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Effects of acorn size and mass on seedling quality of northern red oak (*Quercus rubra*)

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Abstract Oaks are not sustainable in many upland temperate forests because of poor recruitment resulting from natural regeneration. Artificial regeneration is an alternative to natural regeneration, but is difficult, in part, due to large variation in seedling quality. In this study, we examined the effects of acorn size and mass on nursery seedling morphological parameters commonly used to quantify seedling quality, and we determined if genetic factors affected these relationships. Acorns were collected from six open-pollinated orchard trees (i.e., six half-sib families), and were separated into six size classes based on acorn diameter (ranging from 1.3 to 2.5 cm). Samples from each size class were weighed for total fresh mass. Acorns were sown in a commercial bareroot nursery in Polk County, Tennessee, USA, and seedlings were grown for 1 year using nursery protocols to maximize growth. Seedling survival was generally not affected by acorn size class or mass, except one family had higher survival in the larger acorn size classes. Five of the six families had no discernable relationship between acorn size class and seedling size. Acorn mass was positively related to seedling morphology, but relationships were weak ($R^2 \leq 0.11$) and biologically insignificant. Neither acorn size nor mass could be used reliably to predict seedling survival or morphological indicators of seedling quality. We hypothesized that results were affected by an unusually long growing season and advanced fertilization regimes at the nursery, which may have negated acorn size/mass effects on seedling growth. Family affected relationships between acorn size/mass and seedling morphology, indicating that family selections could improve overall seedling quality.

Keywords Artificial regeneration · Half-sib family · Seedling morphology · Bareroot seedling production

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Introduction

Artificial regeneration offers a viable alternative to natural seedling establishment when natural reproduction is absent or does not progress into desired crown positions. Oak (*Quercus* L.) reproduction, in particular, must be present and in a competitive position to become a significant component of the next stand following overstory removal, but this is rarely achieved on moderately to highly productive sites (Johnson et al. 2009). Successive disturbances, such as prescribed burning or herbicide, applied appropriately through time, are required for oak to accumulate in the understory and recruit into the canopy (Arthur et al. 2012). Changes in disturbance regimes, exotic pests and pathogens, and climate change are contributing to oak decline and mortality throughout much of the world (Haavik et al. 2015). These changes, coupled with oak regeneration failures, require novel and refined approaches utilizing artificial regeneration and genetics to maintain or restore species such as oak (Dey et al. 2008; Jacobs et al. 2015; Potter et al. 2017).

Northern red oak (*Quercus rubra* L.) is one of the most desired eastern North American oak species due to its high ecological and utilitarian values, but has been difficult to regenerate through planting (Dey et al. 2008). Northern red oak has exhibited large variability in seedling morphology within and among half-sib families, relatively slow growth of seedlings, and low heritability of desirable seedling traits (Schlarbaum and Bagley 1981; Kriebel et al. 1988; Kormanik et al. 1998). While larger seedlings are recommended for planting on productive sites where competition is the primary limiting factor, nursery prescriptions to produce relatively uniform and large northern red oak seedlings have not been widely tested, particularly for plantings on productive sites in the southern Appalachian region (Dey et al. 2012). Advancements in fertilization and irrigation regimes have improved average size of bareroot northern red oak seedlings (Kormanik et al. 1994; Dey et al. 2008), but variability in seedling quality is still large (Kormanik et al. 1998; Clark et al. 2000; Wilson and Jacobs 2006).

Acorn mass has been positively related to survival and seedling size for many oak species tested on a variety of sites throughout the world (González-Rodríquez et al. 2011; Landergott et al. 2012; Yi et al. 2015), with limited exceptions that were attributed to unknown genetic effects (Long and Jones 1996). Acorn mass is largely associated with the cotyledons, which supply carbohydrate reserves necessary for seedling development after germination, particularly for the root system (Korstian 1927; Jarvis 1963). Acorn mass has been strongly related to acorn size (Ramírez-Valiente et al. 2009), which can be differentiated on a commercial scale with the goal of improving overall seedling size. Previous researchers have used relative acorn size classes (e.g., small, medium, large) and found that acorns from smaller size classes produced smaller seedlings with lower survival than acorns from larger size classes (Kormanik et al. 1998; Navarro et al. 2006; Tilki 2010). Relative size or mass can vary by family, populations, or provenance (González-Rodríquez et al. 2011; Anagiotos et al. 2012; Landergott et al. 2012), and thus has limited practical value in developing a species-specific acorn-sizing criteria for improvement of nursery seedlings. Effects of absolute acorn size on seedling growth has received limited testing in North America (Bonfil 1998), Europe, and Russia (Jarvis 1963; Gómez 2004; Anagiotos et al. 2012; Landergott et al. 2012), and few studies have specifically examined northern red oak (Long and Jones 1996; Yi et al. 2015).

Family effects influenced acorn size and seedling quality even from mother trees within the same provenance (Kriebel 1965; Kormanik et al. 1998; Landergott et al. 2012), and family selections may improve nursery seedling quality and subsequent field performance of artificially regenerated seedlings (Jacobs and Davis 2005). Unfortunately, few studies have tested family influences on oak nursery seedling quality, despite recent recognition that tree improvement and genetic conservation programs will be important for combatting a variety of forest health and sustainability concerns (Jacobs and Davis 2005; Wheeler et al. 2015; Potter et al. 2017).

The goal of this research was to improve efficacy of oak artificial regeneration efforts by testing relationships between acorn and seedling characteristics. This research is particularly pertinent for planting on productive sites where competition from other species is the primary limiting factor inhibiting oak regeneration and recruitment into the canopy. A specific objective was to quantify relationships between absolute acorn size and mass and seedling morphology within six half-sib families of northern red oak. We had two research hypotheses: (1) acorn mass and size would positively relate to seedling survival and size; (2) families would exhibit differences in relationships between acorn attributes and seedling morphological characteristics.

Methods

Experimental material

Acorns were collected from six open-pollinated mother trees from a seed orchard (ca. 1973) on the United States Department of Agriculture, Forest Service, Cherokee National Forest (Lat. 36°07″N, Long. 82°00″W) (Schlarbaum et al. 1994). Acorns were collected daily during peak acorn dispersion in October and November 2011, and were immediately stored at 1–3 °C until sowing to prevent desiccation (Bonner 2008). A float/sink test was performed to improve germination rates (Gribko and Jones 1995). Acorns from each mother tree represented open-pollinated half-sib progeny and are hereafter referred to as a 'family' (Appendix).

Approximately 500 randomly selected acorns from each family were separated into five 0.3 cm diameter classes, based on the width of the acorn, by passing through appropriately sized screens. Size classes are hereafter referred to by the upper limit for that class (e.g., 1.3 includes acorns $\leq$ 1.3 cm, 1.6 includes acorns $>$ 1.3 and $\leq$ 1.6 cm, etc.). No acorns were larger than 2.5 cm diameter. There was a relatively large frequency of acorns in the 2.2 cm size class, so an additional size class of 2.1 cm (acorns $>$ 1.9 and $\leq$ 2.1 cm in diameter) was created to further differentiate the effect of acorn size on seedling performance.

Acorns from each family/size class treatment were evenly divided into one to three replications (Appendix). Treatments with fewer than 30 acorns delivered only one replication. Acorns were exchanged among replications within the same treatment until the difference in total fresh mass among replications was less than 1 g. Hereafter, a 'seed lot' refers to the group of acorns within the same replication and treatment.

A sample of 1–10 acorns from each seed lot were randomly selected and weighed to the nearest 0.1 g to obtain individual acorn fresh mass (IAM), and identification of each weighed acorn and resulting seedling was maintained throughout the study. Each seed lot was weighed for total fresh mass to the nearest 0.1 g and divided by the number of acorns to calculate seed lot acorn mass (SAM).

Experimental design and nursery production

Acorns were manually sown at the East Tennessee State nursery in Delano, TN on 16 February 2012 at a density of 65 per m^2. Seed lots with fewer than three acorns were not included in subsequent analysis of variance (see below); therefore, a total of 49 seed lots were created (Appendix) each separated by 0.6 m buffer of empty bed space. The nursery beds were relatively uniform and areas with distinctly different environmental conditions could not be easily identified; therefore a completely random design with a nested treatment arrangement was used to assign seed lot treatments and replications within the nursery beds. Families did not have all acorn size classes represented; therefore, size class was a fixed effect nested within the fixed effect of family.

Individually weighed acorns were sown in the inner two rows, and the remaining acorns were sown in the outer two rows of each seed lot (each nursery bed contained 4 rows, spaced evenly apart). Weighed acorns were surrounded on each row end by at least one non-weighed acorn to avoid bias associated with edge effects. A metal stake with an identifying tag number were carefully positioned adjacent to each weighed acorn.

We sowed 1.3, 1.6, 1.9, and 2.2 cm-class seed lots with fewer than 20 acorns within the next largest size class for that family to avoid sowing seed lots with low density that might bias seedling size. The 2.5 cm class seed lot with fewer than 20 acorns was sown within the 2.2 cm size class. The smallest seed lot contained 24 acorns and the largest seed lot contained 111 acorns (Appendix).

The resultant seedlings were irrigated as needed and fertilized according to prescriptions to produce large, high-quality seedlings through continuous application of fertilizer during the growing season (cf. Kormanik et al. 1994). During the growing season, tags associated with individually weighed acorns were transferred to the stem of the resulting seedling. On 25 February 2013, a Fobro™ machine lifter was used to undercut seedlings (25–30 cm) and loosen soil around the roots. Seedlings were manually removed from the nursery beds, roots were sprayed with a hydrogel slurry solution to prevent desiccation, and trees were placed in poly-coated paper tree bags in cold storage until measurements were collected.

Seedling data collection

All seedlings were measured for stem height to the nearest cm, and stem root-collar diameter (RCD) to the nearest 0.1 mm. The number of first-order lateral roots (FOLR), defined as a suberized lateral root stemming from the main taproot at least 1 mm in diameter at the proximal end, were counted on seedlings from weighed acorns. Stem diameter at 3 cm below the terminal bud (Dia3cm) was measured to the nearest 0.1 mm on seedlings from weighed acorns. Stems damaged during lifting or processing ($n=50$ of all seedlings, $n=17$ for seedlings from weighed acorns) were not measured for height or top diameter.

Data analyses

Stem volume (cm^3) was estimated by:

$$Volume = \frac{\left(2.5 \times \pi \times Dia3\text{cm}^2\right) + \left[(10\,HT - 30) \times \pi \times \left(\frac{RCD + Dia3\text{cm}}{4}\right)^2\right]}{1000},$$

where Dia3cm = diameter (cm) measured 3 cm from top of the terminal bud, HT = stem height (cm), and RCD = root collar diameter (mm).

SAS Institute (2012) was used to conduct statistical analyses with a confidence level of 5 percent unless otherwise indicated. T-tests were conducted to determine if seedlings from weighed acorns differed from the remaining seedling population for stem height and RCD. This analysis was used to determine if weighing acorns and subsequent tagging of the resulting seedlings biased the results.

General linear mixed models (GLM) were performed to determine treatment effects on each continuous dependent variable (SAM, IAM, and seedling variables). A generalized linear mixed model (GLMM) with LaPlace estimation and a binomial distribution was used to determine treatment effects on seed lot survival, where the syntax was the number of trees lifted (i.e., trees that germinated and survived until lifting) out of the number of acorns sown. GLMM was preferred over a linear model to accommodate for violation of non-normality assumptions (Bolker et al. 2009; Stroup 2014). We considered the GLMM to have an adequate fit when confidence intervals of fixed effect estimates were not exceedingly large (e.g. <6 times the standard error of the estimate) (Bolker et al. 2009). Overdispersion of the residuals was checked using a Pearson Chi square test, and the value was <1, indicating a lack of overdispersion. Comparisons among least-squares means for GLMs and the GLMM were made using Tukey's mean separation test if main effects were significant.

Logistic regression was used to determine if survival of seedlings from weighed acorns could be predicted from IAM and its interaction with family for event = 1 (alive). We used linear regression with indicator variables to determine if the four seedling variables could be predicted from IAM and the interaction with family. Separate linear regression models were run for each seedling variable.

For GLMs and linear regression analyses, we tested normality and equal variance assumptions of residuals using the Shapiro–Wilk test for normality and by examining plots of residuals. Normality was assumed if the Shapiro–Wilk statistic was >0.9. Transformations of independent variables were used when necessary, and stem volume was square root transformed. Because seed lot sizes and survival rates varied, we tested the effect of tree growing space as a covariate in GLMs and linear regression models; growing space was calculated as the seed lot area (length by width) divided by the number of trees lifted in the seed lot (m^2 per tree). The covariate was only included if the corrected Akaike information criterion (AICc) was significantly improved with addition of the covariate in the model. We did not include seed lots with fewer than three acorns in any GLMs or the GLMM (see Appendix).

Results

The tagged seedlings, derived from individually weighed acorns, were 8 cm shorter in stem height than the untagged seedling population ($T = 2.46$, $P = 0.0139$), but root-collar diameter (RCD) was not different ($T = 0.58$, $P = 0.5652$). Survival was approximately 90% when calculated using seed lot means and seedlings from weighed acorns (Table 1). Seedlings were highly variable in size, and averaged 114 cm in stem height and 12.0 mm in RCD.

The seed lot acorn mass (SAM) and individual acorn mass (IAM) were affected by family and acorn size class within family (Table 2). The SAM significantly increased from the smallest to the largest size classes for each family (Table 3), and the IAM had similar values to the SAM (within 0.4 g; data not shown). The acorns of families 4 and

Table 1 Means and associated standard deviations, coefficients of variation (CV), and ranges in seed lot acorn mass (SAM), individual acorn mass (IAM), stem height, root-collar diameter (RCD), number of first-order lateral roots (FOLR), and stem volume for seedlings across all six families

Variable	N	Mean	CV	Minimum	Maximum
SAM (g)[a]	49	7.1 ± 1.7	24.2	2.3	11.2
IAM (g)	476	7.3 ± 1.8	24.4	1.3	11.8
Seed lot density (trees per m^2)[a]	49	63 ± 8	13.0	47	86
Tree growing space per seed lot (m^2 per tree)[a]	49	0.016 ± 0.002	12.1	0.012	0.021
Seed lot survival (percent)[a]	49	86 ± 11	12.5	44	100
Individual seedling survival (percent)	476	88 ± 33	37.3		
Stem height (cm)	2597	114 ± 61	53.8	3	273
RCD (mm)	2647	12.0 ± 4.7	38.9	2.2	25.3
Number of FOLR	418	15 ± 11	73.3	0	49
Stem volume (cm^3)	410	72 ± 72	99.9	1	333

[a]Seed lots with fewer than 3 seedlings were not included (see Appendix)

Table 2 General linear mixed models to test family and acorn size class effects on acorn mass, survival, and seedling morphological variables

Dependent variable	Family		Acorn size class (family)	
	$F_{5, 24}$	*P*	$F_{19, 24}$	*P*
Seed lot acorn mass	27101.5	< 0.01	10624.2	< 0.01
Individual acorn mass	183.8	< 0.01	74.2	< 0.01
Seed lot survival	1.8	0.16	2.3	0.03
Stem height	17.3	< 0.01	2.2	0.03
RCD	9.8	< 0.01	3.4	< 0.01
Number of FOLR	5.9	< 0.01	1.4	0.24
Stem volume[a]	6.2	< 0.01	2.7	0.01

Tree growing space was included as a covariate in the root-collar diameter (RCD) model ($F_{1, 2590} = 3.3$, $P = 0.07$)

FOLR number of first-order lateral roots

[a]Square root-transformed

Table 3 Family and acorn size class least-squares means for seed lot acorn mass (g)

Acorn size class	Family					
	1	2	3	4	5	6
1.3						2.3 s
1.6	5.9 p	6.0 o	5.8 q	5.0 r	6.2 n	5.0 r
1.9	6.7 l	6.1 n	6.8 k	6.0 o	7.1 j	6.0 o
2.1	8.3 f	8.0 g	7.5 i	6.6 m	7.8 h	6.7 l
2.2	9.9 c	9.0 e	9.0 e		9.8 d	
2.5	11.2 a	10.2 b				
Overall	8.4 a	7.9 b	7.3 d	5.9 e	7.7 c	5.0 f

Means followed by the same letter were not significantly different. Overall family means followed by the same letter were not significantly different

6 had consistently lower acorn mass than those of other families within and across size classes. Acorn mass differences between the smallest and largest size classes within a family ranged from 1.6 to 5.3 g for SAM and from 1.6 to 5.2 g for IAM.

Seed lot survival was not affected by family, but was affected by acorn size class (Table 2). Family 1 had increasing survival with increasing size class, but no differences or discernible pattern in survival among classes could be detected in the other five families (Table 4). The results of the logistic regression analysis indicated that survival was not affected by the IAM (Wald Chi square = 0.5374, $P = 0.4635$), family (Wald Chi square = 6.4131, $P = 0.2681$), or their interaction (Wald Chi square = 3.2221, $P = 0.6658$). The IAM did not significantly predict survival if family effects were not included in the model (Wald Chi square = 1.1451, $P = 0.2846$). Logistic regression models adequately fitted the data according to the Hosmer and Lemeshow goodness of fit test ($P > 0.6841$).

All seedling variables were affected by family (Table 2). The values for families 3 and 6 were consistently lower than those recorded for the other four families (data partially shown in Fig. 1), but the values varied little among the other four families. Acorn size class within family significantly affected height, RCD, and stem volume. Seedlings in family 2 generally increased size as acorn size class increased, particularly for stem volume, but differences were not always significant. No discernible relationship between acorn size class and seedling morphology could be detected for the other families. The covariate, tree growing space, significantly improved the RCD model according to the AICc values, and was positively related to RCD. The covariate was not included in any other model.

The relationships between IAM and seedling variables were weak but significant ($0.03 \geq R^2 \leq 0.11$; $P < 0.01$) for all variables tested (Table 5). A 1 g increase in IAM was predicted to increase height by 4 cm, RCD by 0.4 mm, and stem volume by 4–6 cm^3. Family did not affect the relationship between IAM and any seedling variable, except for number of first-order lateral roots (FOLR). Slope parameters in the FOLR model were not significant, except for family 4 where a 1 g increase in IAM increased FOLR by 5. Tree growing space was a significant covariate in the height and stem volume models and was negatively related in both models.

Table 4 Least-squares mean differences in seed lot survival among acorn size classes within families

Acorn size class (cm)	Family					
	1	2	3	4	5	6
1.3						100 ab
1.6	44 c	100 a	89 a	85 a	78 a	88 b
1.9	72 bc	100 a	97 a	94 a	82 a	93 ab
2.1	76 b	94 a	76 b	88 a	85 a	99 a
2.2	81 ab	90 a	92 a		80 a	
2.5	97 a	100 a				

Means followed by the same letter were not significantly different within a family

Fig. 1 Least-squares means for stem height, root-collar diameter (RCD), and stem volume by family and ▸ acorn size classes within each family. Size class means within the same family with the same lowercase letter are not significantly different, and overall family means (white bars) followed by the same uppercase letter are not significantly different

Discussion

We attribute the lack of causal relationships between acorn mass/size and seedling attributes to the relatively large size of seedlings resulting from unusually favorable growing conditions. A long and relatively wet growing season (NCEI 2017), a particularly productive site in the nursery (John Conn, pers. comm.), and advanced nursery protocols designed to promote continuous growth of seedlings (Kormanik et al. 1994) resulted in large seedlings in all treatments. Some seedlings exceeded 2.5 m in height (Table 1), an unusually large size for 1-year-old seedlings of this species (Kormanik et al. 1998; Clark et al. 2015, 2016). Seedlings in this study were 54 cm taller and 3.7 mm larger in RCD than seedlings from Kormanik et al. (1998) grown using similar nursery protocols. Compared to other studies that grew northern red oak seedlings in pots, in a nursery, or by direct seeding in the field, our seedlings were 94–104 cm taller and 5.5–7.0 mm larger in RCD (Korstian 1927; Kriebel 1965; Auchmoody et al. 1994). Our seedlings were also much larger than comparable studies that used other oak species (Jarvis 1963; Bonfil 1998; Navaroo et al. 2006; Landergott et al. 2012).

We suspect that acorns from the larger size classes germinated earlier and the resulting seedlings grew more rapidly than acorns and resulting seedlings from the smaller size classes (Long and Jones 1996; Tilki 2010; Anagiotos et al. 2012), but this effect may have dissipated as favorable weather and the frequent fertilization regime favored growth of all trees. Previous studies have found that carbohydrates in larger acorns may be more important to seedling development when environmental conditions are limiting (Bonfil 1998; Quero et al. 2007; Yi et al. 2015). Monitoring seedling growth regularly throughout the growing season would have allowed examination of temporal relationships between acorn size/mass and seedling attributes, but that was not performed in this study. However, interactions were not found between nut size and daily growth patterns of American chestnut (*Castanea dentata*), a Fagaceae species, in a similar study (Pinchot et al. 2015).

Tagging and associated handling and processing of the acorns to facilitate this study had no significant effects on survival and RCD and a minimal effect on height; therefore, our inferences from data collected from tagged seedlings were assumed to be representative of the remaining population. Acorn mass measured as individual acorn mass (IAM) or as seed lot acorn mass (SAM) was a good proxy for acorn size, as expected from past research findings (Korstian 1927; Jarvis 1963; Ramírez-Valiente et al. 2009), but there was scant evidence to support our first hypothesis that increasing acorn mass/size would improve seedling survival and morphological characteristics commonly associated with seedling quality. IAM was particularly poor at explaining the variation in survival or any seedling morphological variable, counter to previous research results for North American oaks (Bonfil 1998; Kormanik et al. 1998) and European species (Gómez 2004; Ramírez-Valiente

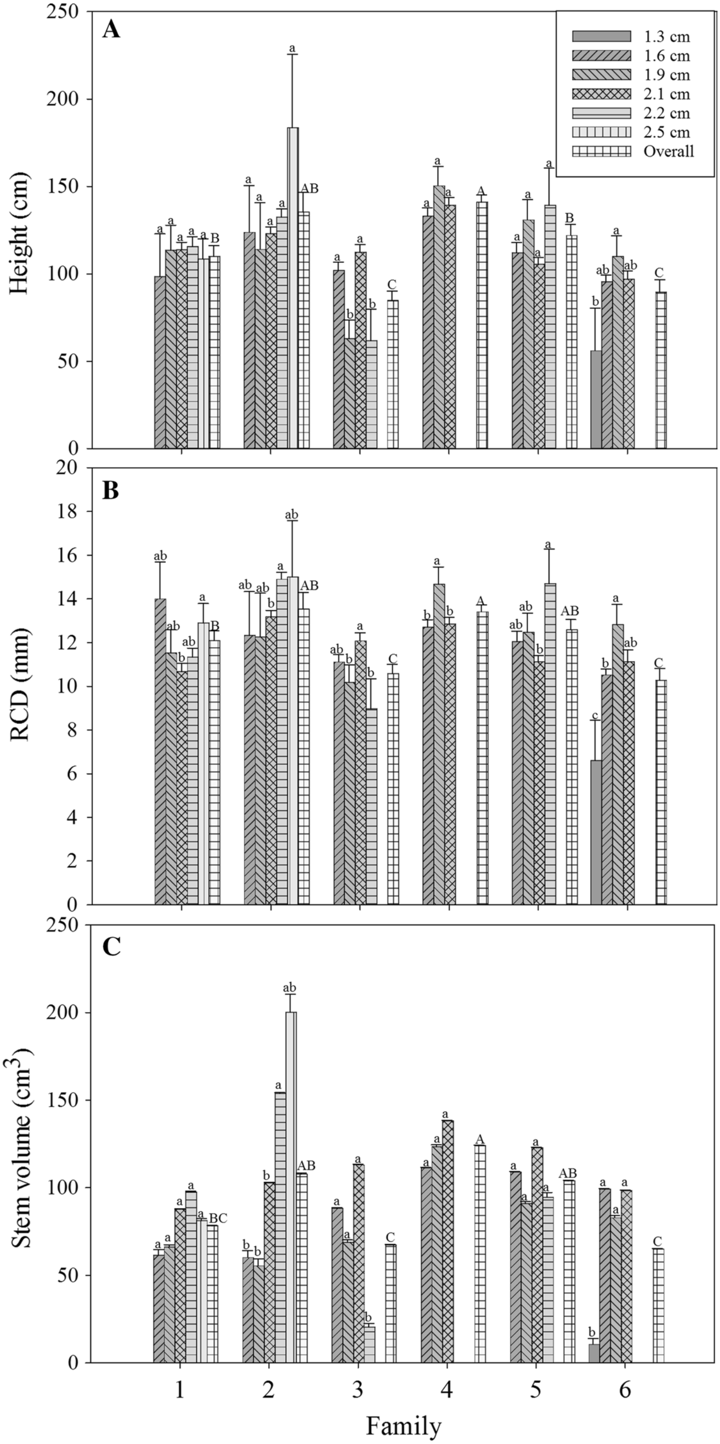
A
B
C
Height (cm)
RCD (mm)
Stem volume (cm3)
Family
1
2
3
4
5
6
1.3 cm
1.6 cm
1.9 cm
2.1 cm
2.2 cm
2.5 cm
Overall
0
50
100
150
200
250
2
4
6
8
10
12
14
16
18
20

Table 5 Parameter estimates from linear regression to predict height, root collar diameter (RCD), number of first-order lateral roots (FOLR), and stem volume from individual acorn mass

Dependent variable	Intercept	Slope	R^2	*P*
Stem height (cm)	139.59	4.15	0.03	< 0.01
Growing space (m^2/tree)		−3926.01		0.01
RCD (mm)	8.65	0.43	0.03	< 0.01
Number of FOLR	−7.94	4.46	0.11	< 0.01
Stem volume (cm^3)[a]	8.11	0.38	0.03	< 0.01
Growing space (m^2/tree)		−229.28		0.04

Tree growing space was included in models when significant. The parameter estimates for the FOLR model include only family 4

[a]Square-root transformed

et al. 2009; González-Rodríquez et al. 2011; Landergott et al. 2012). Our results did agree with a minority of studies that found poor relationships between acorn mass and germination or survival (Landergott et al. 2012; Yi et al. 2015), or between acorn size and growth (Long and Jones 1996). The latter study hypothesized that unaccounted genetic influences were contributing to the poor relationships; however family effects could not be used to explain the poor associations between acorn mass and seedling morphology in our study.

We found strong support for our second hypothesis that family would affect the relationships between acorn attributes and seedling morphological characteristics. Seedlings derived from large acorns were generally bigger than those originating from small acorns for only one of the six families (Family 2). Kriebel (1965) also found a poor relationship between acorn mass and seedling size for most northern red oak families from a broad geographic range, but González-Rodríquez et al. (2011) found families from various oak species generally exhibited positive relationships between seed mass and seedling biomass. Genetic influences cannot be used to explain the poor relationships between acorn mass/size and seedling variables in our study, as Long and Jones (1996) had hypothesized.

Our results suggest that family selections to increase nursery seedling size would be possible, but predicting seedling size based on family acorn attributes may not be reliable. Some mother trees (e.g. families 4 and 6) tended to produce small acorns, but survival and growth following germination was not necessarily lower for these families than for other families. Family differences in northern red oak nursery seedlings has not been widely studied, but our results and others indicate that seedling size can be enhanced through family selections (Kriebel 1965; Kolb and Steiner 1989). Unfortunately, programs that can capitalize on identifying family differences to meet restoration or reforestation goals are largely lacking (Schlarbaum 1999; Wheeler et al. 2015).

Conclusions and practical implications

Acorn mass and size had minimal effects on nursery seedling survival and morphology. This study and many others have demonstrated that variability in oak seedling size, either within or among families, will be large, and acorn grading using mass or size is unlikely to reduce this variability. Unlike the results obtained in many other studies, we did not find that overall seedling size can be improved through sizing acorns or selecting acorns based on mass. Results may have been affected by an unusually long growing season and nursery protocols that enhanced seedling growth, resulting in relatively large seedlings that were less dependent on cotyledon reserves from the acorn.

Family influenced all aspects of this study, and selection for early development of specific nursery traits is possible. However, tree improvement in oak is inherently difficult due to the lack of association between desirable characteristics and temporal changes in family effects (Kriebel et al. 1988). Given the importance of family in this study, we suggest that managers use a diverse mix of material to ensure seedling quality is not heavily influenced by too few families.

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Appendix

See Table 6.

Table 6 Total sample size of acorns in each family and size class, and individually weighed acorns are in parenthesis

Orchard family number	New family number	Acorn size class (cm)						
		1.3	1.6	1.9	2.1	2.2	2.5	Total
528	1	0 (0) $_0$	16 (10) $_1$	13 (10) $_1$	278 (30) $_3$	165 (30) $_3$	29 (10) $_1$	501 (90) $_9$
630	2	0 (0) $_0$	5 (5) $_1$	5 (5) $_1$	271 (30) $_3$	216 (30) $_3$	3 (3) $_1$	500 (73) $_9$
702	3	1 (1) $_0$	199 (30) $_3$	34 (10) $_1$	257 (30) $_3$	12 (10) $_1$	0 (0) $_0$	503 (81) $_8$
1164	4	1 (1) $_0$	217 (30) $_3$	34 (10) $_1$	244 (30) $_3$	2 (2) $_0$	0 (0) $_0$	498 (73) $_7$
2459	5	1 (1) $_0$	141 (30) $_3$	33 (10) $_1$	332 (30) $_3$	10 (10) $_1$	0 (0) $_0$	517 (81) $_8$
2472	6	6 (6) $_1$	324 (30) $_3$	24 (10) $_1$	151 (30) $_3$	2 (2) $_0$	0 (0) $_0$	507 (78) $_8$
Total		9 (9) $_1$	902 (135) $_{14}$	143 (55) $_6$	1533 (180) $_{18}$	407 (84) $_8$	32 (13) $_2$	3026 (476) $_{49}$

The number of replications is in subscript

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