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# Ticks and Tick-borne Diseases

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# TickBot: A novel robotic device for controlling tick populations in the natural environment

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### ABSTRACT

A semi-autonomous 4-wheeled robot (TickBot) was fitted with a denim cloth treated with an acaricide (permethrin $^{\text{TM}}$ ) and tested for its ability to control ticks in a tick-infested natural environment in Portsmouth, Virginia. The robot's sensors detect a magnetic field signal from a guide wire encased in 80 m polyethylene tubing, enabling the robot to follow the trails, open areas and other terrain where the tubing was located. To attract ticks to the treated area, CO<sub>2</sub> was distributed through the same tubing, fitted with evenly spaced pores and flow control valves, which permitted uniform CO<sub>2</sub> distribution. Tests were done to determine the optimum frequency for TickBot to traverse the wire-guided treatment site as well as the duration of operation that could be accomplished on a single battery charge. Prior to treatment, dragging was done to determine the natural abundance of ticks in the test site. Controls were done without CO2 and without permethrin. TickBot proved highly effective in reducing the overall tick densities to nearly zero with the treatment that included both carbon dioxide pretreatment and the permethrin treated cloth. Following a 60 min traverse of the treatment areas, adult tick numbers, almost entirely Amblyomma americanum, was reduced to zero within 1 h and remained at or near zero for 24 h. Treatments without CO also showed reduction of ticks to near zero within 1 h, but the populations were no different than the control sections at 4 h. This study demonstrates the efficacy of TickBot as a tick control device to significantly reduce the risk of tick bites and disease transmission to humans and companion animals visiting a previously tick-infested natural environment. Continued deployment of TickBot for additional days or weeks can assure a relatively tick-safe environment for enjoyment by the public.

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# Introduction

Ticks transmit a greater variety of pathogenic, disease-causing agents than any other blood-sucking arthropod (Jongejan and Uilenberg, 2004). The economic burden to the livestock industry was estimated at more than \$7 billion (McCosker, 1979). The global impact of tick-borne diseases is also enormous, although more difficult to measure (Ginsberg and Faulde, 2008; Jongeian and Uilenberg, 2004). In the United States, Lyme disease is now the most frequently reported vector-borne disease and continues to grow in incidence. Other tick-borne diseases, e.g., Rocky Mountain

http://dx.doi.org/10.1016/j.ttbdis.2014.11.004 1877-959X/© 2014 Published by Elsevier GmbH. spotted fever, human granulocytic anaplasmosis, human ehrlichiosis and babesiosis are also prevalent and present a major public health threat, especially during the spring and summer seasons (Anonymous, 2013; Wormser et al., 2006).

Despite more than a century of efforts, control of ticks and tick-borne diseases remains a daunting challenge throughout the world (Graybill, 1909; Ellenberger and Chapin, 1919; Buxton, 1945; Whitnall and Bradford, 1947). Moreover, the public has become increasingly resistant to large scale release of pesticides, insect growth regulators and other chemicals because of the risks of damage to the natural environment. Clearly, there is a compelling need for the development of novel methods that can mitigate the risk of tick bites and disease transmission to humans and animals so that people can safely enjoy the natural environment.

To address this need, several novel techniques have been introduced in recent years. Among the most popular are the

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host-targeted techniques, wherein small amounts of acaricide are delivered to specific wildlife hosts, e.g., white-tailed deer (*Odocoileus virginianus*), the predominant hosts for lone star ticks (*Amblyomma americanum*) and black-legged ticks (*Ixodes scapularis*). Examples include the "4-poster deer treatment bait station", using medicated foods to attract and treat the animals with sufficient acaricide to kill attached ticks (Schulze et al., 2009; Grear et al., 2014) or the fipronil-impregnated bait box to treat white-footed mice and other small mammal hosts of immature ticks (Dolan et al., 2004).

Although highly effective in the long term, none of these methods achieve immediate relief of the risk of tick attack, even if only for brief periods of hours or days. Consequently, as this is often an important goal, some other method is needed.

To meet the need for immediate relief, we invented and tested a semi-autonomous robotic device (TickBot) that can sweep the vegetation of host-seeking ticks in tick-infested habitats and kill them before they can attack people and/or their pet animals. Following a guide wire and assisted by dispersal of CO<sub>2</sub> along its predetermined pathways, TickBot created a virtually tick-free environment within as little as 1 h following its deployment.

Here we report the efficacy of TickBot for achieving rapid and efficient elimination of ticks, mostly adult *A. americanum*, in a series of experimental trials done in natural habitats in a city park in Portsmouth, Virginia. We suggest that this novel technology may offer a new method for pest-control operators and similar pest management personnel to respond to requests for immediate and efficient reduction of the risk of tick-borne diseases in highly tick-infested environments.

### Materials and methods

Robotics and operation

The robot was a modified remote control car with an attached drag-cloth that followed a wire similar to that used for invisible dog fence (Gehring et al., 2011). The robot's forward velocity is set at a slower-than-walking-speed of 0.3 m/s to maximize the chances that questing ticks will adhere to the drag-cloth. Technical details about the robotics can be found in the on-line technical appendix.

The tick-clearing procedure began with placement of the CO<sub>2</sub>dispensing tube/navigation wire combination around the ecotone of the treated area, whereupon the navigation wire was connected to the base station to provide the navigation signal. The CO master regulator was turned on for 15 min followed by a 15-min diffusion period to attract ticks to the area prior to starting the robot. At that time the drag-cloth was attached to the robot, and the robot was placed over the navigation wire and activated (Fig. 1). The robot was allowed to traverse the wire for 1 h, typically completing the loop 12 times, after which it was turned off. The efficacy experiments were completed along a narrow loop for 1 h; at this same traversal rate, a 0.1 hectare (0.25 acre) circular plot could be treated in slightly less than 2 h. The robot runs for 2–3 h per battery pack depending on the terrain, and new batteries can be swapped in as needed. The base station was operated using the same removable and rechargeable type of battery as the robot, and works for approximately 20 h on a single charge. Setup and teardown time for the system as tested with 73 m of tubing was 30 min each.

The system is designed to be rugged, and no mechanical breakdowns occurred during test operation. The independently articulated suspension and four-wheel drive with a locked differential enabled it to handle terrain with obstacles up to 20 cm high. While the system is not waterproof, the electronics are housed in a rubber-sealed housing with a waterproof power switch and all motors can withstand submersion, making it water-resistant.



**Fig. 1.** Photograph of semi-autonomous robot (Tickbot 2) for killing ticks in vegetation. The robot navigates by following a magnetic signal transmitted along a guide wire placed along trails. The device drags a white denim cloth impregnated with an acaricide (see text for additional details).

The system performed normally during rain and through mud. The battery is designed to be field swapped for continuous operation, but should a malfunction occur, the drive, steering motors, and the entire electronics housing are also capable of being field-swapped.

Design of experimental field trials

Experiments were conducted biweekly from May 21, 2013, through July 3, 2013. For each experimental week, ticks were sampled the day prior to the TickBot deployment by flagging. Flag tick collections were done using a 1 m<sup>2</sup> white denim squares attached to dowel rods which were swept through low vegetation and along the ground to attract questing ticks (Ginsberg and Ewing, 1989). Flags were inspected for ticks every few meters. All adult ticks captured were painted using toothpicks and fingernail polish of various colors. The colors were unique to the experimental plot and the week. The painted ticks were then returned to the areas where they were collected (Fig. 2). The guide/tubing system which guides the Tick-Bot was shortened from the 73 m which was supplied to 61 m to match the three selected transects. The exact path is shown in the specific description of each transect. Two drag-cloths were used for the TickBot and were washed with laundry soap and bleach before initial use and after each field use. One drag-cloth was treated with permethrin as described above, while the second was not treated and was a sham used as described below to demonstrate the impact of the robot itself along with the carbon dioxide. The sham dragcloth was a solid piece of denim cloth with weights sewn into the material while the treated drag-cloth was divided into strips with weights also in each strip (see Fig. 1).



**Fig. 2.** Photographs showing marking of ticks. (A) Ticks captured on set-up days were painted with fingernail polish and returned to the original collection sites. Inset shows enlargement of a male *A. americanum*. (B) Different color paints were used for each of the 4 different trials.

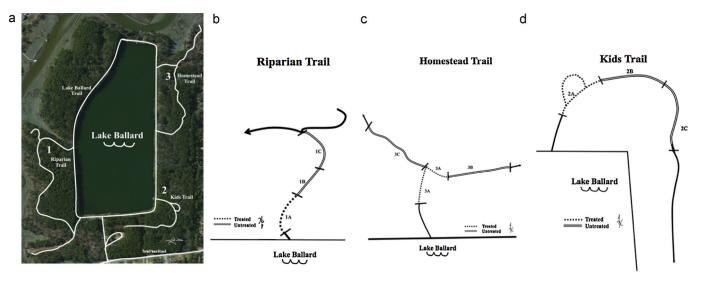


Fig. 3. (a) Map of Hoffler Creek Wildlife Preserve. Experimental plots were established for this project on the Riparian Trail the Homestead Trail and the Kids Trail. (b) Riparian Trail experimental plot. Segment 1A is the treated area, and segments 1B and 1C are the non-treated comparison areas. (c) Homestead Trail experimental plot. Segment 3A is the treated area, and segments 3B and 3C are the non-treated comparison areas. (d) Kids Trail experimental plot. Segment 2A is the treated area, and segments 2B and 2C are the non-treated comparison areas.

The experiments were done along woodland trails, designated experimental plots 1 (Riparian Trail), 2 (Homestead Trail) and 3 (Kids Trail) (Fig. 3a–d). The Lake Ballard trail was used as a reference point (see map, Fig. 3a), and each plot had three sections as noted below. Only one of the sections was treated while the others were untreated references for expected tick densities on that day. Treatments in these plots were done as follows:

# Experimental plot # 1(Riparian Trail)

Three segments (1A, 1B, 1C; see Fig. 3b) were sampled starting approximately 10 m from the Lake Ballard Trail. Segment 1A was fitted with the guide wire set at the trail's edge on each side, constituting a treated area 45 m long. Samples were also taken beyond the treated area in 1B and 1C, two further untreated segments 35 m and 50 m beyond the first one. These untreated segments, 1B and C, served as untreated references for the treated section to provide an expected number of ticks on that particular day along that trail. TickBot runs along 1A were done with permethrin-treated cloth but without CO<sub>2</sub>. The vibrations from the movement of the robot over the terrain did act as a tick attractant to the test areas as it mimics host movement. Set-up of guide wire track in preparation for runs required 15 min to lay-out and stake-down wire. For the first three experiments, the TickBot ran for 60 min while the fourth experiment shortened the run time to 30 min.

# Experimental plot #2 (Homestead Trail)

This trail was divided into three segments: 2A, 2B and 2C (see Fig. 3c). The TickBot was run along the trail in segment 2A, and segments 2B and 2C were not treated and served as untreated references. This site was used to test the effect of  $CO_2$  release, which was used along this trail in all four biweekly experiments. In the first two experiments, TickBot was run for 60 min dragging an untreated cloth, and the results demonstrate the increased tick density from the attraction to  $CO_2$ . Set-up of guide wire track and  $CO_2$  tubing in preparation for runs at this site required approximately 30 min. In the third and fourth experiments, the TickBot was run with permethrin-treated cloth for 60 and 30 min, respectively.

# Experimental plot #3 (Kids Trail)

This trail was divided into three segments: Fig. 3d, A, B and C (see Fig. 3d). TickBot runs were conducted along segment 3A

following dispersal of CO<sub>2</sub> and used a permethrin-treated cloth. For the first three experiments, the TickBot was run for 60 min, and a 4th experiment was done for 30 min. Set-up of guide wire track and CO<sub>2</sub> tubing in preparation for runs at this site required approximately 30 min. Segments 3B and 3C served as an untreated reference for the expected ticks along that trail.

### Ethics statement

This study was conducted at Hoffler Creek Wildlife Preserve with permission granted by the Hoffler Creek Wildlife Foundation through their executive director, Helen Kuhns. Additionally, in accordance with the regulations in the Commonwealth of Virginia, the collection of ticks was granted through Virginia Department of Game and Inland Fisheries Scientific Collection Permit #047118.

# Results

TickBot operated continuously in the treated segments of the three trails for multiple runs without difficulty, successfully overcoming minor obstructions, making turns and generally following the signal from the guide wire throughout its predefined course. The batteries, when fully charged, were able to power the robot for at least 2 h. An intermittent open in the charger's adapter plug occasionally interfered with battery charging. A single battery pack was sufficient to power the wire for the 3 h used for each experiment. Some difficulties were experienced because of the TickBot's asymmetric turning radii. When there were tight left turns, the device sometimes lost the signal, accelerated and wandered off course, so that the operator had to stop it and return it to the wire. Restricting the robot to a clockwise pattern resolved this difficulty in most cases.

# Tests with permethrin and CO<sub>2</sub>

Four 1-h TickBot tests were completed with both  $CO_2$  release and permethrin cloth. One hour following these tests, flagging along the treated trails showed almost no ticks; only one adult tick was captured in one test, while the remaining three tests had zero adult ticks. Repeat flagging at 4 h post treatment found no ticks in the treated plots in all tests. The untreated reference sections were also

**Table 1**Differences in adult tick density for treated and untreated areas.<sup>a</sup>

Event	Treatment	Treated (average, ticks/m²)	Untreated (average, ticks/m²)	Paired <i>t</i> -test <i>p</i> value	Number of samples
Set-up	None	0.0766 <sup>b</sup>	0.0722 <sup>b</sup>	0.41	12
1-h after treatment	CO <sub>2</sub> and permethrin	0.0104	0.0834	0.03°	4
1-h after treatment	Permethrin only	0.0078	0.0421 <sup>b</sup>	0.02*	3
4-h after treatment	CO <sub>2</sub> and permethrin	0.0000	0.0797 <sup>b</sup>	0.04*	4
4-h after treatment	Permethrin only	0.0233	0.0307 <sup>b</sup>	0.27	3
Next day noon	CO <sub>2</sub> and permethrin	0.0457 <sup>b</sup>	0.0794 <sup>b</sup>	0.04*	3
Next day noon	Permethrin only	0.0233	0.0057 <sup>b</sup>	0.25	2
Next day afternoon	CO <sub>2</sub> and permethrin	0.0342	0.0547 <sup>b</sup>	0.30	3
Next day afternoon	Permethrin only	0.0465	0.0115 <sup>b</sup>	0.10	2

<sup>&</sup>lt;sup>a</sup> Tick densities were sampled in both areas the day prior to each treatment, 1-h after treatment, 4-h after treatment as well as twice during the following day. Ticks collected were marked and re-released at the site of collection. Treated and untreated reference sections are compared for each sampling event. Only 60-min robot runs were included in this analysis. The results show that there is no difference between the sections prior to treatment, but there is a statistically significant difference after treatment through the next day for the treatment with CO<sub>2</sub> and permethrin cloth.

sampled 1 and 4 h after the treatment period, and adult ticks were found along these plots in three of the four tests. No ticks were collected 4 h after treatment in one test likely because of a localized rain event at 3 h post treatment. By the following morning, flagging showed no adult ticks in the treated areas in three tests with a small number of ticks (approx. 0.03 ticks/m) in the fourth. However, by the following afternoon, sampling showed the return of adult ticks, but none of the ticks found were painted, which indicates they were not likely active during the TickBot run. Two additional tests were conducted using a 30-min run of the TickBot, and while the 1-h results showed similar results to the full hour run, a significant rain event hampered follow up testing (see Figs. 4 and 5; Table 1).

## Tests with permethrin only

TickBot runs were done with permethrin-treated cloth but without  $CO_2$  for three tests. The results of these tests showed a significant 80% reduction in ticks/min after 1 h, but after 4 h, there was no statistical difference between the treated and untreated sections of the trail. A single 30-min test was also conducted but was also hampered by rain (see Table 1).

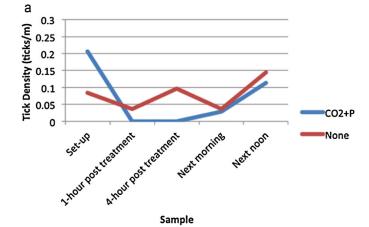
# Tests with CO2 only

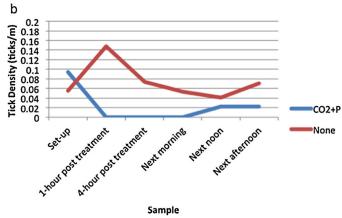
Two test runs were done with  $CO_2$  alone to demonstrate the increases in tick occurrence. TickBot was run for an hour dragging an untreated cloth. Both the treated plots and the surrounding sections had increased numbers of ticks at 1 h post treatment and a return to approximately pre-treatment levels at 4 h post treatment (see Table 1).

Additional evidence of the ability of TickBot to effectively kill the ticks in a given area was the mark-recapture data. A total of 466 adult ticks were painted and released over the course of the entire pilot study. Of that total, no marked ticks were recaptured in the treated areas at the 1-h or 4-h mark. A total of 4 marked ticks were recaptured in the sampling on the day after treatment, but these ticks could have moved into the area from the surrounding untreated reference sections.

## Effects of weather

Rain interfered with our ability to carry out TickBot treatments on many days, and we were unable to obtain some sampling events, as ticks are not likely to be collected in the rain. The rain during the final trial with the 30-min robot runs prevented the collection of meaningful results for that experiment.





**Fig. 4.** Results from combined carbon dioxide and permethrin treated cloth treatment with 60-min robot runs. The graph on the top (A) shows the tick density on Homestead Trail in Week 3, while the graph on the bottom (B) shows the tick density on Kids Trail in Week 2. Both show a drop to zero ticks in the treated areas (blue lines) for the remainder of the treatment day while the untreated areas (red lines) show no such reduction. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

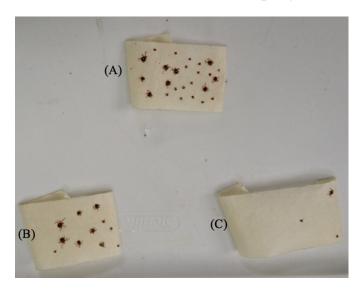
# Statistical analysis

Simple paired *t*-tests comparing the treated and untreated sections for all of the trails demonstrated that the sections were statistically identical each day prior to the experiments (Table 1, set-up). The post-treatment flagging demonstrated a statistically significant reduction in tick density for both permethrin and permethrin plus CO<sub>2</sub> 1 h after treatment. The permethrin plus CO<sub>2</sub>

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<sup>&</sup>lt;sup>b</sup> Indicates recapture of marked ticks.

p value < 0.05.



**Fig. 5.** Ticks collected at 1-h after treatment in Week 3 on Kids Trail. The lower right hand piece of tape (C) is the ticks collected in the treated area (segment 3A) while the lower left (B) is the untreated area closest to the treated area (segment 3B). The ticks on the single piece of tape in the top row (A) were collected in the untreated segment furthest from the treatment area (segment 3C).

treatment continued to show a statistically significant reduction through sampling at noon the following day. Flagging before noon was limited by rain events (Table 1).

### Discussion

The tick control robot, TickBot, proved to be highly efficient in controlling tick populations of A. americanum in the natural environment chosen for the experimental investigations. Ticks were eliminated along the trails treated by TickBot for an entire day; few ticks reappeared the next morning and did not increase to near pre-treatment levels until the following afternoon. Evidence that the ticks eradicated by TickBot were the original, resident individuals was indicated by the virtual absence of marked ticks released before treatment (<0.01%). Although mark-recapture preliminary data focused on adult life stages of A. americanum, a decrease in the abundance of nymphs also was apparent. Further trials will be needed to assess the effectiveness on all A. mericanum life stages as well as other species. Treatments proved most effective when permethrin-treated drag-cloths were used in combination with dispersal of CO<sub>2</sub> in the treatment sites. Based upon these results, we anticipate that Tickbot could be used to control ticks in the environments around private homes (including the lawns, wooded abutments), schoolyards, local parks, woodland trails, campgrounds and military installations.

TickBot provides a method for reducing the risk of tick bite/disease transmission in targeted treatment areas without release of toxic chemicals in the natural environment; all of the pesticide (permethrin) remains on the treated drag-cloth. As noted in the Introduction, other strategies have been proposed to address the need for tick control with little or no environmental damage. Among the most popular is "host-targeted" tick control, using methods in which wild hosts self-treat with acaricides. To treat white-tailed deer, the U.S. Department of Agriculture used acaricide-treated 4-Poster Deer Treatment Bait Stations in five eastern states to control ticks, *I. scapularis* and *A. americanum*, feeding on white-tailed deer and reduce disease transmission risk. However, the 4-Poster technology requires 1 or more years to show efficacy (Pound et al., 2009). Rodent-targeted bait boxes have been used to

kill immature ticks on small mammal hosts before they can mature into human-biting adults (e.g., Dolan et al., 2004; Stafford, 1992).

Allan and Sonenshine (2002) impregnated tick arrestment pheromone and a pesticide into an oily substance which, when sprayed, adhered to vegetation. Ticks were induced to cluster on and around the droplets, imbibed the toxicant and died at a significantly higher percentage than just the toxicant alone. All of these methods, although highly effective, require months or even years to achieve maximum reductions in tick populations and the attendant reduction in disease transmission risk. In contrast, TickBot makes it possible to treat a limited area and reduce the risk of tick bite and disease transmission within hours and, if continued, can extend the safe zones for many days or even weeks. Future research will specifically investigate the relationship between frequency of TickBot use and the reduction of ticks.

An additional consideration for control is the cost of the method relative to the costs of the disease. The worldwide economic loss due to tick-borne diseases, tick infestations along with the costs of vaccination and acaricide treatment, is estimated to be in the billions of dollars annually (Jongejan and Uilenberg, 2004). Treatment of human Lyme disease averages \$281 (Zhang et al., 2006). Tick infestations affect the recreational value of parks, campgrounds, and other natural areas or the willingness of home owners to use their own yards and gardens during the seasons when ticks are active. A typical acaricide treatment for a home environment was reported to cost \$66 every two months, totaling \$396/year (Gaff, Pers. Comm.). There is strong public resistance to widespread dissemination of acaricides in the natural environment and, in many areas, local and/or regional ordinances prevent such use. This treatment leaves a residual of acaricide in the open areas around the home and degrades gradually due to rain or snow. In contrast, the tickbot treatment will required only 2 or 3 brief visits ( $\sim$ 2 h/visit) and will be limited to the tick seasonal activity period (usually about 4 months) and does not leave any residual toxicants in the home

TickBot is also a mimic of the standard dragging method for collecting ticks, and thus could be another potential application for the robot. With an untreated cloth, the TickBot would collect ticks from a given area. This potential application was seen in the experiments with the sham cloth as adult *A. americanum* were seen clinging onto the material.

Difficulties experienced with operation of the robot indicate the need for additional engineering solutions to enhance performance. Especially important is the need to improve TickBot's ability to make tight symmetric turns without running beyond the range of the signal emitted by guide wire. One of the advantages of the wireguided operation is that the range of TickBot is limited only by the distance over which the wire is deployed, potentially over hundreds of meters. Substitution of a wireless transmitter and receiver system instead of the guide wire might also be considered for treating small areas, e.g., a homeowner's yard or a school playground. This substitution would also be beneficial for the convenience of Tickbot's use. Currently a limiting factor for public use is the time and labor included with placement of the guide wire and the polyethylene tubing for the CO<sub>2</sub> attractant. Additionally, the wireless system would allow for the TickBot system to be deployed over a larger area with the use of multiple robots simultaneously.

### **Conclusions**

Overall, TickBot proved highly effective in reducing observable tick densities to nearly zero with the treatment that included both carbon dioxide pretreatment and the permethrin treated cloth. Following a 60-min traverse of the selected trails, adult tick numbers (ticks captured/m), almost entirely A. americanum, were reduced to

zero within 1 h and remained at or near zero for 24 h. Reductions in questing tick numbers to zero after 1 h and continuing at 4-h and 24-h post treatment achieved the goal of protecting habitats for the enjoyment of people visiting these localities. It is likely that the wooded areas adjacent to the treated transects harbor a very large reservoir of ticks that repopulated these localities after 24 h.

Additional studies are needed to test repopulation half-times in a suburban context in which ticks predominantly reside in the relatively narrow ecotone of bushes and shrubs separating property boundaries. Such tests would be useful to determine whether Tick-Bot treatments would last longer than in wooded habitats such as the treatments done at Hoffler Creek. Finally, further studies are needed to test the utility of TickBot in other regions of the United States and to test its efficacy against other vector ticks, especially the Lyme disease vector, *I. scapularis*, and the American dog tick, *Dermacentor variabilis*.

### **Uncited reference**

Stafford et al. (2009).

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### Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.ttbdis.2014.11.004

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