modifying chemicals. All biological methods, such as black light [73], are to some extent species-specific and do not cover all pests associated with a crop, so it will be necessary to develop new mating disruption technology for OFM and other lepidopterous pests in orchards [17, 69, 74].

Conflict of Interests

The authors declare that there is no conflict of interests.

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