

Effects of Exposure to Constant Light and Darkness under Varying Temperatures on *Systema frontalis* (Coleoptera: Chrysomelidae) Adults Feeding on Panicked Hydrangea¹

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Systema frontalis (F.) (Coleoptera: Chrysomelidae) is an insect pest of container-grown ornamental plants in nurseries across the central, midwestern, and eastern regions of the United States (Joseph et al. 2021, J. Integr. Pest Manage. 12: 17; Arshad et al. 2023a, J. Integr. Pest Manage. 14: 7). Adults cause damage to >50 species of ornamental plants in container-grown nurseries, including panicked hydrangea (*Hydrangea paniculata* Siebold; Hydrangeaceae), rose (*Rosa* sp.; Rosaceae), sweetspire itea (*Itea virginica* L.; Iteaceae), and weigela (*Weigela* spp.; Caprifoliaceae) (Arshad et al. 2023a, Joseph et al. 2021). Adults of *S. frontalis* feed on both the upper and lower surfaces of plant leaves, resulting in shot-hole damage and skeletonization of leaves (Arshad et al. 2023a, Joseph et al. 2021). Mated females oviposit in the growing medium of plant containers, where the larvae feed on plant roots and undergo 3 instars before pupating (Arshad et al. 2023a, Joseph et al. 2021). Although larval damage has not been documented, the presence of larvae in the growing medium of containers poses a threat to pest population dynamics in ornamental nurseries (Arshad et al. 2023b, J. Econ. Entomol. 116: 1760–1766).

Insects are ectothermic organisms, thus their body temperature is determined by ambient temperature (Costanzo 2011, Nat. Educ. Knowl. 3: 3). Consequently, temperature changes can affect metabolic rates, feeding activity, growth, development, and behavior. A strong positive correlation between temperature (6–20°C) and feeding activity on oilseed rape flower buds was observed with *Meligethes aeneus* (F.) (Nitidulidae: Coleoptera) (Ferguson et al. 2015, Pest Manage. Sci. 71: 459–466). However, the specific effects of temperature on the feeding activity of *S. frontalis* have not been thoroughly investigated. Similarly, light is a crucial environmental cue

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that influences various aspects of insect biology, including feeding (Constantinou and Cloudsley-Thompson 1980, J. Arid Environ. 3: 319–324; Suárez-Vidal et al. 2017, Front. Plant Sci. 8: 270397). Many insects exhibit phototaxis, being attracted to or repelled by light sources (Kasai and Hironak 2024, Appl. Entomol. Zool. 59: 155–162, van Grunsven, et al. 2014, J. Insect Conserv. 18: 225–231). Light or dark conditions can influence the timing of insect activities, such as feeding (Force et al. 2024, Front. Physiol. 14: 1304626; Saunders 1997, Invertebr. Neurosci. 3: 155–164), as observed in coleopterans (Pszczolkowski and Dobrowolski 1999, Phytoparasitica 27: 19–25).

In wholesale ornamental nurseries, container plants are commonly transported to garden centers, retail nurseries, or landscaping facilities, either within the same state or across state lines, by using 8.3- or 16.2-m-long trailer trucks. Plants are loaded onto rolling racks and moved into trailers from the loading deck. Once inside, plants may remain in the trailer for 6 to 48 h due to logistical delays or scheduling constraints. During such waiting periods, growers may open the rear door of the trailer to improve airflow; however, due to the length of the trailer, only the plants near the door receive light, whereas those further inside remain in complete darkness. The actual transport and unloading process may take an additional ~24 h, extending the period during which plants are subjected to variable temperature and light conditions.

In the nursery, container plants are exposed to *S. frontalis* adults and their growing medium may become infested with eggs, larvae, or pupae. The severity of infestation can vary depending on the duration of exposure, the host plant species, and the management practices. Once loaded onto trailers, these infested containers may serve as a source of newly emerging adults, particularly if pupae complete development and eclose during transit. In addition, adult *S. frontalis* may already be present, concealed in the plant canopy or within the growing medium, before loading. As a result, container plants are vulnerable to feeding damage by *S. frontalis* throughout the entire transportation process.

Currently, little is known about how adult *S. frontalis* respond to the environmental conditions encountered during transport, such as prolonged darkness and fluctuating temperatures. To better inform management strategies during plant shipment, we investigated the feeding behavior of adult *S. frontalis* under controlled combinations of temperature and constant light or dark conditions. Thus, the objective of the study was to determine the effects of high temperatures under light and dark conditions on incidence and severity of feeding damage by adult *S. frontalis*. These conditions simulate transport-relevant environments, providing insight into how these factors influence adult feeding behavior during transit.

In 2021, an experiment was conducted at the University of Georgia's Griffin Campus in Griffin. Adults of *S. frontalis* were collected from the leaves of panicked hydrangea and *Sedum* spp. plants in an ornamental nursery in McDuffie Co., GA. The adults were suctioned with aspirators from mid-May to July. The collected adults were then temporarily housed in a greenhouse at the University of Georgia's Griffin Campus, at ~26°C and 60% relative humidity. They were placed in 2 cages, measuring 47.5 × 47.5 × 93.0 cm (BugDorm, BugDorm-4E4590 Insect Rearing Cage, Taichung, Taiwan) each, that contained an 11.4-L 'Limelight' panicked hydrangea container plant of 60–80 cm sourced from the same nursery. The panicked hydrangea was not exposed to any pesticides for at least 2 mo before the study to ensure minimal residual effects. When ~60% of the leaves of the caged plants was damaged, it was replaced with an

undamaged plant sourced from the same nursery. The *S. frontalis* adults maintained in the cages were used in various experiments within a week. The panicked hydrangea plants were irrigated 3 times daily for 10 min with VibroNet™ sprinkler heads (Green, 12.4 GPH, part no. 0354050-B, Netafim USA, Fresno, CA) installed in the greenhouse.

The panicked hydrangea plants used in the experiment were planted in pine bark, and fertilized with 7.6 kg per m² of 18:9:10 [N:P:K] (Osmocote Pro, Summer-ville, SC) in August. The plants were irrigated for 20 min, 3 times a week by using an overhead irrigation system. The container plants used for the experiment were transported from the nursery to the University of Georgia's Griffin Campus in June 2021 and maintained in a shade house that provided 50% light for ~30 d.

Thirty experimental arenas were constructed using clear Dura-Lar film (Dura-Lar film, Grafix, Maple Heights, OH) with a thickness of 0.381 mm. A cylindrical tube, ~15 cm in diameter and 30 cm in length, was created by rolling a rectangular piece of clear film measuring 29.5 cm × 22.5 cm. The film was rolled end to end with a 2.5-cm overlap and secured lengthwise with masking tape. A no-see-um mesh (20 × 20 cm) with a mesh size of 0.7 × 0.6 mm (catalog no. 7250NSW, BioQuip Inc., Rancho Dominguez, CA) was attached to one end of the cylinder by using masking tape around its circumference. The other end of the cylinder was inserted into the growing medium in a 3.7-L plastic container, forming a cage that sealed the tube and prevented *S. frontalis* adults from escaping. For the experiments, a 30-cm-long panicked hydrangea terminal shoot with 8–14 leaves was collected from hydrangea plants in the shade house by using scissors. The terminal shoots were collected in the early morning (between 7:00 and 9:00 a.m.) on each day of the experiment to minimize water loss and ensure turgor. In total, 30 hydrangea terminal shoots were collected: one for each experimental unit across the 6 treatments (3 temperatures × 2 light conditions × 5 replicates). Each terminal was hydrated with tap water in a 20-ml [6 cm × 7.1 cm, diameter × length] polypropylene sample cup. The excised end of each terminal shoot was inserted through a 5-mm-diameter hole in the lid of the sample cup. The leaves of the terminal shoot remained fresh (no signs of wilting or discoloration and maintained full turgor) for up to 7 d (Arshad and Joseph 2024, J. Econ. Entomol. 117: 251–258). However, the leaves began to wilt at 36°C after 7 d; therefore, the results are not presented. Each sample cup with the terminal shoot was placed in a 3.7-L plastic container filled with growing medium and inserted halfway into the medium. This stabilized the cup upright. The field-collected *S. frontalis* adults were starved for 24 h before use in the experiment. Five starved *S. frontalis* adults were randomly selected and introduced into each experimental arena, containing a terminal shoot as described above. The sex and age of the adults used in the experiments were not determined. This tubular cage setup, containing a terminal shoot and 5 *S. frontalis* adults, constituted a single experimental unit in the feeding assay.

The experiment was conducted in controlled environmental chambers (Intellus control system, Percival Scientific, Perry, IA), and each had internal dimensions of ~91.4 cm (length) × 60.9 cm (width) × 157.5 cm (height) at the University of Georgia's Griffin Campus. As previously described, the plants, cages, and assay setup were used for the experiments. After releasing 5 beetles into each experimental arena, they were placed in the controlled environmental chamber at temperatures of 18, 25, and 36°C, both with and without light. These temperatures were chosen because summer afternoon highs in central Georgia typically exceed the mid-30°C range,

whereas overnight lows vary from 18 to 23°C (National Weather Service). Each treatment (specific temperature and light levels) was replicated 5 times, using 2 chambers.

For evaluation, each leaf was visually assessed for the incidence and severity of feeding damage at 0.5, 1.5, and 3.5 d after exposure. The incidence of damage was determined by counting the number of leaves exhibiting at least 1 instance of discrete or scooped surface damage caused by *S. frontalis* feeding, and the percentages of injured leaves were calculated for each experimental unit. The severity of feeding damage was assessed by visually rating the percentage of leaf area damaged by adult feeding across all leaves in each experimental unit as described in Arshad and Joseph (2024). The percentage severity of damage was then calculated by averaging the ratings from all leaves in each experimental unit.

All data analyses were conducted using SAS 9.3 software (SAS Institute, Cary, NC). Data from the light and temperature treatments were analyzed with a factorial design, incorporating 2 levels of light treatment (light and dark) and 3 temperatures (18, 25, and 36°C). After assessing the residuals for percentages of incidence and severity of damage, as well as examining histograms for normality through the PROC UNIVARIATE procedure, the data underwent arcsine square-root transformation. The transformed data were analyzed using a two-way analysis of variance (ANOVA) with interaction via the general linear model (PROC GLM procedure) for 0.5, 1.5, and 3.5 d. To evaluate the individual treatment effects of light and temperature, a one-way ANOVA was executed using the PROC GLM procedure for 0.5, 1.5, and 3.5 d, and means were separated using Tukey's honestly significant difference test ($\alpha = 0.05$).

The incidence and severity of *S. frontalis* feeding damage did not significantly differ between light and dark conditions at 0.5 and 1.5 d postexposure (Table 1). Significant effects of temperature were observed on both the incidence and severity of *S. frontalis* feeding damage at 0.5 and 1.5 d postexposure; however, at 3.5 d postexposure, only the severity of the damage showed a significant difference (Table 1). The interaction between light and dark conditions and temperature significantly influenced both the incidence and severity of damage at 3.5 d postexposure, but not at 0.5 and 1.5 d postexposure (Table 1).

Because of these interaction effects, a one-way ANOVA was performed at intervals postexposure and during light or dark exposure. Under light conditions, the incidence of *S. frontalis* feeding damage did not differ significantly at any postexposure intervals (0.5 h: $F = 3.7$; $df = 2, 12$; $P = 0.056$; 1.5 h: $F = 1.3$; $df = 2, 12$; $P = 0.317$; 3.5 h: $F = 2.7$; $df = 2, 8$; $P = 0.136$; Fig. 1A–C). The severity of feeding damage was significantly greater in the 25 and 36°C treatments than at the 18°C treatment at 0.5 d postexposure ($F = 42.7$; $df = 2, 12$; $P < 0.001$; Fig. 1D). At 1.5 d postexposure, the severity of feeding damage was significantly greater in the 36°C treatment relative to the 18 and 25°C treatments ($F = 9.3$; $df = 2, 12$; $P = 0.004$; Fig. 1E). There were no significant differences between the 18 and 25°C treatments at 1.5 and 3.5 d postexposure (Fig. 1E, F). Under dark conditions, the incidence and severity of *S. frontalis* feeding damage were significantly greater in the 25 and 36°C treatments than in the 18°C treatment at 0.5 (Fig. 1A [incidence: $F = 12.2$; $df = 2, 12$; $P = 0.001$]; Fig. 1D [severity: $F = 35.5$; $df = 2, 12$; $P < 0.001$]) and 1.5 d postexposures (Fig. 1B [incidence: $F = 4.9$; $df = 2, 12$; $P = 0.028$]; Fig. 1E [severity: $F = 9.3$; $df = 2, 12$; $P = 0.004$]). At 3.5 d postexposure, the incidence and severity of *S. frontalis* feeding damage were significantly greater in the 25°C treatment than in the

Table 1. Two-way analysis of variance of the light or dark, temperature, and their interaction effects on the feeding damage on panicked hydrangea caused by adult *Systema frontalis* feeding in the controlled environmental chamber.

Observation (d)	Light/dark			Temperature			Light-dark × temperature		
	<i>F</i>	df	<i>P</i>	<i>F</i>	df	<i>P</i>	<i>F</i>	df	<i>P</i>
Incidence									
0.5	1.8	1,24	0.195	14.2	2, 24	<0.001	0.9	2,24	0.407
1.5	3.4	1,24	0.078	3.6	2, 24	0.044	2.1	2,24	0.142
3.5	0.2	1,16	0.675	3.3	1, 16	0.088	21.8	1,16	<0.001
Severity									
0.5	0.6	1,24	0.438	76.1	2, 24	<0.001	0.5	2,24	0.600
1.5	1.6	1,24	0.208	16.2	2, 24	<0.001	2.4	2,24	0.110
3.5	4.7	1,16	0.046	21.2	1, 16	<0.001	9.1	1,16	0.008

18°C treatment (Fig. 1C [incidence: $F = 40.8$; $df = 2, 8$; $P < 0.001$]; Fig. 1F [severity: $F = 16.6$; $df = 2, 8$; $P = 0.004$]). The plant terminals in the 36°C treatment were not in good condition after 3.5 d postexposure; therefore, they were excluded from the analysis.

The feeding damage was greater in dark conditions than in light, but only when plants were exposed for an extended period in enclosed controlled chambers. A previous study showed that the weevil *Hylobius abietis* L. was more active in darkness than in illuminated conditions (Suárez-Vidal et al. 2017), whereas the beetle *Sitona gressorius* (F.) demonstrated greater activity at higher temperatures ($> 25^{\circ}\text{C}$) under light conditions in the laboratory (Hannigan et al. 2023, J. Pest Sci. 96: 389–402). The lack of light may have further restricted adult *S. frontalis* movement and increased damage due to their sedentary nature, especially when active during warmer temperatures. Previous research indicates that more plant defense chemicals are produced under light conditions in response to herbivores, resulting in reduced feeding damage compared to dark conditions (Suárez-Vidal et al. 2017). It remains unclear whether these chemical defenses influenced *S. frontalis* feeding on *H. paniculate* in dark conditions. Furthermore, *S. frontalis* adults are polyphagous (Arshad and Joseph 2023a), allowing them to easily locate suitable hosts for feeding in these warm, enclosed environments inside the trailer. Lane and Del-Pozo (2023, Environ. Entomol. 52: 730–739) observed that adult *S. frontalis* were motionless primarily from 7:00 p.m. to 7:00 a.m. as temperatures dropped at night.

In conclusion, our findings demonstrate that *S. frontalis* adults are capable of sustained feeding under extended periods of darkness, particularly at elevated temperatures—conditions that may be encountered during the transportation of containerized plants. This observation suggests a potential risk of feeding damage during transit and underscores the importance of developing mitigation strategies.

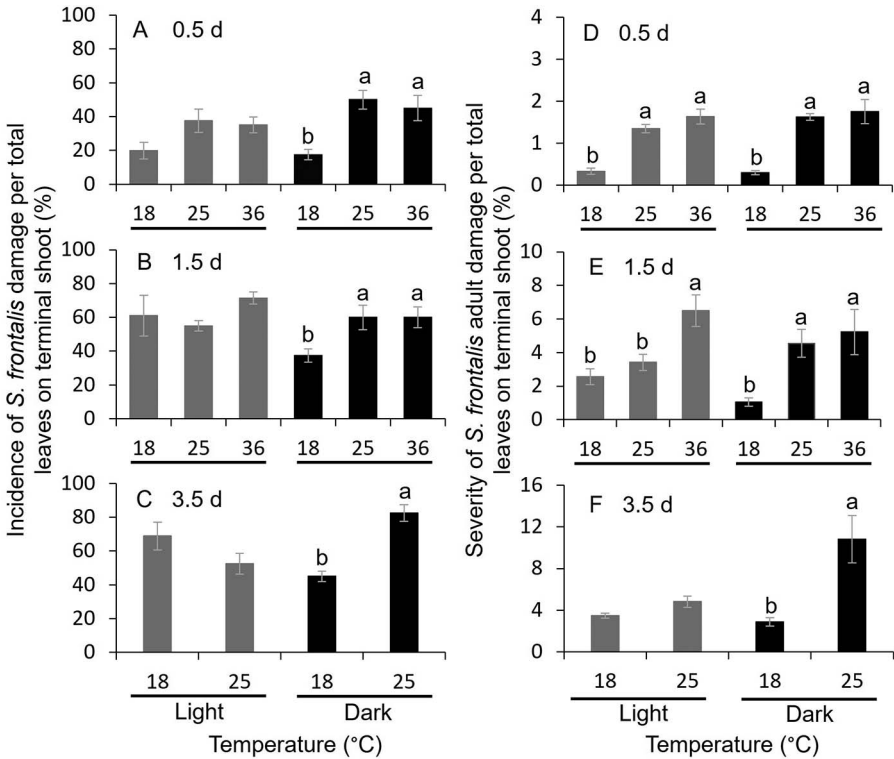


Fig. 1. Mean (\pm SEM) incidence of *Systema frontalis* feeding damage after exposure to 24 h of light (gray bars) or 24 h of dark (black bars) conditions at 18, 25, and 36°C, evaluated at (A) 0.5, (B) 1.5, and (C) 3.5 d postexposure. In addition, the severity of feeding damage after exposure to light-dark conditions at 18, 25, and 36°C was assessed at (D) 0.5, (E) 1.5, and (F) 3.5 d postexposure. Bars with same letters within light or dark conditions (x-axis) are not significantly different according to Tukey's honestly significant difference test ($\alpha = 0.05$).

However, these results should be interpreted with caution due to the limited number of treatments evaluated in this study. Potential management approaches—such as preshipment monitoring of plant leaves for adult presence, application of foliar insecticides or fumigants, and manipulation of shading levels within transport enclosures—warrant further investigation. Future research should focus on assessing the efficacy of these strategies under commercial shipping conditions to inform evidence-based recommendations for protecting ornamental plants during distribution.

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