

The Role of Antennae in Heat Detection and Feeding Behavior in the Bed Bug (Hemiptera: Cimicidae)

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Abstract

The common bed bug (*Cimex lectularius* L.) is an obligate hematophagous ectoparasite that has significant impacts on human health and well-being. All life stages of bed bugs (except eggs) feed solely on blood, which is required to molt and reproduce. Bed bugs use multiple cues to locate their hosts, including heat, CO₂, and body odors. Of these cues, detection of heat appears limited to a short distance of <3 cm. However, it remains unclear if bed bugs can detect radiant heat, what structure(s) are responsible for heat detection, and if heat detection via the antennae is required for feeding. In this study, bed bug response to radiant heat was evaluated using the two-choice T-maze assay with the heat source either in contact with the surface (i.e., conduction) or not in contact (i.e., radiation) in nonantennectomized bed bugs. Further, we systematically ablated the bed bug's antennal segments (distal tip, first segment, and all four segments) and assessed their responses to heat and feeding in a unique two-choice T-maze assay and individual feeding assays, respectively. Our two-choice assays with contact to or no contact with the surface indicated that bed bugs cannot detect radiant heat. Later, we found that the distal tip of the terminal antennal segment is responsible for orientation toward a heat source. However, >50% of the bed bugs fed even when the entire antenna was removed, suggesting redundancy in sensory cues that drive feeding. These results will be used to better understand the role heat plays in bed bug host attraction and design of traps.

Key words: bed bug, heat detection, feeding behavior, antennae, radiant heat

Bed bugs (*Cimex lectularius* L.) are hematophagous ectoparasites that have recently resurged across the globe. Bed Bugs are considered pests of significant public health importance (U.S. CDC and EPA 2010), largely due to their propensity to feed on humans (bites and allergic reactions) (Goddard and de Shazo 2009), the psychological burden they place on those dealing with infestations (Goddard and de Shazo 2012, Susser et al. 2012), and their ability to contaminate the indoor environment (DeVries et al. 2018, Kakumanu et al. 2020). Bed bugs also remain one of the more difficult indoor pests to manage, due primarily to the high levels of insecticide resistance observed toward the most commonly used indoor residual insecticides (Romero 2018).

All life stages of bed bugs (except eggs) feed solely on blood, which is required for growth, development, and reproduction (Usinger 1966). Bed bugs are primarily active during the scotophase, an adaptive behavior that minimizes their detection by the sleeping hosts and facilitates uninterrupted feeding (Romero et al. 2010).

Nocturnal activity necessitates the use of nonvisual cues in host location. As in other hematophagous insects, bed bugs are capable of detecting and responding to CO₂, odors, and heat. Carbon dioxide has previously been shown to be an attractant (Anderson et al. 2009, Wang et al. 2009a, Singh et al. 2012, Aak et al. 2014), although recent findings by Hentley et al. (2017) suggest that CO₂ serves as an activator rather than an attractant. Host odors have also been shown to be attractive to bed bugs (Hentley et al. 2017), with a recent study by DeVries et al. (2019), showing clear attraction to odors alone using a vertical Y-tube olfactometer. One common feature of CO₂ and host odors is their ability to elicit responses over several meters (Anderson et al. 2009, Aak et al. 2014, Hentley et al. 2017). In contrast to CO₂ and host odors, heat detection in bed bugs is limited to short distances of <3 cm (DeVries et al. 2016) and they respond to the heat by directing antennae to the heat source while they are in a hungry state (Rivnay 1932). Despite its short effective range, heat can independently activate bed bugs from an arrested state, guide

orientation toward the heat source, and elicit feeding responses (DeVries et al. 2016). Interestingly, although heat is important for bed bug feeding behavior, it does not appear to be required. DeVries et al. (2016) found that a small proportion of bed bugs were capable of feeding on blood at or below ambient temperature, suggesting feeding can occur independent of heat. Regardless, heat clearly provides a critical short-range cue that is important for host location and feeding.

In the environment, heat is known to transfer by convection (transfer of thermal energy through liquid or gas), conduction (transfer of thermal energy through contact), and radiation (transfer of thermal energy through thermal emission) (Whitaker 1977). At present, it remains unclear how bed bugs perceive host emitted heat. In addition, despite the importance of heat in bed bug host location and feeding (Rivnay 1932, DeVries et al. 2016), the sensory structures responsible for heat detection have not been characterized. Previous studies on bed bugs have shown that receptors responsible for human odor detection and chemical communication are located in the antennae (Herraca et al. 2010, Herraca et al. 2012, Liu and Liu 2015). Based on morphological assessments of bed bugs (Levinson et al. 1974, Steinbrecht and Muller 1976) and other closely related species (Barrozo et al. 2017), the antenna is likely responsible for heat detection, although this has yet to be shown.

We developed a study to determine 1) if bed bugs can detect radiant heat, 2) what parts of the antennae are responsible for detecting heat, and 3) how heat detection influences feeding. To address these questions, we evaluated attraction and orientation toward radiant and conductive heat sources. In addition, we evaluated attraction and orientation toward heat as well as feeding in antennectomized bed bugs. The results from this work are discussed in relation to bed bug host location and pest management.

Material and Methods

Bed Bugs

A laboratory colony of bed bugs was used for all experiments. This population, Harold Harlan (HH), was originally collected in 1973 in Ft. Dix, NJ, USA. Bed bugs were reared in the laboratory as described by DeVries et al. (2016). Briefly, bed bugs were maintained in 168 ml plastic containers (Consolidated Plastics, Stow, OH) on cardstock paper shelters. Bed bugs were reared at 25°C and 50% relative humidity on a 12:12 (L:D) h cycle. Bed bugs were fed defibrinated rabbit blood weekly through an artificial feeding system consisting of a heated water bath (to maintain blood at 37°C) which circulated water through custom-made water-jacketed glass feeders. Glass feeders had an artificial membrane (plant budding tape, A.M. Leonard, Piqua, OH) stretched over the bottom which contained the blood and kept it at 37°C, while simultaneously allowing bed bugs to feed through it. Only adult males were used for all experiments, and all bed bugs were starved 7–10 d prior to behavioral experiments to ensure they were in a host-seeking state.

Radiant Heat Detection

To determine if bed bugs can detect radiant heat, bed bugs with intact antennae were evaluated for orientation toward a heat source using the two-choice T-maze, as described by DeVries et al. (2016). Briefly, bed bugs were introduced individually at the base of a vertically oriented T-shaped arena. The arena was constructed by attaching cardstock paper to an acrylic glass backing, with the paper substrate cut in the following dimensions: 20 mm wide base which narrowed to a width of 3 mm over 50 mm. The two side arms of

the T-maze were each 75 mm long and 20 mm wide. A copper coil (i.d. = 1.5 mm, coiled diameter = 3 cm) was heated via a circulating water bath (RM6 Thermostat, Brinkmann Instruments Inc., Delran, NJ) to 38°C and placed at one side of the ‘T’ 10 mm from the choice point. This assay was modified by either leaving the heat source in contact with the substrate (conduction) or lifting it 1 mm off the substrate (radiation) at the same distance (10 mm) away from the bifurcation point. The heated side was alternated left and right to account for any position biases. Assays were conducted during the scotophase under red light, and included a minimum of 5 min acclimation at the base of the arena before the assay started. The acclimated bed bugs were given a maximum of 15 min to move up the vertical portion of the T-arena and make a choice. A choice was recorded when the bed bug moved 10 mm in one direction at the top of the ‘T’.

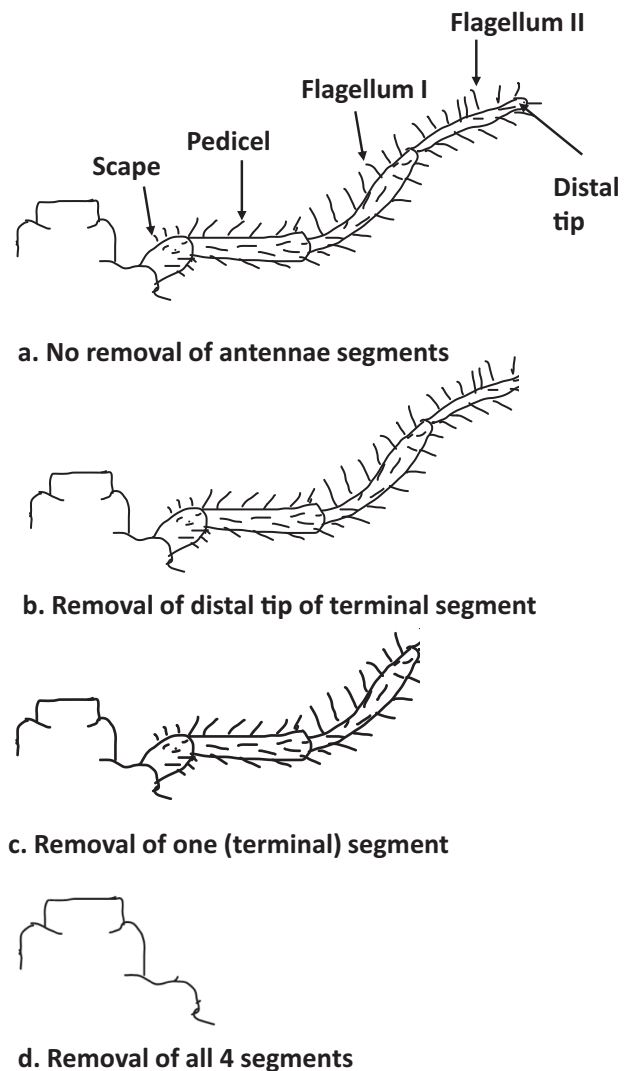


Fig. 1. Bed bug antenna, showing the segments ablated. (a) The right antenna which consists of four segments; scape, pedicel, flagellum I, and flagellum II. Figure (a) was drawn in referenced to Levinson et al. (1974). (b–d) Different segments of both left and right antennae were removed as shown in the above figures for heat choice and feeding assays.

Bed Bug Antennectomies

Bed bugs were anesthetized using CO₂, then antennectomies were performed. Using a fine blade scalpel under a microscope, the antenna was severed at one of three different locations: the tip of the terminal segment (~20 μm, Fig. 1b), between flagellum I and flagellum II (removal of the terminal segment, Fig. 1c), and at the base of the scape (removal of the entire antenna, Fig. 1d). Both antennae from each bed bug were removed at the same location. Control bugs were anesthetized, but no antennal segments were removed. Bed bugs were then placed individually into 7.5 ml glass vials (Thermo Fisher Scientific, Waltham, MA) with a harborage and allowed 24 h to recover prior to initiating any behavioral assays. For feeding assays, each 7.5 ml glass vial was outfitted with a mesh lid (0.3 mm mesh size; BioQuip Products, Rancho Dominguez, CA) and a paper harborage that stretched from the base of the arena to the lid and permitted feeding.

Two-Choice Heat Assays With Antennectomized Bed Bugs

To determine which antennal segments were responsible for heat detection, bed bugs that received different antennectomies were screened for orientation toward a heat source using a two-choice T-maze as described in radiant heat detection section. However, no modifications were done in this assay from the methods described by DeVries et al. (2016).

Feeding Assays With Antennectomized Bed Bugs

To determine if the antennae are needed for feeding behavior, bed bugs that received different antennectomies were screened for feeding using the same feeding setup as used for colony rearing. Bed bugs in individual vials were offered a bloodmeal for 20 min, after which they were evaluated for ingestion. Because lighting can influence feeding behavior (Gaire and DeVries personal observation), feeding assays were conducted during the scotophase under two light conditions: dark room (lights were turned off and a red light was used by the observer, but limited ambient light could get in through windows and doors) and blackout (bugs were covered with a black cloth that prevented ambient light from entering the assay while they were feeding). The number of fully engorged bed bugs was recorded for each antennectomized group.

Statistical Analysis

A chi-square goodness of fit test was used to determine if bed bugs could detect radiant heat, and to evaluate the response of antennectomized bed bugs to heat and, with the null hypothesis that if bed bugs are unable to detect heat they will display a 1:1 preference ratio for both sides of the assay. Feeding responses were evaluated using a chi-square goodness of fit test, where each antennectomized group (distal tip, one segment, four segments) was compared with the intact control group. All tests were conducted in SPSS Version 26 (IBM Corp., Armonk, NY).

Results

Radiant Heat Detection

Bed bugs showed significant preference for the heated side of the assay arena when the heated coil was in contact with the surface ($\chi^2_{1,20} = 7.20$, $P = 0.007$; Fig. 2). When the heated coil was not in contact with the surface, bed bug had no significant preference for either side ($\chi^2_{1,32} = 0.50$, $P = 0.480$; Fig. 2), indicating that under

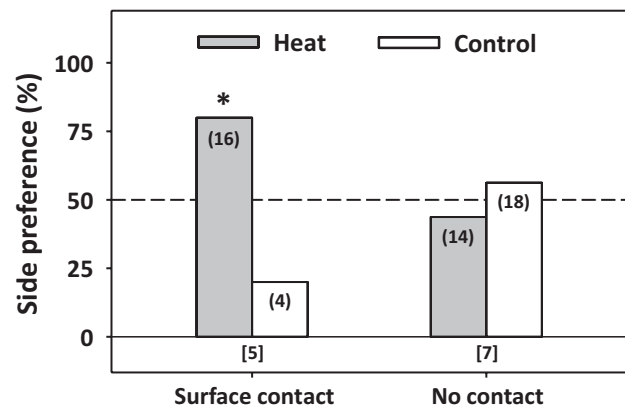


Fig. 2. Response of bed bugs to radiant heat. An asterisk (*) indicates significant preference for one side of the assay ($P < 0.05$; chi-square test). The number of responders is indicated in parentheses for each choice, and the number of nonresponders is indicated in brackets below each set of choices for each assay.

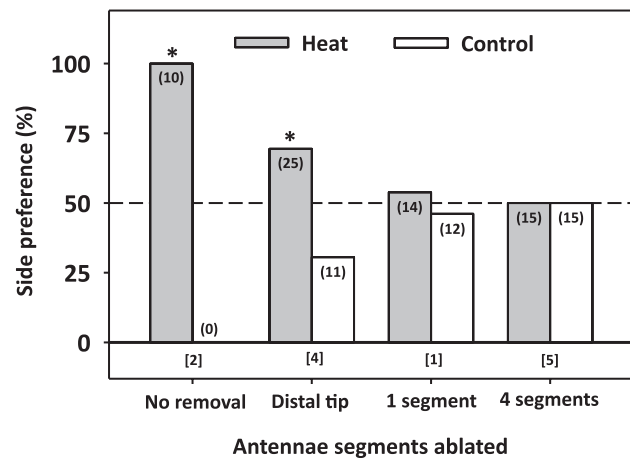


Fig. 3. Response of antennectomized bed bugs to a heated coil. An asterisk (*) indicates significant preference for one side of the T-maze assay ($P < 0.05$; chi-square test). The number of responders is indicated in parentheses for each choice, and the number of nonresponders is indicated in brackets below each set of choices for each assay.

the current experimental conditions they were unable to detect radiant heat.

Responses of Antennectomized Bed Bugs to Heat

Bed bugs with intact antennae (nonantennectomized) showed significant preference for the side with the heated coil ($\chi^2_{1,10} = 10.00$, $P = 0.002$; Fig. 3). However, when we removed either the terminal segment (flagellum II) or all four segments, bed bugs displayed no preference for either side of the T-maze (one segment removal: $\chi^2_{1,26} = 0.154$, $P = 0.695$; four segment removal: $\chi^2_{1,30} = 0.00$, $P = 1.00$; Fig. 3). Interestingly, when we removed only the distal tip of the terminal segment, bed bugs showed a significant preference for the side with the heated coil ($\chi^2_{1,36} = 5.44$, $P = 0.020$; Fig. 3), although the response was lower than in bed bugs with intact antennae.

Blood Feeding Responses by Antennectomized Bed Bugs

Antenna removal significantly effected bed bug feeding. When bugs were fed in dark, but not blackout conditions, antennal removal

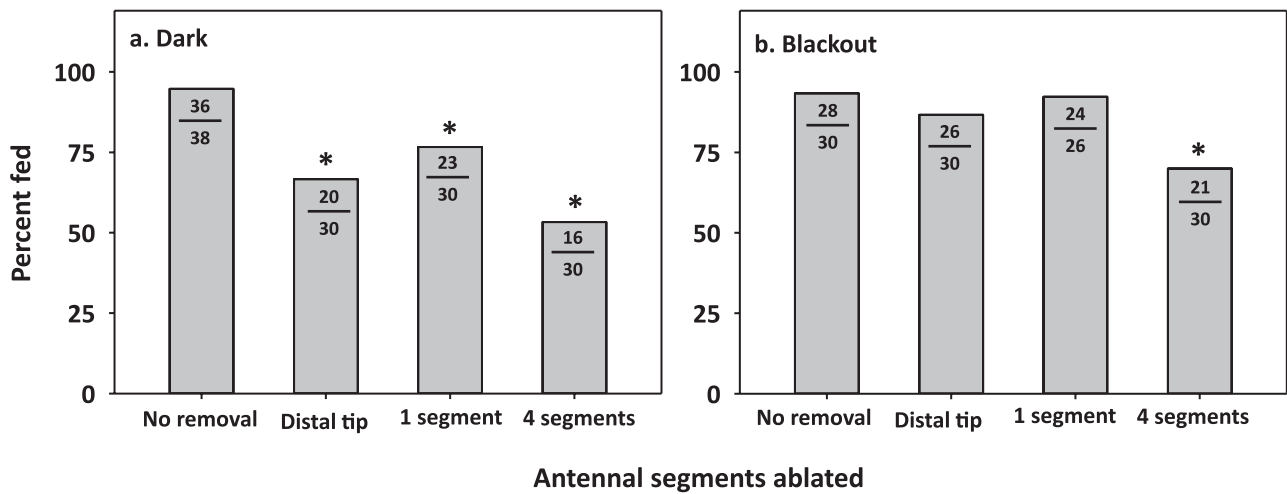


Fig. 4. Feeding response of antennectomized bed bugs fed in (a) dark, and (b) blackout conditions. An asterisk (*) indicates significant reduction in blood feeding in comparison to the intact control group (i.e., no removal) for each lighting condition ($P < 0.05$; chi-square test). The proportion of bed bugs that fed to the total number tested is indicated within each bar.

resulted in a significant decline in the number of bugs that fed (distal tip removal: $\chi^2_{1,30} = 9.08$, $P = 0.003$; one segment removal: $\chi^2_{1,30} = 4.76$, $P = 0.029$; four segment removal: $\chi^2_{1,30} = 15.97$, $P = <0.001$; Fig. 4). However, when bed bugs were fed under blackout conditions, we found no significant difference in feeding response for either the distal tip or one segment removal (distal tip removal: $\chi^2_{1,30} = 0.741$, $P = 0.389$; one segment removal: $\chi^2_{1,26} = 0.022$, $P = 0.882$; Fig. 4) although those with the entire antenna removed showed a significant reduced in feeding response ($\chi^2_{1,30} = 5.45$, $P = 0.020$; Fig. 4). That said, in all assays, >53% of bed bugs fed regardless of antenna removal (Fig. 4).

Discussion

Given the limited distance over which bed bugs can detect heat, it was also important to characterize how they do so. Therefore, we conducted choice assays with the heat source (copper coil) either in contact or not in contact with the surface. When the heat source was in contact with the surface, 80% of the test population chose the heated side; however, when there was no contact between the heat source and the arena surface, bed bugs did not show a significant preference for either side (Fig. 2). These results suggest that bed bugs cannot detect radiant heat, and their ability to detect conductive heat is limited to less than 3 cm (DeVries et al. 2016). In contrast to bed bugs, kissing bugs can detect radiant heat (infrared radiation) and can estimate the temperature from 10 cm at laboratory conditions, especially when in a host-seeking state (Lazzari and Nunez 1989, Schmitz et al. 2000, Barrozo et al. 2017). The thermoreceptors of kissing bug (*Rhodnius prolixus*) present within peg-in-pit sensilla and tapered hairs can respond to both warm airstreams as well as infrared radiation (Zopf et al. 2014). The evolution of radiant heat detection in hematophagous arthropods may be related to their spatial relationship with their hosts. Bed bugs are wingless and are found in close proximity to the host and thus do not need to navigate toward the host over long distances. Kissing bugs on the other hand, are peridomestic, can fly, and often traverse long distances to find their human or animal hosts. Recently, detection of infrared radiation and long distance orientation (1 m) to radiant heat was documented in the ticks (*Amblyomma americanum* and *Dermacentor variabilis*) (Carr and Salgado 2019), which quests on vegetation and needs to be able to detect an approaching potential host.

The behavior assays conducted with antennectomized bed bugs confirmed that the distal tip of the terminal antennal segment is responsible for heat detection. When we further cut the first segment or removed the entire antenna, bed bugs could no longer orient toward the heated coil. This indicates that their ability to orient toward a heat source is reliant upon the distal tip of the terminal segment. The distal tip of the terminal antennal segment of *C. lectularius* houses seven sensilla; bristles (type A1), immersed cones (type B1), plates (type B2), grooved pegs (type C), smooth pegs (type D), and hairs with even (type E1) and uneven (type E2) wall thickness (Levinson et al. 1974). All sensilla can be found in both adult sexes and most are present in nymphs, which indicate that all life stages and both sexes can detect the cues necessary for host location and feeding (Levinson et al. 1974). Among the seven sensilla, porous sensilla (types C, D, and E) are potentially responsible for detecting different cues such as pheromones, odors, and temperature (Steinbrecht and Muller 1976, Liu and Liu 2015). Levinson et al. (1974) showed that type E sensilla are responsible for the detection of alarm pheromones and Herraca et al. (2010) and Liu and Liu (2015) confirmed that D sensilla are responsible for human odorants reception. It still remains unclear, however, which sensilla are responsible for detecting heat. Future work should employ single sensillum recordings on C, D, and E sensilla with a range of temperatures to better understand how bed bugs detect thermal cues. In other hematophagous insects, such as kissing bugs, coeloconic sensilla, tapered hairs, and cave organ located in antennae and some other parts of body are responsible for heat detection (Barrozo et al. 2017). In mosquitoes, small coeloconic sensilla located in antennae are responsible for heat detection (Davis and Sokolove 1975). Advanced molecular work conducted on mosquitoes, kissing bugs, and other insects showed that neurons in thermosensitive sensilla expressed transient receptor potential (TRP) proteins that respond to heat (Wang et al. 2009b, Zermoglio et al. 2015, Corfas and Vosshall 2015, Gonzalez-Tokman 2020). Therefore, it is also important that future work evaluate the functional relationship between thermo-detection and TRP expression in bed bugs.

Heat detection also plays a critical role in bed bug feeding behavior (DeVries et al. 2016). Our results supported previous observations, as feeding responses were significantly reduced when antennal segments were removed. Although there was a significant positive relationship between antennae segments and feeding, more than 53

and 70% of bed bugs were able to feed without all four segments of the antennae in dark and blackout conditions, respectively (Fig. 4). This indicates that although the antennae are critical for orientation toward a heat source, they are not required for feeding. These findings also support previous results from DeVries et al. (2016), which showed bed bugs were capable of feeding even on blood cooled to temperatures below ambient. The gustatory system of insects is broadly distributed in different body parts, with sensilla housing gustatory receptors found on antennae, mouthparts, pharynx, wings, and legs; these receptors detect the presence of phagostimulants and deterrents which signal the presence of nutritious food that promotes feeding and noxious chemicals that signal potential toxins and xenobiotics (Liman et al. 2014). The gustatory receptors respond to different tastants (e.g., bitter, sweet, water, and salts) and convey appetitive or aversive information to the CNS through gustatory neurons (Liman et al. 2014, Freeman and Dahanukar 2015). Romero and Schal (2014) found that adenosine triphosphate (ATP) and sodium chloride (NaCl) are important phagostimulants capable of inducing feeding in bed bugs. However, it remains unclear which stimuli elicit probing in the absence of heat and the antennae, and if other phagostimulants drive ingestion. Future experiments should evaluate natural probing behavior and whether the proboscis can detect heat.

In conclusion, this study documents that bed bugs appear incapable of detecting radiant heat, which is likely why heat detection is limited to short distances. In addition, we found that the distal tip of the terminal antennal segment of bed bugs is responsible for heat detection and orientation toward a heat source. Furthermore, although the presence of the antennae (and thus heat detection) facilitates feeding, the antennae are not required for feeding. This work provides the basis for further physiological and molecular studies to understand not only heat detection and orientation to heat, but host location through the combination of multiple sensory modalities. Future comprehensive behavioral, physiological, and molecular studies should be focused on identifying the sensilla, neurons and receptors that are responsible for heat detection and feeding. In addition, future work should evaluate the responses of different populations, life stages, and sexes to determine their behavior to heat and feeding. With a better understanding of host location, we will be able to develop better traps and predictive models of how bed bugs locate their hosts.

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References Cited

- Aak, A., B. A. Rukke, A. Soleng, and M. K. Rosnes. 2014. Questing activity in bed bug populations: male and female responses to host signals. *Physiol. Entomol.* 39: 199–207.
- Anderson, J. F., F. J. Ferrandino, S. Mcknight, J. Nolen, and J. Miller. 2009. A carbon dioxide, heat and chemical lure trap for the bedbug, *Cimex lectularius*. *Med. Vet. Entomol.* 23: 99–105.
- Barrozo, R. B., C. E. Reisenman, P. Guerenstein, C. R. Lazzari, and M. G. Lorenzo. 2017. An inside look at the sensory biology of triatomines. *J. Insect Physiol.* 97: 3–19.
- Carr, A. L., and V. L. Salgado. 2019. Ticks home in on body heat: a new understanding of Haller's organ and repellent action. *PLoS One.* 14: e0221659.
- Corfas, R. A., and L. B. Vosshall. 2015. The cation channel TRPA1 tunes mosquito thermotaxis to host temperatures. *eLife* 4:e11750.
- Davis, E. E., and P. G. Sokolove. 1975. Temperature responses of antennal receptors of the mosquito, *Aedes aegypti*. *J. Comp. Physiol.* 96: 223–236.
- DeVries, Z. C., R. Mick, and C. Schal. 2016. Feel the heat: activation, orientation and feeding responses of bed bugs to targets at different temperatures. *J. Exp. Biol.* 219: 3773–3780.
- DeVries, Z. C., R. G. Santangelo, A. M. Barbarin, and C. Schal. 2018. Histamine as an emergent indoor contaminant: accumulation and persistence in bed bug infested homes. *PLoS One.* 13: e0192462.
- DeVries, Z. C., A. M. Saveer, R. Mick, and C. Schal. 2019. Bed bug (Hemiptera: Cimicidae) attraction to human odors: validation of a two-choice olfactometer. *J. Med. Entomol.* 56: 362–367.
- Freeman, E. G., and A. Dahanukar. 2015. Molecular neurobiology of *Drosophila* taste. *Curr. Opin. Neurobiol.* 34: 140–148.
- Goddard, J., and R. deShazo. 2009. Bed bugs (*Cimex lectularius*) and clinical consequences of their bites. *JAMA* 301: 1358–1366.
- Goddard, J., and R. de Shazo. 2012. Psychological effects of bed bug attacks (*Cimex lectularius* L.). *Am. J. Med.* 125: 101–103.
- González-Tokman, D., A. Córdoba-Aguilar, W. Dáttilo, A. Lira-Noriega, R. A. Sánchez-Guillén, and F. Villalobos. 2020. Insect responses to heat: physiological mechanisms, evolution and ecological implications in a warming world. *Biol. Rev.* 95: 802–821.
- Harraca, V., R. Ignell, C. Löfstedt, and C. Ryne. 2010. Characterization of the antennal olfactory system of the bed bug (*Cimex lectularius*). *Chem. Senses* 35: 195–204.
- Harraca, V., C. Ryne, G. Birgersson, and R. Ignell. 2012. Smelling your way to food: can bed bugs use our odour? *J. Exp. Biol.* 215: 623–629.
- Hentley, W. T., B. Webster, S. E. F. Evison, and M. T. Siva-Jothy. 2017. Bed bug aggregation on dirty laundry: a mechanism for passive dispersal. *Sci. Rep.* 7: 11668.
- Kakumanu, M. L., Z. C. DeVries, A. M. Barbarin, R. G. Santangelo, and C. Schal. 2020. Bed bugs shape the indoor microbial community composition of infested homes. *Sci. Total Environ.* 743: 140704.
- Lazzari, C. R., and J. A. Nunez. 1989. The response to radiant heat and the estimation of the temperature of distant sources in *Triatoma infestans*. *J. Insect Physiol.* 35: 525–529.
- Levinson, H. Z., A. R. Levinson, B. Muller, and R. A. Steinbrecht. 1974. Structure of sensilla, olfactory perception, and behaviour of the bedbug, *Cimex lectularius*, in response to its alarm pheromone. *J. Insect Physiol.* 20: 1231–1248.
- Liman, E. R., Y. V. Zhang, and C. Montell. 2014. Peripheral coding of taste. *Neuron.* 81: 984–1000.
- Liu, F., and N. Liu. 2015. Human odorant reception in the common bed bug, *Cimex lectularius*. *Sci. Rep.* 5: 15558.
- Rivnay, E. J. P. 1932. Studies in tropisms of the bed bug *Cimex lectularius* L. *Parasitology* 24: 121–136.
- Romero, A. 2018. Insecticide resistance, pp. 273–284. In S. L. Doggett, D. M. Miller and C. Y. Lee (eds.), *Advances in the biology and management of modern bed bugs*. Wiley Blackwell, West Sussex, UK.
- Romero, A., and C. Schal. 2014. Blood constituents as phagostimulants for the bed bug *Cimex lectularius* L. *J. Exp. Biol.* 217: 552–557.
- Romero, A., M. F. Potter, and K. F. Haynes. 2010. Circadian rhythm of spontaneous locomotor activity in the bed bug, *Cimex lectularius* L. *J. Insect Physiol.* 56: 1516–1522.
- Schmitz, H., S. Trenner, M. H. Hofmann, and H. Bleckmann. 2000. The ability of *Rhodnius prolixus* (Hemiptera; Reduviidae) to approach a thermal source solely by its infrared radiation. *J. Insect Physiol.* 46: 747–751.
- Singh, N., C. Wang, R. Cooper, and C. Liu. 2012. Interactions among carbon dioxide, heat, and chemical lures in attracting the bed bug, *Cimex lectularius* L. (Hemiptera: Cimicidae). *Psyche* 102: 1580–1585.
- Steinbrecht, R. A., and B. Moller. 1976. Fine structure of the antennal receptors of the bed bug, *Cimex lectularius*, *Tissue Cell* 8: 615–636.
- Susser, S. R., S. Perron, M. Fournier, L. Jacques, G. Denis, F. Tessier, and P. Roberge. 2012. Mental health effects from urban bed bug infestation (*Cimex lectularius* L.): a cross-sectional study. *BMJ Open* 2: e000838.
- U.S. Centers for Disease Control and Prevention (CDC) and U.S. Environment Protection Agency (EPA). 2010. Joint Statement on Bed Bug Control in the United States from the U.S. Centers for Disease Control and Prevention

- (CDC) and the U.S. Environmental Protection Agency (EPA). Washington, DC, USA <https://stacks.cdc.gov/view/cdc/21750>
- Usinger, R. L. 1966. Monograph of Cimicidae (Hemiptera—Heteroptera). Thomas Say Foundation, College Park, MD.
- Wang, C., T. Gibb, G. W. Bennett, and S. McKnight. 2009a. Bed bug (Heteroptera: Cimicidae) attraction to pitfall traps baited with carbon dioxide, heat, and chemical lure. *J. Econ. Entomol.* 102: 1580–1585.
- Wang, G., Y. T. Qiu, T. Lu, H. W. Kwon, R. J. Pitts, J. J. Van Loon, W. Takken, and L. J. Zwiebel. 2009b. *Anopheles gambiae* TRPA1 is a heat-activated channel expressed in thermosensitive sensilla of female antennae. *Eur. J. Neurosci.* 30: 967–974.
- Whitaker, S. 1977. *Fundamental Principles of Heat Transfer*. Pergamon Press Inc., New York, NY.
- Zermoglio, P. F., J. M. Latorre-Estivalis, J. E. Crespo, M. G. Lorenzo, and C. R. Lazzari. 2015. Thermosensation and the TRPV channel in *Rhodnius prolixus*. *J. Insect Physiol.* 81: 145–156.
- Zopf, L. M., C. R. Lazzari, and H. Tichy. 2014. Differential effects of ambient temperature on warm cell responses to infrared radiation in the blood-sucking bug *Rhodnius prolixus*. *J. Neurophysiol.* 111: 1341–1349.