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Quantifying Corticolous Arthropods Using Sticky Traps

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Dear Editor

JoVe

We are excited to present our manuscript *A method of quantifying corticolous arthropods* for review to *JoVe.* Terrestrial arthropods play an important role in influencing numerous aspects of our environment. Understanding the factors that influence arthropods communities is important form both an economic and environmental standpoint. Studying arthropod communities at a landscape scale requires cost efficient methods of quantifying arthropods with adequate precision to detect variation across spatial, temporal, or environmental characteristics. With this paper, we describe a cost efficient method of quantifying corticolous arthropods with adequate precision proven to detect variation across an environmental gradient.

Thank you for the consideration,

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TITLE:
Quantifying Corticolous Arthropods Using Sticky Traps

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KEYWORDS:
arthropod abundance, arthropod density, bark, capture, community, corticolous, diversity, population, richness, sticky trap, trophic interactions

SUMMARY:
We describe a semi-quantitative approach of measuring characteristics of corticolous (bark-dwelling) arthropod communities. We placed commercially manufactured sticky traps on tree boles to estimate abundance, total length (a surrogate to biomass), richness, and Shannon diversity for comparison among tree species.

ABSTRACT:
Terrestrial arthropods play an important role in our environment. Quantifying arthropods in a way that allows for a precise index or estimate of density requires a method with high detection probability and a consistent sampling area. We used manufactured sticky traps to compare abundance, total length (a surrogate for biomass), richness, and Shannon diversity of corticolous arthropods among the boles of 5 tree species. Efficacy of this method was adequate to detect variation in corticolous arthropods among tree species and provide a standard error of the mean that was <20% of the mean for all estimates with sample sizes from 7 to 15 individual trees of each species. Our results indicate, even with these moderate sample sizes, the level of precision of arthropod community metrics produced with this approach is adequate to address most ecological questions regarding temporal and spatial variation in corticolous arthropods. Results from this method differ from other quantitative approaches such as chemical knockdown, visual inspection, and funnel traps in that they provide an indication of corticolous arthropod activity over a relatively long-term, better including temporary bole residents, flying arthropods that temporarily land on the tree bole and crawling arthropods that use the tree bole as a travel route

from the ground to higher forest foliage. Furthermore, we believe that commercially manufactured sticky traps provide more precise estimates and are logistically simpler than the previously described method of directly applying a sticky material to tree bark or applying a sticky material to tape or other type of backing and applying that to the tree bark.

INTRODUCTION:

Terrestrial arthropods play an important role in our environment. In addition to being of scientific interest in their own right, arthropods can be both detrimental and beneficial to other trophic levels (i.e., crops, horticultural plants, native vegetation, and food for insectivorous organisms[1-4]). Thus, understanding the factors that influence arthropod community development and abundance is critical to farmers[5], pest control managers[6], foresters[4], plant biologists[7], entomologists[8], and wildlife and conservation ecologists that study community dynamics and manage insectivorous organisms[9]. Arthropod communities vary in species composition and abundance both temporally and spatially across a variety of ecological landscapes including plant communities, plant species, and across various regions of individual plants. For example, studies have demonstrated significant differences in arthropod community metrics between the roots, bole and stems, and foliage, within the same individual tree[10,11]. These findings are not surprising considering that different parts of the same plant, e.g., leaves versus barks of a tree, provide different resources for which arthropods have adapted to exploit. Thus, each part of the plant can support a different arthropod community. Because foliage dwelling arthropods can have such a large socioeconomic and environmental impact, substantial effort has been expended to measure community metrics using both qualitative and quantitative approaches[12]. Alternatively, much less effort has been expended to develop approaches of quantifying corticolous (bark-dwelling) arthropod communities.

Like foliage-dwelling arthropod communities, corticolous arthropod communities can be important from both a socioeconomic and environmental viewpoint. Some forest diseases that are caused or facilitated by corticolous arthropods can be detrimental to economically viable timber harvest[4]. Additionally, corticolous arthropods can be an important component of the food chain in forest communities[13,14]. For example, forest dwelling arthropods are the primary food source for many insectivorous bark gleaning song birds[15,16]. Thus, understanding the factors that influence communities of corticolous arthropods is of interest to foresters and both basic and applied ecologists.

Understanding factors that influence arthropod community composition and abundance often requires the capture of individuals. Capture techniques can generally be categorized into qualitative techniques that only detect presence of a species for estimates of species range, richness, and diversity[17], or semi-quantitative and quantitative techniques that allow for an index or estimate of abundance and density of individuals within a taxonomic group[18,19]. Semi-quantitative and quantitative techniques allow researchers to estimate or at least consistently sample a specified sample area and estimate probability of detection or assume detection probability is non-directional and adequate as to not obscure the researcher's ability to detect spatial or temporal variation in abundance. Semi-quantitative and quantitative techniques for quantifying corticolous arthropods include suction or vacuum sampling of a specific area[20-22],

systematic counting of visible arthropods[18,23], sticky traps[24], various funnel or pot-type traps[8,25], and entrance or emergent holes[26,27].

A number of spatial and temporal factors are thought to lead to variation in corticolous arthropod communities[11,14,28,29]. For example, texture of tree bark is thought to influence the community structure of tree-dwelling arthropods[14]. Because of the more diverse surface area of the trunks of trees with more furrowed bark, trees with more furrowed bark are thought to support a greater diversity and abundance of arthropods[14].

With this article we report a new semi-quantitative approach of enumerating corticolous arthropods that could be used to describe and test hypotheses regarding variation in corticolous arthropod communities across time and space with adequate precision to detect differences among tree species. Using sticky traps attached to the trunks of trees, we compared the abundance, total length (a surrogate for body mass), richness, and diversity of the arthropod community on the bole of white oak (*Quercus alba*), pignut hickory (*Carya glabra*), sugar maple (*Acer saccharum*), American beech (*Fagus grandifolia*), and tulip poplar (*Liriodendron tulipifera*) trees, trees that vary in bark texture.

This study was conducted in the Ozark and Shawnee Hills ecological sections of the Shawnee National Forest (SNF) in southwestern Illinois. During July 2015, we identified 18 (9 dominated by oak/hickory and 9 dominated by beech/maple) sites with the USFS stand cover map for the SNF (allveg2008.shp) in ArcGIS 10.1.1. In the xeric sites, the dominant species were pignut hickory and white oak and in mesic sites, the dominant species were American beech, sugar maple, and tulip poplar. To compare bole arthropod community among tree species, at each data collection site, we identified the three of the five (white oak, pignut hickory, sugar maple, American beech and tulip poplar) focal species trees >17 cm diameter at breast height (d.b.h.) closest to the center of a 10 m radial circle. If fewer than three appropriate trees were present, the circle was expanded and the closest tree fitting the criteria was selected. For each tree chosen, we installed four sticky traps at breast height, one facing in each cardinal direction: north, south, east and west.

We collected arthropod data from the boles of 54 individual trees (12 pignut hickories, 15 white oaks, 8 American beeches, 12 sugar maples, and 7 tulip poplars) among the 18 sites. We grouped arthropods according to a simplified guild classification by diagnostic morphological characteristics indicative of closely related orders from current phylogenetic records, similar to that of "operational taxonomic units"[30,31] (**Appendix A**). Based on this classification, we captured representatives of 26 guilds in our traps that were each in place for 9 days (**Appendix A**). Because our study focused on trophic interactions between tree species, corticolous arthropods, and bark-gleaning birds, we removed all arthropods smaller than 3 mm from analysis because their importance as a food resource is minimal for bark-gleaning birds. We used a mixed model that included either arthropod length (surrogate to body mass), abundance, Shannon diversity and, richness as the dependent variable, tree species and effort (proportion of tree covered with traps) as fixed variables, and site as a random variable. Because all traps from a single tree were combined as one sample, individual trees were not included as a random variable.

PROTOCOL:

1. Placement of a trap on the tree

1.1. Measure the diameter of a tree at breast height. At breast height in each cardinal direction, for an area the size of the pre-manufactured sticky trap (glue board), use a bark shaver to remove bark until an area the size for the sticky trap is smooth enough to staple the sticky trap onto the tree so that there is no space for arthropods to crawl under the trap. Label the back of the trap using a dark colored permanent marker with the date, trap number, location and other pertinent information.

1.1.1. To trap arthropods, either (a) capture both flying and crawling arthropods, by opening and removing the sides and cover of the sticky trap by cutting the cardboard along the edge of the sticky material, (b) or exclude flying arthropods from landing directly on the trap, by opening the trap as directed on the box.

1.2. Place one trap on each previously shaved location so that the openings are oriented vertically (one opening facing up, the other opening facing down) to maximize capture of arthropods crawling up and down the tree boles. For traps with the tops removed to capture both flying and crawling arthropods, orient traps so the end that was the opening prior to the removal of the cardboard cover is oriented vertically, to maintain trapping consistency.

1.3. Staple traps to the tree by placing one staple at each corner and one staple in the center bottom and center top of the trap. Start stapling in the bottom right corner, then the bottom center, the top right corner, the top right center, the bottom left corner, and finally the top left corner. Be careful to ensure the entire bottom and top of the traps are flush against the tree to minimize arthropods crawling under the trap.

1.4. Leave traps in place for desired amount of time. Be certain all traps are left in place the same amount of time.

NOTE: In areas where arthropods are extremely abundant, for example during moth outbreaks, traps may become saturated within hour or days. Under these circumstances, traps will need to be regularly replaced prior to be saturated to maintain constant capture probability.

2. Removing the trap from the tree

2.1. After the desired amount of time trapping, cover the entire trap, except for the staples, with polymeric cellulose film (e.g., cellophane).

NOTE: Placing the film on the traps prior to removal will reduce the likelihood of disturbing the trapped arthropods.

2.2. Remove each trap by taking a large flat screwdriver and prying each staple partially from the tree, adequate to facilitate the grasping of the staples using needle nose pliers. Take large needle nose pliers or a similar grasping tool and pull the staples from the tree.

2.3. Place the traps in a rigid box of some type for transportation to a laboratory for analysis. If traps are to be stored for more than 12 h, store traps in a freezer to preserve content.

3. Laboratory analysis

3.1. Using a dissecting scope, examine content of a trap recording the number of individuals to desired taxonomic level.

3.2. Use sorted arthropods to estimate richness (total number of taxonomic groups), diversity indices, or abundance (total arthropods). If estimated biomass is a desired result, measure length and width of arthropods to the nearest mm and use published length/width, biomass regressions to estimate biomass[32,33,34].

3.3. Subtract the total width of the 4 traps from the diameter at breast height for each tree to estimate trapping effort (proportion of tree covered by the traps) for each tree.

3.4. Because samples from multiple traps on the same tree are not independent, either sum samples from the same tree or include individual tree as a random variable in all analysis to avoid pseudo-replication.

REPRESENTATIVE RESULTS:

Based on the mixed model results, the model that included tree species best explained variation in total arthropod length, abundance, and diversity, neither of independent variables explained substantial variation in richness, although the models that included tree species trapping effort were competitive with the null model (**Table 1**). In addition, proportion of the tree trapped appears to have no influence on abundance, total length, and Shannon diversity, with only minimal influence on richness (**Table 1**). The standard error of the mean (SEM) for total arthropod length varied from 4% of the mean in tulip poplar to 17% in sugar maple (**Table 2**). Abundance had similar levels of variation within species where the SEM was 7% of the mean in tulip poplar and 18% in sugar maple (**Table 2**). Conversely, variability in arthropod richness and diversity was much lower within species of tree in that SEM of richness ranged from 4% of the mean for pignut hickory to 9% of the mean in American beech, while diversity ranged from 4% of the mean in American beech to 7% of the mean in tulip poplar.

TABLE LEGENDS:

Table 1: Model results. Results of a mixed model analysis of covariance (ANCOVA) with corticolous arthropod richness, total body length, abundance, or Shannon diversity as the dependent variable, tree species and proportion of tree covered by traps (effort) as the independent fixed variables, and individual site as the independent random variable. K = number

of model parameters, AIC = estimated Akaike's Information Criterion, and ΔAIC = the difference in AIC points form the model to the most parsimonious model.

Table 2: Parameter estimates from the most parsimonious model in Table 1. The mean (X), SEM, and percentage of SEM for each community metric of corticolous arthropods captured on 5 species of trees using commercially manufactured sticky traps in the Shawnee National Forest in southern Illinois.

DISCUSSION:

Although alternative techniques such as suction or sweep nets have been used, most previously published attempts at quantifying arthropods on tree boles used some version of either quantifying arthropods by visually inspecting tree boles in the field, using chemical pesticides to kill arthropods in a specified area then quantifying the recovered arthropods, or placing funnel traps or a sticky substance directly onto the tree[19,23,25,35,36]. Each of these approaches have benefits and shortcomings.

With chemical knockdown, a pesticide is sprayed over a predefined area and arthropods are allowed to drop onto a drop cloth as they die, where they are then collected and quantified[19]. Alternatively, with visual location, live arthropods are located in the predefined area and collected by hand for later quantification[23]. Both of these methods are instantaneous relative to our method, thus provide a more quantifiable estimate of area sampled for use in estimating density. A further attribute to chemical knockdown as well as visual inspection is, because it is somewhat instantaneous, the estimate is limited to the time at which the survey was conducted. Because it only samples arthropods present at the time of sampling, this method provides an accurate estimate of the size of area sampled, facilitating an estimate of density. These approaches, however, often disregard variation in the non-resident arthropod population, arthropods that temporarily inhabit the tree boles such as flying arthropods or arthropods that use the surface of tree boles as travel routes from the ground to higher forest foliage. Because many of the arthropods that influence other trophic levels use bark for short periods as part-time residence, nearly instantaneous samples from the visual observation and chemical knockdown method likely will not adequately depict the entire suit of arthropods that use tree bark as a substrate[8,35,36].

To better depict the corticolous arthropod community that occurs over longer periods, longer-term methods such as funnel and sticky traps have been developed[25-31,35,36]. Funnel traps are attached to tree boles and are designed to funnel arthropods into bottles of preservative, thus are beneficial in that they can be used for long periods of time (weeks to potentially months) while still preserving the arthropods. The limitation of these traps is their limited ability to trap flying arthropods that land on the tree boles. Alternatively, sticky traps are effective at capturing both crawling and flying arthropods.

With the original sticky traps, a sticky material was placed directly onto the tree to trap both crawling and flying arthropods over a predetermined time[37]. While this approach was effective at trapping both crawling and flying arthropods, it is difficult to spread the exact same amount of

material for each trap, thus maintain a consistent sampling area and trapped arthropods have to be identified and quantified in the field under often less than ideal weather conditions, potentially leading to additional variation in the estimates due to misidentification or miscounting. An improvement was offered by Collins et al.[36] when they spread the sticky material on tape, then, after trapping for a predetermined amount of time, covered the tape with cellophane and removed the tape so arthropod identification and quantification could be conducted later in the laboratory, where conditions were much more appropriate for the activity. While this method is an improvement over the previously described methods, it is still messy, and still difficult to consistently spread the same amount of sticky material at each trap. As an improvement to this method, we propose using commercially manufactured sticky traps to address both of these deficiencies.

Commercially produced sticky traps have been used to trap flying arthropods over water[38], at various elevations of vascular vegetation[39], and in the foliage of trees[40], but to our knowledge have not been used to sample arthropods on tree bark. Commercially produced sticky traps provide an improvement over previously used approaches in that the sticky material is adhered to the cardboard backing in the factory and because they are commercially manufactured, the surface area of the material is very consistent. Additionally, the traps can be placed on the trees with the trap intact, preventing flying arthropods from landing directly on the trap, as was done in our study, or the cardboard cover could be removed so the trap is catching both crawling arthropods and flying arthropods landing directly on the trap. Additionally, the traps are easily removed from the tree, covered with cellophane and transported to the laboratory where they can be stored in a freezer and quantified at a later date. The trap's stiff cardboard construction also facilitates viewing the traps in the laboratory under a dissecting microscope allowing for more precise identification, quantification and measurements of arthropods, reducing some of the detection error that would likely occur when conducting this activity in the field. Finally, sticky material on trees can become saturated, reducing the ability of the trap to capture arthropods[41]. The method we describe allows researchers to easily replace the sticky traps to maintain effectiveness, allowing long-term monitoring of individual trees.

As demonstrated by our results, this approach appears to provide adequate precision to address most ecological or environmental questions regarding variation in corticolous arthropod communities. Detection of arthropods from sticky traps used to quantify corticolous arthropods with this method was adequately precise to provide an SEM that was <20% of the mean for all community metrics used in this study. This level of precision was achieved with a reasonable sample sizes of only 7 to 15 individual trees. With this level of precision and moderate sample sizes, we detected differences in total length (a surrogate for biomass), total abundance, total richness, and Shannon diversity among species of trees. We did not partition the variance between measurement error (variance associated with variation in proportion of area trapped among traps or variation in detection probability) and variance among individual trees within a tree species, however, these results clearly indicate that this method has adequate detection probability to prevent measurement error from obscuring results to important ecological or environmental questions.

We describe this method as being semi-quantitative because although we believe our detection probability to be high and provide adequate precision to address most ecological questions, we have no way of estimating detection probability. Thus, we have no way of estimating potential negative bias associated with our point estimates. Additionally, a fully quantitative method that could be used to estimate overall abundance or density, requires an accurate estimate of the sampling area[42]. Unlike the visual inspection or chemical knockdown methods, the sampling area with funnel traps and with this method is uncertain because it is not instantaneous, the traps are placed on the tree for a predetermined amount of time and arthropods going about their normal activities are trapped when they cross the surface of the sticky traps. Thus, the size of area being trapped is dependent on the activity level of the arthropods. Arthropod activity level varies with time of day, by season, by species, or by individual[8]. Because arthropod activity level varies, the sampling area will vary based on activity level. It will be important for researchers to consider how activity level influences inference from results when using both this and the funnel trap method. We argue, however, neither methods that provide a more accurate estimate of the sampling area because they are more instantaneous nor methods that provides a less accurate estimate of sampling area but a better depiction of the arthropod community over time is better. Instead, the two types of methods address different questions. The chemical knockdown and visual inspection methods describe the community during a very specific point in time, while the funnel and sticky trap methods describe the community over a period of hours or days, depending on how long the traps are left in place. We believe, however, when researchers are interested in identifying and describing spatial and temporal variation of corticolous arthropod communities utilizing the bark surface over a substantial time (days to weeks), the method described here is the most convenient and accurate approach.

Finally, the primary objective of our original study was to better understand how mesophication of southeastern deciduous forests is likely to impact forest dwelling insectivorous birds and mammals, thus, we combined arthropods into guilds[43]. We see no reason, however, why these capture techniques could not be used to quantify arthropods at the species or any other taxonomic level.

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DISCLOSURES:

The authors have nothing to disclose.

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Table 2.

Tree species	Richness			Total Length			Shannon Diversity			Abundance		
	X	SE	% of mean	X	SE	% of mean	X	SE	% of mean	X	SE	% of mean
Sugar maple N = 12	8.33	0.59	7%	365.20	63.69	17%	1.59	0.09	6%	45.45	8.15	18%
Pignut Hickory N = 12	7.83	0.30	4%	573.90	81.58	14%	1.24	0.07	6%	70.09	10.10	14%
Tulip Poplar N = 7	8.75	0.49	6%	195.35	7.09	4%	1.73	0.12	7%	25.67	1.87	7%
American Beach N = 8	8.29	0.81	9%	349.91	38.45	11%	1.53	0.06	4%	47.00	5.32	11%
White Oak N = 15	9.07	0.42	4%	407.38	40.16	10%	1.64	0.09	5%	50.57	5.26	10%

Table 1

Dependent variable	Model	K	AIC	ΔAIC
Richness	Null	2	210.56	0
	Tree	7	211.69	1.13
	Effort	3	211.93	1.37
Total body length	Tree	7	719.69	0
	Null	2	727.00	7.31
	Effort	3	728.96	9.27
Abundance	Tree	7	495.55	0
	Null	2	501.04	5.48
	Effort	3	503.04	7.48
Diversity	Tree	7	28.78	0
	Null	2	37.31	8.52
	Effort	3	38.72	9.93

Table 2

Tree species	Richness			Total length			Shannon
	X	SE	% of mean	X	SE	% of mean	X
Sugar maple (N = 12)	8.33	0.59	7%	365.20	63.69	17%	1.59
Pignut Hickory (N = 12)	7.83	0.30	4%	573.90	81.58	14%	1.24
Tulip Poplar (N = 7)	8.75	0.49	6%	195.35	7.09	4%	1.73
American Beach (N = 8)	8.29	0.81	9%	349.91	38.45	11%	1.53
White Oak (N = 15)	9.07	0.42	4%	407.38	40.16	10%	1.64

diversity		Abundance		
SE	% of mean	X	SE	% of mean
0.09	6%	45.45	8.15	18%
0.07	6%	70.09	10.10	14%
0.12	7%	25.67	1.87	7%
0.06	4%	47.00	5.32	11%
0.09	5%	50.57	5.26	10%

Table of Materials

Click here to access/download;Table of Materials;60320_Table_of_Materials.xlsx

Name of Material/Equipment	Company	Catalog Number	Comments/Description
Straight Draw Bark Shaver, 8"	Timber Tuff	TMB-08DS	
PRO SERIES Bulk Mouse & Insect Glue Boards	Catchmaster	#60m	
Staple gun	Stanley	TR45D	

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Author(s): Eichholz, Zari, & Sierzega

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All references to trademarked materials have been removed.

3. Discussion: As we are a methods-based journal, please discuss critical steps in the protocol, modifications and troubleshooting of the method, and limitations of the method.

Done

4. Please upload each Table individually to your Editorial Manager account as an .xlsx file. Avoid any coloring or formatting in the tables.

Done

5. Table of Materials: Please ensure that it has information on all relevant supplies, reagents, equipment and software used, especially those mentioned in the Protocol. Please sort the materials alphabetically by material name.

Done

Reviewers' comments:
Reviewer #1:
Accept

Reviewer #2:
General points
The manuscript describes a modified approach to using sticky traps to estimate community composition of bark-dwelling invertebrates. The method appears to have some merit and is worthy of dissemination. There are some issues with the manuscript, however, that need to be considered. I have listed minor specific points below by line number, but first it is helpful to make some more general observations of a more substantive nature:

a) In several places the manuscript stresses the importance of a high detection probability and known sample area. This seems counter-productive because the manuscript provides no information on detection probability and the sticky trapping method relies on invertebrate activity (and has an unknown effective trap area). Both points are acknowledged in the Discussion. The reader is therefore left wondering why these points are raised and whether the method is useful (given that it fails to address the key requirements highlighted by the authors). The whole manuscript needs editing to focus on the key benefits of the new method and present a more coherent argument.

We agree that we included too much emphasis on known sampling area and have modified the text to decrease that emphasis. One advantage to this method over previously described methods, however, is it provide a more consistent sampling area. Thus we retained some text to recognize the need for consistent sampling areas.

b) The manuscript would benefit from some re-structuring. Specifically, the manuscript includes a case-study, but this is split into two parts. A general description of the study site and tree selection is given (lines 95-110) then the data appears in lines 158-180. The intervening section is the standard protocol and includes some suggestions about alternative placement of the traps which were not utilised in the case-study. It may be better to outline the standard protocol (with options) and then describe the case-study in full (study site, tree selection, trap placement, and analysis) clearly stating the specific options selected from the standard protocol. It would also be useful to give the name of the trap and trap dimensions here. There are numerous types of sticky trap so the protocol will not fit all commercial trap designs. I recommend re-ordering material inline with these comments.

We modified the text as suggested to maintain better congruency within sections.

c) The Discussion is too long. A comparison of the pros and cons of each method could be summarised more succinctly. Overall, the Discussion needs to be edited down by about 50% and checked for internal consistency.

As the reviewer suggests, we have more concisely described other trapping techniques for comparison reducing the length of the discussion.

Ultimately, the conclusion seems to be that the sticky traps are best because they can be left in place for longer (days to weeks; lines 305-308). However, there is some contradiction because funnel traps can be left in place for weeks to months (see lines 228-229) or hours to days (line 304) depending on which section is read. The major justification for the new method, therefore, appears to be flawed.

It appears the reviewer misread or misinterpreted our discussion, potentially because it was too long and convoluted. Our text states that both funnel and sticky traps can be left out for long periods of time, as we state in the text, the benefit of sticky traps over funnel traps is they better trap flighted arthropods that land on tree boles to rest or for concealment.

d) The sampling design in the case-study seems to be sub-optimal in terms of selection of study sites/trees. Trees were selected from 18 sites and not all tree species occurred in all sites. 'Site' is not included in the statistical analysis and hence species of tree may be confounded with 'Site'. I appreciate that the manuscript is not focused on differences between sites or even differences between species of tree, but the case-study should be an exemplar demonstrating successful application of the new method. A simpler experimental design would have done the job more effectively.

Because we didn't consider site relevant to the results for this manuscript (as the reviewer indicates) we did not emphasize the fact that site was controlled for by including it a random variable in the mixed model analysis. We modified the manuscript to make this more clear.

Minor points
Line 36: typo. The second 'the' = 'that'

modified

Line 44: 'our Ecosystem' = 'the environment' (see also line 23)

Modified

Lines 54-57: Clarify this sentence structure

Modified text to: "These finding are not surprising when considering different parts of the same plant, e.g., leaves versus barks of a tree, provide different resources for which arthropods have adapted to exploit. Thus, each part of the plant has the ability to support a different arthropod community."

Lines 75-77: Add a citation to support this statement. To be honest I am not convinced that techniques that only allow inference of presence/absence of a species necessarily have low detection probability. The statement seems to be muddling two different concepts/parameters.

This text was removed based on earlier comments regarding emphasis on probability of detection and area sampled.

Line 113: 'Measure the tree at breast height'?

Clarified text

Line 116: I do not believe that it is possible to staple a sheet of cardboard to a tree so tightly that no invertebrates crawl underneath. Remove this claim.

Modified to: Be careful to ensure the entire bottom and top of the traps are flush against the tree minimize arthropods from crawling under the trap.

Lines 120-123: This section presupposes that all traps have the same design (and have the instructions written on the box). It is also confusing because it is not immediately clear that 1.4a and 1.4b are alternative ways of setting the box and not sequential instructions. It is also not obvious how this relates the sampling approach in the case-study. Did you do both or only one in the case-study?

Clarified to make it clear either 1.4a or 1.4b should be used.

We provide a specific brand and model that provides direction as written. These direction are pertinent to that specific brand and model.

Line 139: 'desired'

modified

Lines 153-154: This estimation of sampling effort makes no sense. The sampling effort was four traps per tree.

Clarified that sampling effort is 4 traps per tree to ensure samples are taken from all 4 cardinal directions.

Lines 164-167: Add a citation to support this statement.

Deleted this statement: "If traps are to be stored for more than 12 hours, store traps in freezer to preserve content."

Lines 168-171: State the independent variables in the sentence. Saying 'neither' is unhelpful because you have not said what they are.

Identified fixed independent variables

Lines 171-172: The 'proportion of the tree trapped' does not seem to be a useful parameter, especially given the emphasis on sampling/estimating density elsewhere. Perhaps this is your measure of 'sampling effort', but the sampling effort was the same for all trees i.e. 4 traps per tree.

We disagree, although effort was not important in our study, previous studies have found proportion sampled impacted estimates using other types of trapping techniques.

Line 188-191: Table 2. State the units used for 'Total length' in the legend of the table. Is it mm?

mm added to legend

Lines 252-254: Commercial sticky traps are probably most widely used in glasshouse production systems. Add a relevant citation here.

Not clear what the reviewer is referring to here, commercial sticky traps were used in all the studies cited as supporting evidence

Line 269: 'long-term'
Corrected

Reviewer #3:
Manuscript Summary:
The authors presented the method of investigating arthropods on tree trunks by using commercially produced sticky traps. This manuscript has some deficiencies. Firstly, the method by using commercially produced sticky traps is common for field entomologists although few papers have been published in methods journals and textbooks. Secondly, for the amount of data obtained, the manuscript is too long, especially the Discussion section which should be reduced by half. Comparison between absolute and relative methods is already done in a classic ecological text (Ecological methods) written by Southwood and Henderson (2009, third edition). In addition, the authors do not obtain the data by using absolute methods in this study. So, half of the discussion (L193-L251) does not make sense.

Southwood, T. R. E., & Henderson, P. A. (2009). Ecological methods. John Wiley & Sons.

Reduced discussion as 2 reviewers suggested.

Minor Concerns:
L99. Insert line feeds

Modified as recommended

L193-L251. I recommend to delete these parts of discussion because these are not based on the results of this study.

Greatly reduced this part of the text as suggested.

L263-L264. "The traps stiff card board construction"
Please check this phrase.

Modified to "The trap's stiff cardboard construction"

L287-L289. "a fully quantitative method … requires an accurate estimate of the sampling area."
Please check and cite Southwood and Henderson (2009).

Modified as suggested

L297-L301. This sentence is too long and complicated.

Restructured to reduce complexity.

L310. "mesophication"
This word is not common. If the authors use this word, cite a reference or define it.

Modified as suggested

Reviewer #4:
Manuscript Summary:
This manuscript details a new method of capturing bark-using arthropods via readily-available sticky traps. Such a method would enable researchers to repeatedly sample over short periods of time (though long than those represented by traditional methods) and maintain the arthropod bodies for identification and measurements.

Major Concerns:
This is a good new method of measuring this. I think the authors should emphasize the fact that this is also much easier to remove and/or replace from the tree than tanglefoot would be.

Done

I would like a bit more detail on the bark shaving as this might not be something that ecologists and entomologists routinely do, especially if they are used to using the other methods to sample bark-dwelling arthropods. Additionally, I think the inclusion of a citation for the body length by

biomass regression calculation would be useful to readers. If possible, include an image of the trap on the tree even though the written description was quite good.

Will be included in the video

I think an addition to the equipment should be whatever was used to shave the bark off of the tree. I will assume needle nose plyers and staples are considered basic lab materials for a lab that does this type of field work. However, the gauge of staple used might be useful to know.

Done

Minor Concerns:
Line 20, abstract - "a surrogate or biomass" needs to be fixed to "a surrogate FOR biomass"

Modified

Line 35, abstract - "long-term" needs the "-" removed or have "period" or similar term placed after it

Modified

Line 44, introduction - "Ecosystem" does not need to be capitalized

Modified

Line 46, introduction - You should add in an example of forest pests here in addition to/instead of one of the listed ones to further emphasize you point in the next sentence and to match the fact that this study is being used in forest systems

Forests are included in native vegetation

Reviewer #5:
Manuscript Summary:
This manuscript concisely and efficiently outlines a new procedure for detecting and quantifying corticolous arthropods. I found the methods description easy to follow and complete. However, I feel that this manuscript lacks a meaningful control. It is difficult to determine how these results compare to other methods, such as pitfall traps, glue spreading, etc. Without the ability to access taxonomic or size biases in collection, as well as direct effort comparisons, it is difficult to contextualize the results. If this data is not available, a literature review should be conducted to provide examples of these characteristics, as well as sampling variability, using other capture techniques.

We do not believe it would be appropriate to include the considerable aunt of text that would be required for this comparison.

Major Concerns:
Line 176- How does this variability compare to other methods of capture?

See above response

Line 283- You need to justify the criterion selected for what constitutes an acceptable amount of error/variability.

It is up to individual researchers to determine what is an acceptable amount based on their study objectives. With this paper we only provide estimates of how much variability is achieved using this method.

Line 313- Many arthropod capture methods are biased (towards specific orders, size classes, etc). Thus, typically one capture method is not universally preferable, and often a combination of methods is used to account for individual biases in methods. A discussion of biases in this manuscript would assist readers in deciding on an appropriate methodology to suit their particular objectives. If these data are not available, I still think that a discussion of possible biases and the biases of other methods is warranted.

The other 3 reviewers recommended we reduce the text of the nature suggested here, we agree with their assessment and did not make this change.

Minor Concerns:
Line 65- Provide a specific example

We provided a citation as an example.

Line 67- This statement is a tautology. Perhaps provide a different example.

We disagree, the first statement emphasizes and provides citations other than the relationship between arthropods and birds, the second sentence emphasizes the arthropod bird relationship.

Line 164- Was each trap out for 9 days, or was this just the total sampling period for all traps? Please clarify.

Modified as suggested.

Appendix A: Classification Guild for Tree Insects

Guild 1: Primitive Wingless Hexapods

- Orders Protura, Collembola, Diplura, Archaeognatha, and Thysanura

Guild 2: Dragonflies and allies

- Anisoptera (Dragonflies) and Zygoptera (Damselfies)

Guild 3: Orthopterans

- Plecoptera (Stoneflies), Dermaptera (Earwigs), Ensifera (Crickets, Katydids, Grasshoppers), Caelifera (Short-horned Grasshoppers and Locusts), Phasmatodea (Walking Sticks), Blattodea (Cockroaches), Isoptera (Termites), and Mantodea (Mantids)

Guild 4: True Bugs 1 (Hemiptera)

- Pentatomidea (Stink Bugs and Relatives), other true bugs (Assassin, Lace, Ambush, Stilt, Plant, Bark, etc. Bugs)

Guild 5: True Bugs 2

- Auchenorrhyncha (Cicadas, Treehoppers, Leafhoppers, Froghoppers, etc.) and Sternorrhyncha (Scale Bugs, Mealybugs, Aphids, Whiteflies, etc.)

Guild 6: True Bugs 3

- Psocoptera (Barklice and Booklice), and Thysanoptera (Thrips)

Guild 7: Beetles 1

- Adephaga (Carabid and Tiger Beetles)

Guild 8: Beetles 2

- Scarabaedoidea (Scarabs Beetles, Stag Beetles, etc.)

Guild 9: Beetles 3

- Elateroidea (Fireflies, Click Beetles, etc.)

Guild 10: Beetles 4

- Cucujoidea (Lady Beetles, etc.) and Chrysomeloidea (Leaf Beetles, Boring long-horned Beetles, etc.)

Guild 11: Beetles 5

- Curculionoidea (Weevils, etc.)

Guild 12: Beetles 6

- Miscellaneous Beetles (families not in the other 6 groups.)

Guild 13: Lepidoptera

- Lepidoptera (Butterflies and Moths)

Guild 14: Diptera 1

- Nematocera (Midges, Crane Flies, Mosquitoes, etc.)

Guild 15: Diptera 2

- Brachycera (Short-horned Flies: Horse, House, Deer, Bot, etc. Flies)

Guild 16: Hymenoptera 1

- Apocrita (Parasitic Wasps and Gall Wasps)

Guild 17: Hymenoptera 2

- Aculeata (Cuckoo wasps, Dirtdobbers, Bees, Ants, Sweat Bees, and Wasps)

Guild 18: Hymenoptera 3

- Symphyta (Sawflies, Horntails, and Wood Wasps)

Guild 19: Miscellaneous Holometabola

- Strepsiptera (Twisting-winged Parasites), Megaloptera (Dobsonflies, Alderflies, etc.), Neuroptera (Lacewings, Antlions, etc.), and Trichoptera (Caddisflies)

Guild 20: Parasitic Insects

- Siphonaptera (Fleas), Mecoptera (Scorpionflies), and Phthiraptera (Lice)

Guild 21: Araneae mites

Guild 22: Araneae

Guild 23: Opilionids

Guild 24: Gastropods

Guild 25: Polydesmida and Julida

Guild 26: Oligachaeta worms