

**Effects of Forest Features and Location on Emerald Ash Borer (*Agrilus planipennis*)
Infestation Severity of Ash (*Fraxinus spp.*) Trees**

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Abstract

The Emerald Ash Borer (*Agrilus planipennis*; EAB) is a well-studied invasive species that was first discovered in the United States in 2002 (Herms & McCullough, 2014), and has spread quickly since, infesting all ash (*Fraxinus spp.*) trees native to the U.S (Cappaert et al., 2005). Once symptoms are observed, infested trees die within 3-4 years (Herms & McCullough, 2014). Difficulty in detecting new infestations has made them difficult to contain.

EAB prefers *Fraxinus spp.* stems grown individually in sunlight (Chen & Poland, 2009). Previous studies have also shown that lakes can act as barriers restricting insect pest dispersal (Kim & McPheron, 1993). Additionally, there are conflicting results in previous literature regarding *Fraxinus spp.* density and EAB's rate of infestation (Knight et al., 2013; Chen et al., 2009). In order to further investigate these variables' effects on infestation, we considered three questions: (1) Does the density of surrounding *Fraxinus spp.* stems affect the severity of an individual's EAB infestation; (2) Does the distance between a *Fraxinus spp.* stem and the shoreline affect the severity of its EAB infestation; (3) Does the level of canopy cover surrounding an individual *Fraxinus spp.* stem affect the severity of its EAB infestation?

We created three transects on the University of Michigan's Biological Station (UMBS) property. In each transect we recorded canopy cover, density, and severity of infestation for each *Fraxinus spp.* stem. Distance to shoreline and density were mapped in ArcGIS. The three *Fraxinus spp.* found in northern Michigan, *F. americana*, *F. nigra*., and *F. pennsylvanica* are all affected by EAB equally (Agius et al., 2005), therefore differentiation by species was deemed irrelevant to this study.

We found a significant inverse correlation with a weak R^2 value between *Fraxinus spp.* stem density and severity of infestation on individual stems. Our data support the hypothesis that increased *Fraxinus spp.* density within a stand is associated with slower mortality due to EAB infestation.

We found no significant correlation between EAB infestation severity and canopy cover or distance to shoreline. In forest stands, the canopy cover of each stem does not affect whether or not it suffers more severe infestations. We hypothesize that Douglas and Burt Lakes are either not significant barriers to the spread of EAB, or that they are no longer effective barriers.

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Methods

Our study took place at three transects on UMBS property: one on Grapevine Point and two in the Gorge (Figure 10). Study sites were chosen based on presence of *Fraxinus spp.* and proximity to the water. We surveyed for *Fraxinus spp.* within a 50 m transect extending 10m on each side. The Grapevine Point Transect and Gorge Transect #2 began at the water of Douglas Lake and Burt Lake, respectively. At these transects, 0 m on the transect tape was the shoreline. Gorge Transect #1 was placed offshore in a stand located along the Little Carp River with many *Fraxinus spp.* present (Table 2). Within all transects, we recorded measurement and data for every *Fraxinus spp.* stem above breast height in ArcGIS Collector.

We measured percent canopy cover using a densiometer in the four cardinal directions 1 m away from the base of the tree and subtracted the average of these four readings from 100, giving the percent canopy cover for that specific tree. We took the diameter at breast height (DBH) with a DBH tape or calipers depending on the size and accessibility of the tree. We estimated the height of each tree using a sectional pole, and recorded by bins at breast height-5m, 5.1-10 m, and 10.1-15 m. *Fraxinus spp.* density per hectare around each stem was calculated by counting all *Fraxinus spp.* stems in a circular 100 m² plot centered on that stem. Severity of infestations were measured on a 0-10 scale indicating crown health, twig dieback, and bark health (Murfitt et al.,2016). 0 is the most severe and 10 indicates a healthy tree (Table 1).

We measured distance to the shoreline in ArcGIS using the Near (analysis) tool. At the two shoreline transects, the distance to shore was the same as the distance of the tree along the transect. The map on ArcGIS also shows the density of *Fraxinus spp.* in the plot.

We used SPSS for statistical analysis. Distance to shoreline, percent canopy cover, surrounding *Fraxinus spp.* density, and DBH were compared to severity of infestation in individual regressions and ANOVA tests.

Results and Discussion

We found no significant correlation between distance to shoreline and infestation severity in the transects near the shore (Grapevine Point and Gorge #2) (ANOVA, $p = 0.535$; $y = -0.0796x + 9.46$, $R^2 = 0.087$; Figure 2), nor in the Gorge #1 transect (ANOVA, $p = 0.215$; $y = -0.033x + 26.958$, $R^2 = 0.029$; Figure 3). Even between all three transects there was no correlation (ANOVA, $p = 0.061$).

We found significant correlation between number of surrounding *Fraxinus spp.* stems within 100 m² and EAB infestation severity (ANOVA, $p = 0.002$; $y = 0.3281x + 5.2078$, $R^2 = 0.087$; Figure 4).

We failed to reject the null hypothesis that canopy cover does not affect severity of infestation in sub-canopy *Fraxinus spp.* stems (ANOVA, $p = 0.451$; $y = 0.0308x + 3.6376$, $R^2 = 0.007$; Figure 5). Overstory *Fraxinus spp.* stems had infestation severity ranging from 0-10 (Figure 6), indicating that being in full sun does not exclude individual stems from infestation, nor does it guarantee that they will be affected by EAB.

The correlation between stem DBH and infestation severity (ANOVA, $p < 0.001$; $y = -0.3992x + 8.1483$, $R^2 = 0.404$; Figure 6) matched the findings of previous studies (Knight et al., 2014). Larger *Fraxinus spp.* stems tended to have more severe EAB infestations.

The data supported previous findings that larger DBH is associated with more severe EAB infestation (Knight et al., 2014; Figure 7). This is expected, as more EAB adults can emerge per square meter from trees ≥ 13 cm DBH than from smaller *Fraxinus spp.* stems (McCullough & Mercador, 2012). EAB feed and live in the phloem, thus availability of phloem determines the number of EAB that can develop within a host (Herms & McCullough, 2014). Phloem volume is proportional with tree size, so larger trees allow more EAB to colonize leading to an increase in severity of infestation (Hölttä et al., 2013).

The lack of significant correlation between distance to shoreline and infestation severity failed to support our hypothesis that *Fraxinus spp.* stems located closer to the lake have lower infestation severity. Based on these results, we concluded that Douglas and Burt Lakes are not acting as geographic barriers to EAB spread. We had proposed that the lakes may serve as barriers based on broader descriptions of impediments to insect dispersal laid out in Kim and McPheron (1993). Geographic barriers, including bodies of water like lakes, can stop or restrict spread of insects (Kim & McPheron, 1993). The failure of our data to show significant lake impact on infestation severity, however, can be explained by EAB flight capabilities. Female EAB adults can fly up to 2 km a day, with a cumulative flight distance of up to 9.8 km over a lifespan (Herms & McCullough, 2014). Douglas and Burt Lakes are not large enough to prevent EAB from travelling around or across narrow stretches of water, and thus fail to have an effect on pest dispersal and ensuing severity of infestation.

The lack of correlation is relevant to conservation efforts. It indicates that *Fraxinus spp.* stems on lakeshores are equally vulnerable to EAB as those not on lakeshores. There would be no immediate benefits to the survival of *Fraxinus spp.* if lakes were used as geographical barriers for conservation. New *Fraxinus spp.* stems planted on lakeshores would not have an overall higher chance of survival. Douglas and Burt Lakes are both smaller inland lakes (Figure 1), however, and it is worth further study to determine whether larger bodies of water such as the Great Lakes, or physiographic barriers such as mountains prevent EAB dispersal.

Number of *Fraxinus spp.* stems surrounding an individual tree was inversely correlated with severity of infestation of that tree. Our results are consistent with those of Knight, Brown, and Long (2013), who observed more rapid mortality in stands with lower density of *Fraxinus spp.* Knight et al. (2014) theorized that this is an extension of the resource dilution hypothesis, in which high concentration of EAB on a few trees leads to more rapid decline of those hosts. In areas with more *Fraxinus spp.* stems, EAB are distributed across more trees, resulting in a lower concentration of EAB larvae per individual. The inverse-density dependence of *Fraxinus spp.* mortality can be used to inform management of at-risk forests. Assuming this density model of mortality is valid, thinning *Fraxinus spp.* stands will not protect against infestation, and instead may actually shorten time to tree death.

Further research to confirm and expand upon these findings may be relevant to management techniques slowing the spread of EAB in *Fraxinus spp.* Given the mortality rate and spread of infestation, however, it is unlikely that prevention of *Fraxinus spp.* death or eradication of EAB is viable, at least in the short term. Regionally, EAB has inflicted a higher degree of mortality than other forest insect outbreaks such as gypsy moth or hemlock wooly adelgid because of low level of resistance present (Morin et al., 2017), making control of the pest a high priority.

EAB reportedly colonizes non-*Fraxinus* genera (i.e. *Ulmus* L., *Juglans* L., and *Pterocarya* Kunth.) in Asia, but is unable to produce viable larvae on synonymous non-*Fraxinus* logs in southeastern Michigan. EAB is currently only able to create galleries in *J. nigra* (Anulewicz et al., 2008). EAB-host expansion to non-*Fraxinus* species is of great ecological concern, since it was so devastating to *Fraxinus*, and could have destructive effects on other genera. Understanding EAB and host interactions will allow for more informed management and conservation decisions, potentially preventing further spread of EAB.

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Tables and Figures

Table 1

Health Class	Parameters for General Health Assignment
9 or 10	Crown is in good health, no evidence of twig dieback or slight, bark is healthy, no evidence of obvious EAB infestation.
7 or 8	Crown is slightly different from a tree that received a 9 or 10. There may be small gaps in the crown (larger gaps for a 7). Bark may be peeling lightly and there may be trace amounts of evidence of EAB infestation.
5 or 6	Crown is less full. There are medium-sized gaps in the crown and significant twig dieback. There is evidence of epicormic branching on the lower trunk of the tree, and moderate evidence of EAB infestation.
3 or 4	There is significant crown thinning and twig dieback. There is obvious evidence of EAB.
1 or 2	There is significant crown thinning and twig dieback. There is significant evidence of EAB.
0	0 is only given if the tree is dead.

Table 1. Ash tree classes for general tree health taken directly from Murfitt et al. (2016). Severity of infestations were measured on a 0-10 scale indicating crown health, twig dieback, and bark health.

Table 2

Site Location	Coordinates (decimal degrees)	
	Transect Start Point	Transect End Point
Grapevine Point	45.56481, -84.67684	45.56481, -84.67749
Gorge Transect #1	45.54419, -84.68092	45.54374, -84.68092
Gorge Transect #2	45.53734, -84.67883	45.53779, -84.67883

Table 2. Geographic coordinates of 50 m transect start and end points. Grapevine Point and Gorge Transect #1 ran perpendicular to Douglas and Burt Lake shorelines, respectively.



Figure 1. Location of transects along Grapevine Point and the Gorge (Geographic Coordinate system: GCS North American 1983).

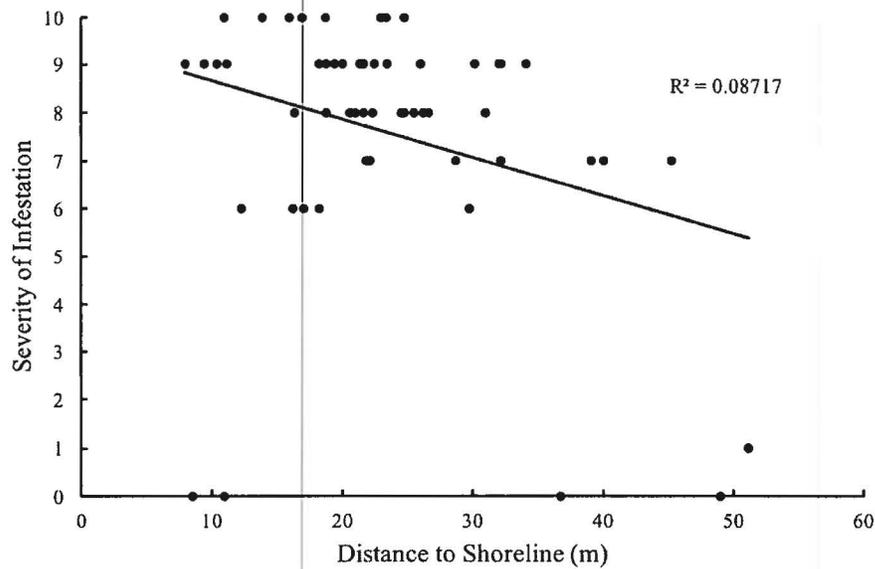


Figure 2. Line of best fit for relationship between distance to shoreline and severity of infestation of EAB on *Fraxinus spp.* stems (determined by crown dieback, bark health, etc.) at the Grapevine Point site and Gorge Transect #2. Correlation was not significant ($p = 0.535$).

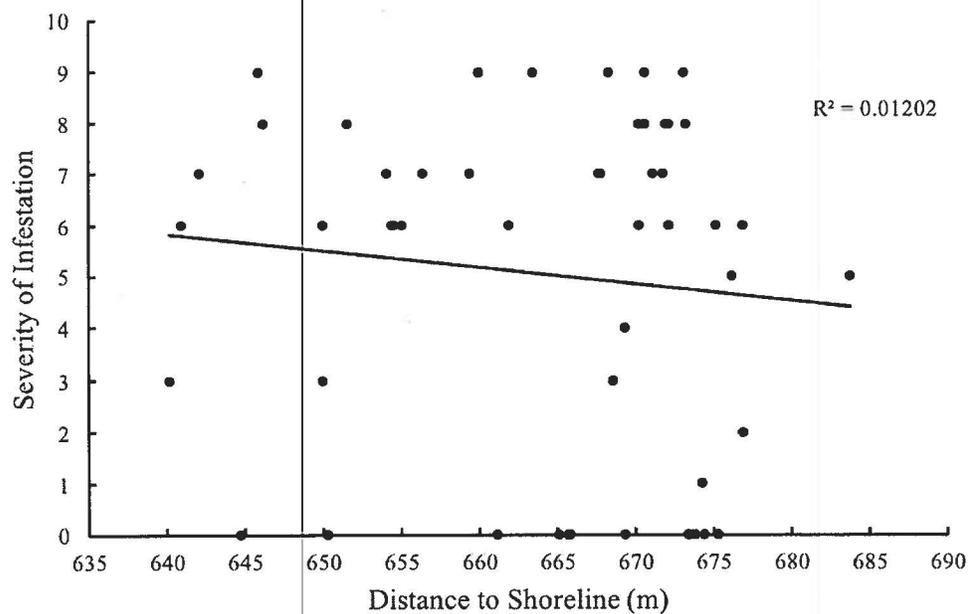


Figure 3. Line of best fit for relationship between distance to shoreline and severity of EAB infestation on *Fraxinus spp.* stems (determined by crown dieback, bark health, etc.) at Gorge Transect #1. Correlation was not significant ($p = 0.215$).

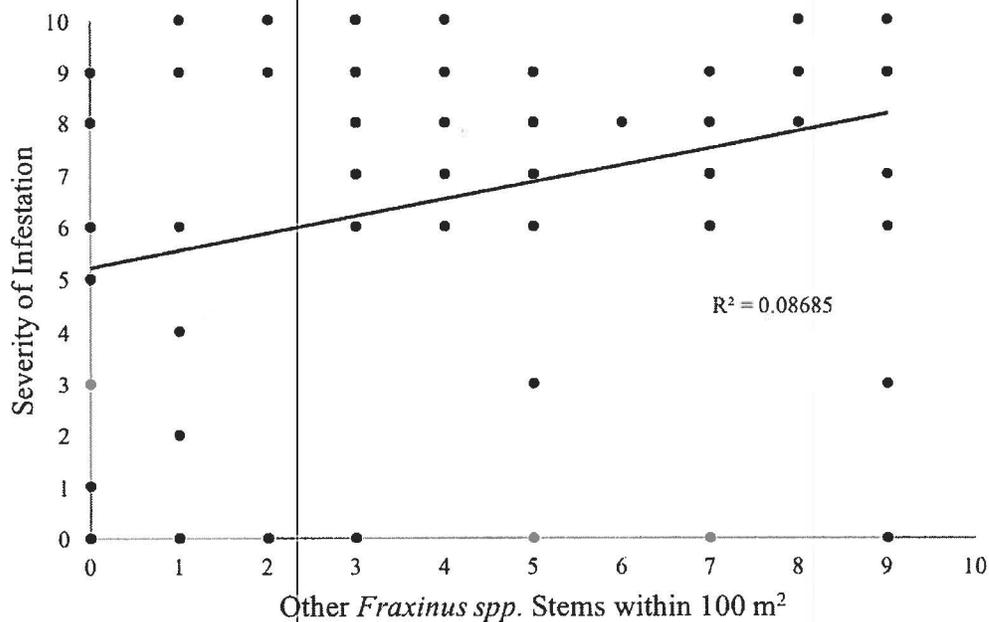


Figure 4. Line of best fit between severity of infestation (determined by *Fraxinus spp.* crown dieback, bark health, etc.) and amount of surrounding *Fraxinus spp.* stems within 100 m². The ANOVA test found a negative correlation between the two variables ($p = 0.002$).

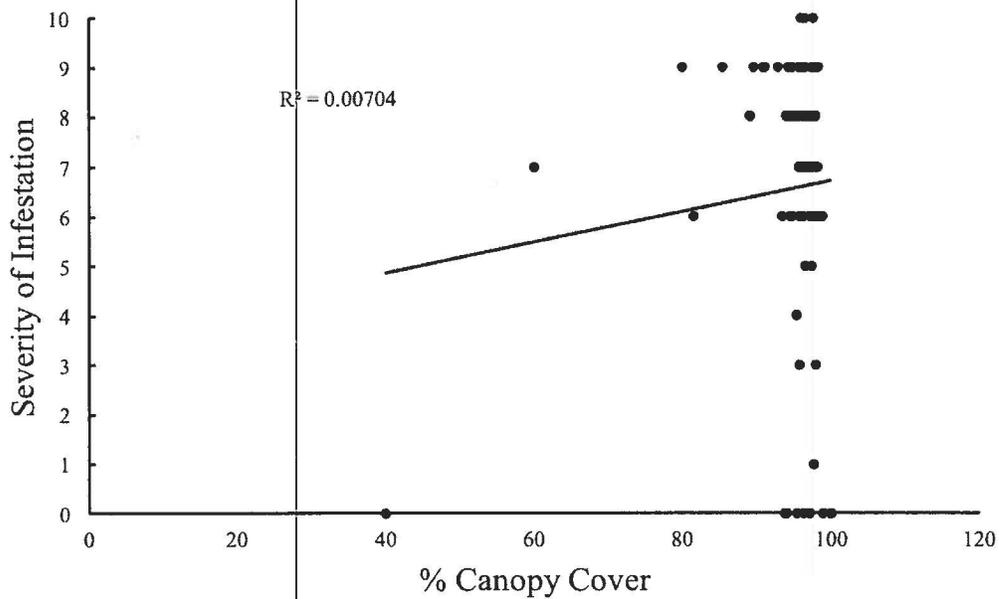


Figure 5: Line of best fit for relationship between % canopy cover for individuals in the understory and severity of infestation (determined by *Fraxinus spp.* crown dieback, bark health, etc.). No correlation was found ($p = 0.451$).

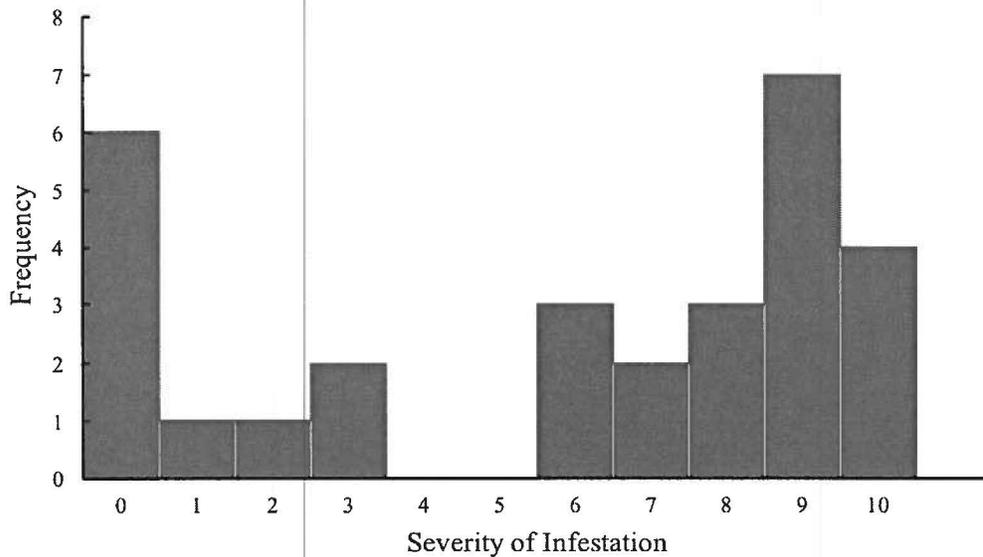


Figure 6. Illustrates the frequency of different infestation severities (determined by *Fraxinus spp.* crown dieback, bark health, etc.) for *Fraxinus spp.* individuals observed in the canopy.

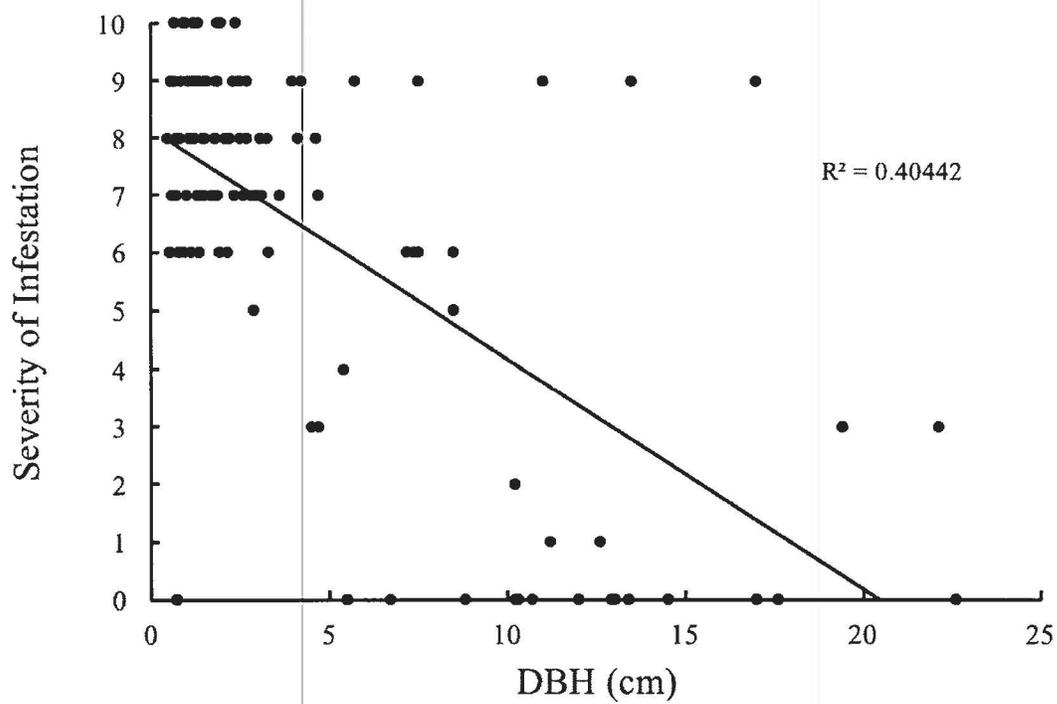


Figure 7. Line of best fit between severity of infestation (determined by *Fraxinus spp.* crown dieback, bark health, etc.) and DBH of *Fraxinus spp.* stems. The ANOVA test found a negative correlation between the two variables ($p < 0.0001$).