Journal of the Torrey Botanical Society 147(1): 000–000, 2020.

Honeysuckle leaf blight reduces the growth of infected Amur honeysuckle (*Lonicera maackii*, Caprifoliaceae) seedlings in a greenhouse experiment[1]

Richard L. Boyce,[2] Steven M. Castellano, Andrea N. Marroquin, Onyinye M. Uwolloh, Sarah E. Farrar, and Kimberly P. Wolfe

Department of Biological Sciences, Northern Kentucky University, 1 Nunn Drive, Highland Heights, KY 41099, USA

Abstract. Amur honeysuckle (*Lonicera maackii*) is an important invasive plant species in the Ohio River Valley. Previous work has shown extensive dieback of honeysuckle in the region, coupled with the appearance of the native fungal pathogen, honeysuckle leaf blight (*Insolibasidium deformans*). Our goal was to find if the blight causes growth decline or mortality in Amur honeysuckle. Seedlings were grown under greenhouse conditions in 2017. Treated seedlings were sprayed with a spore solution prepared from blighted leaves that were collected from the field. They were placed into a growth chamber with conditions set for optimum spore growth and then returned to the greenhouse after leaf blight began to develop. Growth (height, total stem length, leaf area, and leaf number) and dark-adapted chlorophyll fluorescence (F_v/F_m) were measured periodically over the growing season. A repeated-measures analysis of aboveground growth indicated that larger, faster-growing plants were more likely to be infected, but their growth rates were subsequently reduced much more than uninfected treated plants and controls. There were positive correlations between F_v/F_m and RGR (relative growth rate). Blighted leaves had lower values of F_v/F_m than uninfected leaves. No infected plants died, but this experiment supports our hypothesis that leaf blight causes a significant growth decline in Amur honeysuckle. Future work will determine if the patterns seen under greenhouse conditions hold in the field.

Key words: chlorophyll fluorescence, *Insolibasidium deformans*, invasive plants, *Lonicera maackii*, relative growth rate

Amur honeysuckle (*Lonicera maackii* (Rupr.) Herder) was introduced in the 19th century as an ornamental shrub (Luken and Thieret 1996) and has since spread to at least 34 states (EDDMapS 2019, PLANTS 2019). Many studies have shown that it is an important invasive plant species in the Ohio River Valley. A review by McNeish and McEwan (2016) describes many of the detrimental effects caused by this species. Its extended-deciduous leaf habit (McEwan *et al*. 2009) gives it a competitive advantage over native plants (Chen and Matter 2017). It has allelopathic effects on herbaceous species (Dorning and Cipollini 2006; Cipollini *et al*. 2008a; Cipollini *et al*. 2008b) and shows resistance to herbivory (Cipollini *et al*. 2008b, Lieurance and Cipollini 2013). It has also been shown to have negative effects on both herbaceous and woody species (Hutchinson and Vankat 1997; Luken *et al*. 1997a; Medley 1997; Gould and Gorchov 2000; Collier *et al*. 2002; Gorchov and Trisel 2003; Hartman and McCarthy 2004, 2007; Miller and Gorchov 2004; McKinney and Goodell 2010; Loomis *et al*. 2015). Honeysuckle presence is associated with other invasive species, suggesting it is an invasive facilitator (Cully *et al*. 2016). Amur honeysuckle also affects animal (Christopher and Cameron 2012; Loomis and Cameron 2014; Loomis *et al*. 2014) and microbial (Arthur *et al*. 2012; Shannon *et al*. 2014; Ali *et al*. 2015) communities. It changes both water and nutrient dynamics in ecosystems (Arthur *et al*. 2012; Boyce *et al*. 2012; McEwan *et al*. 2012; Poulette and Arthur 2012; Kolbe *et al*. 2015; Pfeiffer and Gorchov 2015; Hopfensperger *et al*. 2017). Honeysuckle's effects also extend to aquatic communities (Watling *et al*. 2011; McNeish *et al*. 2012, 2017; Borth *et al*. 2018).

Previous work (Boyce *et al*. 2014; Boyce 2018) has shown extensive dieback of honeysuckle in the region, coupled with the appearance of the native fungal pathogen honeysuckle leaf blight (*Insolibasidium deformans* (C.J. Gould) Oberw. &

[1] We would like to thank the UR-STEM program, NKU CINSAM (Center for Integrative Natural Science and Mathematics), and the Northern Kentucky University Department of Biological Sciences for funding this project. In addition, we thank Andrew Schille and Adam O'Brien for their assistance with this project.

[2] Author for correspondence: boycer@nku.edu

doi 10.3159/TORREY-D-19-00025.2

Received for publication May 31, 2019, and in revised form October 29, 2019; first published Month 00, 2020.

Bandoni). Honeysuckle leaf blight is native to North America and infects most species in the genus *Lonicera*, including both native and introduced species (Gould 1945, Riffle and Peterson 1986, Cordell *et al*. 1989). Widespread outbreaks were observed in the Cincinnati, OH, region beginning in 2012, and they have continued every summer since then (R. Boyce, personal observation).

However, this co-occurrence in time cannot by itself be used to show that the leaf blight was responsible for the decline in Amur honeysuckle. One way to show the effect of a plant disease is to grow the plant under controlled conditions, where infected plants can be compared against noninfected controls. Here, we describe an experiment involving the effect of leaf blight on honeysuckle seedlings under growth chamber and greenhouse conditions. Our goal was to find if the blight causes growth decline or mortality in honeysuckle.

Material and Methods. SEEDLING GROWTH. We collected honeysuckle seeds from local northern Kentucky populations in 2016, and we cold-stratified them over the winter at 4 °C. Seedlings were then grown under greenhouse conditions in a standard potting mix with slow-release fertilizer in spring 2017. Blighted leaves were collected from the field at NKU-REFS (Research and Education Field Station) in Melbourne, KY, on May 16, 2017, and a spore inoculant was prepared by placing leaves in distilled water, then gently rubbing the leaves to dislodge spores into the water. The 35 treated seedlings were sprayed with the spore inoculant until all leaf surfaces were dripping, and they were placed into a growth chamber at 16° C and close to 100% relative humidity for six days, conditions that promote infection (Gould 1945). The 35 control seedlings were sprayed with distilled water in a similar manner and placed into another growth chamber under similar conditions. Spraying of inoculum and distilled water, as applicable, was repeated once per day while seedlings were in the growth chambers. Treated and control plants were then returned to the greenhouse and monitored for the development of blight symptoms. To prevent contamination of control seedlings, control and treated seedlings were kept in different parts of the greenhouse. Roughly half (17 of 35) of the treated seedlings developed blight symptoms and were designated "blighted"; treated seedlings that did not develop symptoms were designated as "clear."

MEASUREMENTS. We measured plant size on May 19, 2017, June 22, 2017, and July 27, 2017 (hereafter referred to as May, June, and July). On each plant, we measured stem height (leader length), total stem length (leader + side branches), number of leaves, and leaf area. Area of each leaf was estimated by assuming they were ellipses (*i.e*., area = π(length/2*width/2)^2. This species has opposite leaves, so we measured length and width of one member of the pair and assumed the other had the same dimensions. Relative growth rates (RGR) were calculated for the May–June and June–July periods as $RGR = [\ln(G_2) - \ln(G_1)]/(t_2 - t_1)$, where G_1 and G_2 are growth measurements at time 1 and time 2 (t_1 and t_2). Plants were harvested on September 8, 2017 and separated into leaves, shoots, and roots. Roots were washed in tap water to separate them from soil. After oven drying at 70° C for three days, parts were weighed.

Dark-adapted chlorophyll fluorescence (F_v/F_m) was measured on all plants on May 25, 2017, June 9, 2017, June 20, 2017, and July 27, 2017 with an Opti-Science OS5-FL chlorophyll fluorometer (Opti-Sciences, Hudson, NH). On June 20, 2017, we also compared F_v/F_m of blighted and clear leaves on the same blighted plants.

DATA ANALYSIS. Because data did not meet the assumptions required for parametric data analysis (*i.e*., normality, equal variances, etc.), we used nonparametric techniques in our data analysis. Permutation repeated-measures analysis of variance (PRMANOVA), using the package permuco in R (R Core Team 2019), was performed on all growth measurements taken for all plants from May, June, and July for the comparison of their growth patterns. The treatment had three levels: control, blighted, and clear. Permutation tests are a type of randomization test where the data are permuted and the procedure is run to generate the test statistic, usually a large number (hundreds to thousands) of times. The statistic generated by the original data is then compared against the distribution of statistics from the permuted data to generate the probability levels (*P* values) that are used to test the null hypothesis, as in all statistical procedures (Manly 2006). Relative growth rates, which were based on growth over specific time periods, were analyzed with a nonparametric Kruskal-Wallis test, followed by a

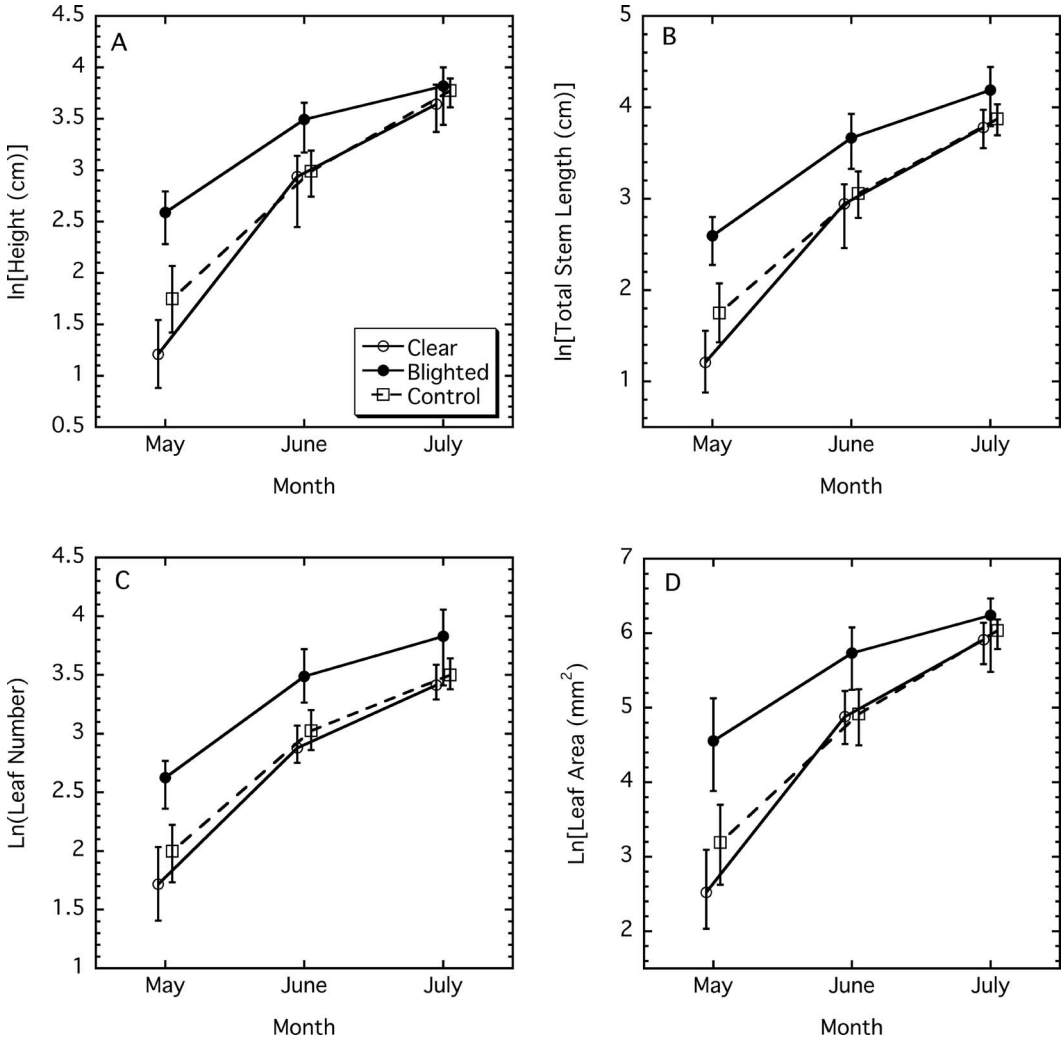

FIG. 1. Mean growth of seedlings in May, June, and July 2017, showing (A) height (leader length), (B) total stem length (including side branches), (C) leaf number, and (D) leaf area. Error bars are bootstrapped 95% confidence intervals, and means where error bars do not overlap are considered to be significantly different. Statistics from the permutation RMANOVA time × treatment interaction terms are as follows: height, $F_{4,124} = 14.934$, $P = 0.0001$; total stem length, $F_{4,124} = 8.518$, $P = 0.0001$; leaf number, $F_{4,124} = 3.292$, $P = 0.0127$; and leaf area, $F_{4,124} = 12.830$, $P = 0.0001$.

post hoc Tukey-like test (Zar 2010). For a comparison of growth over the entire season, final harvested root, shoot, and leaf weights were also analyzed with a *post hoc* Tukey-like test performed after a nonparametric Kruskal-Wallis test. Spearman rank correlations between RGRs and F_v/F_m taken at the end of each RGR period were run to examine the relationship between chlorophyll fluorescence and growth. A nonparametric Wilcoxon paired-sample test was used to compare blighted and clear leaves on blighted plants on June 20, 2017. The bootstrapped 95% confidence intervals for the mean values shown in our results were generated by sampling with replacement 10,000 times to generate a distribution of means, and limits that included 95% of the means were set using the accelerated bias-corrected technique outlined in Manly (2006).

Results. For seedlings that were treated with the spore solution, roughly half (17 of 35) developed blight symptoms (blighted). Blighted seedlings had

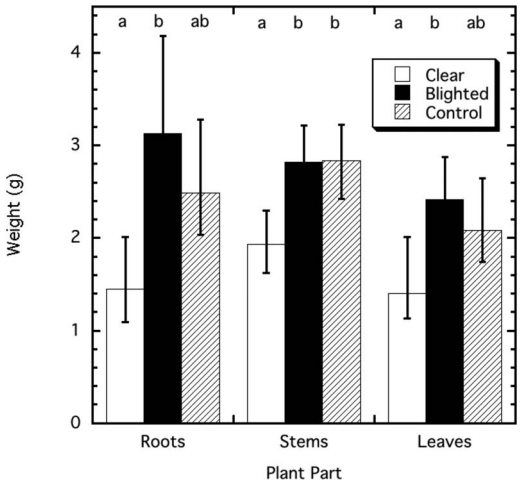

FIG. 2. Final dry weights for roots, stems, and leaves of plants harvested in September. Error bars are bootstrapped 95% confidence intervals. Letters denote differences in means as determined by a *post hoc* Tukey-like test performed after a nonparametric Kruskal-Wallis test (Zar 2010).

$\sim$ 7% of their leaves showing blight symptoms in June (data not shown). At the start of the experiment in May, treated seedlings that later developed blight symptoms (blighted) were significantly larger than those that did not (clear), as shown in Fig. 1; this was true for height, total stem length, leaf number, and leaf area. Control seedlings fell in between, although their bootstrapped 95% confidence intervals overlapped those for clear seedlings and did not overlap those for the blighted seedlings. Growth measurements of control and clear seedlings caught up with blighted ones by July, and there were no significant differences on the last sampling date. All time $\times$ treatment interaction terms in the PRMANOVAs had P values < 0.05, indicating that growth rates differed. The final weights, when seedlings were harvested in September, reflected the aboveground plant dimensions that were seen in May. Blighted seedlings had significantly mean higher root, stem, and leaf weights than clear seedlings, while control seedlings generally had mean weights that, while not differing significantly from blighted seedlings, fell between clear and blighted (Fig. 2).

Measurements of relative growth rate (RGR) over the May–June 2017 and the June–July 2017 periods showed that although the clear seedlings were initially the smallest (Fig. 1), they had the highest RGRs in the May–June 2017 period (Fig. 3). Although RGRs both decreased and converged during the June–July 2017 period, those for the blighted seedlings remained significantly lower than for the clear seedlings.

Dark-adapted chlorophyll fluorescence (F_v/F_m) declined from May 25, 2017 to June 20, 2017, then increased on July 27, 2017 (Fig. 4). On most dates, there were no significant differences; on July 27, 2017, however, there was some indication that the clear values exceeded those of the blighted and controls. On June 20, 2017, a Wilcoxon paired-sample test showed that, on blighted plants, blighted leaves had lower values than clear leaves (Fig. 5). There were positive correlations between RGRs and the F_v/F_m measurements taken at the end of each RGR (Table 1). Most of those values were significant at the $P \leq 0.05$ level.

Discussion. In June, we found about 7% of leaves on blighted plants of *L. maackii* seedlings exhibited blight symptoms (it was difficult to determine in July because some blighted leaves had fallen off), which is similar to the 11% that Gould (1945) found to develop infections. About 50% (17 out of 35) of treated plants developed leaf blight at some point during the experiment. It is clear that faster-growing plants were more likely to be infected by blight (Fig.1). Gould (1945) found that only leaves $<$ 20 days old were infected by *I. deformans* in his experiments, and infection was more likely to occur in the younger leaves. Fast-growing plants will have both a greater number and proportion of younger leaves, which may explain our findings of more infection in them.

The relationship between chlorophyll fluorescence (F_v/F_m) and growth is not completely clear. Leaf blight clearly stresses infected leaves, as shown by reduced F_v/F_m in blighted *vs*. clear leaves in the blighted group of seedlings (Fig. 5). There are also correlations between F_v/F_m and RGR (Table 1), but relationships between F_v/F_m and actual amount of growth are less clear. Chlorophyll fluorescence (F_v/F_m) declined slightly from May to the end of June in clear leaves for all seedlings (Fig. 4), perhaps reflecting the increase in temperature experienced by the seedlings as the summer progressed; stress of any kind generally causes a decline in this parameter, although the values measured in this experiment are typical of healthy plants (Bohlàr-Nordenkampf and Öquist 1993). During the May–June period, there did not appear to be any significant differences among

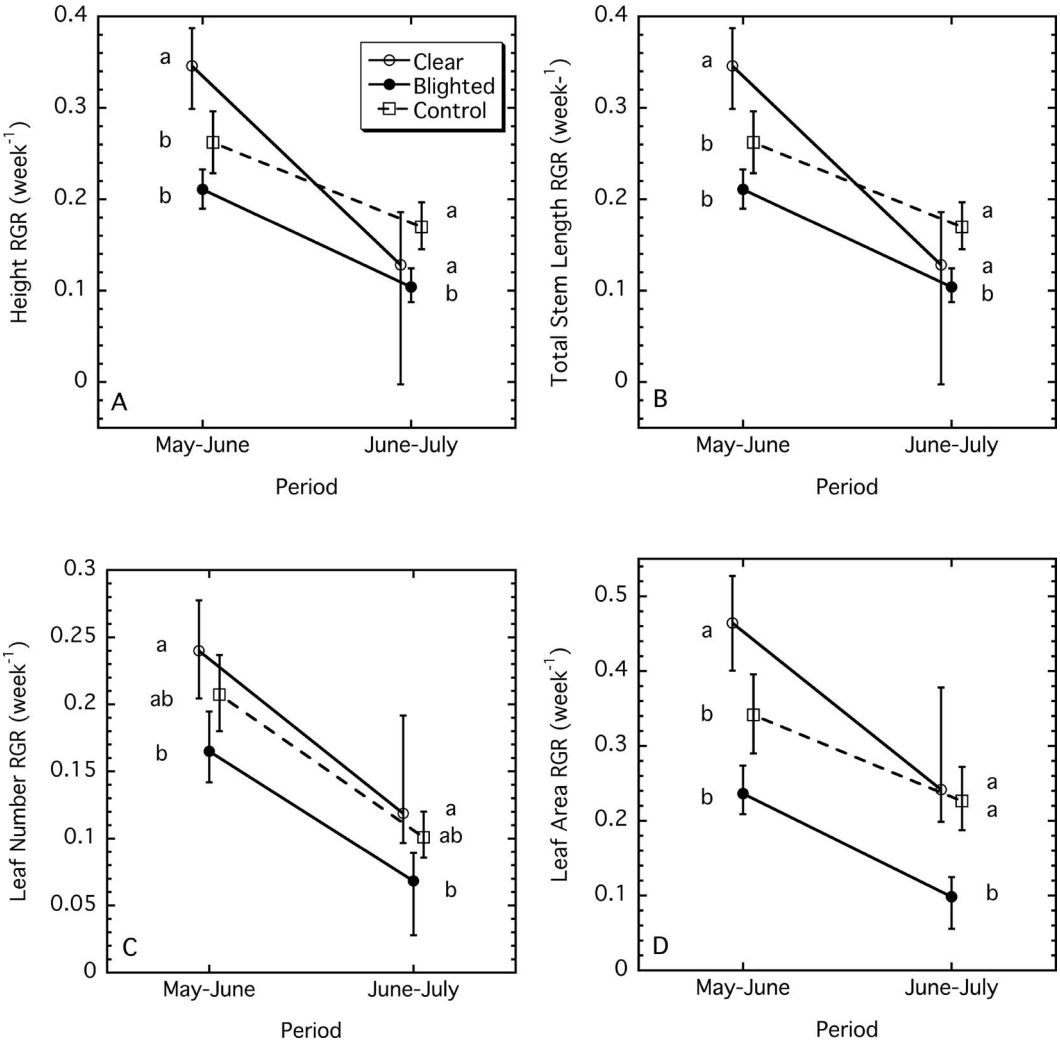

FIG. 3. Relative growth rates (RGR) of (A) height, (B) total stem length, (C) leaf number, and (D) leaf area. Data shown are for two periods, May–June and June–July. Error bars are bootstrapped 95% confidence intervals. Letters denote differences in means as determined by a *post hoc* Tukey-like test performed after a nonparametric Kruskal-Wallis test (Zar 2010).

blighted, clear, or control groups (Fig. 4). In July, while all treatment groups showed a recovery, the clear seedlings were significantly higher than blighted and control seedlings. These differences in F_v/F_m are not a reflection of greater overall growth by clear seedlings, however (Fig. 1). Thus, the relationship between growth and F_v/F_m is not completely clear.

Relative growth rate (RGR) in our study was higher in the blighted seedlings than either control or clear seedlings during the May–June period. However, over the June–July period, blighted RGR fell so that it was significantly lower than those of clear and (usually) control seedlings (Fig. 3), which allowed growth of these latter two to catch up by the July measurement period (Fig. 1). Nevertheless, the initial size advantage that infected seedlings had over clear seedlings still appeared in the harvest weights in September (Fig. 2), although blighted seedlings were not significantly greater than control seedlings. Final root, stem, and leaf weights for control seedlings did exceed those for combined treated (blighted + clear) seedlings, although not significantly (data not shown).

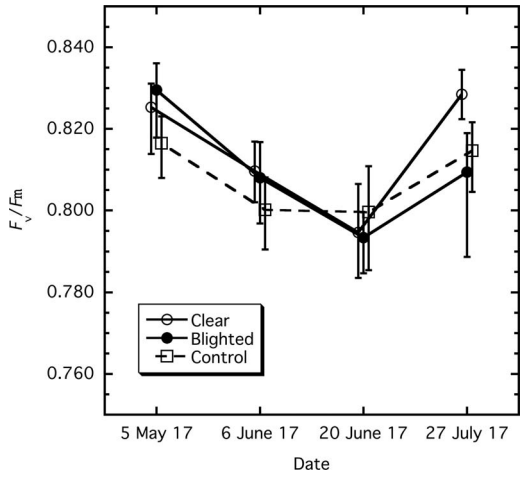

FIG. 4. Dark-adapted chlorophyll fluorescence (F_v/F_m) of seedlings measured on four dates in 2017. Error bars are bootstrapped 95% confidence intervals, and means where error bars do not overlap are considered to be significantly different. There was not a significant treatment × time interaction, $F_{6,180} = 1.995$, $P = 0.0686$, or treatment effect, $F_{2,60} = 1.676$, $P = 0.1957$, but there was a significant time effect, $F_{3,180} = 14.698$, $P < 0.0001$.

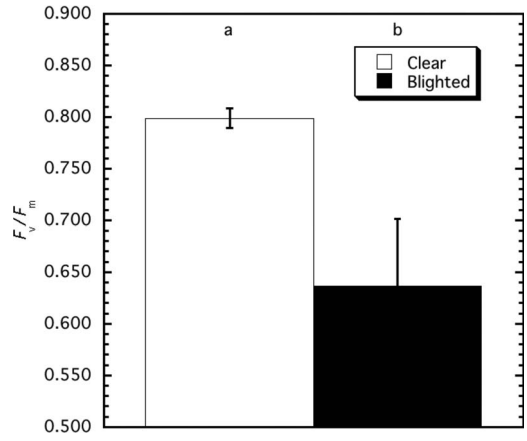

FIG. 5. Comparison of dark-adapted chlorophyll fluorescence (F_v/F_m) of blighted and clear leaves from the blighted group of seedlings, measured on June 20, 2017. Error bars are bootstrapped 95% confidence intervals. A paired-sample Wilcoxon test found a significant difference, $T_{16} = 3$, $P < 0.001$.

Values of RGR for total stem length (Fig. 3B) were similar to those measured by Luken *et al*. (1995) for Amur honeysuckle seedlings that were grown in outdoor shade houses with light levels ranging from 25% to 100% of full sunlight; they also showed a decline in RGR from May through July. Another study by Luken *et al*. (1997b) found that total stem length RGR of honeysuckle seedlings was much greater than that of the native spicebush (*Lindera benzoin* L.) at 25% and 100% full sunlight—RGR of spicebush was roughly 20% to 50% of that of honeysuckle—and they attributed this trait to the success of honeysuckle in outcompeting spicebush, especially when light is less limiting. We found that RGR of blighted seedlings was 60% and 83% of that of clear seedlings in May–June and June–July, respectively; because clear seedlings were grown next to the blighted seedlings in the greenhouse, they are a better group for comparison than the control seedlings. Care must be taken when extrapolating results from greenhouse-grown seedlings to mature plants in the field; however, our results suggest that these blight-induced reductions in RGR may make honeysuckle a less robust competitor than it has been in the past.

Our study shows that honeysuckle leaf blight clearly reduces growth of those seedlings it infects, especially fast-growing ones. We found no evidence of mortality induced by the blight; however, growing conditions for mature plants in the field are quite different than for seedlings in a greenhouse setting. Growth declines under field conditions would certainly reduce the competitive abilities of honeysuckle; it could also potentially lead to the increased mortality observed by Boyce *et al*. (2014) and Boyce (2018) in open-grown stands coinciding with the widespread appearance

Table 1. Spearman rank correlations (r_s) between measurements of dark fluorescence (F_v/F_m) and measurements of RGR. *P* values are given in parentheses. Numbers in bold are statistically significant at the $P \leq 0.05$ level.

F_v/F_m Measurement date	Height	Total stem length	Number of leaves	Leaf area
June 20, 2017	0.201	0.231	**0.332**	**0.252**
	(0.120)	(0.073)	(0.009)	(0.050)
July 27, 2017	**0.391**	**0.359**	**0.314**	**0.406**
	(0.002)	(0.005)	(0.014)	(0.001)

of leaf blight, beginning in 2012. It remains to be seen if the growth declines seen in this study also occur under field conditions in infected mature plants.

Literature Cited

Ali, J., D. Lieurance, and D. Cipollini. 2015. Soil biota affect mycorrhizal infection and growth of *Impatiens capensis* and alter the effects of *Lonicera maackii* rhizosphere extracts. Journal of the Torrey Botanical Society 142: 1–11.

Arthur, M. A., S. R. Bray, C. R. Kuchle, and R. W. McEwan. 2012. The influence of the invasive shrub, *Lonicera maackii*, on leaf decomposition and microbial community dynamics. Plant Ecology 213: 1571–1582.

Bolhàr-Nordenkampf, H. R. and G. Öquist. 1993. Chlorophyll fluorescence as a tool in photosynthesis research, pp. 193–206. *In* D. O. Hall, J. M. O. Scurlock, H. R. Bolhàr-Nordenkampf, R. C. Leegood, and S. P. Long, eds., Photosynthesis and Production in a Changing Environment: A Field and Laboratory Manual. Chapman & Hall, London, UK. 512 pp.

Borth, E. B., K. W. Custer, and R. W. McEwan. 2018. Lethal effects of leaf leachate from the non-native invasive shrub Amur honeysuckle (*Lonicera maackii*) on a model aquatic organism (*Hyalella azteca*). Écoscience 25: 189–197.

Boyce, R. L. 2018. High mortality seen in open-grown, but not forest-understory, Amur honeysuckle (*Lonicera maackii*, Caprifoliaceae) stands in northern Kentucky. Journal of the Torrey Botanical Society 145: 21–29.

Boyce, R. L., R. D. Durtsche, and S. L. Fugal. 2012. Impact of the invasive shrub *Lonicera maackii* on stand transpiration and ecosystem hydrology in a wetland forest. Biological Invasions 14: 671–680.

Boyce, R. L., S. N. Brossart, L. A. Bryant, L. A. Fehrenbach, R. Hetzer, J. E. Holt, B. Parr, Z. Poynter, C. Schumacher, S. N. Stonebraker, M. D. Thatcher, and M. Vater. 2014. The beginning of the end? Extensive dieback of an open-grown Amur honeysuckle stand in northern Kentucky, USA. Biological Invasions 16: 2017–2023.

Chen, H. and S. F. Matter. 2017. Quantification of changes in light and temperature associated with invasive Amur honeysuckle (*Lonicera maackii*). American Midland Naturalist 177: 143–152.

Christopher, C. C. and G. N. Cameron. 2012. Effects of invasive Amur honeysuckle (*Lonicera maackii*) and white-tailed deer (*Odocoileus virginianus*) on litter-dwelling arthropod communities. American Midland Naturalist 167: 256–272.

Cipollini, D., R. Stevenson, and K. Cipollini. 2008a. Contrasting effects of allelochemicals from two invasive plants on the performance of a nonmycorrhizal plant. International Journal of Plant Sciences 169: 371–375.

Cipollini, D., R. Stevenson, S. Enright, A. Eyles, and P. Bonello. 2008b. Phenolic metabolites in leaves of the invasive shrub, *Lonicera maackii*, and their potential phytotoxic and anti-herbivore effects. Journal of Chemical Ecology 34: 144–152.

Collier, M. H., J. L. Vankat, and M. R. Hughes. 2002. Diminished plant richness and abundance below *Lonicera maackii*, an invasive shrub. American Midland Naturalist 147: 60–71.

Cordell, C. E., R. L. Anderson, W. H. Hoffard, T. D. Landis, R. S. Smith, Jr., and H. V. Toko. 1989. Forest nursery pests. Agriculture Handbook No. 680. USDA Forest Service, Washington, DC, USA. 212 pp.

Culley, T., G. N. Cameron, S. E. Kolbe, and A. I. Miller. 2016. Association of non-native Amur honeysuckle (*Lonicera maackii*, Caprifoliaceae) with other invasive plant species in eastern deciduous forests in southwestern Ohio. Journal of the Torrey Botanical Society 143: 398–414.

Dorning, M. and D. Cipollini. 2006. Leaf and root extracts of the invasive shrub, *Lonicera maackii*, inhibit seed germination of three herbs with no autotoxic effects. Plant Ecology 184: 287–296.

[EDDMapS] Early Detection & Distribution Mapping System. 2019. EDDMapS. Retrieved May 16, 2019 from Center for Invasive Species and Ecosystem Health, University of Georgia. <http://www.eddmaps.org>.

Gorchov, D. L. and D. Trisel. 2003. Competitive effects of the invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae), on the growth and survival of native tree seedlings. Plant Ecology 166: 13–24.

Gould, A. M. A. and D. L. Gorchov. 2000. Effects of the exotic invasive shrub *Lonicera maackii* on the survival and fecundity of three species of native annuals. American Midland Naturalist 144: 36–50.

Gould, C. J., Jr. 1945. The parasitism of *Glomerularia lonicerae* (Pk.) D. and H. in *Lonicera* species. Iowa State Coll Journal of Science 19: 301–331.

Hartman, K. M. and B. C. McCarthy. 2004. Restoration of a forest understory after the removal of an invasive shrub, Amur honeysuckle (*Lonicera maackii*). Restoration Ecology 12: 154–165.

Hartman, K. M. and B. C. McCarthy. 2007. A dendroecological study of forest overstorey productivity following the invasion of the non-indigenous shrub *Lonicera maackii*. Applied Vegetation Science 10: 3–14.

Hopfensperger, K. N., R. L. Boyce, and D. Schenk. 2017. Removing invasive *Lonicera maackii* and seeding native plants alters riparian ecosystem function. Ecological Restoration 35: 320–327.

Hutchinson, T. F. and J. L. Vankat. 1997. Invasibility and effects of Amur honeysuckle in southwestern Ohio forests. Conservation Biology 11: 1117–1124.

Kolbe, S. E., A. Towsend-Small, A. I. Miller, T. M. Culley, and G. N. Cameron. 2015. Effect of *Lonicera maackii* on soil carbon and nitrogen in southwestern Ohio forests. Invasive Plant Science and Management 8: 375–384.

Lieurance, D. and D. Cipollini. 2013. Exotic Lonicera species both escape and resist specialist and generalist herbivores in the introduced range in North America. Biological Invasions 15: 1713–1724.

Loomis, J. D. and G. N. Cameron. 2014. Impact of the invasive shrub Amur honeysuckle (*Lonicera maackii*)

on shrub-layer insects in a deciduous forest in the eastern United States. Biological Invasions 16: 89–100.

LOOMIS, J. D., G. N. CAMERON, AND G. W. UETZ. 2014. Impact of the invasive shrub *Lonicera maackii* on shrub-dwelling Araneae in a deciduous forest in eastern North America. American Midland Naturalist 171: 204–218.

LOOMIS, J. D., S. F. MATTER, AND G. N. CAMERON. 2015. Effects of invasive Amur honeysuckle (*Lonicera maackii*) and white-tailed deer (*Odocoileus virginianus*) on survival of sugar maple seedlings in a southwestern Ohio forest. American Midland Naturalist 174: 65–73.

LUKEN, J. O. AND J. W. THIERET. 1996. Amur honeysuckle, its fall from grace. BioScience 46: 18–24.

LUKEN, J. O., L. M. KUDDES, AND T. C. THOLEMEIER. 1997a. Response of understory species to gap formation and soil disturbance in *Lonicera maackii* thickets. Restoration Ecology 5: 229–235.

LUKEN, J. O., L. M. KUDDES, T. C. THOLEMEIER, AND D. M. HALLER. 1997b. Comparative responses of *Lonicera maackii* (Amur honeysuckle) and *Lindera benzoin* (spicebush) to increased light. American Midland Naturalist 138: 331–343.

LUKEN, J. O., T. C. THOLEMEIER, L. M. KUDDES, AND B. A. KUNKEL. 1995. Performance, plasticity, and acclimation of the nonindigenous shrub *Lonicera maackii* (Caprifoliaceae) in contrasting light environments. Canadian Journal of Botany 73: 1953–1961.

MANLY, B. F. J. 2006. Randomization, bootstrap, and Monte Carlo methods in biology, 3rd ed. Chapman & Hall/CRC, London, UK. 480 pp.

MCEWAN, R. W., M. A. ARTHUR, AND S. E. ALVERSON. 2012. Throughfall chemistry and soil nutrient effects of the invasive shrub *Lonicera maackii* in deciduous forests. American Midland Naturalist 168: 43–55.

MCEWAN, R., M. BIRCHFIELD, A. SCHOERGENDORFER, AND M. ARTHUR. 2009. Leaf phenology and freeze tolerance of the invasive shrub Amur honeysuckle and potential native competitors. Journal of the Torrey Botanical Society 136: 212–220.

MCKINNEY, A. AND K. GOODELL. 2010. Shading by invasive shrub reduces seed production and pollinator services in a native herb. Biological Invasions 12: 2751–2763.

MCNEISH, R. E. AND R. W. MCEWAN. 2016. A review on the invasion ecology of Amur honeysuckle (*Lonicera maackii*, Caprifoliaceae) a case study of ecological impacts at multiple scales. Journal of the Torrey Botanical Society 143: 367–385.

MCNEISH, R. E., M. E. BENBOW, AND R. W. MCEWAN. 2012. Riparian forest invasion by a terrestrial shrub (*Lonicera maackii*) impacts aquatic biota and organic matter processing in headwater streams. Biological Invasions 14: 1881–1893.

MCNEISH, R. E., M. E. BENBOW, AND R. W. MCEWAN. 2017. Removal of the invasive shrub, *Lonicera maackii* (Amur honeysuckle), from a headwater stream riparian zone shifts taxonomic and functional composition of the aquatic biota. Invasive Plant Science and Management 10: 232–246.

MEDLEY, K. E. 1997. Distribution of the non-native shrub *Lonicera maackii* in Kramer Woods, Ohio. Physical Geography 18: 18–36.

MILLER, K. E. AND D. L. GORCHOV. 2004. The invasive shrub, *Lonicera maackii*, reduces growth and fecundity of perennial forest herbs. Oecologia 139: 359–375.

PFEIFFER, S. S. AND D. L. GORCHOV. 2015. Effects of the invasive shrub *Lonicera maackii* on soil water content in eastern deciduous forest. American Midland Naturalist 173: 38–46.

[PLANTS] USDA NATURAL RESOURCES CONSERVATION SERVICE. 2019. Welcome to the PLANTS Database | USDA PLANTS. Retrieved May 15, 2019 from U.S. Department of Agriculture. <http://plants.usda.gov/>.

POULETTE, M. M. AND M. A. ARTHUR. 2012. The impact of the invasive shrub *Lonicera maackii* on the decomposition dynamics of a native plant community. Ecological Applications 22: 412–424.

R CORE TEAM. 2019. R: A language and environment for statistical computing. Retrieved March 9, 2018 from R Foundation for Statistical Computing, Vienna, Austria. <https://www.R-project.org/>.

RIFFLE, J. W. AND G. W. PETERSON. 1986. Diseases of trees in the Great Plains. USDA Forest Service General Technical Report RM-129: 1–161.

SHANNON, S. M., J. T. BAUER, W. E. ANDERSON, AND H. L. REYNOLDS. 2014. Plant–soil feedbacks between invasive shrubs and native forest understory species lead to shifts in the abundance of mycorrhizal fungi. Plant and Soil 382: 317–328.

WATLING, J. I., C. R. HICKMAN, E. LEE, K. WANG, AND J. L. ORROCK. 2011. Extracts of the invasive shrub *Lonicera maackii* increase mortality and alter behavior of amphibian larvae. Oecologia 165: 153–159.

ZAR, J. H. 2010. Biostatistical Analysis, 5th ed. Prentice Hall, Upper Saddle River, NJ, USA. 756 pp.