ABSTRACT: A greater than 98% decline in *Arundinaria gigantea* (Walt.) Muhl. (giant cane or river cane) canebrake communities have resulted in a critically endangered ecosystem. Historical accounts suggest loss of canebrake habitat has resulted in the extirpation (and perhaps extinction) of many species. Thus, canebrake restoration is necessary for maintaining and enhancing biodiversity in the southeastern United States. The purpose of this study is to facilitate reestablishing *A. gigantea* canebrakes by optimizing growth and survivorship conditions for seedlings. To this end, we developed a lab experiment to test the effects of soil moisture (periodic drought, periodic flooding, and well-drained) and nitrogen fertilization on *A. gigantea* seedling growth and physiology, *Arundinaria gigantea*, while able to survive both drought and flooding, had significantly greater growth under well-drained conditions. Shoot growth averaged 28 cm and root growth averaged 11 cm in well-drained pots. Seedlings grown in flooded and drought treatments were not significantly different, averaging 20 cm in shoot growth and 4 cm in root growth. Biomass results mirrored shoot and root growth; biomass of roots and shoots was 30% greater under well-drained conditions. Nitrogen fertilization did not significantly affect seedling growth or physiology. Neither stomatal conductance nor net photosynthesis was significantly affected by moisture regime or nitrogen fertilization. However, results imply soil moisture regime as a significant factor controlling the growth and distribution of *A. gigantea*, and thus canebrake restoration will likely be most successful on well-drained soils.

Index terms: *Arundinaria gigantea*, cane, community restoration, ecology, seedling physiology

INTRODUCTION

Historical accounts suggest a 98% decline in *Arundinaria gigantea* (Walt.) Muhl. canebrake communities, resulting in an ecosystem determined to be endangered (Noss et al. 1995). Loss of canebrake habitat has resulted in the extirpation (and perhaps extinction) of several faunal species (Platt et al. 2001). Thus, canebrake restoration is necessary for maintaining and enhancing biodiversity in the southeastern United States. Transplantation attempts to reintroduce cane, however, have met with limited success (Feeback and Luken 1992; Platt and Brantley 1993). In addition, our understanding of the environmental constraints of cane growth is hampered by its fragmented distribution. The goal of our research was to determine the effects of moisture regime and supplementary nitrate-nitrogen on *A. gigantea* (giant cane or river cane) seedling growth for use in management and restoration of canebrakes.

Extensive canebrakes of *A. gigantea*, North America’s only native bamboo, were historically found throughout the southeastern United States, either as the understory of large widely spaced trees (Platt and Brantley 1997) or as treeless areas sometimes extending several miles (Nuttall 1821, Roosevelt 1908). Although historical accounts of canebrakes suggest *A. gigantea* tolerated a variety of environmental conditions (Fralish and Franklin 2002), soil moisture and soil nutrients were posited as two important growth parameters.

The largest canebrakes and areas of best growth occurred with moist soils (Feeback and Luken 1992, Platt and Brantley 1997). Most accounts suggest canebrakes were restricted to seepages, stream terraces, or ridges and natural levees of stream bottoms (Schneider and Sharitz 1986, Nelson 1997). Platt and Brantley (1997) suggested these periodically flooded areas maintained moist soils but not prolonged periods of inundation. Propagation methods corroborate an affinity for aerated soil, clearly showing the need for good drainage (Judziewicz et al. 1999, Meredith 2001). However, canebrake soils of the Kentucky karst plain ranged from good drainage to poor drainage (Baskin et al. 1997), again suggesting a wide range of tolerance. The U.S. Department of Agriculture (Natural Resources Conservation Service (NCRS) Plants Profile) classified *A. gigantea* as having medium drought tolerance and medium anaerobic tolerance, but we have found no clear studies of these soil factors.

Feeback and Luken (1992) and Platt and Brantley (1992) suggest canebrakes were found in rich (high nutrient) soils. Settlers regarded canebrakes as fertile areas, a factor leading to their demise. However, Farrelly (1984) found canebrake soils ranged from sandy to highly acidic, suggesting a
wide tolerance to soil nutrient conditions. Cultivation of bamboo also suggests nutrients may be a factor in growth and distribution of bamboo taxa in general. Meredith (2001) advises fertilizing similar to other grasses with relatively higher amounts of nitrogen than other nutrients. Judziewicz et al. (1999) also suggests regular, monthly fertilization. Reecht and Wetterwald (1999) suggest periodic fertilization (especially nitrogen and silica) based on the typical growth characteristics of bamboo; fertilize when culms sprout and three months later when rhizomes substantially develop. The implication of these studies is that nutrients may be a major constraining factor on A. gigantea growth and, thus, an important component of restoration efforts. Indeed, Hughes (1951) found that seedlings grew better under fertilized treatments, and Feeback and Luken (1992) advise fertilizing transferred rhizomes when restoring canebrakes.

Because canebrakes provide a habitat for a diversity of fauna, including endangered butterflies (Platt et al. 2001) and avifauna, such as Swainson’s warbler (Limnothlypis swainsonii), and because so little is known about the physiological and ecological requirements of cane, research is needed to determine factors affecting this unique ecosystem (Thomas et al. 1996). However, most of the canebrake habitat has been lost due to lack of fire disturbance, dampening of a flood cycle (perhaps a nutrient subsidy, Odum 1979), replacement by cultivated fields, or being eaten by domestic livestock (Hughes 1966, Platt and Brantley 1997). Thus, the current distribution of cane does not necessarily reflect the physiological or ecological tolerances for various environmental conditions that impact growth and survivorship of this species. To gain a greater understanding of the environmental constraints on canebrakes, we developed an experiment to test the effects of moisture regime (flood, drought, and moist/well-drained) and nitrogen fertilization (supplementary nitrogen and no nitrogen) on A. gigantea seedling growth and physiology. We hypothesized that A. gigantea seedling biomass and height growth would be greatest under moist, well-drained soils with enhanced nitrate, and that moisture would be the main factor controlling seedling growth. We further hypothesized that physiological parameters of stomatal conductance and net photosynthesis would be reduced under flood and drought conditions. An underlying theory was that A. gigantea has adapted to the natural pulse of flooding (Middleton 1999) and the simultaneous nutrient pulse (Odum 1979), and would grow best under these mimicked conditions.

**METHODS**

We set up a two-factor, completely crossed experimental design to examine moisture regime (three levels) and nitrogen fertilization (two levels) effects on A. gigantea seedling growth. For the moisture regime, we mimicked three conditions: (1) moist, well-drained soils, (2) flooded soils, and (3) drought conditions. To accomplish these treatments, nested pipes were prepared consisting of a 46 cm section of 5.08 cm PVC pipe for the inner pipe (planting pot) and 51 cm section of 10.16 cm PVC pipe for the outer pipe. Both inner and outer pipes were capped at the base and sealed to retain water, but three holes were drilled in the lower 6 cm of the inner pipe for drainage. The inner pipes were filled to approximately 10 cm from the top with a soil mixture of 60% play sand (for drainage) and 40% soil taken from a natural cane site (to mimic natural soil nutrient processes) at the Edward J. Meeman Biological Field Station. Soil series used for the canebrake plantings were Falaya Silt Loam, described as somewhat poorly drained and very silty (Sease et al. 1989).

We grew Arundinaria gigantea seedlings from seed (supplied by Adam Turtle, Summertown, TN) to 5 to 15 cm in height. We measured two randomly chosen individual plants for shoot length, root length, and number of leaves prior to planting and planted in each pot. There were sixty pots with ten replicates for each of the six treatments (n = 10). All pots were fertilized with ~50 ml of Peter’s 20-20-20 Professional Plant Food (1 tsp gal⁻¹) and then watered every other day for two weeks to standardize conditions prior to beginning treatments; treatments began on day 14. We grew the plants under laboratory conditions of 16 h light - 8 h dark, and 26.0 °C. We measured shoot length monthly to the nearest 0.01 cm and included the total length from all shoots of each individual. We calculated average shoot length from the two individual plants in each pot prior to statistical analyses. In addition, we measured stomatal conductance and net photosynthesis every two weeks using a portable gas exchange analyzer (Model Ciras 1, PPSystems, Havervile, MA, USA), and we used repeated measures analysis of variance to examine effects of moisture regime and nitrogen fertilization on shoot growth and physiological parameters.

At the completion of the experiment, we extracted, separated, and dried individual shoots at 60 °C for 48 hours, and then weighed to the nearest 0.001 g oven dry weight (O.D.W. biomass). We counted the number of leaves on each individual and subtracted from the initial number of leaves. We calculated total growth by subtracting initial root and shoot lengths...
from final root and shoot lengths. Before statistical analyses, we calculated, from the two individual plants in each pot, the average shoot and root length, number of leaves, and shoot and root biomass. We performed two-factor analysis of variance (ANOVA) tests followed by a post-hoc Student-Newman-Keuls procedure to test for significant differences in total length growth, biomass and in number of leaves due to moisture regime and nitrogen fertilization, as well as any interaction. All statistical tests were performed in SAS (SAS Inst. 1999).

RESULTS

Seedling shoot heights were significantly affected by the interaction of moisture regime and nitrogen fertilization (Table 1, Figure 1). Well-drained treatments had higher shoot growth with nitrogen fertilization than without fertilization, while nitrogen had no effect on plants in drought and flood treatments. Moisture significantly affected shoot heights during the last time period, where seedlings in well-drained conditions grew significantly more than seedlings in drought and flood treatments.

Nitrogen had no effect on number of leaves, biomass of roots and shoots, or lengths of roots and shoots after 88 days (Table 2). However, moisture regime significantly affected seedling growth, both length and biomass. Well-drained conditions rendered significantly greater biomass (nearly two times) of both shoots (mean = 0.79 g O.D.W.) and roots (mean = 0.76 g O.D.W.) than drought and flooding treatments, while drought and flooding were not significantly different (Table 2, Figure 2a). The number of leaves also was significantly affected by moisture regime (Table 2); well-drained treatments had the greatest number of leaves (mean = 23) and drought treatments the fewest (mean = 15, Figure 2b). Seedling shoot length and root length mirrored biomass results, both significantly greater under well-drained conditions (p (> F) < 0.001). Seedlings under well-drained conditions had significantly greater shoot (mean growth = 28.58 cm) and root growth (mean growth = 11.16 cm) over the course of the study than seedlings in drought and flood treatments (Figure 3); lengths of seedling roots and shoots under drought and flood conditions were not significantly different.

Table 1. Effects of nitrogen fertilization (two levels, no fertilization and fertilization) and moisture regime (three levels, well-drained, drought, and flooded) on Arundinaria gigantea (giant cane) cumulative seedling height growth and gas exchange over an 88-day period.

<table>
<thead>
<tr>
<th>Variable</th>
<th>F</th>
<th>p (&gt; F)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shoot Height</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Time</td>
<td>187.8</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Time * Nitrogen</td>
<td>1.26</td>
<td>0.281</td>
</tr>
<tr>
<td>Time * Moisture</td>
<td>2.42</td>
<td>0.031</td>
</tr>
<tr>
<td>Time * Moisture * Nitrogen</td>
<td>3.93</td>
<td>0.013</td>
</tr>
<tr>
<td>Stomatal Conductance</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Time</td>
<td>3.29</td>
<td>0.015</td>
</tr>
<tr>
<td>Time * Nitrogen</td>
<td>0.99</td>
<td>0.439</td>
</tr>
<tr>
<td>Time * Moisture</td>
<td>1.54</td>
<td>0.143</td>
</tr>
<tr>
<td>Time * Moisture * Nitrogen</td>
<td>0.54</td>
<td>0.856</td>
</tr>
<tr>
<td>Net Photosynthesis</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Time</td>
<td>3.57</td>
<td>0.010</td>
</tr>
<tr>
<td>Time * Nitrogen</td>
<td>0.44</td>
<td>0.817</td>
</tr>
<tr>
<td>Time * Moisture</td>
<td>1.64</td>
<td>0.110</td>
</tr>
<tr>
<td>Time * Moisture * Nitrogen</td>
<td>1.03</td>
<td>0.428</td>
</tr>
</tbody>
</table>

Figure 1. Effects of nitrogen fertilization (two levels, no fertilization and fertilization) and moisture regime (three levels, well-drained, drought, and flooded) on Arundinaria gigantea (giant cane) seedling cumulative height growth over an 88-day period. Dotted line indicates day 14 when treatments were started. MOIST = there was a significant interaction of moisture and nitrogen treatments based on repeated measures analysis, but only moisture had a significant effect among time periods, and only during the third time period.
trends that matched biomass and growth results. Stomatal conductance ranged from 18 mmol m$^{-2}$ sec$^{-1}$ to 62 mmol m$^{-2}$ sec$^{-1}$. Net photosynthesis ranged from 1.8 µmol m$^{-2}$ sec$^{-1}$ to 4 µmol m$^{-2}$ sec$^{-1}$. Elevated levels of stomatal conductance equated to higher net photosynthesis.

DISCUSSION

Although canebrakes represent one of the most rare communities of the southeastern United States, they have received little attention (Brantley and Platt 2001), and land stewards lack information on their structure, composition (flora and fauna), distribution, and function. As hypothesized, our results suggest moisture is a major factor controlling the growth and thus distribution of canebrakes, but we failed to find a significant effect on seedling physiology. Nitrogen fertilization at the levels applied did not significantly affect growth or physiology of *Arundinaria gigantea* seedlings, perhaps due to low nitrogen level and low light intensities in the growth room.

It was evident from both the biomass data and growth data that well-drained moisture condition provided the best environment for seedling growth. Growth, recorded by biomass, shoot and root length, and number of leaves, was approximately 30% greater in seedlings grown under well-drained conditions as compared to those grown in flood and drought conditions. This corroborates the current distribution of canebrakes, found generally on stream terraces and ridges of stream bottoms (Nelson 1997), but not in areas permanently inundated (Platt and Brantley 1997). The results also support advice for propagation that specify good drainage (Judziewicz et al. 1999, Meredith 2001).

Contrary to our hypotheses, and contrary to the obvious morphological changes, we found no significant differences in net photosynthetic rates or stomatal conductance of *A. gigantea* seedlings among the moisture regime treatments. Little is known of *A. gigantea* physiology except that it uses a C$_3$ photosynthesis pathway, as do all bamboo taxa (Judziewicz et al. 1999). One conclusion is that *A. gigantea* has adapted to fluctuating moisture environments of
floodplains through morphological changes rather than a plastic physiological response. For example, seedlings in well-drained conditions averaged 22% more leaves than seedlings in flooded conditions and they averaged 35% more leaves than seedlings in drought conditions. If greater number of leaves equates to greater total leaf surface area, seedlings in well-drained soils would ultimately yield greater growth if photosynthetic rates were similar among treatments. The results help explain the significant difference found in biomass between well-drained plants and those in flooded and in drought conditions and the insignificant, but slightly better, growth in flooded pots compared to growth in drought pots.

However, growth was obviously inhibited under flooded and drought conditions. Several species exhibit similar responses morphologically and physiologically to flood and drought conditions, including loss of leaves, decreased stomatal conductance, and decreased net photosynthesis (Kozlowski and Pallardy 1997, Pezeshki 1994, 2001, Elcan and Pezeshki 2002). Unfortunately, the extent of drought (drop in soil or plant water potential) is not known in the present study; thus, the threshold water potential at which plant’s gas exchange will respond has not been identified. In addition, the intensity of soil reduction in flooded conditions was not quantified.

The lack of a nitrogen fertilization effect was surprising, given that both restoration (Feeback and Luken 1992) and horticultural (Meredith 2001) researchers suggest its importance to cane growth and distribution. Contrary to our hypotheses, nitrogen fertilization had minimal impact on seedling shoot growth and no effect on seedling physiology. There are three potential reasons for the lack of significant effects: (1) *A. gigantea* has a wide tolerance of soil nitrogen levels, (2) nitrogen was not the limiting factor, and (3) the level of nitrogen added was too low to significantly affect *A. gigantea* seedling growth. There is little doubt that cane is widely distributed under a variety of soils (Luken 1989, Fralish and Franklin 2002), and it likely tolerates a wide range of soil nutrient conditions. A wide tolerance may be beneficial due to the notoriously variable soil nutrient pools of bottomland forests (Jones et al. 1994). The limiting resource may not be nitrogen, but instead one of the other nutrients suggested by researchers. Recht and Wetterwald (1999) suggest silica may be limiting, and Judziewicz et al. (1999) suggest iron may limit bamboo growth in general. Phosphorus may also limit the distribution of cane since phosphorus is a typical limiting nutrient in wetland ecosystems (Mitsch and Gosselink 1993).

Another possible reason for the lack of nitrogen fertilizer effects on *A. gigantea* seedling growth was that our nutrient treatment mimicked a natural nutrient pulse (Odum 1979, Middleton 1999) from an adjacent stream rather than a strong fertilization treatment. The difference in our natural fertilization versus a typical application of fertilizer may explain the inconsistency in experimental results given by Hughes (1951) – who found increased growth of cane seedlings with fertilization – and results given in the present study. In a previous study, however, we found full fertilization (as recommended by manufacturers of fertilizers) had deleterious effects on cane seedling growth (Cirtain et al. 2003), resulting in about one-half the growth compared to seedlings subjected to a diluted nutrient solution. The two studies suggest some optimal fertilization may occur between full nutrient additions and highly diluted conditions (as in the present study). Comparing our control and nitrogen fertilization treatments under well-drained conditions, the nitrogen-supplemented treatment did have significantly greater cumulative shoot growth. For the practice of restoration, perhaps a greater nitrogen supplement would yield increased growth and vigor, but this has yet to be shown experimentally.

A final point to discuss is the use of seedlings with a species that seldom flowers...
Rhizome growth shoots from rhizomes may grow > 30 cm (Hughes 1951). Indeed, we observed no seedling root systems may remain simple conditions with adequate light. In addition, (Cirtain et al. 2003) under well-drained study and > 23 cm in a previous study better growth than Hughes found; shoot development, but still only reached heights < 30 cm after three years of growth. Artificially propagated seedlings had faster heights < 30 cm after three years of growth. Hughes (1951) found that natural much greater number of plants, it is unprobable through micropropagation. Virtually impossible by transplanting but

While micropropagation will provide a much greater number of plants, it is unclear if the overall brake restoration will be faster. Hughes (1951) found that natural seedlings developed very slowly, with heights < 30 cm after three years of growth. Artificially propagated seedlings had faster development, but still only reached heights of 8 cm to 13 cm after the first season’s growth. Data from our research suggests better growth than Hughes found; shoot height growth averaged > 28 cm in this study and > 23 cm in a previous study (Cirtain et al. 2003) under well-drained conditions with adequate light. In addition, seedling root systems may remain simple (no rhizome development) for several years (Hughes 1951). Indeed, we observed no rhizome development in the present study or in a previous study (Cirtain et al. 2003). Converse to seedling development, new shoots from rhizomes may grow > 30 cm day⁻¹ (Meredith 2001). Rhizome growth may also be dramatic. Measurements taken at the Edward J. Meeman Biological Field Station in the loess soils of western Tennessee (near Memphis, Tenn.) show cane invading old fields at a rate of 2-3 m year⁻¹ (Personal observation, SBF). Without rhizome development, i.e., vegetative propagation, restored canebrakes would fail to expand or pervade.

CONCLUSIONS FOR CANEBRAKE RESTORATION

Based on the present study, moisture regime is a major factor controlling the growth and propagation of Arundinaria gigantea. A second major factor, based on previous research (Cirtain et al. 2003) and horticultural advice (Recht and Wetterwald 1999), is adequate light. The current ubiquitous occurrence of cane along forest edges, and lack of cane under full forest canopies, suggests A. gigantea is intolerant of shade. We believe these optimum seedling conditions will hold for rhizome transplants as well. Thus, successful establishment of canebrakes will require well-drained soil conditions with adequate light. There are two viable alternatives for canebrake restoration. First, as suggested by Platt and Brantley (1992), cane rhizomes could be transplanted to old fields. The present research suggests that transplant areas should have good drainage conditions, potentially a problem with post-agriculture soils. A quick analysis of the bulk density, porosity, or infiltration of the restoration area prior to significant effort may minimize failed restorations. Poor-drainage soils should be avoided or somehow altered to alleviate drainage problems.

A second alternative would be to release remnant stands of canebrakes currently found in the understories of dense forest by thinning the canopy. In this case, a possible assumption may be made that adequate drainage exists since cane is already present and the major factor limiting growth is light. The thinning method would offer a different microclimate for cane growth than field restorations, with less potential wind desiccation and higher humidity compared to open field conditions. Due to the general sensitivity of bamboo taxa to humidity and wind desiccation (Judziewicz et al. 1999, Recht and Wetterwald 1999, Meredith 2001), these conditions may prove better for growth. In addition, no transplanting of cane rhizomes may be necessary, depending on the current density of stems in the remnant brake.

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Margaret Cirtain, a doctoral graduate student with Dr. Scott Franklin as major professor, is researching propagation and establishment of Arundinaria gigantea canebrakes.

Scott Franklin, an Associate Professor at the University of Memphis, is a community ecologist. He studies disturbance and vegetation dynamics with interest in spatial and temporal dynamics.

Reza Pezeshki, a Full Professor at the University of Memphis, is a plant ecophysiologist. The major focuses of his research are the physiological ecology of wetland plants and the effects of natural and anthropogenic factors on plant survival, productivity, and distribution.

LITERATURE CITED


