Effects of climate change on seed germination may contribute to habitat homogenization in freshwater forested wetlands

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Abstract

Climate changes are expected to result in warmer, shorter winters in temperate latitudes. These changes may have consequences for germination of plant species that require a period of physiological dormancy. The effect of cold duration on seed germination has been investigated in a number of plant taxa, but has not been well studied in wetland and bottomland forest tree species, an ecosystem that is threatened by habitat homogenization. Our work sought to test the role of changing winter temperatures on seed germination in specialist (*Nyssa aquatica* and *Taxodium distichum*) and generalist (*Acer rubrum* and *Liquidambar styraciflua*) tree species within forested wetlands throughout the eastern U.S.. The experiment was conducted in an environmental chamber in Norfolk, VA, USA. Seeds of *T. distichum, N. aquatica, A. rubrum*, and *L. styraciflua* were exposed to each of pre-germination cold exposure durations (0, 15, 30, 45, 60, 75, and 90 days) and observed for germination for 30 days. *C*old stratification duration positively impacted total percent germination in *N. aquatica* ($p < 0.0001$) as well as *A. rubrum* ($p = 0.0008$) and *T. distichum* ($p = 0.05$). *Liquidambar styraciflua* seeds exhibited more rapid rates of germination with increasing cold exposure duration and greater percent germination compared to the others regardless of cold stratification duration. Our results provide insight into how community dynamics and biodiversity of wetland and bottomland trees may shift with a changing climate. Further, this work emphasizes the importance of understanding the role of plant functional traits in early life stages in community dynamics and has implications for management practices.

Introduction

Biotic homogenization in plant communities is the process in which the composition of an ecological community is altered to favor one or a few organisms (Tilman et al., 1997; McKinney and Lockwood, 1999). Homogenization within a habitat may occur genetically, taxonomically, and/or functionally and results in lower genetic, species, and/or functional diversity within the community (Olden et al., 2004; Olden 2006). Drivers of biological homogenization include both natural and anthropogenic factors including climate change, disturbance events, habitat fragmentation, and invasive species introduction (Baruah et al., 2017; Bilyaminu et al., 2020). These processes can overlap and compound over time (Olden and Rooney, 2004). For example, Abadie et al. (2011) found that generalist species replaced specialist species in urban plant communities with increasing habitat fragmentation. Lougheed et al. (2008) found that anthropogenic nutrient enrichment led to lowered plant taxonomic richness in degraded wetlands. Additionally, a recent meta-analysis of 200,000 plant species showed global widespread decreases in species diversity throughout the anthropocene (Daru et al., 2021). Disturbances not only contribute to homogenization, but the consequences of species convergence may alter the natural functions of an ecosystem thus reducing the system's resilience to future disturbance events (Tilman et al., 1997; Hooper et al., 2005; Olden, 2006).

Habitat homogenization is a prevalent phenomenon in bottomland forests and freshwater forested wetlands throughout the southeastern United States, partially due to longstanding history of disturbance to these systems (Brinson, 1990). Examples of reduced species diversity can be found throughout the

range of charismatic forested wetland specialist species such as bald cypress (*Taxodium distinchum*) and water tupelo (*Nyssa aquatic*a). Near the northernmost historic range of bald cypress, within Virginia's Great Dismal Swamp, specialist obligate wetland tree species (e.g., *Taxodium distichum* and *Nyssa aquatic*a, and *Quercus* spp.) have been replaced by generalist facultative wetland communities dominated by red maple (*Acer rubrum*) and sweet gum (*Liquidambar styraciflua*), termed maple-gum (Levy, 1991). The Dismal Swamp has a long history of disturbance and today ~ 60% of the Dismal Swamp is characterized as maple-gum habitat (Ludwig et al., 2021; Speiran and Wurster, 2021). Toward the southernmost range of bald cypress (i.e., Texas, Louisiana, Alabama), mixed bottomland forests subjected to frequent anthropogenic and climatic disturbances are now characterized by reduced species diversity and dominant representation (i.e., frequency, basal area, relative importance) by invasive generalist species such as sweet gum (Hart and Holmes, 2013), red maple (Abrams, 1998), and invasive Chinese tallow (*Triadica sebifera*) (Henkel, 2012).

Homogenization, such as that observed in the freshwater forested swamps and bottomland forests through the southeast U.S., is often the result of changing environmental conditions that favor the traits of competitive generalist species compared to specialist species (Naaf and Wulf, 2010). Generalist and/or invasive plant species often possess functional traits (e.g., morphological, phenological, or physiological characteristics) favored by disturbance (Holl et al., 2022) and a wider range of abiotic tolerances compared to specialist species (Qian and Guo, 2010). Plant functional traits provide information about species' life strategies (Condit et al., 2006) and responsiveness to changing environmental cues; thus traits have been used to understand a variety of dynamics related to habitat homogenization, including differences in ecological niches (Silvertown, 2004), environmental partitioning (Drake and Franks, 2003), and relationships between traits and ecosystem function (Carlucci et al., 2020). The most commonly measured plant functional traits across plant taxa include a mix of early- and mature-life stage traits (e.g., seed mass, plant height, specific leaf area, and wood density) (Moles, 2017). Early life-stage traits (e.g., seed rain, seed dormancy, germination percentage and rate, and seedling growth rate) are important first indicators of the breadth of environmental conditions under which a species may establish and spread. Additionally, the processes that drive community assembly of the soil seed bank following a disturbance are poorly understood compared to drivers of aboveground plant community assembly (Helsen et al., 2015).

Seed dormancy, or the innate delay of germination when environmental stimuli would not support plant survival (Carta et al., 2014), is one early-life stage trait that differs greatly between species and influences germination dynamics and other early-life traits. In temperate climates with cyclical temperature and precipitation regimes, seed dormancy and germination dynamics of many species are synchronized with environmental cues (Baskin and Baskin, 2014), with some species requiring a period of cold exposure, or cold stratification, to overcome dormancy (Angevine and Chabot, 1979). As a result, changes to the environmental stimuli that influence germination (light, temperature, moisture, etc.) due to disturbances or climate change will likely play an important role in differentially limiting species depending on their species-specific germination traits and dormancy requirements (Walck et al. 2010). As global temperatures increase, plant community structure is likely to shift if species specific

germination requirements are not met, potentially resulting in increased homogenization by generalist species (Qian and Guo, 2010). Daily minimum winter temperatures throughout the geographic range of swamp specialist species *T. distichum* and *N. aquatica* show a warming trend over the past century, though the rate of warming varies slightly by location (Fig. 1, NOAA, 2023a).

Here, we explore the extent to which early life traits, specifically seed germination and germination rate, may explain observed current patterns of species homogenization in freshwater forested swamp and bottomland hardwood forests in the United States. Although the relationship between germination niche and ecological niche has been explored in other systems, to our knowledge, few studies exist that explore this in forested wetlands which are uniquely subject to strong ecological drivers (e.g., hydrology) that limit species establishment at the seedling life stage. This study also investigates the influence of changing climate scenarios on the establishment dynamics, viability, and permanence of key species in these forested wetland habitats using pre-germination cold stratification treatments of variable durations (0 days to 90 days). Species addressed in this study include two forested wetland specialist species (*Taxodium distichum* and *Nyssa aquatica)* and two generalist species (*Acer rubrum* and *Liquidambar styraciflua*) characteristic of the increasingly prevalent maple-gum habitat observed throughout freshwater forested wetlands and bottomland forests of the U.S. The objective of this work is two-fold: 1) to inform our understanding of the role of early life traits on the permanence of specialist and generalist species in freshwater forested wetlands, and 2) understand whether warming winters characteristic of climate change may further exacerbate plant community homogenization by differentially limiting germination of specialist and generalist species. The ultimate goal of this work is to increase our understanding of species-specific physiological demands at early life-stages and inform habitat management actions that may aid in preserving biodiversity and reducing species conversion in forested wetlands.

Methods

Experimental Design

Target conservation tree species in freshwater forested wetlands within the United States include *Taxodium distichum* (bald cypress) and *Nyssa aquatic*a (water tupelo). Forested wetland communities may include a number of other tree species, but here we focus on generalist species prevalent in maple-gum habitats, *Acer rubrum* (red maple) and *Liquidambar styraciflua* (sweet gum). The seeds of four tree species (*L. styraciflua, A. rubrum, N. aquatica, T. distichum)* were obtained from Sheffield's Seed Company, Inc. in Fall 2021. Seeds were kept in dry cool (22° C) conditions prior to pre-germination treatments. Previous research on *Taxodium distichum* seed germination showed that the species exhibits better germination when seeds were soaked in 1% NaOH for five minutes and then in water for the next 24 hours; this processes neutralizes the seed's resin coating ex-situ that would be neutralized under natural swamp conditions (Liu et al., 2009). Thus, we conducted a fully factorial experiment, consisting of five species groups (*L. styraciflua, A. rubrum, N. aquatica*, non-treated *T. distichum*, and NaOH-treated *T. distichum)*, seven pre-germination cold stratification durations (0, 15, 30, 45, 60, 70 and

90 days), and five replicates of each combination. Each replicate consisted of ten seeds, for a total of 350 seeds.

Pre-germination cold stratification treatments, with the exception of the 0-day duration treatment, consisted of placing seeds in air-tight plastic tupperware containing moist builders' sand and exposing them to cool (4° C), dark conditions for their respective cold duration treatments in a refrigerator. Following each replicate's cold stratification treatment, seeds were soaked in deionized water in dark conditions at room temperature for 24 hours. The NaOH-treated *T. distichum* seeds were placed in a 1% NaOH solution for five minutes directly after refrigeration, followed by soaking in deionized water for 24 hours (after Lui et. al., 2009). After soaking, all seeds were placed on Whatman 3 filter paper in a square clear plastic Petri dish (10 seeds per replicate Petri dish) with 5 mL of deionized water. Petri dishes were kept closed under a clear lid and the filter paper was kept moist with deionized water throughout the duration of observation. All Petri dishes were placed in an environmental chamber (CONVIRON Gen1000, Controlled Environments Ltd., Winnipeg, Manitoba, CAN) with alternating photoperiod and thermal conditions that mimic general springtime conditions (e.g., 12 hr light at 25℃, 12 hr darkness at 20℃). Seeds were observed daily for germination (indicated by the emergence of radicle) for 30 days. Germinating seeds were recorded each day and removed from the Petri dish. Fungus was observed during both cold stratification and germination. Fungal growth was mitigated by washing affected seeds with deionized water. Whatman filter paper was replaced as necessary. Sterilizing chemicals were not used to avoid possible influences on seed germination. The length and mass of an additional 25 seeds from each species (not used in germination trials) were measured using a digital caliper and analytical balance respectively to obtain average seed traits per species.

Statistical Analysis

Each replicate Petri dish for all treatment combinations was measured for total percent germination at the end of 30-days as well as germination rate. The effect of species, cold stratification duration, and their interaction on percent germination was tested using a two-way analysis of variance (ANOVA). A one-way ANOVA with Tukey posthoc test of pairwise comparisons was performed for each individual species to determine the effect of cold stratification within species. A two-way ANOVA was used to test the effect of NaOH treatment, and cold stratification duration, on *T. distichum* only.

Germination rate was analyzed as a time-to-event, in which seed germination was the event. Non-parametric and semi-parametric time-to-event analyses are recommended as a statistical technique for investigating incidence and timing of germination (Peréz and Kettner 2013). Seeds that germinated during the 30-day experiment were coded as "1" and seeds that did not germinate by day 30 were right-censored and assigned as "0". No seeds were lost due to random censoring. Kaplan-Meier estimates of survivor functions (i.e., germination event) were generated using the survival and survminer packages in R (version 4.2.3). Survivor functions were stratified by cold exposure duration and generated for each species separately. Kaplan-Meier curves were generated using ggsurvplot with "event" function. Few species and cold stratification combinations reached 25, 50, or 75% germination, therefore the Kaplan-

Meier estimator for these data never reached a failure probability > 0.25, 0.50, or 0.75; resulting in inestimable quartile values. A Cox proportional hazards model was generated using the survival (v.3.5-5, Therneau, 2023) and survminer (v.0.4.9, Kassambara, 2021) packages in R to determine the effect of species and cold stratification treatment on germination rate.

Results

Percent Germination

Overall, *L. styraciflua* exhibited superior percent germination compared to other species tested and showed no impact of cold stratification duration on total percent germination. *Acer rubrum* seed germination was ~ 25% for most cold stratification treatments with significantly greater germination (> 50%) occurring in seeds exposed to 75 days of cold. No *A. rubrum* seeds exposed to 90-day cold stratification germinated, however we acknowledge some of this loss may have been due to fungal growth that occurred. *Nyssa aquatica* seeds exposed to 15 days or fewer of cold exposure showed low percent germination (< 10%). Percent germination increased significantly (> 50%) with increasing cold stratification duration up to 75 days. Germination of *T. distichum* was consistently low (< 20%) compared to other species. However, germination in non-treated *T. distichum* seeds was greatest when seeds were exposed to 60 days of cold stratification. Contrarily, cold stratification duration had no impact on NaOH-treated *T. distichum* seeds. A separate one-way ANOVA comparing non-treated *T. distichum* seeds with NaOH-treated *T. distichum* seeds showed that overall germination was greater with pre-germination NaOH treatment ($F_{1,56}$=4.840, p = 0.0320). A two-way ANOVA showed a significant interaction between cold stratification duration and species on total percent germination of seeds ($F_{24,140}$=6.019, p < 0.0001) (Fig. 2, Supplementary table 1). ANOVAs conducted within-species showed an effect of cold stratification on germination in *A. rubrum, N. aquatica*, and non-treated *T. distichum* seeds (Fig. 2, Supplementary table 2).

Germination Rate

Liquidambar styraciflua seeds exposed to cold stratification durations of 15 days, or 45 + days resulted in more rapid germination compared to the control (0 days cold) (Fig. 3, Table 1). Although all *L. styraciflua* cold treatments resulted in similar germination percentages, the rate at which that germination occurred was more rapid with increasing cold duration (Fig. 3, Table 1). *Acer rubrum* seeds exposed to 75-days of cold exposure showed a significant increase in rate of germination compared to the control, though other cold treatment durations had no effect. *Nyssa aquatica* seed germination was faster than the control treatment when cold exposure treatments lasted 30 + days. Germination of *N. aquatica* generally began after 10 days regardless of cold treatment. Rate of germination was not impacted by cold stratification duration in either the treated or non-treated *T. distichum* seeds (Fig. 3, Table 1).

Table 1
Results of Cox Proportional Hazards test for each species compared to 0 days cold stratification.

Cold duration	coef	exp(coef)	se(coef)	Pr(>|z|)
Liquidambar styraciflua				
15 Days	1.0050	2.7319	0.2301	< 0.0001***
30 Days	0.2777	1.3201	0.2431	0.25
45 Days	1.3280	3.7733	0.2260	< 0.0001***
60 Days	1.7595	5.8094	0.2277	< 0.0001***
75 Days	1.9253	6.8572	0.2374	< 0.0001***
90 Days	1.7812	5.9367	0.2328	< 0.0001***
Acer rubrum				
15 Days	0.3670	1.443	0.3970	0.34428
30 Days	-0.8212	0.4399	0.5394	0.1279
45 Days	0.2272	1.255	0.4097	0.57919
60 Days	0.1818	1.199	0.4175	0.66311
75 Days	0.1011	2.748	0.3669	0.00587**
90 Days	0.1828	< 0.0001	0.2709	0.99462
Nyssa aquatica				
15 Days	0.4212	1.5238	0.9129	0.6445
30 Days	3.0673	21.4840	0.7331	< 0.0001***
45 Days	3.3585	28.7447	0.7273	< 0.0001***
60 Days	3.5189	33.7475	0.7272	< 0.0001***
75 Days	3.8535	47.1556	0.7233	< 0.0001***
90 Days	1.9211	6.8285	0.7638	0.0119*
Taxodium distichum - untreated				
15 Days	0.1786	< 0.0001	0.0053	0.997
30 Days	0.1787	< 0.0001	0.0053	0.997
45 Days	0.1827	< 0.0001	0.0053	0.997
60 Days	0.1931	< 0.0001	0.0053	0.997

Cold duration	coef	exp(coef)	se(coef)	Pr(>|z|)
Liquidambar styraciflua				
75 Days	0.1717	< 0.0001	0.0053	0.997
90 Days	0.1716	< 0.0001	0.0053	0.997
Taxodium distichum -NaOH treated				
15 Days	-0.2431	0.7842	0.6708	0.717
30 Days	-0.9385	0.3912	0.8367	0.262
45 Days	0.2260	1.2535	0.6055	0.709
60 Days	-0.9429	0.3895	0.8367	0.260
75 Days	0.5344	1.7065	0.5701	0.349
90 Days	-0.2293	0.7951	06708	0.732

Discussion

The results of this study show important differences both within and between forested wetland and bottomland forest species regarding optimal conditions for seed dormancy break and germination. Most notably, generalist species that have been associated with rapid colonization of bottomland forests throughout the southeast U.S. (*A. rubrum* and *L. styraciflua*) showed markedly greater germination success compared to specialist species (*T. distichum* and *N. aquatica*) across a wide range of seed cold exposure treatments. The impact of longer pre-germination cold exposure was most important for *N. aquatica* seeds as longer cold stratification (> 15 days) significantly increased overall germination success. Although there was limited effect of cold stratification on germination for other species, the rate of germination showed a positive correlation with duration of cold exposure for both *N. aquatica* and *L. styraciflua*. Our results provide important information about each species' germination niche and development under a changing climate. Additionally, our results have implications for explaining and predicting shifts in community composition where these species co-occur as well as informing their management.

Variability in early life-history traits is important to understanding shifts in plant community structure and functional diversity in response to changing environmental conditions. Several studies have investigated the relationship between breadth of seed germination dynamics and ecological niche and found that generalist germination strategies, such as not requiring specific cues to germinate, may have evolved in response to higher spatial variation (Marques et al., 2014; Barga et al., 2017). Wetlands are characterized by specialist species that are uniquely adapted to the variable physiological stressors inherent to inundated and or saline soils; in fact, in the U.S., plant species are assigned a wetland indicator status which indicates the species' likelihood for occurrence in wetlands (i.e., wetland specialist vs. generalist)

(USACE, 2020). In forested wetlands and bottomland forests, existing research regarding species' niche and physiological thresholds, specifically as it pertains to habitat suitability, focuses on post-germination life history stages. Seed traits, such as seed size and dispersal mode, have frequently been used to explain functional diversity of a variety of forest types (Swenson et al., 2012; Lamanna et al., 2014; Andrew et al., 2022), including forested wetlands and bottomland forests. Generally, seeds with smaller mass are more likely to have dormancy and greater seed production per unit canopy area, but less likely to survive as seedlings than seeds with greater mass (Moles, 2017). Inclusion of seed germination dynamics as a predictor of functional diversity or community complexity is less common, despite an abundance of research investigating seed ecology of forested wetland and bottomland forest tree species. Our findings bunk common trends in predictive seed traits, as the smaller-seeded generalist species (Fig. 1) did not show a strong requirement for dormancy. We did not follow survival beyond germination, but total seed production and seedling survival are important to understanding trends in habitat homogenization.

Previous studies and planting recommendations for these species support our findings that *A. rubrum* and *L. styraciflua* have high rates of fecundity. Abrams (1998) documented that requirements for *A. rubrum* seed germination are broad and seeds may germinate immediately following dispersal or in the second year after they are produced. Moore and Lacey (2009) found that *Liquidambar styraciflua* show rapid germination rates (first germination in less than 15 days) and high overall percent germination (60–83%). Species-specific forestry guides (USDA, 1965) provide broad recommendations for cold stratification requirements for these species (e.g., 30 to 90 days for *A. rubrum*, 30 to 120 days for *N. aquatica*, and 30 to 90 days for *T. distichum*). Our study found increased germination results within these broad cold stratification ranges, but also show that germination may be more variable, with some species showing no increase in germination within this range. Our findings, coupled with these broad recommendations, demonstrate there may be significant variability within species regarding germination requirements, potentially depending on population, phenotype, or climate/geographic range. Additionally, it is important to note that the majority of information on the basic ecology for these species is sourced from studies originating more than 50 years ago (USDA, 1965). Although many aspects of individual species' traits may not change over, the large gap in time from when these species' reproductive ecologies were last analyzed provides impetus to revisit basic phenology and reproductive dynamics given the impact of a rapidly changing climate.

Implications of climate change on community structure

The cumulative effects of climate change and other anthropogenic influences on community composition have been studied across a variety of ecosystems. Resulting shifts toward redundant functional diversity have the ability to reduce the plasticity of ecosystems, leaving them more vulnerable to stress events as global change continues (Olden et al., 2004). In the southeastern United States, shifts in local climate have altered soil chemistry and hydrology, creating favorable conditions for generalist, non-native, and invasive species (Mulholland et al., 1997). Shortened winters due to climatic shifts may have differential impacts depending on plant species and life stage. Seeds and seedlings are especially

sensitive to changes in temperature and water availability and they often rely on these factors to signal the shift from dormancy to germination (Walck et al., 2010). A change in these cues may cause change in community composition.

Liquidambar styraciflua's superior germination percentages, regardless of cold stratification duration, paired with its extended habitat range suggest that this species will exhibit similar germination success under shorter, warmer winters. However, *Nyssa aquatica* showed comparable germination percentages only after cold exposure durations of more than 30 days. Although *A. rubrum* showed maximum germination when exposed to 75 days of cold stratification, this species was able to germinate successfully (~ 25%) regardless of cold duration. By contrast, *T. distichum* showed maximum percent germination when exposed to 60 days of cold stratification, but even maximum germination was < 25%. Additionally, for *L. styraciflua* and *N. aquatica*, the rate of germination increased with increasing cold exposure. Speed of germination may have conflicting effects on establishment and survival. Strong competition dynamics observed between new seedlings may favor the first to germinate, as has been observed in grasslands (Louda et al., 1990). However, variable springtime temperatures where snap freezes may occur may favor the survival of slower germination mechanisms that allow the plant to avoid tissue damage from freeze events. Other studies show that some plant species demonstrate different germination strategies along abiotic gradients, suggesting that slower and asynchronous germination may increase seedling survival under unpredictable environmental conditions (Sales et al., 2013).

Our results show that replacement of freshwater forested specialist species, such as *Taxodium distichum* and *Nyssa aquatica*, by generalists with broad germination requirements may be exacerbated by warming winter temperatures, further accelerating ongoing habitat homogenization. Though many factors contribute to shifts in species abundance, rising winter temperatures throughout the range of these species may contribute to the limitations on *N. aquatica*'s presence. This should be of concern for habitat managers as changes in freshwater forested wetland and bottomland forest community composition can challenge long standing ecosystem functions and services, including biodiversity, carbon sequestration and storage, nutrient cycling, and flood mitigation (Hooper et al., 2005; Lougheed et al., 2008).

Opportunities for future inquiry

From this study we have identified several areas of opportunity for future inquiry. First, we recognize that although germination percentage and rates are an important part of a species' reproductive success; to best predict community composition dynamics we also need information about total seed rain. Similar to other aspects of a species' reproductive ecology, seed production per tree may vary across geographic ranges, environmental conditions, or even within the individual's life-span. Available information on average seed production for the species addressed in this study is limited, variable, and often sourced from older field studies. For example, Abbott (1974) found that *A. rubrum* can yield annual seed crops between 12,000 to 1,000,000 seeds depending on tree size. For *L. styraciflua*, seed balls may

contain between 8 to 56 sound seeds per ball, though there is little data available on the number of seed balls produced per tree per year or factors that predict seed fecundity (USDA, 1965; USDA, 1974). *Taxodium distichum* trees average 16 viable seeds per cone (Faulkner and Toliver, 1983), and while some seeds/cones are produced every year, good seed crops only occur at 3- to 5-year intervals (USDA 1974). Data on the actual quantity of seed/cone production for these species is obfuscated by forestry conventions to report number of seeds/cones per bushel rather than annual production by tree. We believe there is value in revisiting some basic descriptive ecology for species under current climate conditions with consideration of local- to landscape-scale phenological variability.

Next, cold stratification studies performed on many species use constant pre-germination cold exposure, albeit of variable durations. Climate data from throughout temperate regions show that not only will winters be of shorter lengths but may be interrupted (NOAA, 2023a). Weather station data sourced from the Great Dismal Swamp in Suffolk, VA, one of the northernmost range limits for all four species, showed that the maximum number of consecutive days where temperatures did not exceed 4° C was 18 days over a 30-year period (NOAA, 2023b). Consistent and consecutive cold winter days may not be a reality in many of the regions in which these species co-occur, thus research on the effects of interrupted cold exposure may be equally informative. It's important to recognize that the effects of cold stratification may depend on incubation temperatures during germination. Weber and Sorensen (1990) found that the effectiveness of stratification duration on *Pinus ponderosa* seed germination was correlated with incubation temperature (e.g., higher incubation temperatures promoted germination when paired with shorter stratification periods, whereas the effects of stratification were greater when paired with lower incubation temperatures). Germination responses to climate change may be generational (Fernández-Pascual, 2018). Chen et al. (2011) showed that in *Arabidopsis* spp. the climate conditions experienced by the mother plant is integrated into the fruit bearing its seeds (thermotolerance), which controls seed dormancy. The presence of this phenomena may aid species in adjusting to changing winter conditions, but deserves further study across taxa.

Finally, our results have implications for forest management. The range of species' hydrothermal germination niches has been used to explain overall species range limits and inform the conservation of threatened species and habitats (Rajapkshe et al., 2022). To operationalize this information, and promote biodiversity, forest managers are encouraged to explore controlled seed preparation (i.e., cold stratification) and manual seed dispersal in habitats where seed rain for desired species may be limited. Although using seed for forest enhancement is not a common practice in freshwater forested wetlands compared to seedling transplantation, which ideally has higher rates of survival, it may be a cost effective and labor-efficient option. Of course, successful seed dispersal must also consider other environmental factors that may limit seed germination and survival (e.g., light, hydrology, soil pH, etc.). Direct seeding has been employed as a successful restoration technique in neotropical savanna (Sampaio et al., 2019) and tropical forests (Palma and Laurance, 2015). This type of forest restoration or enhancement technique may be particularly useful in bottomland forests that have been severely disturbed or selectively logged.

Conclusion

Many factors contribute to biotic homogenization, including propagule pressure, dispersal abilities, life history traits, and tolerance of species to the biophysical attributes of their surrounding habitat (Qian and Guo, 2010). Worldwide, across multiple taxa, disturbances to habitat and climate have contributed to the replacement of specialist species by generalists (Clavel et al., 2010). The causes for species convergence may be attributed to a myriad of functional traits throughout an individual's life span, but factors that control seed germination dynamics are one of the first filters on community composition. Climate change brings about more variable temperature regimes, and with that, conditions that will favor species with broader germination requirements. Understanding the differential effects of changing winter temperatures on early life stage traits of temperate forest species is important to understanding the multiple interacting factors driving observed patterns of habitat homogenization, particularly in systems like wetlands characterized by species with broad tolerances to other abiotic stressors.

Declarations

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Competing Interests

The authors have no relevant financial or non-financial interests to disclose.

Author Contributions

All authors conceptualized the research and wrote and edited the original draft. Kori Carr developed the methodology, acquired research funds, and collected data. Kori Carr and Taylor Sloey analyzed data. Taylor Sloey prepared the figures. All authors read and approved the final manuscript.

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Figures

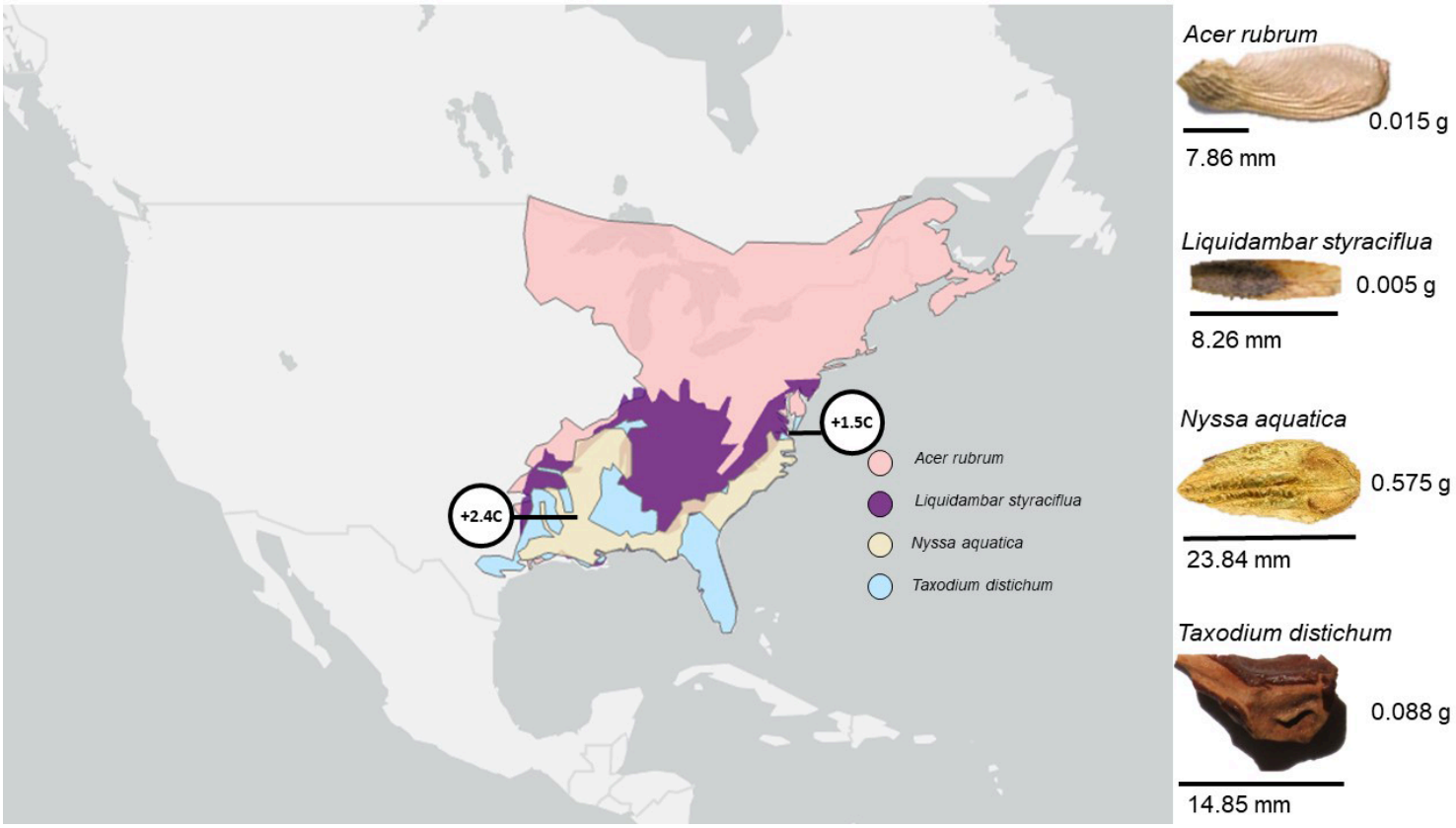

Figure 1

Native range limits of *A. rubrum* (red), *L. styraciflua* (violet), *N. aquatica* (yellow), and *T. distichum* (blue) in the United States and century trends in winter minimum temperatures (November - March) at two latitudinal range limits at which all species co-occur (Norfolk, VA, and Shreveport, LA). Temperature data were sourced from NOAA (2023), and species range limits were provided by USDA (2023). Right panel: mean length and mass of seeds from each species.

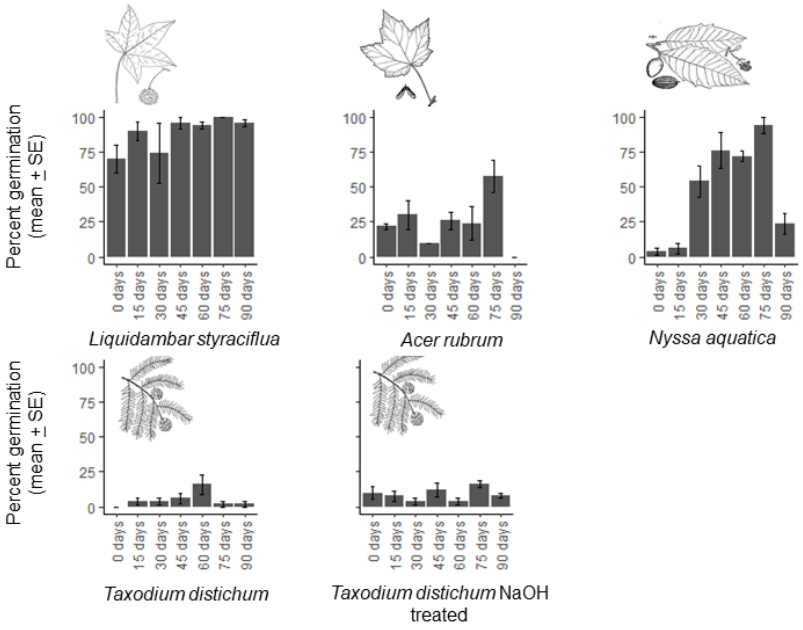

Figure 2

Percent germination (mean ± SE) of seeds from freshwater forested wetland tree species (*L. styraciflua, A. rubrum, N. aquatica, T. distichum,* and NaOH-treated *T. distichum*) exposed to cold stratification durations ranging from 0 days to 90 days. Symbols above bars indicate significant differences within species revealed by species-specific Tukey posthoc analyses. Different letters indicate significant ($p<0.05$) differences between groups. Vector images were sourced from open access image sources: Vectorstock, Alamy Stock Photo, and Vecteezy.

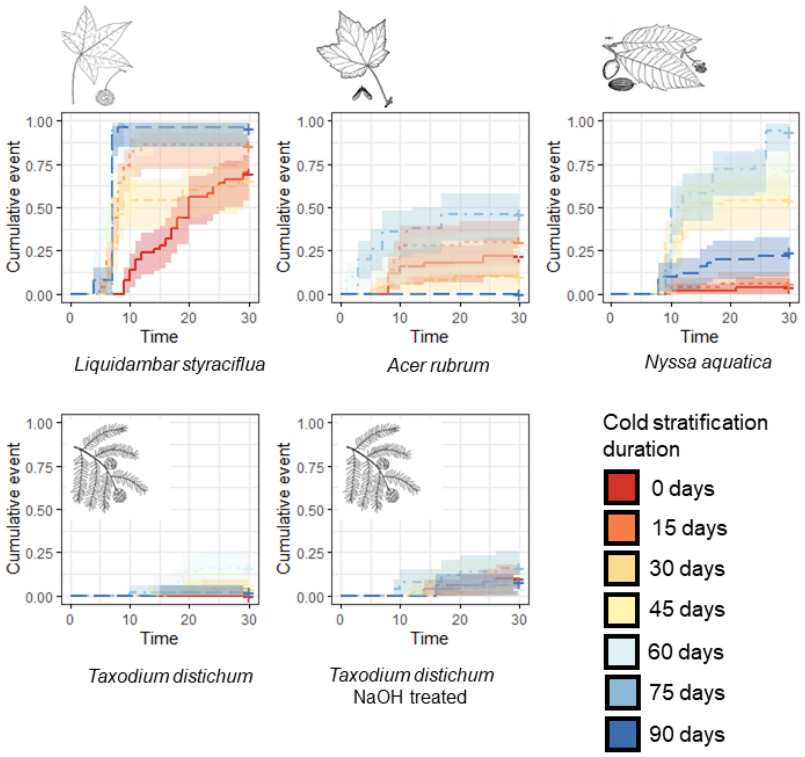

Figure 3

Kaplan-Meier estimates of event functions (i.e., germination) for *L. styraciflua, A. rubrum, N. aquatica, T. distichum*, and treated *T. distichum* seeds exposed to varying durations of pre-germination cold stratification over a 30-day observation period. Shading represents the 95% confidence interval.

Supplementary Files

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- SupplementaryTables.docx