THE UTILITY OF FOUR PROTOCOLS FOR ASSESSING WHITE-TAILED DEER BROWSING SEVERITY AS INDICATORS OF ECOLOGICAL CHANGE: THE NEED FOR VALIDATION AND ACCOUNTABILITY

A Thesis
Presented to the Faculty of the Graduate School of Cornell University
In Partial Fulfillment of the Requirements for the Degree of Master of Science in Natural Resources

by
Brendan Richard Quirion
May 2022
ABSTRACT

Escalating global environmental change resulting from multiple anthropogenic stressors has necessitated the development and use of robust ecological indicators that can accurately and efficiently inform ecosystem management decisions and actions. High deer populations are a primary driver of ecosystem degradation on multiple continents, but proposed indicators of deer impacts largely lack experimental evaluation to determine whether they meet the eight criteria of an ecological indicator defined by Dale and Beyeler (2001). Using a network of large white-tailed deer (Odocoileus virginianus) exclosures established in 2013, we evaluated whether three protocols meant to assess deer browsing severity: Assessing Vegetation Impacts of Deer (AVID), the Ten-tallest protocol, and the Twig Age protocol, met the criteria of an ecological indicator, and whether three common understory herbaceous species: blue-stemmed goldenrod (Solidago caesia), white wood aster (Eurybia divaricata), and zigzag goldenrod (Solidago flexicaulis) could strengthen a partially evaluated Sentinel protocol. We found that all the evaluated protocols could meet at least one of the criteria of an ecological indicator, but an expanded Sentinel protocol had the greatest potential to meet all criteria. Further research is needed to evaluate the ability of these protocols to be anticipatory of and sensitive to variations in deer browsing pressure. Using defined scientific criteria and experimentation to evaluate whether protocols meant to measure deer impacts can serve as robust indicators is necessary to improve deer and ecosystem management decisions, actions, outcomes, and accountability.
BIOGRAPHICAL SKETCH

Brendan Quirion was born in Schenectady, New York and grew up in Schoharie, New York. He attended Schenectady Christian School in Scotia, New York through elementary, middle, and high school, receiving his high school diploma with honors. He then attended the State University of New York (SUNY) at Cobleskill where he majored in Wildlife Management and received his Bachelor of Technology degree in Animal Science (magna cum laude). Following graduation, he worked ten years for the Adirondack chapter of The Nature Conservancy in Keene Valley, New York, first as Terrestrial Invasive Species Project Coordinator and then as Program Director for the award-winning Adirondack Partnership for Regional Invasive Species Management.

Brendan’s passions for deer management and improving outcomes of natural resource management through ecological monitoring eventually led him to pursue a graduate degree at Cornell. Graduate study was made possible through the support of family, friends, and colleagues, but most importantly his loving wife, Brianna Quirion. Brendan now works as the deer and forest management biologist for the New York State Department of Environmental Conservation in the lower Hudson Valley and Catskills region. He currently lives with his wife and son, Luke Quirion, in Freehold, New York.
I dedicate this research thesis to my wife, Brianna Quirion, and to my son, Luke Quirion. Bri, I could not have undertaken graduate study without your love, support, and encouragement. Luke, your arrival brought me so much joy and motivated me to finish strong. I love you both and will continue to work hard to make you proud.
ACKNOWLEDGMENTS

My advisor and Committee Chair, Dr. Bernd Blossey, supported my work, provided this opportunity, and challenged me to reach higher levels of excellence throughout my graduate career. Committee Member, Dr. Paul Curtis, provided additional guidance and support over the course of my research program. Victoria Nuzzo, Audrey Bowe, Dr. Bernd Blossey, and Ben Hopkins helped implement the protocols evaluated in this study and assisted with data collection. Dr. Erika L. Mudrak, Dr. Stacey Endriss, and Dr. Andrea Davalos provided guidance on statistical analysis. All members of Dr. Bernd Blossey’s lab provided feedback and support along the way. To all, I express my deepest gratitude.
# TABLE OF CONTENTS

**BIOGRAPHICAL SKETCH** ................................................................. iii

**DEDICATION** ................................................................................ iv

**ACKNOWLEDGEMENTS** .................................................................. v

**TABLE OF CONTENTS** .................................................................... vi

**LIST OF FIGURES** .......................................................................... vii

**LIST OF TABLES** ........................................................................... viii

**MAIN TEXT**

1. **Introduction** ............................................................................. 1

2. **Methods & Materials** .............................................................. 5

   2.1 **Study Sites & Experimental Design** .................................... 5

   2.2 **Proposed Protocols Meant to Assess Deer Browsing Severity** ...... 6

   2.3 **Sentinel Protocol** ............................................................... 9

   2.4 **Statistical Analysis** ............................................................ 12

3. **Results** ..................................................................................... 14

   3.1 **Proposed Protocols Meant to Assess Deer Browsing Severity** ...... 14

   3.2 **Sentinel Protocol** ............................................................... 21

4. **Discussion** ................................................................................ 27

**REFERENCES** ................................................................................ 39
LIST OF FIGURES

Figure 1: Boxplots of *A. pensylvanicum*, *F. grandifolia*, and *Fraxinus spp.* seedling twig ages (years) in fenced and unfenced plots at two sites, four sites, and one site respectively using the Twig Age protocol…………………………………….. 16

Figure 2: Boxplots of *F. grandifolia*, *Trillium spp.*, and *S. caesia* heights in fenced and unfenced plots at four sites, two sites, and one site respectively using the Ten-tallest and/or AVID protocols……………………………………………………………….. 17

Figure 3: Barplots of number and proportion of flowering *Trillium spp.* individuals in fenced and unfenced plots at two sites using the Ten-tallest and AVID protocols…… 18

Figure 4: Proportion of planted *Q. rubra*, *S. caesia*, *S. flexicaulis*, and *E. divaricata* browsed by deer in unfenced plots at five sites using the Sentinel protocol………… 23

Figure 5: Proportion of planted *Q. rubra*, *S. caesia*, *S. flexicaulis*, and *E. divaricata* attacked by rodents or invertebrates and that survived for one year in fenced and unfenced plots at five sites using the Sentinel protocol……………………………… 24

Figure 6: Proportion of planted *S. caesia* flowering in fenced and unfenced plots at five sites using the Sentinel protocol………………………………………………………….. 25

Figure 7: Line plots of vertical growth of planted *Q. rubra*, *S. caesia*, *S. flexicaulis*, and *E. divaricata* (N = 249 - 336) in fenced and unfenced plots over two years at five sites using the Sentinel protocol………………………………………………………….. 26
LIST OF TABLES

Table 1: Results of linear mixed models for effect of fencing on twig age, plant height, and flowering under four different protocols meant to assess deer browsing severity (Twig age, Ten-tallest, AVID, and Sentinel)......................................................... 19

Table 2: Results of generalized linear mixed models with binomial errors on proportion of planted *Q. rubra, S. caesia, S. flexicaulis,* and *E. divaricata* that were browsed by deer, attacked by rodents and invertebrates, survived for one year, and flowered in fenced and unfenced plots at five sites under the Sentinel protocol, and proportion of *Trillium spp.* that flowered under the AVID protocol....................... 20

Table 3: Ability of four different protocols meant to assess deer browsing severity (AVID, Ten-tallest, Twig Age, and Sentinel) to meet eight criteria established for selecting ecological indicators (Dale and Beyeler 2001)................................. 38
1. Introduction

Terrestrial ecosystems are under escalating stress from climate change, biological invasions, pollution, and habitat loss, challenging the ability of societies to effectively respond (Lovett et al., 2009; Malhi et al., 2020; Simberloff et al., 2013; Wilson, 1989). Ecosystem management and policy must increasingly rely on indicators of change that provide simple and efficient measurements of key conditions, processes, and biota that characterize ecosystem composition, structure, and function (Angermeier and Karr, 1994; Cairns et al., 1993; Karr, 1991, 1981). The challenge is to develop a manageable set of ecological indicators that are: (1) easily measured, (2) sensitive to stresses on the system, (3) respond to stress in a predictable manner, (4) signify an impending change in the ecosystem under consideration that (5) can be averted by management interventions, (6) provide coverage of the key ecosystem conditions and resources that could be compromised, (7) have a known response through time, and (8) have low variability of that response (Dale and Beyeler, 2001). Failure to use a defined and scientifically-rigorous selection process when identifying ecological indicators can result in misguided management and policy decisions and actions (Brice et al., 2021; Dale and Beyeler, 2001).

Early warnings of threats posed by increasing deer (family Cervidae) populations were raised in the late 1800s in Europe, and early 1900s in North America (Leopold, 1933; Hare et al., 2021). Since then, continued extirpations of predators, changes in land use, transitions from market to regulated recreational hunting, intercontinental introductions, and shifts in societal norms and values, have allowed
deer populations to grow to levels considered historically unprecedented and become major drivers of ecosystem degradation on multiple continents (Côté et al., 2004; Nishizawa et al., 2016; Reimoser et al., 1999; Rooney and Waller, 2003). High deer populations exert strong top-down consumptive and non-consumptive effects on vegetation with cascading impacts to many other biota including birds, small mammals, and invertebrates (Côté et al., 2004; Ramirez et al., 2020; Rooney and Waller, 2003; Tilghman, 1989). Legacies of altered plant reproduction and recruitment, soil structure and chemistry, and nutrient dynamics can allow these degraded systems to persist for decades, even after deer populations have been reduced (Harada et al., 2020; Nuttle et al., 2014). Substantial economic and societal impacts from deer-vehicle collisions, agricultural, forestry, and landscape damage, spread of tick-borne diseases, and compromised ecosystem services are also commonplace where high deer populations and human development intersect (Curtis, 2020; Kilpatrick et al., 2014).

In response to voluminous and growing evidence of ecosystem degradation, researchers and managers have developed and promoted a variety of assessment protocols and associated metrics as indicators of deer densities and impacts (Putman et al., 2011). Proposed indicators of deer health, population abundance, and habitat quality, have been developed to link deer and ecosystem management objectives and inform adaptive management decisions in Europe (Chevrier et al., 2012; Morellet et al., 2007; Rhodes and St. Clair, 2018), New Zealand (Sweetapple and Nugent, 2004), Asia (Iijima and Nagaike, 2015), and North America (Bachand et al., 2014; Blossey et al., 2019; Curtis et al., 2021; Frerker et al., 2014; Kraft et al., 2004; Pierson and De
Calesta, 2015; Rawinski, 2018; Royo et al., 2016; Waller et al., 2017; Williams et al., 2000). However, many proposed protocols have not been experimentally evaluated to determine whether they meet the eight criteria of an ecological indicator as defined by Dale and Beyeler (2001) or should be relied upon to accurately inform ecosystem management decisions and actions.

Here we evaluate three proposed protocols and associated metrics: the Twig Age Method (Waller et al., 2017), the Ten-tallest Method (Rawinski, 2018), and Assessing Vegetation Impact from Deer (AVID) (Curtis et al., 2021) to gauge their utility as ecological indicators of white-tailed deer (*Odocoileus virginianus*) browsing severity to forest understory vegetation. Using a network of five sites in central New York State where paired 2-ha plots (one fenced plot paired with an unfenced adjacent plot) have been maintained since 2013, we hypothesized that (1) tree seedlings and herbaceous plants would be taller; (2) flowering of herbaceous plants would be greater; and (3) twig ages would be older in fenced plots when compared to adjacent unfenced plots.

Blossey et al. (2019) previously demonstrated the utility and sensitivity of annually planted red oak (*Quercus rubra*) seedlings as robust indicators of deer browse severity under a Sentinel protocol. This approach documents deer browsing severity over a single growing season to inform near real-time management decisions. *Quercus rubra* is a species of intermediate preference/palatability to deer (Averill et al., 2016) and a portfolio of regionally-relevant plant species is needed to gauge deer browsing severity to more preferred herbaceous species. We therefore assessed whether three herbaceous species (*Solidago caesia*, blue-stemmed goldenrod; *Eurybia*
*divaricata*, white wood aster; and *Solidago flexicaulis*, zigzag goldenrod) could be used to complement *Q. rubra* seedlings as useful indicators of deer browsing severity. We hypothesized that these herbaceous species would (1) be browsed more readily by deer than *Q. rubra* in unfenced plots; (2) be affected similarly by rodents and invertebrates across the fence; and (3) flower more frequently; (4) achieve greater height; and (5) have greater one-year survival in fenced plots when compared to unfenced plots.
2. Methods & Materials

2.1 Study Sites & Experimental Design

We used an established network of paired unfenced and fenced 2-ha plots in five upland, closed-canopy forests in the Finger Lakes Region of central New York State (Bobolink Hill, Ellis Hollow, Polson, Ringwood, and Sapsucker Woods) comprised of maple (*Acer spp.*), American beech (*Fagus grandifolia*), ash (*Fraxinus spp.*), birch (*Betula spp.*), red oak (*Quercus rubra*), shagbark hickory (*Carya ovata*), American hornbeam (*Carpinus caroliniana*), black cherry (*Prunus serotina*), tulip tree (*Liriodendron tulipifera*), eastern white pine (*Pinus strobus*), and eastern hemlock (*Tsuga canadensis*). Within a site, paired plots were established in areas with similar slope, aspect, elevation, canopy cover, and species composition, and were permanently grided into 49 equally-spaced cells (20m x 20m), each marked with a central stake. In 2013, one of each pair of plots was fenced to exclude deer (Trident extruded deer fence, 2.3-m-high fence, DeerBusters.com, Maryland, USA). Fence breeches due to treefalls occurred periodically, allowing deer access for short periods of time. Therefore, we investigated greatly reduced deer browsing by fencing since 2013 rather than complete deer exclusion. A deer breech of the fenced plot at Ellis Hollow in late winter 2019/2020 resulted in browsing damage to most of the understory woody vegetation within. Therefore, only the Sentinel protocol and herbaceous species components of the Ten-tallest and AVID protocols were attempted at this site during the 2020 growing season, and only after fences had been mended.
2.2 Proposed Protocols Meant to Assess Deer Browsing Severity

The Twig Age protocol (Waller et al., 2017) measures time (in years) that a twig of a deciduous tree seedling has grown without being browsed by deer or experienced other sources of twig death. The protocol involves counting the number of terminal bud scale scars extending from the tip of a lateral twig back to a branching point to record its age. Twig ages are recorded along randomly selected straight line or belt transects on two random twigs per seedling and on 50 to 60 seedlings per species (height: 20 - 180 cm). We randomly established two 100m x 2m belt transects (one in each fenced and unfenced plot), using the outside edge of each row of grid cells to avoid walking through sampling locations used for the other protocols. We attempted to perform the Twig Age protocol on seedlings of two different tree species per site, but resorted to utilizing a single species if only one was present in sufficient abundance in both plots. We recorded twig ages before leaf-out in April/May 2020, which allowed for unobstructed visualization of bud scale scars. For seedlings with only a leader but no lateral twigs, we only recorded the number of bud scale scars along the leader.

The Ten-tallest protocol (Rawinski, 2018) records plant heights (≤122 cm) of the 10 tallest stems and the total number of fertile stems, if present, for one or more native or non-native tree or herbaceous species in an unbounded number of permanent or nonpermanent circular (5.64 m radius) sampling locations. Sampling locations and species are typically selected based on availability of vegetation. To meet assumptions of statistical analysis and avoid bias in selection, we attempted to randomly select six sampling locations in each plot using a random number generator assigned to the
center stake for each cell within a plot’s spatial grid. However, we reverted to subjectively selecting locations if the first five random locations did not contain at least 10 stems of a single species in the appropriate size class. We ran a tape measure from each center stake to delineate the circumference of the sampling location and selected species to assess based on which had at least 10 stems present. We measured the height of every stem of the target species to ensure that the 10 tallest were recorded, and documented evidence of flowering or fruiting for each to determine the total number of fertile stems.

AVID (Curtis et al., 2021) tracks heights (15.24 - 152.4 cm) of five to six permanently marked tree seedlings or stems of specific herbaceous plants (i.e. *Trillium spp.*, *Medeola virginiana*, or *Arisaema triphyllum*), and proportion flowering, in six permanent circular (1.83 m radius) sampling locations per site. Because each sampling location selected for the Ten-tallest protocol contained at least 10 stems of a species, we nested all AVID sampling locations within these utilizing the same center stake. We performed the Ten-tallest and AVID protocols for tree seedlings from May through November 2020. Herbaceous species were assessed from June 29 through July 9, 2020 to document flowering.

For each protocol, we recorded time (in minutes) required for two people (a data collector and a data recorder) to implement the protocol in each plot, excluding plots where sampling locations had to be selected subjectively. This included time spent navigating between sampling locations and individual plants as well as evaluating whether randomly selected sampling locations could be utilized. Total time was recorded using a stopwatch when measurements in a plot began and ended. Since
we performed the Ten-tallest and AVID protocols simultaneously using nested sampling locations, we recorded the amount of time required to perform each separately.
2.3 Sentinel Protocol

Blossey et al. (2017, 2019) developed a sentinel protocol using red oak (*Q. rubra*) seedlings to facilitate and standardize assessments, allowing comparisons across sites with different understory plant communities. Individual seedlings are propagated, planted in spring, and tracked for a year to assess survival, growth, browsing by deer, attack by rodents or invertebrates, and overwinter mortality. Cohorts of seedlings are planted annually facilitating near real-time (i.e., same growing season) assessment of changes in deer browsing pressure, for example in response to management activities such as deer culls.

To expand the species portfolio of the Sentinel protocol to include herbaceous plants, we collected seeds of *Q. rubra, S. caesia, E. divaricata,* and *S. flexicaulis* in fall 2019 near Ithaca, New York. These species are favored by deer at our study sites, relatively simple to collect and propagate, and fast growing. For oaks we followed the propagation procedure outlined in Blossey et al. (2017). For herbaceous species we stratified seeds over winter and sowed them into flats of potting soil (BX Mycorrhizae General Purpose Pro-mix; Premier Brands, Riviere-du-Loup, Quebec, Canada) in March 2020. We maintained flats in a greenhouse (12:12 day:night, 20° C) until seedlings produced 2 – 3 true leaves. We then individually transplanted seedlings into 72-cell flats and moved flats outdoors in late May to allow for hardening. We stratified all seedlings by height before planting to ensure initial height distributions were similar for plants across all study sites.

Within each plot, we selected a starting point and used a measuring tape to establish a 100 – 150m transect that did not interfere with other experimental
investigations or the other protocols evaluated in this study. We used a cordless impact
driver (Milwaukee M18 series, milwaukeetool.com) with a wood-boring bit (Speedbor
Max 3.175 cm x 15.24 cm, irwin.com) to create planting holes at 1-m increments
along the transect, excluding unsuitable planting locations (e.g., boulders, blowdowns,
wet depressions, dense vegetation, etc.) that could potentially prevent browsing by
deer or negatively affect plant growth or survival. We planted 20 individuals of each
species from 15 – 24 June 2020 in a repeating pattern (i.e. *Q. rubra*, *S. caesia*, *E.
divaricata*, *S. flexicaulis*; repeat) for a total of 80 individuals per plot. We firmly
planted each seedling, adding surrounding soil to planting holes as needed to ensure
adequate root contact and avoid root desiccation. We pressed a steel framing nail (10.2
cm long) inserted through a steel washer (diameter 2.5 cm or 3.8 cm) into the ground
immediately adjacent to each plant to allow for relocation of planted individuals using
a metal detector (ACE 350, Garret Electronics Inc., Garland, Texas, USA) and to
avoid visual markers that could attract deer or other species (Cohen et al., 2014).
Immediately after planting we recorded plant height (cm).

We revisited all sentinel transects two weeks after planting and replaced eight
individuals that had died due to transplant shock; 2 *Q. rubra*, 2 *E. divaricata*, 3 *S.
caesia*, and 1 *S. flexicaulis*. Thereafter, we revisited transects monthly through October
2020 and recorded plant presence, attack incidence and type, and flowering incidence.
We did not record invertebrate attack at the end of the growing season as elevated
invertebrate attack correlates with plant senescence and tissue decay in the fall. We
recorded overwinter survival in June 2021 based on the presence of live aboveground
plant tissue. We recorded plant heights through the end of August 2021.
We recorded time (in minutes) required for three people to plant each transect and for two people (a data collector and a data recorder) to implement the Sentinel protocol over four subsequent revisits throughout the 2020 growing season. This included time spent navigating between individual plants but did not include time spent propagating species before planting. Total time was recorded using a stopwatch which we started upon the first planting or when measurements began on a transect and ended after the last planting or when measurements ended on that transect.
2.4 Statistical Analysis

We analyzed data in R and considered $P < 0.05$ statistically significant (R Core Team, 2020). For data collected under the Twig Age, Ten-tallest, and AVID protocols, we fitted linear mixed effects models for twig age and plant heights, modeling each species separately and including fencing treatment as a fixed effect. The seedling on which twigs were measured was included as a random effect for twig age. The Ten-tallest/AVID sampling location in which plants were measured was included as a random effect for plant heights. For all linear mixed effects models, site was also included as a random effect if a species could be assessed at all four sites, and as a fixed effect if it could only be assessed at three or fewer sites. We fitted a linear model for the total number of flowering herbaceous plants per sampling location recorded by the Ten-tallest protocol, including fencing and site as fixed effects. We fitted a generalized linear mixed effects model with binomial errors for flowering of *Trillium spp.*, *Medeola virginiana*, or *Arisaema triphyllum* recorded by AVID, including fencing and site as fixed effects and sampling location as a random effect.

For the Sentinel protocol, we first applied data exclusions to restrict our analysis to those plants that would have been available to be browsed by deer and attacked by rodent or invertebrates. We excluded plants that were dug up or had an unverifiable attack type that resulted in premature plant death from all analyses. We also excluded plants that were browsed by deer inside fenced plots due to fence breeches from all analyses except one-year survival. For deer browsing, we excluded plants that remained below browsing height (<5cm) throughout the entire 2020 growing season, were absent after planting, or had compromised leaf tissue that did
not recover by the end of the 2020 growing season. For invertebrate attack, we excluded plants that were absent after planting or had lost their leaves, and that had a lack of vegetative regrowth by the end of the 2020 growing season. For rodent attack, we excluded plants that were absent after planting, and that had a lack of vegetative regrowth by the end of the 2020 growing season. For heights of plants by the end of August 2021, we excluded plants that did not survive to the end of the 2020 growing season or that did not overwinter into 2021.

We fitted a generalized linear mixed effects model with binomial errors for deer browsing incidence of unfenced plants, including species as a fixed effect and site as a random effect. We fitted generalized linear mixed effects models with binomial errors for rodent and invertebrate attack incidence, one-year survival, and flowering of plants, including fencing treatment and species as fixed effects, and site as a random effect. The initial height of each individual plant at planting was also included as a fixed effect for one-year survival. For height of plants by the end of August 2021, we fitted a linear mixed effects model including fencing treatment, species, planting height, and days since planting as fixed effects and site as a random effect.

We used the packages “lmerTest” (Kuznetsova et al., 2017) for linear models and “brglm2” (Kosmidis and Firth, 2021) for generalized linear models. We examined effects of experimental variables by conducting a type III analysis of variance, applying Bonferroni corrections as necessary to account for multiple comparisons. For models with multiple comparisons, we report Bonferroni-corrected $P$-values.
3. Results

3.1 Proposed Protocols Meant to Assess Deer Browsing Severity

*Fagus grandifolia* seedlings were present in sufficient abundance to assess at all sites using all three protocols. Seedlings of *A. pensylvanicum* were available in sufficient abundance at two sites, and *Fraxinus spp.* (most likely *Fraxinus americana*, white ash) at one site to perform the Twig Age protocol. Both *S. caesia* and *Trillium spp.* were present and in sufficient abundance at one and two sites, respectively, to perform the Ten-tallest protocol for herbaceous plants. We also assessed *Trillium spp.* at two sites using the AVID protocol.

We recorded twig ages on seedlings of two tree species at every site except Sapsucker Woods where only *F. grandifolia* could be assessed. The only tree species we could assess through random selection of Ten-tallest/AVID sampling locations was *F. grandifolia*, but we had to reject as many as 14 random sampling locations in a plot before we could identify six with enough seedlings. In only one instance (unfenced plot at Polson) could we establish all six sampling locations from the first six random locations we visited. We had to subjectively select six sampling locations in the fenced plot at Polson after not finding enough seedlings in the first five locations we visited. We also subjectively selected all Ten-tallest/AVID sampling locations for *Trillium spp.* and *S. caesia* to meet the minimum number of plants required per sampling location by each protocol. We could only identify four Ten-tallest/AVID sampling locations with enough *Trillium spp.* plants in both plots at Ringwood Preserve.

For all species, twig ages were significantly older in fenced plots when compared to unfenced plots: *F. grandifolia* ($F = 33.77$, $P = <0.001$), *A. pensylvanicum*


\( F = 16.44, P < 0.001 \), \textit{Fraxinus spp.} (\( F = 46.66, P < 0.001 \)) (Fig. 1, Table 1). \textit{Fagus grandifolia} seedlings were significantly taller in fenced plots when compared to unfenced plots using the AVID protocol (\( F = 8.21, P = 0.006 \)), but not using the Ten-tallest protocol (\( F = 0.68, P = 0.414 \)) (Fig. 2, Table 1). \textit{Trillium spp.} were significantly taller in fenced plots when compared to unfenced plots using both the AVID protocol (\( F = 8.14, P = 0.011 \)) and Ten-tallest protocol (\( F = 17.16, P = 0.001 \)), but we found no significant difference in S. caesia heights across the fence using the Ten-tallest protocol (\( F = 0.27, P = 0.616 \)) (Fig. 2, Table 1). A significantly greater number and proportion of \textit{Trillium spp.} flowered in fenced plots when compared to unfenced plots using the Ten-tallest protocol (\( F = 11.44, P = 0.004 \)) and AVID protocol (\( z = -2.88, P = 0.004 \)) (Fig. 3, Table 1, Table 2). None of the \textit{S. caesia} assessed by the Ten-tallest protocol flowered.

Two people spent on average 91 minutes per plot implementing the Twig Age protocol when two tree species could be assessed, and 59 minutes per plot when only one species could be assessed. Two people spent on average 110 minutes per plot implementing the Ten-tallest protocol for a single species and 62 minutes per plot implementing the AVID protocol for a single species.
Figure 1: Boxplots of *A. pensylvanicum*, *F. grandifolia*, and *Fraxinus spp.* seedling twig ages (years) in fenced and unfenced plots at two sites, four sites, and one site respectively using the Twig Age protocol. Boxplots show medians, ranges, and error bars representing 95% confidence intervals. Data points represent individual twig ages (N = 90 – 500 per species/plot combination). Asterisks indicate statistically significant differences (* = P <0.05, ** = P <0.01, *** = P <0.001) between fenced and unfenced plots.
Figure 2: Boxplots of *F. grandifolia*, *Trillium spp.*, and *S. caesia* heights in fenced and unfenced plots at four sites, two sites, and one site respectively using the Ten-tallest and/or AVID protocols. Boxplots show medians, ranges, and error bars representing 95% confidence intervals. Data points represent individual plant heights (N = 60 – 240 per species/plot combination). Asterisks indicate statistically significant differences (* = P < 0.05, ** = P < 0.01, *** = P < 0.001) between fenced and unfenced plots.
Figure 3: Barplots of number and proportion of flowering *Trillium spp.* individuals in fenced and unfenced plots at two sites using the Ten-tallest and AVID protocols. Asterisks indicate statistically significant differences (* = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$) in the proportion and number of individuals flowering between fenced and unfenced plots.
Table 1: Results of linear mixed models for effect of fencing on twig age, plant height, and flowering under four different protocols meant to assess deer browsing severity (Twig age, Ten-tallest, AVID, and Sentinel). Site is included as a fixed effect if a plant species could be evaluated by a protocol at three or fewer sites and as a random effect if at four or five sites.

<table>
<thead>
<tr>
<th>Response</th>
<th>Predictors</th>
<th>df Num</th>
<th>df Den</th>
<th>Sum sq</th>
<th>Mean sq</th>
<th>F</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Twig age (years)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>F. grandifolia</em>¹</td>
<td>Fencing</td>
<td>1</td>
<td>493</td>
<td>82.8</td>
<td>82.8</td>
<td>33.77</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>A. pensylvanicum</em>²</td>
<td>Fencing</td>
<td>1</td>
<td>228</td>
<td>42.9</td>
<td>42.9</td>
<td>16.44</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>Site</td>
<td>1</td>
<td>228</td>
<td>20.4</td>
<td>20.4</td>
<td>7.83</td>
<td>0.006</td>
</tr>
<tr>
<td>*Fraxinus spp.*²</td>
<td>Fencing</td>
<td>1</td>
<td>119</td>
<td>43.4</td>
<td>43.4</td>
<td>46.66</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>Ten-tallest</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>F. grandifolia</em>³</td>
<td>Fencing</td>
<td>1</td>
<td>46</td>
<td>180.0</td>
<td>180.0</td>
<td>0.68</td>
<td>0.414</td>
</tr>
<tr>
<td>*Height (cm)*³</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>*Trillium spp.*⁴</td>
<td>Fencing</td>
<td>1</td>
<td>17</td>
<td>136.9</td>
<td>136.9</td>
<td>17.16</td>
<td>0.001</td>
</tr>
<tr>
<td>*Height (cm)*⁴</td>
<td>Site</td>
<td>1</td>
<td>17</td>
<td>27.8</td>
<td>27.8</td>
<td>3.49</td>
<td>0.079</td>
</tr>
<tr>
<td>*Trillium spp.*⁴</td>
<td>Fencing</td>
<td>1</td>
<td>116</td>
<td>1125.0</td>
<td>1125.0</td>
<td>11.44</td>
<td>0.004</td>
</tr>
<tr>
<td><strong>Total fertile stems (N)</strong></td>
<td>Site</td>
<td>1</td>
<td>116</td>
<td>45.6</td>
<td>45.6</td>
<td>0.46</td>
<td>0.505</td>
</tr>
<tr>
<td><em>S. caesia</em>⁴</td>
<td>Fencing</td>
<td>1</td>
<td>10</td>
<td>15.8</td>
<td>15.8</td>
<td>0.27</td>
<td>0.616</td>
</tr>
<tr>
<td><strong>AVID - Height (cm)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>F. grandifolia</em>³</td>
<td>Fencing</td>
<td>1</td>
<td>46</td>
<td>6106.8</td>
<td>6106.8</td>
<td>8.21</td>
<td>0.006</td>
</tr>
<tr>
<td><strong>Sentinel - Height (cm)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>by August end 2021⁵</td>
<td>Fencing</td>
<td>1</td>
<td>1206</td>
<td>254.6</td>
<td>254.6</td>
<td>6.59</td>
<td>0.010</td>
</tr>
</tbody>
</table>

¹Includes seedling and seedling within site as random effects
²Includes seedling as a random effect
³Includes sampling location and sampling location within site as random effects
⁴Includes sampling location as a random effect
⁵Includes site as a random effect and species, planting height, and days since planting as fixed effects
Table 2: Results of generalized linear mixed models with binomial errors on proportion of planted *Q. rubra*, *S. caesia*, *S. flexicaulis*, and *E. divaricata* that were browsed by deer, attacked by rodents and invertebrates, survived for one year, and flowered in fenced and unfenced plots at five sites under the Sentinel protocol, and proportion of *Trillium spp.* that flowered under the AVID protocol. Site is included as a fixed effect if a plant species could be evaluated by a protocol at three or fewer sites and as a random effect if at four or five sites.

<table>
<thead>
<tr>
<th>Response</th>
<th>Predictors</th>
<th>Estimates</th>
<th>SE</th>
<th>z</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Sentinel</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Deer browse¹</td>
<td>Intercept (Q. rubra)</td>
<td>-2.22</td>
<td>0.43</td>
<td>-5.22</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td><em>S. caesia</em></td>
<td>1.18</td>
<td>0.44</td>
<td>2.72</td>
<td>0.007</td>
</tr>
<tr>
<td></td>
<td><em>S. flexicaulis</em></td>
<td>-0.14</td>
<td>0.51</td>
<td>-0.27</td>
<td>0.789</td>
</tr>
<tr>
<td></td>
<td><em>E. divaricata</em></td>
<td>0.83</td>
<td>0.50</td>
<td>1.66</td>
<td>0.098</td>
</tr>
<tr>
<td>Rodent attack¹</td>
<td>Intercept (Fenced, Q. rubra)</td>
<td>-2.84</td>
<td>0.74</td>
<td>-3.84</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>Unfenced</td>
<td>-17.29</td>
<td>40.48</td>
<td>-0.43</td>
<td>0.669</td>
</tr>
<tr>
<td></td>
<td><em>S. caesia</em></td>
<td>0.76</td>
<td>0.45</td>
<td>1.68</td>
<td>0.094</td>
</tr>
<tr>
<td></td>
<td><em>S. flexicaulis</em></td>
<td>1.15</td>
<td>0.45</td>
<td>2.57</td>
<td>0.010</td>
</tr>
<tr>
<td></td>
<td><em>E. divaricata</em></td>
<td>1.70</td>
<td>0.44</td>
<td>3.90</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>Unfenced * S. caesia</td>
<td>16.39</td>
<td>40.48</td>
<td>0.41</td>
<td>0.686</td>
</tr>
<tr>
<td></td>
<td>Unfenced * S. flexicaulis</td>
<td>15.89</td>
<td>40.48</td>
<td>0.39</td>
<td>0.695</td>
</tr>
<tr>
<td></td>
<td>Unfenced * E. divaricata</td>
<td>16.57</td>
<td>40.48</td>
<td>0.41</td>
<td>0.682</td>
</tr>
<tr>
<td>Invertebrate attack¹</td>
<td>Intercept (Fenced, Q. rubra)</td>
<td>-2.45</td>
<td>0.56</td>
<td>-4.41</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>Unfenced</td>
<td>0.54</td>
<td>0.46</td>
<td>1.16</td>
<td>0.245</td>
</tr>
<tr>
<td></td>
<td><em>S. caesia</em></td>
<td>-0.67</td>
<td>0.56</td>
<td>-1.21</td>
<td>0.228</td>
</tr>
<tr>
<td></td>
<td><em>S. flexicaulis</em></td>
<td>2.33</td>
<td>0.43</td>
<td>5.43</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td><em>E. divaricata</em></td>
<td>1.99</td>
<td>0.43</td>
<td>4.59</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>Unfenced * S. caesia</td>
<td>-0.69</td>
<td>0.79</td>
<td>-0.87</td>
<td>0.383</td>
</tr>
<tr>
<td></td>
<td>Unfenced * S. flexicaulis</td>
<td>-1.04</td>
<td>0.57</td>
<td>-1.83</td>
<td>0.068</td>
</tr>
<tr>
<td></td>
<td>Unfenced * E. divaricata</td>
<td>0.71</td>
<td>0.58</td>
<td>1.22</td>
<td>0.222</td>
</tr>
<tr>
<td>One-year survival¹</td>
<td>Intercept (Fenced, Q. rubra)</td>
<td>1.12</td>
<td>0.49</td>
<td>2.27</td>
<td>0.023</td>
</tr>
<tr>
<td></td>
<td>Unfenced</td>
<td>-0.32</td>
<td>0.39</td>
<td>-0.84</td>
<td>0.401</td>
</tr>
<tr>
<td></td>
<td><em>S. caesia</em></td>
<td>0.27</td>
<td>0.47</td>
<td>0.56</td>
<td>0.576</td>
</tr>
<tr>
<td></td>
<td><em>S. flexicaulis</em></td>
<td>-0.49</td>
<td>0.38</td>
<td>-1.31</td>
<td>0.192</td>
</tr>
<tr>
<td></td>
<td><em>E. divaricata</em></td>
<td>-1.33</td>
<td>0.36</td>
<td>-3.69</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>Planting Height</td>
<td>0.06</td>
<td>0.03</td>
<td>2.13</td>
<td>0.033</td>
</tr>
<tr>
<td></td>
<td>Unfenced * S. caesia</td>
<td>-0.80</td>
<td>0.58</td>
<td>-1.39</td>
<td>0.164</td>
</tr>
<tr>
<td></td>
<td>Unfenced * S. flexicaulis</td>
<td>0.06</td>
<td>0.50</td>
<td>0.12</td>
<td>0.903</td>
</tr>
<tr>
<td></td>
<td>Unfenced * E. divaricata</td>
<td>-0.51</td>
<td>0.49</td>
<td>-1.04</td>
<td>0.297</td>
</tr>
<tr>
<td><em>S. caesia</em> flowering¹</td>
<td>Intercept (Fenced)</td>
<td>-0.01</td>
<td>0.40</td>
<td>-0.03</td>
<td>0.973</td>
</tr>
<tr>
<td></td>
<td>Unfenced</td>
<td>0.25</td>
<td>0.31</td>
<td>0.82</td>
<td>0.413</td>
</tr>
<tr>
<td><strong>AVID</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Trillium spp.</em> flowering²</td>
<td>Intercept (Fenced, Bobolink Hill)</td>
<td>-1.38</td>
<td>1.05</td>
<td>-1.31</td>
<td>0.190</td>
</tr>
<tr>
<td></td>
<td>Unfenced</td>
<td>-4.65</td>
<td>1.62</td>
<td>-2.88</td>
<td>0.004</td>
</tr>
<tr>
<td></td>
<td>Ringwood</td>
<td>1.34</td>
<td>1.30</td>
<td>1.03</td>
<td>0.301</td>
</tr>
</tbody>
</table>

¹Model includes site as a random effect
²Model includes sampling location as a random effect
3.2 Sentinel Protocol

Tree falls on fences allowed deer access at Ellis Hollow and Ringwood during the 2020 and 2021 growing seasons, resulting in browsing damage to 3 S. caesia, 12 S. flexicaulis, and 1 E. divaricata planted at Ellis Hollow and 11 S. caesia, 9 S. flexicaulis, 4 E. divaricata, and 10 Q. rubra planted at Ringwood. We excluded these individuals from our analyses, except for one-year survival, as they represented a failure of our fencing treatment. We did not observe evidence of recent deer browsing to plants measured within fenced plots under the Twig Age, Ten-tallest, or AVID protocols at any of our study sites since data collection for these protocols occurred prior to these fence breeches.

A significantly greater proportion of planted S. caesia were browsed by deer when compared to planted Q. rubra in unfenced plots ($z = 2.72, P = 0.007$). There was no significant difference in the proportion of planted S. flexicaulis ($z = -0.27, P = 0.789$) or E. divaricata ($z = 1.66, P = 0.098$) browsed by deer in comparison to planted Q. rubra in unfenced plots (Fig. 4, Table 2). Solidago flexicaulis was attacked significantly more by rodents in fenced plots when compared to unfenced plots ($z = 2.92, P = 0.013$) with all other species planted showing a consistent, but non-significant pattern of rodent attack (Fig. 5, Table 2). A significantly greater proportion of planted E. divaricata were attacked by invertebrates in unfenced plots when compared to fenced plots ($z = -3.53, P = 0.002$), but patterns of invertebrate attack for other planted species were not consistent or significant (Fig. 5, Table 2). Solidago caesia ($z = 2.62, P = 0.035$) and E. divaricata ($z = 2.78, P = 0.005$) had significantly greater survival in fenced plots when compared to unfenced plots, with planted Q.
rubra and S. flexicaulis showing a consistent, but non-significant pattern of one-year survival (Fig. 5, Table 2). Only S. caesia flowered in sufficient abundance for analysis, but there was no significant difference in the proportion that flowered between fenced and unfenced plots ($z = 0.82, P = 0.413$) (Fig. 6, Table 2). Solidago caesia planted in fenced plots were significantly taller than S. caesia planted in unfenced plots by the end of August 2021 ($t = 4.50, P < 0.001$), but there was no significant difference in heights of the other species planted across the fence by the end of August 2021 (Fig. 7, Table 1).

Three people spent on average 78 minutes per plot planting each transect for the Sentinel protocol. Two people spent on average 112 minutes per plot implementing the Sentinel protocol for four species over a single growing season.
Figure 4: Proportion of planted *Q. rubra*, *S. caesia*, *S. flexicaulis*, and *E. divaricata* browsed by deer in unfenced plots at five sites using the Sentinel protocol. Numbers above individual bars represent the number of plants browsed out of the pool of available individuals (see text for details). Asterisks indicate statistically significant differences (* = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$) in proportion of individuals browsed between herbaceous species and *Q. rubra*. 
Figure 5: Proportion of planted *Q. rubra*, *S. caesia*, *S. flexicaulis*, and *E. divaricata* attacked by rodents or invertebrates and that survived for one year in fenced and unfenced plots at five sites using the Sentinel protocol. Numbers above individual bars represent the number of individuals attacked or surviving out of the pool of available individuals (see text for details). Asterisks indicate statistically significant differences (* = *P* < 0.05, ** = *P* < 0.01, *** = *P* < 0.001) in proportion of individuals attacked or surviving between fenced and unfenced plots.
Figure 6: Proportion of planted *S. caesia* flowering in fenced and unfenced plots at five sites using the Sentinel protocol. Numbers above individual bars represent the number of plants flowering out of the pool of available individuals (see text for details). Asterisks indicate statistically significant differences (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$) in proportion of individuals flowering between fenced and unfenced plots.
Figure 7: Line plots of vertical growth of planted *Q. rubra*, *S. caesia*, *S. flexicaulis*, and *E. divaricata* (N = 249 - 336) in fenced and unfenced plots over two years at five sites using the Sentinel protocol. Dashed lines represent changes in height of individual plants. Solid, colored lines represent mean changes in height for each species in fenced and unfenced plots. Asterisks indicate statistically significant differences (* = P < 0.05, ** = P < 0.01, *** = P <0.001) in mean height (cm) between fenced and unfenced plots for individual species by the end of August 2021.
4. Discussion

Our results offer reason for caution when selecting a particular protocol to assess deer browsing severity to inform deer and ecosystem management decisions. None of the protocols we evaluated met all the criteria of an ecological indicator as defined by Dale and Beyeler (2001), casting doubt on whether these protocols can serve as robust and reliable indicators of deer browsing severity in all contexts. The Twig Age protocol met five of the criteria of an effective ecological indicator, while the AVID and Ten-tallest protocols met three and two of the criteria respectively. An expanded Sentinel protocol that incorporates a portfolio of regionally-relevant tree and herbaceous species has potential to meet all the criteria of an ecological indicator of deer browsing severity, but the ability of all sentinel species to be anticipatory, and new sentinel species to be sensitive to changes in deer browse severity, requires further investigation.

We were able to readily implement the Twig Age protocol within randomly-selected transects, record information for multiple tree species at all but one site, and achieve statistically robust sample sizes. The twig age metric consistently and predictably detected reductions in deer browsing severity across the fence and produced data distributions with low variance for all tree species utilized, reinforcing the findings of others (Waller, 2018; Waller et al., 2017). However, because the protocol could only be utilized on seedlings of deciduous tree species, it does not meet the criteria of integration because it cannot measure deer browsing severity to herbaceous or evergreen tree species. We were also unable to record twig ages for the same tree species across all our study sites, except for *F. grandifolia*, which could
limit standardization of assessments to inform management decisions beyond the site scale (Frerker et al., 2013). Because twig tip death can originate from a variety of sources other than deer browsing (Franklin et al., 1987), the twig age metric may also be susceptible and sensitive to natural background variation, but we did not detect evidence of this. Therefore, the Twig Age protocol met five of the criteria of an ecological indicator: easily measured, responds to stress in a predictable manner, predicts changes that can be averted by management actions, has a known response, and has low variability in the response.

We were able to implement the Ten-tallest protocol for *F. grandifolia* seedlings at every site, but had to reject numerous randomly selected sampling locations, and in one plot resorted to subjective selection of locations before identifying six with enough seedlings. We had to establish all sampling locations for herbaceous species subjectively and could not establish any sampling locations at one site due to a low abundance of herbaceous species. When many plants of similar height were present, we had to measure each to identify the 10 tallest. This became tedious, especially for herbaceous species when there could be hundreds of individual plants present in a single sampling location. The protocol did not detect a significant difference in the tallest *F. grandifolia* or *S. caesia* across the fence (Fig. 2). However, the tallest *Trillium spp.* and total flowering appeared responsive to reductions in deer browsing severity when *Trillium spp.* were present and could be assessed (Fig. 2, Fig. 3), reinforcing the findings of others (Anderson, 1994; Rooney and Gross, 2003). We could find no evidence that a consistent pattern of response for the ten-tallest metric had been previously established in the peer-reviewed scientific literature, but the
metric produced data distributions with low variance for all species utilized. Therefore, the Ten-tallest protocol met two of the criteria of an ecological indicator: integrative, because it could record information for both trees and herbaceous species at all but one of our sites, and low variability in the response.

We faced similar challenges with randomly establishing sampling locations for the AVID protocol, especially for herbaceous species. We emphasize that none of the herbaceous species targeted by the AVID protocol (Trillium spp., Medeola virginiana, or Arisaema triphyllum) were present in sufficient abundance to randomly establish sampling locations at any of our study sites. These species are highly preferred by deer and have likely been eliminated or substantially reduced from many forests across eastern North America, handicapping the AVID protocol’s ability to record deer browsing severity to herbaceous vegetation (Anderson, 1994, Dávalos et al., 2014, Frerker et al., 2014, Begley-Miller et al., 2014). When Trillium spp. were present, we struggled to establish the required number of sampling locations, even when attempting to do so subjectively. The protocol could not meet the criteria of being easily measured or integrative because of these challenges. However, the AVID protocol detected significant differences in F. grandifolia and Trillium spp. heights, and Trillium spp. flowering across the fence with low variability in these response metrics (Fig. 2, Fig. 3). A consistent pattern of response for heights of tree seedlings measured in paired plots using AVID has been previously established (Curtis et al., 2021), but that assessment utilized subjective selection of sampling locations, and we could find no evidence that the herbaceous species that the protocol targets have been similarly evaluated. Finally, T. grandiflorum and T. erectum cannot be reliably
identified to species unless flowers or fruits are present, and as a result the AVID protocol often combines performance and demographic parameters for both species when present which may yield inconclusive or misleading results (Dávalos et al., 2014; Rooney and Gross, 2003). Therefore, the AVID protocol met three of the criteria of an ecological indicator: responds to stress in a predictable manner, predicts changes that can be averted by management actions, and has low variability in the response.

Deer have been filtering plant communities across eastern North America over the past century and impacts to vegetation at our study sites are chronic rather than incipient (Begley-Miller et al. 2014, Leopold, 1933; Miller and McGill, 2019). Therefore, we could not evaluate whether any of the protocols could meet the criterion of being anticipatory (i.e., signify an impending change in key characteristics of the ecological system). We could also not evaluate whether the proposed protocols were sensitive to variations in deer browsing severity because our study did not directly manipulate and measure deer abundance. Future studies focused in areas where deer are likely to expand their ranges due to climate change (Dawe and Boutin, 2016; Harsch et al., 2009), or have been newly introduced (Casabon and Pothier, 2008), and utilizing experimental deer enclosures (Horsley et al., 2003), or known population variation resulting from differences in management intensity (Blossey et al., 2019) could help determine whether these protocols or others meet these two criteria.

One of the greatest concerns we have with the AVID and Ten-tallest protocols is how prone both are to sampling bias. Brice et al. (2021) found that targeting only the tallest plants overestimated regeneration of overstory aspen (*Populus tremuloides*)
by a factor of 4-7 compared to random sampling because this practice favors plants that are over the preferred browsing height of ungulates. We believe this to be the primary reason why the AVID protocol could detect significant differences in \( F.\) \textit{grandifolia} heights across the fence when the Ten-tallest protocol could not, because the AVID protocol records heights from a more representative sample of plants (Fig. 2). However, we also violated basic sampling principles for both protocols when we had to establish sampling locations subjectively based on the availability of vegetation, which has the potential to produce fundamentally flawed and misleading results (Brice et al., 2021).

The AVID protocol recommends that sampling be conducted in forests with less than 50\% canopy closure, which increases the amount of vegetation available for sampling thereby decreasing the likelihood that sampling locations need to be selected subjectively (Curtis et al., 2021). Our results were more indicative of how the AVID protocol would perform in closed-canopy forests where light limitations and chronic browsing by deer have substantially reduced the amount of vegetation available in the understory for sampling as well as plant growth rates (Alverson et al., 2019; Nagy et al., 2022). Seedlings of gap-phase or shade-tolerant species may not exhibit substantial increases in height until a canopy opening occurs due to tradeoffs in resource allocations required for growth versus survival in the understory (Seiwa, 2007). The AVID protocol’s 50\% canopy closure recommendation will likely limit where the protocol can be effectively implemented, and its ability to inform regional deer management decisions, especially in forests that are not prone to large-scale natural disturbance events or where timber harvest is limited or prohibited (Brown et al.,
2018; Masek et al., 2013). Because the twig age metric also accounts for small changes in horizontal twig expansion, such as in response to sunflecks on the forest floor, we suspect, and our results suggest, that it is a more sensitive metric than plant heights in closed-canopy forest settings (Fig. 1).

There may be little added value in monitoring deer browsing severity using *F. grandifolia, A. pensylvanicum*, and *Fraxinus spp.* when we consider the primary goals of forest management. These species are often considered weed trees by foresters, have low economic value, can inhibit the regeneration of more desirable tree species, and are prone to pests and disease (Busby and Canham, 2011; Forrester et al., 2003; Herms and McCullough, 2014; Nyland et al., 2006). If these are the only species available to be monitored by a particular assessment protocol, as was the case for the three protocols we evaluated, then that information is likely of little relevance to forest managers because these species are more likely to be managed against than for even if they express increased growth following reductions in deer browsing severity. It may also be impossible to deduce a forest’s trajectory and ecosystem integrity using only the vegetation that remains after chronic browsing by deer, especially if all that remains are species considered to be of low palatability to deer or the system has transitioned to an alternative stable state (Augustine et al., 1998; Healy, 1971; Royo et al., 2017). These challenges compound the limitations of protocols meant to assess deer browsing severity that rely on existing vegetation.

A previous assessment of the Sentinel protocol using planted *Q. rubra* was found to meet most of the criteria of an ecological indicator, even the criterion of sensitivity to variations in deer abundance and browsing severity (Blossey et al.,
However, the protocol could not yet integrate information on browsing severity to herbaceous vegetation more highly preferred by deer. Through our evaluation of planted herbaceous species that could complement *Q. rubra*, we identified *S. caesia* as a potentially viable ecological indicator of deer browsing severity to herbaceous vegetation. Of all the species planted, unfenced *S. caesia* were browsed significantly more than unfenced *Q. rubra* (Fig. 4), and fenced *S. caesia* grew significantly taller by the end of August 2021 and had significantly greater one-year survival than unfenced *S. caesia* (Fig. 5, Fig. 7). It was the only planted herbaceous species to flower in sufficient quantities for analysis and its high one-year survival rates may provide opportunity to monitor the same individuals over multiple growing seasons as opposed to replanting annually. We suspect that significant differences in these metrics would have been even more prevalent if we had not had to exclude individuals from analysis due to fence breeches by deer at two of our study sites. We encourage further assessment of *S. caesia* to evaluate its ability to be an anticipatory and sensitive ecological indicator of deer browsing severity. Neither planted *E. divaricata* nor *S. flexicaulis* showed as much promise.

Our observations of increased one-year survival and prevalence of rodent attack for all species planted inside fenced plots suggests that deer may also be having indirect, non-consumptive effects at our study sites (Rooney and Waller, 2003). Elevated or chronic browsing by deer can modify rates of leaf litter accumulation, decomposition, and nutrient cycling as well as reduce available resources for plants and small mammals (Bardgett and Wardle, 2003; Dávalos et al., 2015; Nuttle et al., 2011; Ramirez et al., 2020). This has important implications for higher trophic levels,
especially mesopredators, birds of prey, and reptile communities, which remain largely unevaluated (Côté et al., 2004). These findings also suggest that the Sentinel protocol could detect changes in ecological conditions beyond vegetation, whereas the other protocols we evaluated could not. Combined, our results suggest that an expanded Sentinel protocol that incorporates a portfolio of regionally-relevant tree and herbaceous species meets the criterion of integration and may provide a greater measure of coverage of key ecological gradients beyond vegetation.

A primary criticism of the Sentinel protocol has been the time and effort required to propagate, plant, and assess the fate of seedings semi-monthly throughout a growing season (Curtis et al., 2021). Contrary to these concerns, our results suggest that the Sentinel protocol could be more efficient in closed-canopy forests, and on an effort-per-species basis, than both the Ten-tallest and AVID protocols. When the basic principles of random sampling were adhered to, sampling locations established under the AVID and Ten-tallest protocols could only capture information for a single species at all our study sites. Although we did not record the amount of time required to propagate the seedlings used in this study, seedlings can be purchased from local suppliers, or mass-reared by existing nurseries administered by management agencies to limit the time and effort required to advance the Sentinel protocol for multiple species.

We cannot recommend the Ten-tallest protocol to managers or policymakers for assessing deer browsing severity because the protocol did not consistently detect greatly reduced deer browsing pressure inside our fenced plots when compared to unfenced plots. The Ten-tallest protocol also inherently promotes sampling bias by
targeting the tallest plants in sampling locations that are selected subjectively based on availability of vegetation, and required the most time to implement in the field. The AVID protocol may have utility in open canopy forests or where tree regeneration is abundant, but will be difficult to implement through random sampling in closed canopy forests, especially where chronic browsing by deer has greatly reduced the number of tree seedlings and herbaceous species the protocol targets. The herbaceous species targeted by the AVID protocol have not been fully validated to confirm that they can serve as indicators of deer browsing severity and often require subjective sampling when present which will also limit the protocol’s ease of use, reliability, and scalability. The Twig Age protocol is likely to perform well in both open and closed canopy forests because it utilizes transects which capture enough vegetation to avoid subjective sampling, and a metric that responds even when vertical growth of seedlings is resource limited. The Twig Age protocol’s primary limitation is in not being able to record information on deer browsing severity to herbaceous vegetation or in areas where tree seedlings are depauperate.

Data generated from protocols that utilize existing vegetation will be difficult to standardize across sites with differing vegetation composition. Because the Sentinel protocol relies on planted seedlings rather than existing vegetation, it can be readily implemented in any context where browsing by deer is occurring, even areas completely devoid of understory vegetation, and standardized across sites. However, additional research is needed to identify and validate a portfolio of regionally-relevant sentinel species that can be used across the white-tailed deer’s range. Mass-rearing and dissemination of sentinel seedlings to volunteers or management agency staff will also
require additional funding, logistics, and oversight. Dedicated and concerted funding generated from excise taxes on purchases of outdoor recreation related equipment, habitat/access stamps, or hunting license sales could facilitate validation and implementation of protocols meant to assess deer browsing severity to inform deer management decisions at regional scales.

Viewing our results through the lens of an effective ecological indicator magnifies the importance of using defined scientific criteria and experimentation to evaluate any protocol and associated metrics proposed to measure deer impacts (Table 3). The protocols we evaluated represent a small fraction of the number that currently exist and are being utilized to inform deer and ecosystem management decisions across North America. Many have been touted for their simplicity and effectiveness, and some have begun to be incorporated into management plans, without having passed this critical evaluation. This is a major disservice to deer and ecosystem management and policy which increasingly must rely on information generated from such protocols. Echoing the concerns of others (Curtis, 2020; Waller et al., 2017), we caution management agencies to avoid hastily adopting protocols that have not been thoroughly evaluated as this could result in misguided management decisions and actions. By allocating and prioritizing funding for additional experimental evaluations, management agencies could facilitate the expanded use of protocols for deer impact assessment while ensuring that the products and information they generate are accurate and relevant from a deer and ecosystem management context.

Deer management is in desperate need of increased scientific validity, accountability, and tools to inform local, real-time, and adaptive management
decisions. Efforts to advance ecological, economic, and societal objectives through deer management remain largely disconnected and antagonistic in practice (Beguin et al., 2016; DeCalesta and Stout, 1997; Mcshea, 2012); ecological data with high spatial and temporal resolution to inform deer management decisions remains limited (Lesser et al., 2019; Miller and McGill, 2019); and challenges and costs associated with censusing cryptic deer populations requires the development and adoption of alternative impact-monitoring approaches and metrics (Abolaffio et al., 2019; Putman et al., 2011). The expanded use of protocols for assessment of deer impacts holds much promise to address these challenges. However, that promise will only be realized if the protocols utilized can stand up to the scrutiny of experimental evaluation to determine whether they meet the criteria of a robust and reliable indicator of deer impacts.
Table 3: Ability of four different protocols meant to assess deer browsing severity (AVID, Ten-tallest, Twig Age, and Sentinel) to meet eight criteria established for selecting ecological indicators (Dale and Beyeler 2001). (✓) indicates that criteria is met, (−) indicates that criteria is not met, and (?) indicates that criteria has not been evaluated.

<table>
<thead>
<tr>
<th>Indicator selection criteria</th>
<th>AVID</th>
<th>Ten-tallest</th>
<th>Twig Age</th>
<th>Sentinel</th>
</tr>
</thead>
<tbody>
<tr>
<td>Easily measured¹</td>
<td>-</td>
<td>-</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Sensitive to stresses on system²</td>
<td>?</td>
<td>?</td>
<td>?</td>
<td>✓*</td>
</tr>
<tr>
<td>Responds to stress in predictable manner³</td>
<td>✓</td>
<td>-</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Anticipatory⁴</td>
<td>?</td>
<td>?</td>
<td>?</td>
<td>?</td>
</tr>
<tr>
<td>Predicts changes that can be averted⁵</td>
<td>✓</td>
<td>-</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Integrative⁶</td>
<td>-</td>
<td>✓</td>
<td>-</td>
<td>✓</td>
</tr>
<tr>
<td>Known response⁷</td>
<td>-</td>
<td>-</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Low variability of response⁸</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
</tbody>
</table>

¹Easy to understand, simple to apply, scientifically sound, easily documented, & cost-effective
²Displays high sensitivity to deer browse and limited sensitivity to natural variation
³Unambiguous and predictable
⁴Measurable before substantial change in ecological system occurs
⁵Relevant to management
⁶Provides a measure of coverage of key gradients across ecological systems
⁷Extensively studied with clearly established pattern of response established in scientific literature
⁸Small range in response allows changes to be distinguished from background variability

*Criterion of sensitivity of *Q. rubra* met (Blossey et al., 2019), but other species require assessment
REFERENCES


Dawe, K.L., Boutin, S., 2016. Climate change is the primary driver of white-tailed deer (Odocoileus virginianus) range expansion at the northern extent of its range; land use is secondary. Ecol. Evol. 6, 6435–6451. https://doi.org/10.1002/ece3.2316


