

# Supporting Information

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## Methods for Culture of Organisms

**CMV Inoculation Procedures.** CMV strain FNY was obtained from John Murphy at the University of Auburn and maintained in *C. pepo* cv. Dixie. Plants used for infected treatments were inoculated mechanically to isolate the effects of the virus and because mechanical inoculations are logistically simpler to perform in an insect-free environment. Examining the effects of infection in the absence of aphid feeding was desirable, because our field data (Fig. 2) indicate that infected plants host aphids less frequently than healthy plants. To generate the inoculum, one or two leaves of young, highly symptomatic tissue from a CMV-infected *C. pepo* plant were ground in 15 mL of 0.1 M potassium phosphate buffer, pH 7.0. To inoculate, the cotyledons of 2- to 3-day-old *C. pepo* plants were dusted with carborundum powder, and the inoculum was spread over the cotyledons using a cotton-tipped applicator. Mock-inoculated plants were generated at the same time by applying clean buffer to cotyledons.

**Plants.** *Cucurbita pepo* cv. Dixie seeds (Willhite Seed Co.) were planted in 140 cm<sup>3</sup> of autoclaved Pro-Mix potting soil with 5 g of Osmocote slow-release fertilizer (NPK:14-14-14). Plants were grown in a climate-controlled, insect-free growth chamber under fluorescent and incandescent lights at a 16:8-h (light:dark) photoperiod at 24 °C. CMV experiences seasonal amplification via wide-scale infection of cultivated cucurbit crops following movement from perennial reservoir hosts on field edges (1). Thus, CMV frequently experiences transmission, and presumably selection, within cultivated cucurbit hosts.

**Insects.** *Aphis gossypii* were collected from field-grown *C. pepo* (in Pennsylvania) and maintained on *C. pepo* cv. Dixie. *Myzus persicae* were obtained from the Pennsylvania State University Plant Pathology Department and maintained on turnips (*Brassica rapa* cv. Purple Top White Gold). Aphids were reared at 23 °C on a 14:10-h (light:dark) photoperiod. Wingless individuals in behavioral experiments were third or fourth instar. The ages of wingless individuals in growth experiments were standardized within 1 day by placing fourth-instar aphids on 2-week-old *C. pepo* plants for 36 h, then removing these individuals and rearing the offspring produced.

## Detailed Methods for Greenhouse and Field Volatile Collections

Volatile collections in the greenhouse employed a closed push/pull system with a filtered air input. We placed 2.5-week-old plants (inoculated 2 weeks previously) in the center of a Teflon base, which closed around the stem. Closed glass chambers (9-L capacity) containing ports for air input and output were then fitted over the plants. Filtered air was pushed into the chamber at a rate of 5.0 L

per min through a Teflon tube fitted to a port at the top of the chamber and was pulled through Super-Q (Alltech) traps at a rate of 1.0 L per min. The higher airflow into the chambers with excess air vented at the bottom of the system prevented contamination by outside air (via positive pressure), avoided artificial elevation of volatile concentrations (which can influence plant chemistry), and minimized plant stress from overheating or high humidity. Volatiles were sampled during three 4-h intervals over a 14-h daylight period. Following collections, plants were dried at 50 °C for 7 days and then weighed. In the field, individual leaves of 6-week-old plants (inoculated 5.5 weeks previously) were enclosed in small glass/Teflon chambers and sampled using a portable push/pull system using Super-Q traps at a rate of 1.0 L of air per min for 30 min. Following collection, leaves were traced on paper while still attached to plants to obtain a nondestructive estimate of their area. The tracings were scanned, and their area was calculated using SigmaScan Pro-5 (Image Analysis).

Compounds were eluted from the filters using 150 µL of dichloromethane with 5 µL of an internal standard added after eluting (80 ng/µL nonyl acetate, 40 ng/µL n-octane). Samples were injected in 1-µL aliquots into an Agilent model 6890 gas chromatograph fitted with a flame ionization detector (column: Agilent 19091Z-331, 0.25 mm internal diameter, 0.1-µm film thickness). The column was held at 35 °C for 0.5 min then increased by 4 °C per min to 160 °C and further increased by 20 °C per min to a maximum temperature of 220 °C. Total volatiles produced per gram of dry weight were quantified using MSD Chemstation (Agilent Technologies 2003) by measuring volatile output in nanograms relative to the internal standard and dividing this value by the dry weight of the particular sample (in grams). It was not necessary to calculate response factors for our quantification because we were interested only in relative amounts of compounds and because all behavioral assays were conducted using live plants. Tentative identifications were made by calculation of Kovats indices for unidentified compounds and comparison of these values with a previously compiled list of known compounds and indices (run on the same type of column) and by retention time comparisons with known standards as well as comparison of mass spectra with a library of known compounds (Agilent Technologies 2003). Quantitative comparisons among individual volatiles were made by comparing means and assessing overlap of standard errors (2). For field-collected volatile samples, amounts were divided by leaf area in square centimeters. Total size-corrected volatiles for all treatments were log transformed and analyzed using a General Linear Model with collection date as a random blocking factor (Minitab v.14). Field-collected volatiles and greenhouse-collected volatiles were analyzed separately.

1. Gallitelli D (2000) The ecology of Cucumber mosaic virus and sustainable agriculture. *Virus Res* 71:9–21.

2. Eigenbrode SD, Ding H, Shiel P, Berger PH (2002) Volatiles from potato plants infected with potato leafroll virus attract and arrest the virus vector, *Myzus persicae* (Homoptera; Aphididae). *Proc R Soc Lond B Biol Sci* 269:455–460.

