



Article

# Unattended Trapping of Whiteflies Driven out of Tomato Plants onto a Yellow-Colored Double-Charged Dipolar Electric Field Screen

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**Abstract:** An unattended pest control system was developed to eliminate whiteflies (*Bemisia tabaci*) that settled on greenhouse tomato plants. The system exploited the whitefly's habit of flying up from a plant that was mechanically tapped and then heading toward yellow objects. Remote-controlled dollies with arms that tapped plants and yellow-colored double-charged dipolar electric field screens (YDD-EFSs) (oppositely electrified transparent insulator tubes filled with yellow-colored water) attracted and trapped the whiteflies. The whiteflies flew up when the plants were mechanically tapped with the dolly's arms during reciprocating movements and were subsequently trapped by YDD-EFSs that were automatically translocated to the target plants. The system was applied to rows of whitefly-infested tomato plants. Almost all whiteflies transferred to plants were successfully recovered by two dollies moving on either side of the plants, approaching all plants individually (via programmed movement). In summary, we present an efficient unattended method for controlling whiteflies on tomato plants in greenhouses.

**Keywords:** attraction; electric field; greenhouse tomato; pest control; phototactic insect; yellow sticky trap

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# 1. Introduction

We observed that tomato plants in our greenhouses were frequently attacked by *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae), tomato leaf miner ((syn. vegetable leaf miner) *Liriomyza sativae* Blanchard (Diptera: Agromyzidae)), and a phytophagous ladybird beetle ((syn. Hadda beetle) *Epilachna vigintioctopunctata* (syn. *Henosepilachna vigintioctopunctata*) (Fabricius) (Coccinellidae: Coleoptera)). Two or even all three of these pets can occur together. Infestation by viruliferous whiteflies causes very severe disease; viruses of the TYLCV family are transmitted. Although several insecticides have been used for a long time, insecticide resistance has developed in whiteflies [1–4], tomato leaf miners [5,6], and ladybird beetles [7–9]. Given public health concerns, we aimed to develop insecticide-independent methods. Biological control is a non-chemical component of integrated pest management (IPM) during greenhouse tomato cultivation. Our effective insecticide-free pest biocontrol approaches include spraying entomopathogenic [10] or chitinolytic [11,12] phylloplane bacteria onto tomato leaves; this kills phytophagous ladybird beetles. However, we failed to isolate microorganisms that kill the whitefly and tomato leaf miner. We thus turned our attention to physical techniques. Electrostatic killing is

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largely unaffected by biological and environmental phenomena, and we tested its effectiveness in our greenhouses [13].

We devised systems generating an electric field (EF) in the space surrounding an electric charge, which imparts a force to another electric charge [14]. Our first EF-generating apparatus was a single-charged dipolar electric field screen (SD-EFS) with a layer comprising many iron insulated conductor wires (ICWs) arranged in parallel according to a defined inter-wire interval and a grounded metal net (G-MN) lying parallel to the ICW layer [15]. An EF formed in the space between the negatively charged ICWs (N-ICWs) and the positively polarized metal net. Nonomura et al. [16] found that whiteflies reaching the outside surface of the G-MN did not enter the SD-EFS. Matsuda et al. [17] reported that this avoidance behavior is seen in many insects (13 orders, 45 families, and 62 genera). Thus, the EFS repelled insects that reached the EFS. EFS screens were thus installed in the side windows of a greenhouse to exclude flying insects such as whiteflies, western flower thrips (Frankliniella occidentalis (Pergande) (Thysanoptera: Thripidae)), green peach aphids (Myzus persicae Sulzer (Hemiptera: Aphididae)), and shore flies (Scatella stagnalis (Fallén) (Diptera: Ephydridae)), which can all readily pass through conventional woven insectproof nets [18]. However, although the apparatus was highly effective, production and installation costs were too high for non-research use [19]. To reduce costs, Nonomura et al. [20] modified the EFS; an electrostatic soil cover trapped adult tomato leaf miners emerging from underground pupae. The insects were killed and free electrons were drained from their bodies [13].

Our second EF-generating apparatus was a double-charged dipolar EFS (DD-EFS) that captured insects entering the EF [21]. The apparatus had paired ICWs that were oppositely charged (linked to negative and positive voltage generators, respectively). The EF formed in the space between the oppositely charged ICWs. When an insect entered the EF, it was captured either due to a loss of free electrons, thus becoming positively electrified in the space near the N-ICW (negative pole) followed by attraction to the negative pole, or by receiving electrons that had accumulated around the positively charged ICW (P-ICW; positive pole), thus becoming negatively electrified, followed by attraction to the positive pole [22]. We prepared a bamboo-blind type DD-EFS that could be fabricated easily and cheaply by greenhouse workers [23].

The DD-EFS effectively captured insects that entered the EF [21,23]. However, the DD-EFS did not attract insects to the EF. Recently, Takikawa et al. [24] found that a colored DD-EFS lured phototactic insects such as whiteflies, tomato leaf miners, and western flower thrips. The conductor material (in a transparent insulating tube) was changed from an iron wire to yellow-colored water. The EF was generated by oppositely charging waterfilled paired tubes. In the present study, we fabricated a yellow-colored DD-EFS (YDD-EFS) to attract whiteflies that live on greenhouse tomato plants. Adult whiteflies fly up from a plant when it is gently tapped or shaken and then quickly return [24]. We exploited this habit to drive whiteflies out of the plant; they then became attracted to a YDD-EFS located nearby and died before they could return to the plant. Here, we present an unattended pest-control system. Tomato plants were mechanically tapped at defined times, at which the YDD-EFS was automatically moved near each tapped plant. To move the YDD-EFS, we added a non-grounded circuit to the DD-EFS [25]. In this circuit, free electrons from the conductor (yellow-colored water) were directly supplied to another conductor using the voltage produced by two generators in a single box. In such a circuit, the DD-EFS did not require a ground line and was thus mobile. The voltage applied to the DD-EFS was important [26]. A voltage generator enhanced the initial voltage (12 V) to the desired levels using a transformer and integrated Cockcroft circuit [27]. The tubes containing the yellow-colored water were thus appropriately electrified. As the strength of the force imparted by the DD-EFS depended on the applied voltage [26], we determined the optimum voltage for capturing all insects blown toward the screen. Based on the results, we considered automatic (unattended) elimination of whiteflies from tomato plants; i.e., an insecticide-independent method of pest control in a greenhouse.

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#### 2. Materials and Methods

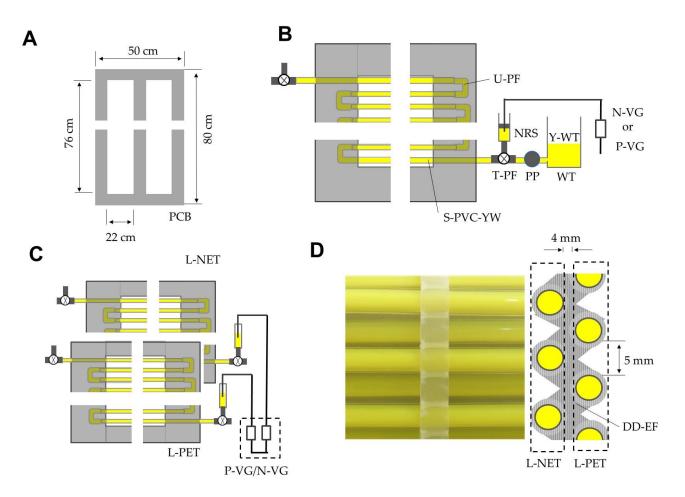
#### 2.1. Insect Management

Pupae of the whitefly ( $B.\ tabaci$  type G) were purchased from Sumika Technoservice (Hyogo, Japan) and placed under non-heading cabbage plants in a phytotron (25.0  $\pm$  0.5 °C) as described previously [28]. Emerged adults deposited their eggs on the leaves. After hatching and subsequent pupation, adult whiteflies that emerged from pupae were collected using an insect aspirator (Science Lab Supplies, Navasota, TA, USA).

#### 2.2. Electric Field Screens

The YDD-EFS was fabricated using a slightly modified version of a method described previously [24]. We used plastic (polypropylene) cardboard (thickness, 5 mm; Toyo Label, Kyoto, Japan) to construct a frame to hold transparent, soft polyvinyl chloride (PVC; an insulator) tubes (wall thickness, 2 mm; inner diameter, 3 mm; resistivity, 10° Ωcm; Junkosha Inc. Tokyo, Japan), arranged in parallel at intervals of 5 mm. Thus, two identical pieces  $(22 \times 76 \text{ cm}^2)$  were cut out of cardboard  $(50 \times 80 \text{ cm}^2)$ ; margins = 2 cm) using a laser cutter (Speedy 100; Trotec Laser GmbH, Marchtrenk, Austria) (Figure 1A), and 50 PVC tubes (length, 45 cm) were passed through the inner holes (hole diameter, 5 mm) of the cardboard (every other hole) and interlinked using U-shaped pipe fittings (Figure 1B). The open ends of the highest and lowest tubes were connected to T-shaped pipe fittings with a channel-switching cock. Yellow-colored water (in a tank) was sent to the tube via a needle-free injector syringe (inner dimeter, 1 cm; length, 5 cm) with a silicone stopper. The fluid traveled through a T-shaped pipe fitting linked to the lowest tube with a peristaltic pump. The silicone stopper was pierced with an iron wire (diameter, 0.5 mm; length, 3 cm) linked to a negative or positive voltage generator (Figure 1B). After the tube and syringe were filled with water, the pump and water tank were removed from the pipe fitting. The water was colored yellow using a watercolor paste (Turner Color Works Ltd., Osaka, Japan); the Munsell hue/value/chroma index [29] was 7Y8.5/11 (yellow), corresponding to the color of a commercial yellow sticky trap (Horiver, Tokyo, Japan). A colorant concentration of 750 mg/100 mL of water afforded the yellow index mentioned above. The color was measured using an RGB-1002 color analyzer (Sato Shoji Inc., Kanagawa, Japan). Two identical tube layers (50 × 80 cm²) were coupled in parallel at a 4 mm interval to create the DD-EFS (Figure 1C). The tubes of the two layers were offset such that the DD-EFS formed between all tubes (Figure 1D).

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**Figure 1.** (**A**) A plastic cardboard frame holding soft polyvinyl chloride (S-PVC) tubes. (**B**) S-PVC tubes linked to each other using U-shaped pipe fittings and filled with yellow-colored water negatively or positively charged by an iron wire. One end was linked to a negative or positive voltage generator and the opposite end was in contact with the water via a needle-free syringe. (**C**) Two layers of S-PVC tubes with opposite electrifications were coupled to construct the yellow-colored double-charged dipolar electric field screen (YDD-EFS). The negative and positive voltage generators were combined in a box that obviated the need for a ground line. (**D**) Photograph of the YDD-EFS (left) and schematic of the electric field in the space between the oppositely electrified tubes (right) (cross-sectional view). Abbreviations: PCB, polypropylene cardboard; U-PE, U-shaped pipe fitting; S-PVC-YW, soft PVC chloride tube filled with yellow-colored water; NRS, needle-free syringe; T-PF, T-shaped pipe fitting; PP, peristaltic pump; WT, water tank; Y-WT, yellow-colored water; L-PET, layer of positively electrified tubes; L-NET, layer of negatively electrified tubes; P-VG/N-VG, positive and negative voltage generators in one box, respectively; DD-EF, double-charged dipolar electric field.

As a second modification, a non-grounded circuit was used for charging (Figures 1C and S1B). The two tube layers were linked to negative and positive voltage generators, respectively. In this non-grounded circuit, free electrons from the conductor (water) of one tube layer were supplied directly to the conductor of the other tube layer using the voltages produced by the two voltage generators housed in a single box (P-VG/N-VG) (Figure 1C). In this circuit, the YDD-EFS did not require a ground line, thus the DD-EFS was mobile. We explored whether the YDD-EFS could attract and capture whiteflies.

We fabricated the SD-EFS as described previously [15]. The ICWs were arrayed in parallel in a polypropylene cardboard frame ( $90 \times 45 \text{ cm}^2$ ) and linked to a negative voltage generator (Figure S2A). Two G-MNs were placed on either side of the ICW layer (Figure S2B). The ICWs were negatively charged to 1.2 kV. The EF formed in the space between the ICW layer and G-MN (Figure S2B). The SD-EFS repelled insects that reached the G-

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MN [15,16] and thus served as the lateral windows of a cabinet. The SD-EFSs prevented entry by insects on the outside and egress by whiteflies inside. The SD-EFS-containing cabinet was placed inside a greenhouse (Figure S2C).

To trap whiteflies flying over tomato plants, we fabricated an electrostatic flying insect catcher (EFIC) as reported previously [30]. The EFIC was a racket-shaped DD-EFS (Figure S3A); two layers of oppositely charged ICWs were integrated into the racket ring and the box containing the two voltage generators and a storage battery was placed inside the hand grip (Figure S3B). The ICWs were charged (negatively and positively) to the same voltage (1 kV). Whiteflies colonizing tomato plants were forced to fly up by tapping the plants and were then caught by the EFIC (waved once or twice in the air above the plants) (Figure S3C).

# 2.3. Capturing and Attracting Whiteflies

The first experiment used an insect-capturing assay. Adult whiteflies were collected using an insect aspirator and blown toward the tubes of an YDD-EFS that was oppositely charged (with different voltages) to determine the lowest voltage that captured all insects. Twenty insects were used to test each voltage; the experiment was repeated five times.

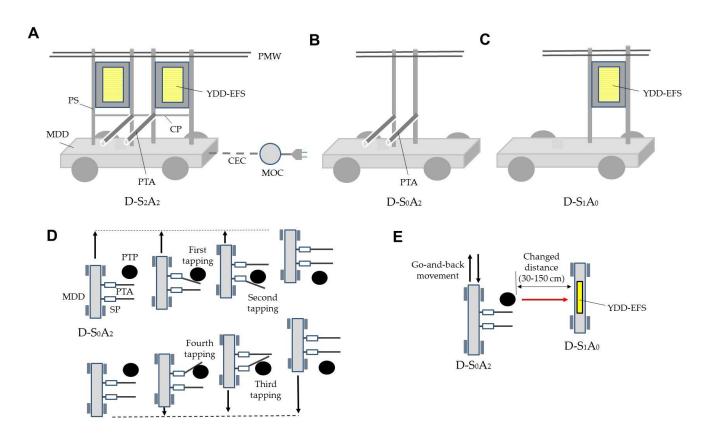
The second experiment used an insect-attracting assay. The YDD-EFS (a yellow sticky plate the same size as the screen) and potted tomato plant (a 1-month-old seedling) were placed along three lateral faces of an SD-EFS-installed cubic cabinet (floor area,  $2 \times 2$   $m^2$ ; height, 2 m), and a vessel containing 50 whiteflies was placed in the center to determine to which item the whiteflies were most attracted to. After 30 min, we counted the whiteflies captured by the YDD-EFS and yellow sticky plate, as well as those resting on the leaves of the tomato plant and various other places (i.e., in the vessel and on the floor, walls, and ceiling of the cabinet). The experiment was repeated five times.

# 2.4. Attraction of Whiteflies Driven out of Tomato Plants by Mechanical Tapping

# 2.4.1. Construction of a Moving YDD-EFS with and without Plant-Touching Arms

To move the YDD-EFS, we fabricated a motor dolly; i.e., a four-wheeled flat polypropylene box (floor area;  $120 \times 20 \text{ cm}^2$ ; height, 20 cm) in which a motor and gears were installed to rotate the wheels (at 0.4 km/h) and on which four supporters held two YDD-EFSs (Figure 2A). In addition, a crosspiece (90 cm in length) was mounted over the four supporters, and two arms that touched the tomato plants were installed vertical to the center of the crosspiece (Figure 2A). The arm was constructed by joining short and long urethane rods (1 cm in diameter) with a spring to impart elasticity. The motor was connected to a controller with a coiled electric extension cable; dolly operation (forward or backward movement and stopping) was manually remote-controlled. Apart from the dolly with two screens and two arms (D-S<sub>2</sub>A<sub>2</sub>), we fabricated two other dollies: one with no screen and two arms (D-S<sub>0</sub>A<sub>2</sub>) (Figure 2B) and one with a screen but no arms (D-S<sub>1</sub>A<sub>0</sub>) (Figure 2C).

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**Figure 2.** (**A** – **C**) Three types of motor-driven dollies that moved two yellow-colored double-charged dipolar electric field screens (YDD-EFSs): a dolly with two arms for plant touching (D-S<sub>2</sub>A<sub>2</sub>) (**A**), a dolly with no YDD-EFS and two arms (D-S<sub>0</sub>A<sub>2</sub>) (**B**), and a dolly with one YDD-EFS and no arm (D-S<sub>1</sub>A<sub>0</sub>) (**C**). The dolly motor was manually remote-controlled. (**D**) Step-by-step forward (upper) and backward (lower) D-S<sub>0</sub>A<sub>2</sub> motion with tapping of a potted tomato plant by the two arms. The plant was tapped four times with the two elastic arms via reciprocating movements of the D-S<sub>0</sub>A<sub>2</sub> (bird's-eye view). (**E**) Experimental assay of flying whitefly attraction by the YDD-EFS on the D-S<sub>1</sub>A<sub>0</sub> dolly placed at various distances from a tomato plant to which whiteflies had been transferred. Black and red arrows represent the direction of dolly movement and path of the whiteflies attracted to the YDD-EFS, respectively. Abbreviations: MDD, motor-driven dolly; PS, polypropylene supporter; PTA, plant-touching arm; YDD-EFS, yellow-colored double-charged dipolar electric field screen; CP, crosspiece; CEC, coiled extension electric cable; MOC, motor-operation controller; PMW, paired metal wires; PTP, potted tomato plant; SP, spring-jointed short and long arms.

# 2.4.2. Tracking of Whiteflies Driven out of Tomato Plants by Mechanical Tapping

In the present study, a potted seedling (1 month-old, 90–100 cm high (from pot bottom to plant top)) of tomato (*Solanum lycopersicum* L. cv. Moneymaker) propped up with a pole was used. Whiteflies were placed on the plant and tracked when driven out by mechanical tapping. Twenty adult whiteflies were transferred to random abaxial leaf surfaces using an insect aspirator. After 30 min, all insects had settled on the leaves; the D-S<sub>0</sub>A<sub>2</sub> tapped a plant twice (with the two arms) as the dolly moved forward and backward (Figure 2D). The dolly moved 1 m in either direction at 0.4 km/h with a 1 min pause between movements. The spring-jointed arm was sufficiently elastic to not damage the plant, which was simply shaken as the dolly moved. The fixed course of the dolly was guided by two metal wires strung above it (Figure 2B). After tapping, the whiteflies flew up and then rapidly returned to the plant (within 10–40 s); the whiteflies exhibited this behavior on every occasion. We tapped the plant 2–10 times (via 0.5–2.5 reciprocating dolly movements). The whiteflies that flew up after the final tapping were caught by the

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EFIC. We confirmed that no whiteflies remained on the plant and counted those that returned to the plant after each tapping.

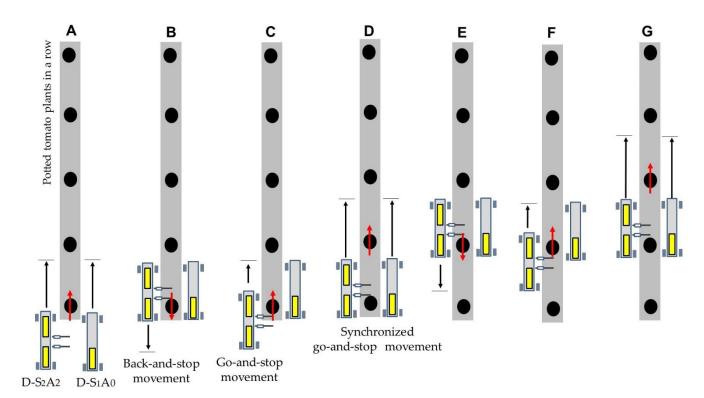
# 2.4.3. Attraction of Flying Whiteflies to the YDD-EFS

The D-S<sub>0</sub>A<sub>2</sub> was placed 50 cm from a potted plant on which 20 whiteflies had settled and the D-S<sub>1</sub>A<sub>0</sub> was placed on the opposite side of the plant at a distance of 30, 90, or 150 cm (Figure 2E). The plant was repeatedly tapped by the two arms of the D-S<sub>0</sub>A<sub>2</sub> (via several reciprocating movements under the conditions mentioned above). At each distance, the whiteflies captured by the YDD-EFS were counted to determine the most effective distance between the plant and YDD-EFS. The experiment was repeated five times at each distance.

# 2.4.4. Automatic Trapping of Whiteflies Driven out of Tomato Plants

Five potted tomato plants (1-month-old seedlings) were arrayed in a row in an SD-EFS-installed cabinet (floor area, 2 × 6 m<sup>2</sup>; height 2 m). Twenty whiteflies settled on each plant. Two dollies (D-S<sub>2</sub>A<sub>2</sub> and S<sub>1</sub>A<sub>0</sub>) were placed on either side of the plants (Figure 3). Both dollies were manually remote-controlled with a single controller. First, the two dollies were synchronously moved to the first plant (Figure 3A); the D-S<sub>2</sub>A<sub>2</sub> was then moved backward (Figure 3B) and then to the same position as the D-S<sub>1</sub>A<sub>0</sub> (Figure 3C). Next, the two dollies were synchronously moved to the second plant (Figure 3D). In the same manner, the two dollies were moved to the third plant (Figure 3E-G). During the dolly movements, each plant was tapped six times by the two arms of the D-S<sub>2</sub>A<sub>2</sub> (four times when the D-S<sub>2</sub>A<sub>2</sub> moved back and forward along the plant and two more times when the dolly moved to the next plant). A 1 min pause was scheduled after each forward or backward movement. The whiteflies that were driven out were attracted to and captured by the YDD-EFSs that faced the plant. In this manner, all plants were tapped in turn (the first insect-trapping approach). After the final tapping of the last plant, we counted the whiteflies captured by the three YDD-EFSs and carefully checked whether any whiteflies remained on the plants. If there were none remaining, we determined the recovery rates of the whiteflies used and the numbers that flied to other places (if necessary). If some whiteflies remained on the plants, we repeated the entire tapping sequence, but in reverse (the second insect-trapping approach). The analyses were similar to those performed after the first approach. The experiment was repeated 10 times and the whitefly recovery rates were determined.

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**Figure 3.** Step-by-step synchronous movements of two dollies (D-S<sub>2</sub>A<sub>2</sub> and D-S<sub>1</sub>A<sub>0</sub>) from plant to plant. Five potted tomato plants were arrayed in a row and 20 whiteflies were transferred to the leaves of each plant. The D-S<sub>2</sub>A<sub>2</sub> was placed on one side of the plant and the D-S<sub>1</sub>A<sub>0</sub> on the other. Both dollies were manually remote-controlled. The D-S<sub>2</sub>A<sub>2</sub> moved forward and backward along one plant and then moved to the next plant; the D-S<sub>1</sub>A<sub>0</sub> moved forward in step with the forward movement of the D-S<sub>2</sub>A<sub>2</sub>. Each plant was tapped six times by the two arms of the D-S<sub>2</sub>A<sub>2</sub> (via the go-backgo movement). All plants were treated in the same manner. Black and red arrows represent the direction of dolly movement and plant tapping by the arms, respectively. The whiteflies captured by three YDD-EFSs were counted after the final tapping of the fifth plant. The second approach was similar but proceeded in the opposite direction; it trapped whiteflies that remained on the plants.

### 2.5. Statistical Analysis

All experiments were repeated five times; all data are presented as means with standard deviations. We tested for significant differences among the various conditions with a Tukey's test performed using EZR software (ver. 1.54; Jichi Medical University, Saitama, Japan).

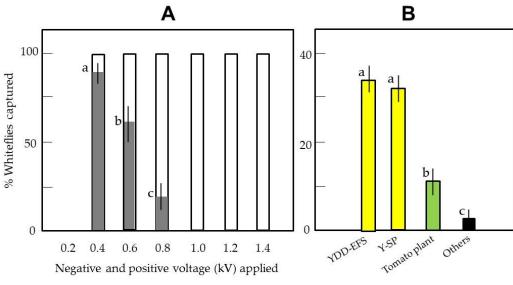
# 3. Results and Discussion

#### 3.1. Ability of the YDD-EFS to Attract and Capture Whiteflies

The photoselective behavior of insects has encouraged the use of colored sticky traps [31]. Yellow sticky traps are highly effective for attracting and trapping whiteflies in greenhouses and fields [32–34]. We colored the DD-EFS yellow and lured whiteflies to the EF of the apparatus. In the first experiment, we optimized the voltage applied to the YDD-EFS and aimed to capture all insects blown toward the charged tubes of the YDD-EFS. In the YDD-EFS, the EF formed between tubes that were oppositely charged (Figure 1D). The forces drew a whitefly to the nearest tube when the insect entered the EF [22]. A higher voltage imparted a greater insect-capture force in the EF [26]. Figure 4A shows the relationship between the insect-capture rate and applied voltage. When the voltage was too low (0.4–0.8 kV), the force generated did not confine the whiteflies to the tubes after they had been drawn to the tubes; the insects twisted strenuously and escaped the trap (Video S1A). However, the escape rate decreased as the voltage increased. Negative and positive

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charging of 1 kV confined all whiteflies to the tubes (Video S1B) (Figure 4A). Thus, we applied these charges in the subsequent experiments.



Destination of whiteflies released

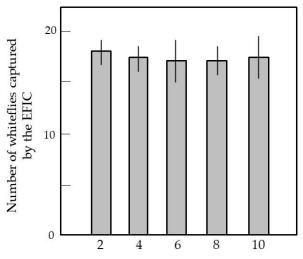
**Figure 4.** (**A**) The numbers of adult whiteflies captured by the yellow-colored double-charged dipolar electric field screen (YDD-EFS) oppositely charged with different voltages. At each voltage, the paired tubes of the YDD-EFS were oppositely charged with the same negative and positive voltages. Open and gray columns denote whiteflies captured by the charged tubes and those that escaped after capture, respectively. Twenty adult whiteflies were blown toward the apparatus at each voltage. (**B**) The destinations of whiteflies released at the central position among the YDD-EFS, a yellow sticky plate (Y-SP), and a potted tomato plant. Other whiteflies were in the vial or on the floor, walls, or ceiling of the SD-EFS-installed cabinet. A total of 50 whiteflies (in a vessel) were released, and 30 min later, the whiteflies in various places were counted. In both experiments, the means and standard deviations were calculated using five experimental replicates. The letters (a–c) in each vertical column indicate significant differences (p < 0.05) as revealed by a Tukey's test.

In the second experiment, we examined the photoselective behavior of whiteflies; the targets were the YDD-EFS, a yellow sticky plate, and a tomato plant placed around the whiteflies (Figure 4B). The YDD-EFS was highly attractive, comparable to the commercial yellow sticky plate. More importantly, the whiteflies selected the yellow traps more often than the tomato plant. Small numbers of whiteflies remained in the vessel or escaped to the cabinet floor, walls, and ceiling. After 1 day, however, these whiteflies were also found on the YDD-EFS or sticky plate. Thus, the YDD-EFS-trapped whiteflies were driven out of the tomato plants.

### 3.2. Flush-and-Return Behavior of Plant-Residing Whiteflies after Plant Tapping

During our routine care of greenhouse tomato plants, we frequently noted that adult whiteflies living on the plants flushed when the plants were gently shaken; most whiteflies returned to their former plants after flying over the plants for 10–40 s. We sought to exploit this habit to kill the pests. First, we derived a method of constant plant shaking. Our motor dolly (D-S<sub>0</sub>A<sub>2</sub>) was moved backward and forward (0.5–2.5 times) and tapped the plants with the arms. Whiteflies that flew up were collected by the EFIC to confirm that they had all returned to the same plants after every tapping. Figure 5 shows that there was no significant difference in the number of whiteflies captured according to the number of tappings. Over 90% of whiteflies exhibited flush-and-return behavior in response to tapping regardless of the number of tappings.

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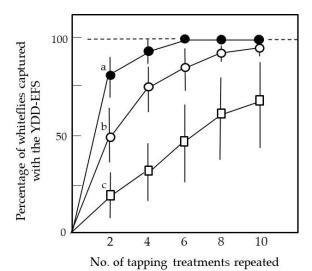
No. of tapping conducted before capturing

**Figure 5.** Capture of whiteflies that flew up after final tapping of a tomato plant. Twenty adult whiteflies were transferred to a potted plant and forced to fly up by tapping, which was repeated every time whiteflies returned to the plant. The whiteflies exhibited repeated flush-and-return behaviors in response to successive tappings. After the final tapping, the whiteflies that flew up were caught by the electrostatic flying insect catcher (EFIC). The means and standard deviations were calculated from five experimental replicates. There was no significant difference in the number of whiteflies captured according to the number of tappings prior to capture.

#### 3.3. Distance between the YDD-EFS and an Infested Tomato Plant

We explored whether the YDD-EFS attracted whiteflies driven out of tomato plants by tapping. We presumed that distance would be important for whiteflies to recognize a yellow-colored object. We compared the number of whiteflies captured by an YDD-EFS among the different distances from the plant (Figure 6). The whiteflies that flew up were the most attracted to the nearest YDD-EFS (30 cm); over 80% of whiteflies were attracted after the first two taps. A very small number flew up on the opposite side and returned to the plant. However, these whiteflies were attracted to the YDD-EFS when they flew up at the next tapping. The capture rate was 100% after the sixth tapping. As the distance became greater, the number of whiteflies that returned to the plant increased. At 90 cm, it was necessary to tap the plant >10 times to capture all whiteflies. At 150 cm, the efficiency of attraction was very low; >80% of the whiteflies that flew up returned to the plant, and approximately 30% were still on the plant even after 10 tappings (Figure 6). We thus optimized the experimental set-up (Figure 3). We placed YDD-EFSs on both sides of the plant (i.e., on the D-S<sub>2</sub>A<sub>2</sub> and D-S<sub>1</sub>A<sub>0</sub> dollies) at a distance of 30 cm from the plant, followed by go-back-stop-and-go movement of the D-S2A2 and go-stop-and-go movement of the D-S<sub>1</sub>A<sub>0</sub>. Below, we describe the application of this system to an unattended method in which whiteflies were largely eliminated from tomato plants.

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**Figure 6.** The numbers of whiteflies captured by the yellow-colored double-charged dipolar electric field screen (YDD-EFS) placed at different distances (30 (closed circle), 90 (open circle), and 150 (open square) cm) from a potted tomato plant. Adult whiteflies settled on the leaves and the plant was repeatedly tapped by the arms of the dolly (D-S<sub>0</sub>A<sub>2</sub>) via reciprocating movements. The whiteflies that flew up were attracted to the YDD-EFS on the second dolly (D-S<sub>1</sub>A<sub>0</sub>) placed on the opposite side of the plant. The whiteflies captured by the YDD-EFS were counted. Twenty whiteflies were used to test each distance, and the means and standard deviations were calculated from five experimental replicates. Letters (a–c) in each vertical column indicate significant differences (p < 0.05) as revealed by a Tukey's test.

### 3.4. Automatic Control of Whiteflies on Tomato Plants

The most important finding of this study was that an YDD-EFS close to a tomato plant was optimal for attracting whiteflies driven from the plant (to the non-sticky surfaces of electrified YDD-EFS tubes). Each tube had a negative or positive charge imparted by dielectric polarization [35] of negatively or positively charged water inside the tube. Matsuda et al. [22] found that an insect invading space near a negatively charged insulated conductor (a negative pole) was deprived of free electrons (and was thus positively electrified) and attracted to the negative pole, while an insect that entered space near a positively charged pole received electrons that had accumulated around the positive pole (and was thus electrified negatively) and was attracted to the positive pole. Thus, the tubes generated forces that attracted insects but the tube surfaces were not sticky, unlike the yellow trap plate. Usually, yellow sticky traps are placed away from cultivated plants to ensure that leaves do not stick to the traps; the traps are often hung from lateral pillars near greenhouse windows and crossbeams. More importantly, even if the YDD-EFS touched a tomato leaf, the force generated by the YDD-EFS was too low to draw the leaf. We were thus able to trap whiteflies within the vicinity of plants.

The aim of the final experiment was to develop an unattended pest control system; tomato plants were mechanically tapped to drive out whiteflies that were then attracted to YDD-EFSs automatically translocated near the target plant. The system was successfully applied to a row of five potted tomato plants (Figure 3); i.e., a scaled-down version of plants in a greenhouse row. Table 1 lists the numbers of whiteflies attracted to and captured by the YDD-EFSs, those that remained on the plants and fled to other places during the first round of insect-trapping, and those captured during the second round of trapping. The system effectively eliminated whiteflies; over 95% were recovered after the first round of trapping (Table 1). More importantly, all whiteflies that evaded the first trapping and remained on the plants were trapped during the second round (Table 1). A small proportion (<2%) of whiteflies flied to other places (Table 1). Although we did not pursue these whiteflies, they remained inside the cabinet because the open windows had SD-EFSs that repelled nearby whiteflies [16]. These whiteflies could have been the source

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of a second infestation; indeed, they had moved back to the test plants by the following day. Fortunately, these whiteflies were successfully trapped by a third round of trapping (data not shown). Thus, periodic application of the system reliably controlled whiteflies on plants.

**Table 1.** Numbers of whiteflies captured by yellow-colored double-charged dipolar electric field screens on motor dollies during the first and second rounds of automated insect-trapping from a row of five tomato plants.

Experiments	First Round			Second Round		
	A	В	С	D	E	<b>A</b> + <b>E</b>
1	94	2	4	2	2	96
2	92	8	0	8	8	100
3	100	0	0	n.c.	n.c.	n.c.
4	100	0	0	n.c.	n.c.	n.c.
5	90	2	8	2	2	92
6	93	6	1	6	6	99
7	100	0	0	n.c.	n.c.	n.c.
8	88	9	3	9	9	97
9	96	4	0	4	4	100
10	100	0	0	n.c.	n.c.	n.c.
Average % and S.D.	$95.3 \pm 1.3$	$3.1 \pm 0.3$	$1.6 \pm 0.1$	-	100	97.3 ± 1.3

A, number of whiteflies captured; B, number of whiteflies that remained on the plants; C, number of whiteflies that flied elsewhere; D, whiteflies targeted in the second round of capture; E, captured whiteflies; n.c., not conducted.

As the yellow sticky trap is highly attractive to insects, almost all tomato growers in our district (Nara Prefecture, Japan) who cultivate plants organically in large greenhouses use them to reduce whitefly populations. Many traps are hung from lateral pillars near greenhouse windows and crossbeams. However, the stickiness of the surface is reduced as increasing numbers of insects are trapped, so farmers must frequently replace their traps during peak pest seasons. The farmers in our district have requested a less expensive, reusable trap with a non-sticky surface that can attract and capture insects. The YDD-EFS is a promising candidate, as it is both washable and easily repaired; moreover, the structure is simple and the materials are inexpensive. More importantly, because the apparatus moves along the plants, the hardware requirements are low.

The most important advantage of our system is that the labor required to control plant pests is reduced; this is very important at larger scales. If many tomato plants are grown in large greenhouses, our system could be completely automated by: (1) making the dolly wireless by adding a storage battery for motor operation; (2) making the dolly radio-controllable by installing a receiver; and (3) using a radio transmitter to program all dolly movements. Thus, we have provided an experimental basis for unattended control of whiteflies during greenhouse tomato cultivation.

### 4. Conclusions

We devised an unattended pest control system that eliminates whiteflies from green-house tomato plants and thus reduces labor requirements. The components included plant-tapping arms, a yellow-colored insect-attracting and -capturing device, and a motor-driven dolly, all of which can be simply and inexpensively fabricated by tomato growers. The pest-control principle was based on the habitual behavior of the whitefly and the ability of the EF-forming apparatus to capture insects that entered the EF. The system exhibited highly efficient and stable pest-control performance; this study thus provided an experimental basis for automatic eradication of whiteflies that infest tomato plants in greenhouses.

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Supplementary Materials: The following are available online at www.mdpi.com/article/10.3390/horticulturae8090764/s1, Figure S1: Schematic representations of the grounded (A) and non-grounded (B) circuits integrated into the double-charged dipolar electric field screen (DD-EFS), Figure S2: (A) Schematic representation of a single-charged dipolar electric field screen (SD-EFS), which consisted of a layer of insulated iron wires (ICWs) and grounded metal nets (G-MNs) on each side of the layer. (B) Single-charged dipolar electric field (SD-EF) formed in the space between the negatively charged ICWs (N-ICWs) and G-MNs (cross-sectional view). (C) A cabinet with lateral windows furnished with the SD-EFSs, Figure S3: (A) Diagram of the racket-shaped electrostatic flying insect catcher (EFIC). (B) Two layers of insulated conductor wires (ICWs) oppositely charged with two voltage generators. (C) The EFIC was portable and easy to operate on site in a greenhouse, Video S1: Capture of adult whiteflies with oppositely electrified insulator tubes of the yellow-colored double-charged dipolar electric field screen (YDD-EFS).

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## References

 Prabhaker, N.; Coudriet, D.L.; Meyerdirk. D.E. Insecticide resistance in the sweetpotato whitefly, Bemisia tabaci (Homoptera: Aleyrodiae). J. Econ. Entomol. 1985, 78, 748–752.

- Palumbo, J.C.; Horowitz, A.R.; Prabhaker, N. Insecticidal control and resistance management for *Bemisia tabaci. Crop Prot.* 2001, 20, 739–765.
- 3. Horowitz, A.R.; Kontsedalov, S.; Ishaaya, I. Dynamics of resistance to the neonicotinoids acetamiprid and thiamethoxam in *Bemisia tabacci* (Homoptera: Aleyrodidae). *J. Econ. Entomol.* **2004**, *97*, 2051–2056.
- 4. Nauen, R.; Denholm, I. Resistance of insect pests to neonicotinoid insecticides: Current status and future prospects. *Arc. Insect Biochem. Physiol.* **2005**, *58*, 200–215.
- 5. Ferguson, J.S. Development and stability of insecticide resistance in the leafminer *Liriomyza trifoli*i (Diptera: Agromyzidae) to Cyromazine, Abamectin, and Spinosad. *J. Econ. Entomol.* **2004**, 97, 112–119.
- 6. Wei, Q.-B.; Lei, Z.-R.; Nauen, R.; Cai, D.-C.; Gao, Y.-L. Abamectin resistance in strains of vegetable leafminer, *Liriomyza sativae* (Diptera: Agromyzidae) is linked to elevated glutathione S-transferase activity. *Insect Sci.* **2013**, *13*, 243–250.
- 7. Surjeet, K.; Jitender, K. Comparative biology of resistant and susceptible strains of *Epilachna vigintioctopunctata* (Fabricius) to malathion and endosulfan. *J. Entomol. Res.* **1997**, 21, 303–306.
- 8. Sheikh, K.; Desh, R. Efficacy of insecticides and biopesticides against hadda beetle, *Henosepilachna vigintioctopunctata* (fabricius) (coleoptera: Coccinellidae) on bitter gourd. *Ind. J. Entomol.* **2013**, *75*, 163–166.
- 9. Hanif, M.U.; Raza, A.B.M.; Majeed, M.Z.; Arshad, M.; Ullah, M.I. Laboratory evaluation of selected differential chemistry and botanical insecticides against hadda beetle *Epilachna vigintioctopunctata* Fabricius (Coleoptera: Coccinellidae). *Punjab Univ. J. Zool.* **2021**, *36*, 185–191.
- 10. Otsu, Y.; Matsuda, Y.; Mori, H.; Ueki, H.; Nakajima, T.; Fujiwara, K.; Matsumoto, M.; Azuma, N.; Kakutani, K.; Nonomura, T.; et al. Stable phylloplane colonization by entomopathogenic bacterium *Pseudomonas fluorescens* KPM-018P and biological control of phytophagous ladybird beetles *Epilachna vigintioctopunctata* (Coleoptera: Coccinellidae). *Biocontrol Sci. Technol.* **2004**, *14*, 427–439.
- 11. Otsu, Y.; Matsuda, Y.; Shimizu, H.; Ueki, H.; Mori, H.; Fujiwara, K.; Nakajima, T.; Miwa, A.; Nonomura, T.; Sakuratani, Y.; et al. Biological control of phytophagous ladybird beetles *Epilachna vigintioctopunctata* (Col., Coccinellidae) by chitinolytic phylloplane bacteria *Alcaligenes paradoxus* entrapped in alginate beads. *J. Appl. Entomol.* **2003**, 127, 441–446.
- 12. Otsu, Y.; Mori, H.; Komuta, K.; Shimizu, H.; Nogawa, S.; Matsuda, Y.; Nonomura, T.; Sakuratani, Y.; Tosa, Y.; Mayama, S.; et al. Suppression of leaf feeding and oviposition of phytophagous ladybird beetles *Epilachna vigintioctopunctata* (Coleoptera: Coccinellidae) by chitinase gene-transformed phylloplane bacteria and their specific bacteriophages entrapped in alginate gel beads. *J. Econ. Entomol.* **2003**, *96*, 555–563.
- 13. Matsuda, Y.; Nonomura, N.; Toyoda, H. Physical methods for electrical trap-and-kill fly traps using electrified insulated conductors. *Insects* **2022**, *13*, 253.

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14. Jones, E.; Childers, R. Electric charge and electric field. In *Physics*, 3rd ed.; McGraw-Hill: Boston, MA, USA, 2002; pp. 495–525.

- 15. Matsuda, Y.; Nonomura, T.; Kakutani, K.; Takikawa, Y.; Kimbara, J.; Kasaishi, Y.; Kusakari, S.; Toyoda, H. A newly devised electric field screen for avoidance and capture of cigarette beetles and vinegar flies. *Crop Prot.* **2011**, *30*, 155–162.
- 16. Nonomura, T.; Matsuda, Y.; Kakutani, K.; Kimbara, J.; Osamura, K.; Kusakari, S.; Toyoda, H. An electric field strongly deters whiteflies from entering window-open greenhouses in an electrostatic insect exclusion strategy. *Eur. J. Plant Pathol.* **2012**, *134*, 661–670.
- 17. Matsuda, Y.; Nonomura, T.; Kakutani, K.; Kimbara, J.; Osamura, K.; Kusakari, S.; Toyoda, H. Avoidance of an electric field by insects: Fundamental biological phenomenon for an electrostatic pest-exclusion strategy. *J. Phys. Conf. Ser.* **2015**, *646*, 0120031–0120034.
- 18. Kakutani, K.; Matsuda, Y.; Nonomura, T.; Kimbara, J.; Kusakari, S.; Toyoda, H. Practical application of an electric field screen to an exclusion of flying insect pests and airborne conidia from greenhouses with a good air penetration. *J. Agric. Sci.* **2012**, *4*, 51–60
- 19. Toyoda, H.; Kusakari, S.; Matsuda, Y.; Kakutani, K.; Xu, L.; Nonomura, T.; Takikawa, Y. Practical implementation of single-charged dipolar electric field screen. In *An Illustrated Manual of Electric Field Screens: Their Structures and Functions*; Toyoda, H., Ed.; RAEFSS Publishing Department: Nara, Japan, 2019; pp. 41–49.
- 20. Nonomura, T.; Toyoda, H. Soil surface-trapping of tomato leaf-miner flies emerging from underground pupae with a simple electrostatic cover of seedbeds in a greenhouse. *Insects* **2020**, *11*, 878.
- 21. Nonomura, T.; Matsuda, Y.; Kakutani, K.; Takikawa, Y.; Kimbara, J.; Osamura, K.; Kusakari, S.; Toyoda, H. Prevention of white-fly entry from a greenhouse entrance by furnishing an airflow-oriented pre-entrance room guarded with electric field screens. *J. Agric. Sci.* **2014**, *6*, 172–184.
- Matsuda, Y.; Kakutani, K.; Nonomura, T.; Kimbara, J.; Kusakari, S.; Osamura, K.; Toyoda, H. An oppositely charged insect exclusion screen with gap-free multiple electric fields. J. Appl. Phys. 2012, 112, 116103.
- 23. Takikawa, Y.; Matsuda, Y.; Nonomura, T.; Kakutani, K.; Okada, K.; Shibao, M.; Kusakari, S.; Miyama, K.; Toyoda, H. Exclusion of whiteflies from a plastic hoop greenhouse by a bamboo blind-type electric field screen. *J. Agric. Sci.* **2020**, *12*, 50–60.
- 24. Takikawa, Y.; Nonomura, T.; Sonoda, T.; Matsuda, Y. Developing a phototactic electrostatic insect trap targeting whiteflies, leafminers, and thrips in greenhouses. *Insects* **2021**, *12*, 960.
- 25. Kakutani, K.; Matsuda, Y.; Nonomura, T.; Takikawa, Y.; Osamura, K.; Toyoda, H. Remote-controlled monitoring of flying pests with an electrostatic insect capturing apparatus carried by an unmanned aerial vehicle. *Agriculture* **2021**, *11*, 176.
- 26. Toyoda, H.; Matsuda, Y. Basic concepts for constructing an electric field screen. In *Electric Field Screen: Principles and Applications*; Toyoda, H., Ed.; Nobunkyo Production: Tokyo, Japan, 2015; pp. 3–17.
- 27. Wegner, H.E. Electrical charging generators. In *McGraw-Hill Encyclopedia of Science and Technology*, 9th ed.; Geller, E., Moore, K., Well, J., Blumet, D., Felsenfeld, S., Martin, T., Rappaport, A., Wagner, C., Lai, B., Taylor, R., Eds.; The Lakeside Press: New York, NY, USA, 2002; pp. 42–43.
- 28. Xie, W.; Wu, Q.; Wang, S.; Jiaol, X.; Guo, L.; Zhou, X.; Zhang, Y. Transcriptome analysis of host-associated differentiation in *Bemisia tabaci* (Hemiptera: Aleyrodidae). *Front. Physiol.* **2014**, *5*, 487.
- 29. Munsell Color Company. Munsell Hue Circle Poster. Available online: https://munsell.com/color-blog/munsell-hue-circle-poster/ (accessed on 21 August 2022).
- 30. Takikawa, Y.; Matsuda, Y.; Nonomura, T.; Kakutani, K.; Okada, K.; Shibao, M.; Kusakari, S.; Toyoda, H. Elimination of white-flies colonizing greenhouse tomato plants using an electrostatic flying insect catcher. *Int. J. Curr. Adv. Res.* **2017**, *6*, 5517–5521.
- 31. Shimoda, M. Recent advances in the optical control of insect pests using light and color. In Proceedings of the 2018 International Symposium on Proactive Technologies for Enhancement of Integrated Pest Management of Key Crops, Taichung, Taiwan, 3–5 April 2018; pp. 87–98.
- 32. Hoelmer, K.M.; Simmons, A.M. Yellow sticky trap catches of parasitoids of *Bemisia tabaci* (Hemiptera: Aleyrodidae) in vegetable crops and their relationship to in-field populations. *Environ. Entomol.* **2008**, *37*, 391–399.
- 33. Moreau, T.L.; Isman, M.B. Trapping whiteflies? A comparison of greenhouse whitefly (*Trialeurodes vaporariorum*) responses to trap crops and yellow sticky traps. *Pest Manag. Sci.* **2011**, *67*, 408–413.
- 34. Lu, Y.; Bei, Y.; Zhang, J. Are yellow sticky traps an effective method for control of sweet potato whitefly, *Bemisia tabaci*, in the greenhouse or field? *J. Insect Sci.* **2012**, *12*, 113.
- 35. Halliday, D.; Resnick, R.; Walker, J. Electric discharge and electric fields. In *Fundamentals of Physics*; Johnson, S., Ford, E., Eds.; John Wiley & Sons: New York, NY, USA, 2005; pp. 561–604.