Development and field evaluation of a motion sensor activated suction trap to study vector–host interactions

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Abstract

1. Researchers elucidating vectors of zoonotic diseases encounter problems with inefficient surveillance techniques leading to underestimation of the importance of some species, and the overestimation of the importance of others. Carbon dioxide-baited light traps are the most widely used traps for sampling vector groups. However aspirating directly from the hosts is the most accurate method to incriminate vectors.

2. A novel vector trapping system was developed, consisting of a suction trap, activated by a motion sensor, and controlled by a microcontroller, which activates automatically when host animals approach. The prototype was tested in two field experiments with ungulates and biting midges (Diptera: Ceratopogonidae) at a preserve in Florida. We measured the biting midge community collected at traps near and far from hosts and compared communities using diversity metrics and abundance curves.

3. Traps activated in the presence of host animals with 94% accuracy. Diversity and richness of Culicoides species differed between sensor and control traps with 11 species captured by control traps and seven species by sensor traps. Vector species were captured in significantly greater numbers in sensor traps, while more non-vector species were caught in control traps.

4. Results confirm that vector species can be underrepresented in light trap collections, likely due to their tight associations with vertebrate hosts, a finding that should be taken into consideration when incriminating arbovirus vectors. Our novel trap system was a first attempt at solving the issue of collecting vector species from non-tame animals, effectively aspirating questing midges. Simple modifications of the system could be made to target other vector–host systems.

Keywords

Culicoides, hosts, trapping, vectors

1 | INTRODUCTION

Researchers elucidating vectors of zoonotic diseases encounter problems with inefficient surveillance techniques leading to underestimating the importance of some species, while overestimating the importance of others. Aspirating directly from the host is the most accurate method to incriminate vector species (Silver, 2008), but is tedious for tame animals, and intractable with non-tame animals. The light trap, baited with carbon dioxide, is the most widely used trap for sampling vector groups, and has largely replaced the use of human and animal-baited traps for assessing vectors (Silver, 2008). However, several studies have demonstrated the light
trap’s sampling biases for certain hematophagous Diptera, often resulting in inaccurate representation of the host-seeking population (Acuff, 1976; Brown et al., 2008; McDermott & Mullens, 2018). New innovations in animal-baited traps are needed, as they better represent the vector community, avoiding failures of light traps to accurately portray the vector population and their omission of putative vector species.

Battery-powered aspirators can be used to sample directly from tame animals and are important for investigating the vectors and/or transmission ecology of zoonotic or veterinary pathogens. However, these methods expose researchers and animal subjects to vector-borne pathogens, are time intensive and do not afford the ability to sample from wild or non-tame animals (Tangena et al., 2015). Several traps have been developed to study the arthropods feeding upon nonhuman vertebrates (Lima et al., 2014; Tangena et al., 2015). The Trinidad No. 10 trap (Davies, 1971), for example, utilizes small, caged animals, such as rodents, opossums or birds, as bait to attract and collect hematophagous insects (Davies, 1978; Emord & Morris, 1982; Ferro et al., 2003; Lepore et al., 2004). Traps for collecting vectors from larger hosts include the drop-net trap, wherein a large, fine-mesh net is lowered over a tame animal and biting insects are aspirated (Carpenter et al., 2008), or a sticky-cover trap, capturing insects which land on the host (Viennet et al., 2011). These methods present challenges in that they are time consuming, do not typically provide robust samples (Moncayo et al., 2009), and either require trapping of live, wild animals or use of tame animals.

Here we report development of a novel system which actively samples potential vectors around host animals, utilizing a passive infrared (PIR) motion sensor and microcontroller, activating a suction trap. The system was field tested on Culicoides vectors of Orbiviruses at a preserve in Florida with numerous native and non-native, free-roaming and penned, ungulate species. Objectives of this study were to (a) develop a motion sensor-activated trap to automatically sample Culicoides vectors from host animals and (b) to determine whether insects trapped from host animals are representative of the overall insect community in the area.

2 | MATERIALS AND METHODS

2.1 | Trap development

A motion sensor suction trap device was developed for the collection of host-seeking Culicoides biting midges from captive, non-tame ungulates. The device consists of three major commercially available components, including (a) a miniature Centers for Disease Control and Prevention (CDC) light trap (Model #: 2836BQ) from BioQuip Products Inc.: Rancho Dominguez, CA, (b) Arduino Uno from Arduino.cc: Scarmagno, Italy and (c) a passive infrared (PIR) motion sensor (Model #: HC-SR501) from Ardest Electronics: Gauteng, South Africa (Figure 1). Total approximate cost of the device including items 1–6 (Figure 1) is $275.82 USD.

The suction trap consists of a CDC Miniature Light Trap with UV/LED-array (BioQuip Products Inc., Model # 2790V390). The light trap was attached, using a standard PVC pipe-coupler, to a fine stainless-steel wire mesh funnel and 50 ml conical tube (Figure 1). The PVC pipe-coupler was attached to a wooden shaft and steel shepherds hook for stability, and was positioned ~1.5 m above the ground. The light trap and Arduino Uno were powered by separate 6V 5Ah batteries. The motion sensor used to turn the suction trap and light on/off was manually calibrated using time (90 s) and sensitivity (lowest setting) dials built into the sensor.

**FIGURE 1** Schematic of the motion sensor trap: (1) 6V DC power supply ($19.99), (2) Arduino Uno ($20.49), (3) breadboard ($1.30), (4) power inverter ($2.60), (5) PIR motion sensor ($2.60), (6) light trap without motion sensor ($202.18), (7) light trap with motion sensor ($202.18), (8) pipe coupler ($5.30), (9) wire-mesh funnel, (10) collection tube with 90% EtOH. Not to scale. Dollar amounts in USD.
2.2 | Study sites and locations

Field work was conducted at a 200-ha big game preserve in Gadsden County, Florida. Three locations were chosen based on the diversity of ungulate species present (Table 1). Locations within the property where traps were set are referenced in the results by the diversity of host species visiting each site, according to the Morisita-Horn index in the R package SpadeR (Chao et al., 2015; Table 1). Sites were named such that the site named ‘High diversity’ has the highest diversity of ungulate species, ‘Low diversity’ for the white-tailed deer pen and finally, ‘Medium diversity’ for the location with intermediate diversity.

2.3 | Experimental design

Two traps were set at each location, with one trap located directly next to a feeder (motion sensor) and the other located 10 m distant from the feeder (control), but in an otherwise equivalent habitat (Figure 2). Feeders consisted of a wooden trough 1.5 m from the ground, with an aluminum roof and filled with one 22.7-kg bag of Sportsman Choice™ Record Rack® (Cargill, Incorporated) deer feed daily. Both traps were connected to the same motion sensor, such that both actuated for the same time and duration. The motion sensor trap was located next to the feeder in order to capture biting midges from approaching animals. The control trap was positioned to capture biting midges that were active in the environment.

Two short-term field experiments were conducted to determine whether midge species captured in motion sensor traps were different from those in control traps. In Experiment 1 (23–27 July 2018) the motion sensor and control traps used only light to attract biting midges. In Experiment 2 (25–29 August 2018) the control trap was augmented with 2 L of dry ice to simulate an important host cue. In both experiments, traps were operated from approximately 1 hr before sunset until 1 hr after sunrise. Midges were captured directly into 90% EtOH in a 50 ml conical tube. Culicoides species were identified to species using morphological characteristics (Blanton & Wirth, 1979).

|                | High diversity | Medium diversity | Low diversity |
|----------------|----------------|------------------|---------------|
| High diversity | —              | 4                | 2             |
| Medium diversity | 0.891          | —                | 1             |
| Low diversity  | 0.297          | 0.606            | —             |

**TABLE 1** Results of the Morisita-Horn similarity index using SpadeR package in R. Cells in yellow represent shared species between sites, while green cells represent similarity between sites, with a corresponding list of species observed at each site.

**FIGURE 2** Generalized experimental set-up for experiment 1. Experiment 2 was set up similarly, with the addition of CO₂ at the control trap. (1) Feeder, (2) game camera, (3) motion sensor trap, (4) motion sensor, (5) microcontroller, (6) 10-m wire connecting motion sensor and control traps, (7) control trap.
Two motion activated game cameras (Model # G42NG, Stealth Cam) were set up at each experimental location in order to monitor trap functionality and to quantify feeder visits by ungulate species. Cameras were pointed towards the motion sensor traps with the control traps in the background, and set to take 10-s videos with a 30-s delay between videos. Host species abundance, date, time, and whether the trap appeared ‘on’ or ‘off’ in animal presence, was recorded with the videos.

2.4 | Statistical analysis

Chi-squared tests of independence were carried out in Microsoft Excel 2013 (v15.0; Microsoft Corporation) to test the independence of distributions of Culicoides species composition between the control and motion sensor traps at each site (McDonald, 2014). This test was carried out for four vector species for experiments 1 and 2, including Culicoides insignis, Culicoides debilipalpis, Culicoides stellifer Culicoides venustus and one non-vector species during experiment 2 only, Culicoides torreyae.

A generalized linear mixed model using glmmADMB r package (Bolker et al., 2012) was used to determine differences between numbers of Culicoides species found between sensor and control traps using r version 3.6.1 (R Development Core Team, 2019). Variation between sites and sampling dates were accounted for in the model as random effects using the negative binomial family of models (O’Hara & Kotze, 2010). A sample size-based diversity accumulation curve and rarefaction analysis was carried out using the iNEXT r package (Chao et al., 2014) on data from the second experiment to allow for the simulation of expected number of species trapped beyond the data provided.

3 | RESULTS

In total, 13,659 Culicoides females were captured: 1,978 females from experiment 1, and 11,681 from experiment 2 (Table 2). Some males were also captured but numbers were not included in the analyses as males do not take blood meals. Wildlife cameras show motion sensor traps functioning in 94% of the recordings according to 3,774 10-s recordings.

Control traps collected a wider variety of species with diverse host affinities than motion sensor traps, which primarily collected putative vector species. This was demonstrated by the presence of a greater number of species captured in control traps \( n = 11 \) than motion sensor traps \( n = 7 \). Species captured in control traps but not in motion sensor traps (Table 2) are not considered to be major vector species. Only one species, Culicoides torreyae Blanton and Wirth was collected in greater numbers in the control trap (Table 3), while still being present in motion sensor traps.

Motion sensor traps collected primarily Culicoides species known to feed on ungulate hosts, and in higher numbers than control traps. During experiment 1, motion sensor traps captured 3.6 times as many total female Culicoides from five different species and 39.2 times as many blood-engorged females as control traps (Tables 2 and 3). In experiment 2, motion sensor traps collected 1.9 times as many females and 16.7 times as many blood-engorged specimens as control traps. Non-vector species including C. torreyae, were captured in significantly greater numbers in the control traps during experiment 2 (Table 3; Figure 3). In general, vector species were caught in greater numbers in motion sensor traps, and non-vector species were caught in greater numbers in control traps.

The species composition of Culicoides was different at each site during both experiments. During both experiments, the distribution of all four vector species, including C. debilipalpis, C. insignis, C. stellifer and C. venustus were significantly different between all three sites (Table 4), where all species except C. stellifer were found in highest numbers in the control trap at the white-tailed deer pen. The distribution of non-vector species C. torreyae was also significantly different between each site, and was primarily found at the low ungulate diversity site.

| TABLE 2 | Abundances of Culicoides collected for various physiological states across experiments 1 and 2, separated by control or sensor traps |
| Control | Blood-engorged | Total females | Sensor | Blood-engorged | Total females |
|---------|----------------|----------------|---------|----------------|----------------|
| Experiment 1 (no CO₂) | | | | | |
| C. debilipalpis | 0 | 13 | 7 | 248 |
| C. insignis | 0 | 48 | 41 | 156 |
| C. stellifer | 4 | 336 | 92 | 1,063 |
| C. variipennis | 0 | 2 | 1 | 6 |
| C. venustus | 0 | 32 | 16 | 74 |
| Total Experiment 1 | 4 | 431 | 157 | 1,547 |
| Experiment 2 (CO₂ added to control) | | | | | |
| C. debilipalpis | 2 | 162 | 5 | 284 |
| C. furens | 0 | 0 | 0 | 1 |
| C. insignis | 9 | 512 | 114 | 795 |
| C. mulrennani | 0 | 1 | 0 | 0 |
| C. pallidicornis | 0 | 1 | 0 | 0 |
| C. paraensis | 0 | 5 | 0 | 0 |
| C. pusillus | 0 | 1 | 0 | 0 |
| C. spinosus | 0 | 2 | 0 | 0 |
| C. stellifer | 17 | 2,312 | 336 | 4,940 |
| C. torreyae | 1 | 95 | 0 | 14 |
| C. variipennis | 0 | 3 | 7 | 57 |
| C. venustus | 1 | 219 | 40 | 299 |
| Total Experiment 2 | 30 | 3,313 | 502 | 6,390 |
| Total both experiments | 34 | 3,744 | 659 | 7,937 |
control traps did not become asymptotic at the high or medium ungulate diversity sites (Figure 4). Curves level-off as they reach the maximum number of species expected to be sampled by each trap, providing an estimation for trap efficiency in comparing vector richness between traps. The motion sensor traps captured five species at the high and medium ungulate diversity sites, and six at the low ungulate diversity site. Despite the control traps requiring more sampling to reach asymptote, the rarefaction/extrapolation curves demonstrate that control traps with CO₂ drew in and captured a greater diversity of Culicoides species.

### DISCUSSION

Using a novel trapping system with a PIR motion sensor to collect Culicoides species in the presence of ungulate hosts, we demonstrated that vector species are collected in much higher numbers from the vicinity of the host animals. In contrast, disproportionately high numbers of non-vector species were collected from light traps that sample the

| Table 3 | Results of negative binomial regression of total female abundances of Culicoides species separately as well as total female abundances captured during both experiments 1 and 2. p-values signify significant differences in total female abundance between motion sensor and control traps.

| Species          | SE  | z-value | p-value | SE  | z-value | p-value |
|------------------|-----|---------|---------|-----|---------|---------|
| C. insignis      | 0.557 | 2.116   | 0.034   | 0.564 | 0.780   | 0.435   |
| C. stellifer     | 0.536 | 2.148   | 0.032   | 0.396 | 1.917   | 0.055   |
| C. torreyae      | —    | —       | —       | 0.738 | −2.593  | 0.010   |
| C. venustus      | 0.765 | 1.096   | 0.273   | 0.480 | 0.649   | 0.517   |
| Total abundance  | 0.441 | 2.833   | 0.005   | 0.341 | 2.155   | 0.031   |

### Table 4

Chi-squared test of independence between five Culicoides species by site for experiments 1 and 2.

| Species          | Experiment 1 | Experiment 2 |
|------------------|--------------|--------------|
|                  | $\chi^2$     | p-value      | $\chi^2$ | p-value |
| C. debilipalpis  | 34.729       | <0.001       | 62.671   | <0.001  |
| C. insignis      | 28.396       | <0.001       | 11.8     | 0.003   |
| C. stellifer     | 133.909      | <0.001       | 44.117   | <0.001  |
| C. torreyae      | —            | —            | 11.046   | 0.004   |
| C. venustus      | 8.147        | 0.017        | 21.288   | <0.001  |

### Figure 3

Biting midges trapped in relation to trap type and available hosts. Motion sensor traps sampled midges from approaching animals. Control traps used only light (Experiment 1) or light and carbon dioxide (Experiment 2). Ungulate observations determined from motion activated wildlife cameras.
general environment. We found that motion sensor traps captured fewer species \((n = 7)\) than control traps \((n = 11)\) and also those species in the motion sensor traps primarily consisted of putative vector species *C. venustus*, *C. stellifer*, *C. insignis* and *C. debilipalpis* (McGregor et al., 2019).

The finding that motion sensor traps collected greater numbers of females of vector species, as well as a higher number of blood-engorged females indicates that motion sensor traps preferentially collect host-seeking *Culicoides*. This corroborates findings by Viennet et al. (2011) which showed that host-based traps, including vacuum aspirations, sticky-cover traps and drop-traps, collected between six and 10 *Culicoides* species over the duration of the study. In contrast, light traps caught 15 different *Culicoides* species, six of which were completely absent in animal-based collection methods, and not implicated as orbivirus vectors in Europe. Similarly, Carpenter et al. (2008) found that fewer *Culicoides* species were captured using drop-net traps than with light traps, but that most of these were probable vectors. Together, these studies indicate that light trap captures are more representative of the actual *Culicoides* community, but are not necessarily an accurate representation of the mammalian host-seeking segment of the population.

Our finding that the putatively autogenous species, *C. torreyae*, (Blanton & Wirth, 1979) was the only species collected in greater numbers in the control trap supports the assertion that primarily vector species congregate around host animals. In our study, *C. torreyae* was almost exclusively captured by control traps with CO₂. The difference in the patterns of capture between *C. torreyae* in control traps, and known mammal feeding *Culicoides* species, found primarily in motion sensor traps, is unambiguous. This provides evidence that conventional light traps are relatively indiscriminate in which species they capture, and may even over-represent non-vector species if not strategically placed near target host animals.

Observing differences in the ungulate host compositions and comparing collections of vector species between sites can help to further elucidate host-use by *Culicoides* species to confirm vector/host associations. Motion sensor traps collected significantly greater number of females than control traps, regardless of the vertebrate composition (Figure 3). They also provide some evidence that non-native ungulate hosts could alter the transmission cycles of native orbiviruses. For example, elk and fallow deer are both highly competent hosts of epizootic hemorrhagic disease virus (EHDV) (Gibbs & Lawman, 1977; Hoff & Trainer, 1973) and are preferentially fed upon by *C. stellifer* (McGregor et al., 2018). Our study shows that the number of *C. stellifer* captured in motion sensor traps increased slightly at sites where elk and sika dear were more common (Figure 3). Therefore, if *C. stellifer* is a competent vector of EHDV, the presence of non-native ungulates could increase transmission.

The most notable disparity between motion sensor and control traps was observed with *C. variipennis*, a species rarely collected during previous studies at this location (McGregor et al., 2018, 2019). Although *C. variipennis* is not currently considered a major vector species in most parts of the United States, its close phylogenetic relationship with *C. sonorensis*, which is a highly competent vector of Orbiviruses, raises concern that it could be contributing to the transmission cycle of orbiviruses in Florida (Holbrook et al., 2000). The collection of this species by our motion sensor traps led us to conclude that some species such as *C. variipennis*, may appear to be rare when using UV-light or UV-light and CO₂ baited CDC light traps, however this species may effectively avoid traps by congregating around host animals. This possibility makes host-baited traps more important for surveillance of vector species and virus-infected individuals.

This study is a first attempt to solve the problem of collecting putative vector arthropods from non-tame animals by using motion
sensors attached to UV-CDC light traps. We show that automatic animal-baited traps can be developed to improve efficiency of trapping. In this study, traps focused on collecting Culicoides species by using UV/LED arrays, a wavelength of light most useful in capturing Culicoides species (Sloyer et al., 2019). Traps could easily be modified for capturing mosquitoes simply by changing the bulb out for an incandescent one and collecting into a bag rather than an ethanol-filled tube. Traps could be improved by modifications to materials, taking animal behaviour into consideration, and allowing for a wider variety of fauna from varying systems to be sampled. For example, pressure plates could be used in place of motion sensors, requiring animals to walk closer to the collection device than was possible in this study. Either pressure plates, or motion sensors used here, could be particularly useful for collecting mosquitoes from hosts in systems such as West Nile virus, or Eastern equine encephalitis virus, for which birds are major amplification hosts. Importantly, further modification to trap design could be made to ensure animals are not able to interfere with the trap, such as by placing a cage around the important components of the trap. In this system, traps could be improved by placing them lower such that they collect more readily from the belly and legs, where high Culicoides activity has been documented previously (Nielsen, 1971). Finally, although animal encounters activated the trap 94% of the time, there were a few instances where the traps were not activated even though the game cameras were recording animals feeding. This occurred primarily because the sensors did not have a 360° range. This problem is avoided with the addition of another sensor to cover a full 360°. Future studies to test the efficacy of this trap for collecting vector species could focus on direct comparison of this trap to other methods of collection from host animals, including hand-aspirators, drop-traps and sticky-cover traps.

The finding that our novel motion sensor traps were successful in capturing a focused group of vector Culicoides species, while also excluding known non-vector species, is an important advancement for surveillance and vector ecology. Future traps based off our trap design which utilize the automatic aspiration from host animals have the potential to save vector ecologists time and resources. In the same sense, this study broadens our understanding of the importance of host availability in the strategic placement of commercially available traps.

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AUTHORS’ CONTRIBUTIONS
K.E.S. and N.D.B.-C. conceived the ideas and designed methodology for the study; K.E.S. developed and wrote the code for the novel trap, collected and analysed data and led the writing of the manuscript; N.D.B.-C. acquired funding, provided supervision and revised the manuscript. Both authors contributed substantially to the drafts and gave final approval for publication.

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DATA AVAILABILITY STATEMENT
All data and motion sensor codes are available on Dryad Digital Repository https://doi.org/10.5061/dryad.ncjxskssf (Sloyer & Burkett-Cadena, 2020).

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