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Biomass Yield and Nutrient Removal Rates of Perennial Grasses under Nitrogen Fertilization

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Abstract Perennial grasses may provide a renewable source of biomass for energy production. Biomass yield, nutrient concentrations, and nutrient removal rates of switchgrass (*Panicum virgatum* L.), giant miscanthus (*Miscanthus* x *giganteus*), giant reed (*Arundo donax* L.), weeping lovegrass [*Eragrostis curvula* (Shrad.) Nees], kleingrass (*Panicum coloratum* L.), and Johnsongrass (*Sorghum halepense* (L.) Pers.) were evaluated at four N fertilizer rates (0, 56, 112, or 168 kg Nha^{-1}) on a Minco fine sandy loam soil in southern Oklahoma. Species were established in 2008 and harvested for biomass in winter of 2009 and 2010. Biomass yield (dry matter basis) did not show a strong relationship with N fertilizer rate (p=0.08), but was affected by year and species interactions (p<0.01). Weeping lovegrass and kleingrass produced 29.0 and 16.0 Mg ha^{-1} in 2009, but only 13.0 Mg ha^{-1} and 9.8 Mg ha^{-1} in 2010, respectively. Biomass yields of giant reed, switchgrass, and Johnsongrass averaged 23.3, 17.8, and 6.0 Mg ha^{-1}, respectively. Giant miscanthus established poorly, producing only 4.7 Mg ha^{-1}. Across years, giant reed had the highest biomass yield, 33.2 Mg ha^{-1} at 168 kg Nha^{-1}, and the highest nutrient concentrations and removal rates (162 to 228 kg Nha^{-1}, 23 to 25 kg Pha^{-1}, and 121 to 149 kg Kha^{-1}) among the grasses. Although giant reed demonstrated tremendous biomass production, its higher nutrient removal rates indicate a potential for increased fertilization requirements over time. Switchgrass had consistently high biomass yields and relatively low nutrient removal rates (40 to 75 kg Nha^{-1}, 5 to 12 kg P ha^{-1}, and 44 to 110 kg Kha^{-1}) across years, demonstrating its merits as a low-input bioenergy crop.

Keywords Biomass yield · Biomass quality · Nitrogen fertilizer · Nutrient removal · Warm-season grasses

Introduction

Research sponsored by the US Department of Energy in the 1990s identified switchgrass (*Panicum virgatum* L.) as a model feedstock for energy production [1]. Switchgrass, a perennial grass native to North America, contained desirable agronomic and feedstock characteristics including high biomass yield potential, prolific seed production, adaptation to marginal environments, and high N-use efficiency [1, 2]. Studies have demonstrated origin, and ecotype of switchgrass cultivars affects its biomass yield potential [3, 4]. In general, cultivars selected from plant materials originating from northern latitudes flower earlier, produce less biomass, and have a longer winter dormant period than cultivars derived from southern latitudes when grown in the same environment [3, 4]. Lowland ecotypes of switchgrass tend to have bunch-type growth forms, thicker stems, shorter rhizomes, and more biomass production than upland ecotypes [3, 4]. Maximum yields of switchgrass in single harvest per year, biomass for energy production systems have been obtained with N fertilizer rates typically ranging from 120 to 168 kg ha^{-1}, depending on cultivar, age of stand, and harvest time [5–7]. Several studies support harvesting of switchgrass after frost to maximize transloca-

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tion of nutrients to stem bases, rhizomes, and roots before harvest [5, 6, 8].

Despite potential of switchgrass as a bioenergy crop, other perennial grasses, including giant miscanthus (*Miscanthus x giganteus*) and giant reed (*Arundo donax*), show promise for biomass energy production [2]. Miscanthus, a C-4 species native to Southeast Asia, has produced dry matter yields ranging from 10 to 25 Mg ha^{-1} in central and northern Europe and above 30 Mg ha^{-1} under irrigated conditions in southern Europe [2]. In the USA, dry matter yields of miscanthus and switchgrass in side-by-side trials averaged 30 and 10 Mg ha^{-1} [9]. Giant reed, a C-3 species native to Europe, produced 23 Mg ha^{-1} across a 6-year study in Italy [10]. In another study in Italy, giant reed produced 38 Mg ha^{-1} by the third year after establishment compared to 27 Mg ha^{-1} produced by miscanthus [11]. Under non-fertilized conditions in Georgia, USA, dry matter yields of giant reed and switchgrass were similar, averaging 6.4 and 8.6 Mg ha^{-1} [12].

Biomass for energy production also may come from locally adapted, perennial forage grasses. Dry biomass yields of switchgrass, bermudagrass (*Cynodon dactylon*), flaccidgrass (*Pennisetum flaccidum*), and weeping lovegrass increased with applications up to 134 kg Nha^{-1}, averaging 12.3, 10.5, 9.7, and 9.2 Mg ha^{-1}, respectively, in a single harvest per year system in Oklahoma, USA [13, 14]. Perennial grasses, such as kleingrass, weeping lovegrass, and Johnsongrass, have demonstrated high forage yield potential on marginal soils in this region [15]. A limitation to understanding the value of such grasses for biomass energy production systems is that their high biomass yields are often achieved through multiple defoliations per year. Fertilization requirements and biomass yield potential of these grasses under a single harvest system for biomass energy production has not been documented.

Sustainability of biomass energy production systems also depends on how fertilization rates affect concentration and removal of nutrients in harvested biomass [16–18]. Mineral concentrations affect biomass quality [19–21] and greater rates of removal in biomass harvests drive up fertilizer input costs [13, 14]. Biomass quality depends on whether conversion systems use biochemical, thermochemical, or direct combustion processes [19, 20]. Perennial grasses remobilize nutrients from above to belowground structures across the growing season, which, depending on harvest period, has an effect on the levels of N, P, and K in harvested material [19, 22]. Harvesting after plant senescence reduces mineral concentrations in biomass, desirable characteristics for direct combustion and thermochemical conversion systems [19, 20, 22]. Harvesting during early winter after a killing frost was recommended to minimize mineral concentrations and optimize biomass yields in perennial grass stands [8, 18, 22].

Although a number of studies [5, 18] have evaluated biomass yields and fertilizer responses of switchgrass managed for biomass energy production, research on biomass yields, nutrient concentrations, and nutrient removal rates of other perennial grasses remains limited. Thus, the overall objective of this research was to begin to address this knowledge gap for the southern Great Plains region of USA. Specific aims within this study were to: (1) quantify effects of N fertilizer rate on biomass yields of locally adapted forage and promising biomass energy grasses including switchgrass, giant miscanthus, giant reed, weeping lovegrass, kleingrass, and Johnsongrass harvested once per year under a single, after-frost system; (2) document changes in nutrient concentrations among these grasses across the growing season; and (3) determine nutrient removal rates from these grasses in the single, after-frost harvest system.

Materials and Methods

Experimental Design and Grass Establishment

The research was conducted at The Samuel Roberts Noble Foundation Red River Research and Demonstration Farm near Burneyville, OK (33°53′ N, 97°16′ W) from 2007 to 2010. Beginning in winter 2007, more than 600 individuals of the perennial grasses were established in a greenhouse with Metro-mix 350 rooting media (BWI Companies Inc., TX, USA). The media contained 45–55% horticultural grade vermiculite, bark, Canadian sphagnum peat moss, coarse perlite, bark ash, starter nutrient charge, gypsum and slow release nitrogen, and dolomitic limestone. Grass species included "EG1101" switchgrass (a selection derived from the cultivar Alamo), giant miscanthus, common giant reed, "Ermelo" weeping lovegrass, "Selection 75" kleingrass, and common Johnsongrass. Individuals of giant reed were propagated in 10.16-cm diameter pots from rhizomes, tillers, and stem cuttings collected from a local population near Ardmore (34°10′ N, 97°8′ W). Individuals of giant miscanthus were propagated in 10.16-cm diameter pots from rhizomes obtained from John Caveny (Monticello, IL, USA). All other species were started from seed in flats with 6.45 cm^2 cells. As these species outgrew their cells, they were transferred to 10.16-cm pots. Growth was kept in check throughout spring 2008 with constant trimming at a 30.5-cm height.

Following propagation in the greenhouse, the grasses were transplanted on 12 May 2008 into a tilled and disked Minco fine sandy loam soil (coarse-silty, mixed, superactive, thermic Udic Haplustolls). Six hundred individuals of each species were transplanted across 120, 9.3-m^2 plots. The plots were arranged in a randomized complete block design to accompany the six grass species, four nitrogen fertilizer rates (0, 56, 112, and 168 kg Nha^{-1}) and six replications. Twenty-five individuals of each species were transplanted in the monoculture plots at a 76.2-cm spacing within and between rows. During transplanting, plants were placed in shallow holes, and soil was firmed around the roots to improve root–soil contact. Plants were watered before transport to the field and irrigated the morning after transplanting with 25.4 mm of water to ensure stand establishment.

Stand Maintenance

The seedbed was weed-free at the time of transplanting, but crabgrass (*Digitaria sanguinalis* (L.) Scop) became problematic during the first few weeks thereafter. Mechanical weed control measures done in early July of 2008 included a combination of mowing and tilling between plants. Due to the absence of labeled herbicides and a desire to avoid losing stands, chemical weed control was not used during this study. Ants were controlled using a granulated pesticide, Amdro (active ingredient hydramethylnon, Ambrand Inc., Atlanta, GA, USA), on 29 August 2008. Two months after transplanting, percentage survival of individual plants averaged 95% for giant reed, 98% for switchgrass, 100% for kleingrass, 91% for Johnsongrass, and 97% for weeping lovegrass, and 55% for giant miscanthus. Dead plants were replaced during this time with new transplants.

Soil samples were collected from 0- to 15-cm depths in February 2009 and analyzed for pH at a 1:1 soil to water ratio [23], organic matter by high-temperature combustion [24], P by the Mehlich-3 procedure [25], and K via ammonium acetate extraction [26]. Soil had a pH of 5.3, 1.0% organic matter, 4 mg $\mathrm{N kg^{-1}}$, 54 mg $\mathrm{P kg^{-1}}$ and 60 mg $\mathrm{K kg^{-1}}$. In mid-March of each year, potassium chloride (0–0–60) at 134 kg $\mathrm{K_2O\ ha^{-1}}$ was applied to all plots. To minimize competition from weeds, N fertilizer rate treatments were not applied during establishment year of 2008. Urea (0–0–46) was applied at 0, 56, 112, or 168 kg $\mathrm{N ha^{-1}}$ to the assigned experimental units on 27 Mar 2009 and 20 Apr 2010.

Biomass Harvesting and Analysis

Whole-plot biomass yields were determined for each species during winter on 22 December 2009 and 26 January 2011. First fall frosts (< −2.5°C) occurred on 26 November 2009 and 25 November 2010. Whole-plot biomass was harvested at a 10-cm height from a 0.91× 3.05-m strip through the center of each plot with a Carter flail harvester (Carter Mfg Co., Inc., Brookston, IN, USA). Subsamples of the harvested biomass were then removed for determination of dry matter yield, analysis of nutrient concentrations (N, P, K, Ca, and Mg), and calculation of nutrient removal rates. Subsamples of biomass also were collected by clipping one individual plant of each species outside of the center strip at a 10-cm height in May, June, July, October, and December to determine how concentrations of N, P, K, Ca, and Mg varied across the growing season. In the October and December biomass collections, whole-plant subsamples of switchgrass and giant reed were further separated into stem, leaf, and inflorescence fractions to determine how nutrient concentrations varied among these fractions. Following all harvests, biomass samples were dried at 60°C in a forced-air oven for 3–4 days and then ground to pass a <1 mm screen using a Wiley Mill (Thomas Scientific, Swedesboro, NJ, USA).

Biomass samples were analyzed for N, P, K, Ca, and Mg using the Foss 6500 near infra-red reflectance spectroscopy (NIRS) instrument. The samples were scanned using Foss ISI Scan software and prediction equations developed by the NIRS Forage and Feed Testing Consortium (Hillsboro, WI, USA). The N concentration mean, standard error of validation, and r^2 for the equation were 19.9, 1.3, and 0.98 $\mathrm{g kg^{-1}}$, respectively. The P mean, standard error of validation, and r^2 for the equation were 1.9, 0.4, and 0.73 $\mathrm{g kg^{-1}}$, respectively. The K mean, standard error of validation, and r^2 for the equation were 16, 2.8, and 0.85 $\mathrm{g kg^{-1}}$, respectively. The Ca mean, standard error of validation, and r^2 for the equation were 4.9, 0.9, and 0.84 $\mathrm{g kg^{-1}}$, respectively. The Mg mean, standard error of validation, and r^2 for the equation were 2.6, 0.5, and 0.91 $\mathrm{g kg^{-1}}$, respectively. These equations were then used to predict N, P, K, Ca, and Mg for all samples.

An analysis of repeated measures data was conducted using the PROC MIXED procedure in SAS [27] to determine main effects and interactions of N rate, grass species, and year since data were collected on the same experimental unit across the 2 years of the experiment. Grass species, N rate, and year were considered fixed effects, and replications were considered random effects. Significance was determined at the $P<0.05$ level. The PDIFF feature of the LSMEANS procedure was used to compare means. Single degree of freedom contrasts were used to evaluate linear, quadratic, and cubic effects of nitrogen fertilizer on biomass. A repeated measures analysis using the mixed-models procedure also was conducted to determine main effects and interactions of N rate, species, and year on nutrient concentrations at the different biomass sampling periods (May, June, July, October, and December) and on nutrient concentration in the leaves and stems of switchgrass and giant reed (October and December). The statistical models applied the autoregressive (AR1) spatial power covariance structure to account for temporal data collection across years.

Results

Growing Conditions

During the establishment year of 2008, precipitation was lower (30–88%) than the long-term 30-year average for all months from April through December, except for August (Table 1). While it was higher in 2009, precipitation in 2010 was comparable to long-term 30-year average (Table 1). Due to lower than average monthly precipitation in June (16% and 64%) and August (41% and 49%) during the production years, supplemental irrigation (25.4 mm day^{-1}) was carried out on appropriate days during these months. Total annual amount (precipitation+irrigation) was 1,404 mm in 2009 and 1,040 mm in 2010. Although mean temperature in June during production years was 1.6°C higher than long-term 30-year average, mean annual temperature was 0.6°C lower.

Biomass Yield

Species

Year by species interactions for biomass yield were significant ($P<0.01$); therefore, means were reported by species. Biomass yield of giant reed increased by 18% from 21.4 Mg ha^{-1} in 2009 to 25.3 Mg ha^{-1} in 2010, whereas, biomass yield of all other species declined from 2009 to 2010 (Table 2). Biomass yield of switchgrass and Johnsongrass did not differ in 2009 and 2010 (17.8 and 6.0 Mg ha^{-1}, respectively). Whereas, biomass yield of kleingrass and weeping lovegrass declined from 16.0 and 29.0 Mg ha^{-1} in 2009 to 9.8 and 13.0 Mg ha^{-1} in 2010 (39% and 55%, respectively). Due to poor establishment after transplanting in 2008, giant miscanthus produced only 3.4 and 6.0 Mg ha^{-1} in 2009 and 2010, respectively. Giant miscanthus may need more precipitation or it may not be well adapted to sandy soils compared to these other species.

N Rate

Year by N rate interactions were not significant ($P=0.78$), but species by N rate interactions were significant ($P=0.02$), therefore, means were pooled across years and reported by species. Biomass yield of giant reed increased in a linear manner from 19.4 Mg ha^{-1} at 0 kg Nha^{-1} to 33.2 Mg ha^{-1} at 168 kg Nha^{-1} (Table 2). Biomass yield of switchgrass increased in a linear manner from 14.7 Mg ha^{-1} at 0 kg Nha^{-1} to 19.7 Mg ha^{-1} at 112 kg Nha^{-1}. Biomass yield of Johnsongrass increased quadratically with N fertilizer rate up to 112 kg Nha^{-1}. Weeping lovegrass, giant miscanthus, and kleingrass showed no response to N fertilizer rate.

Nutrient Concentrations

Year by species and year by sampling period interactions were not significant; however, sampling period by species interactions were significant for N, P, K, Ca, and Mg concentrations. Therefore, means were reported by species and by sampling period averaged across years (Table 3). Nutrient concentrations declined across the growing season in all species. In May, giant reed had 33.8 gN kg^{-1} compared to 23.7, 20.3, 18.9, and 18.6 gkg^{-1} found in

Table 1 Precipitation and temperature across 2008, 2009, 2010, and 30-year average for Burneyville, Oklahoma, USA

Month	Precipitation				Temperature			
	Average –(mm)–	2008	2009	2010	Average –(°C)–	2008	2009	2010
January	43	3	9	46	5.5	4.8	5.1	4.0
February	53	32	40	71	8.1	8.2	11.0	3.6
March	86	142	48	71	12.4	12.9	13.6	11.0
April	84	59	390	74	17.5	16.8	16.8	17.3
May	130	90	125	107	21.7	21.7	20.2	21.7
June	107	61	63	55	26.0	27.3	27.2	28.0
July	56	15	85	129	28.6	29.0	27.9	27.9
August	69	90	58	25	28.2	26.8	27.2	29.5
September	102	36	181	173	24.1	21.8	21.9	24.2
October	112	32	204	74	18.4	17.4	14.6	17.0
November	69	13	6	35	11.9	11.6	12.9	12.2
December	61	7	93	51	6.8	6.0	2.8	6.5
Total (mm) or mean (°C)	972	580	1,302	912	17.4	17.0	16.8	16.9

Table 2 Total annual biomass yield in response to N fertilizer rate of perennial grass species at Burneyville, Oklahoma, USA

Variable	Giant reed –Mg DM ha^{-1}–	Switchgrass	Kleingrass	Johnsongrass	Weeping lovegrass	Miscanthus
Year						
2009	21.4bY	19.5bcY	16.0cY	6.9dY	29.0aY	3.4eY
2010	25.3aY	16.0bY	9.8cdZ	5.0dY	13.0bcZ	6.0dY
N rate						
0	19.4abZ	14.7abY	13.7bcY	4.1cY	21.0aY	4.2cY
56	23.4aZ	17.2abY	9.6bcY	6.4cY	22.5aY	4.8bY
112	17.2abZ	19.7aY	12.5bcY	7.4cY	22.3aY	4.7dY
168	33.2aY	19.5bY	15.8bY	6.1cY	18.2bY	5.2cY
–P-value–						
Linear	0.08	0.09	0.25	0.07	0.49	0.43
Quadratic	0.16	0.55	0.09	0.04	0.43	0.46

Upper and lower case letters are for column and row comparison, respectively. For each variable (Year and N rate), values with same letter on a row or column are not significantly different at $P=0.05$

Table 3 Seasonal changes in whole-plant N, P, K, Ca, and Mg concentration of perennial grasses pooled across 2009 and 2010 seasons at Burneyville, Oklahoma, USA

Sampling period	Giant reed	Switchgrass	Kleingrass	Johnsongrass	Weeping lovegrass	Miscanthus
–g Nkg^{-1}–						
May	33.8a	18.9d	23.7b	24.2b	18.6d	20.3c
June	24.7a	13.1c	13.9bc	15.0b	13.9bc	14.9b
July	16.6a	9.6c	10.4bc	11.1b	11.3b	10.3bc
October	15.1a	6.9d	12.2b	12.2b	12.8b	8.0c
December	9.3b	3.7d	7.3c	11.1a	10.5a	7.9c
–g Pkg^{-1}–						
May	2.83a	2.38c	2.47b	2.48d	2.03d	2.33c
June	2.56a	2.08b	2.02bc	1.96c	1.7d	1.99c
July	2.21a	1.91b	1.75c	1.77c	1.52d	1.77c
October	1.82a	1.31d	1.51b	1.34 cd	1.38bc	1.77a
December	1.13a	0.92c	0.86cd	0.84 d	1.04b	0.72e
–g Kkg^{-1}–						
May	19.9cd	20.8bc	21.6ab	19.6d	15.9e	22.7a
June	17.4ab	18.3a	17.7ab	16.7b	14.6c	18.6a
July	15.6a	15.3ab	14.5b	15.4ab	12.3c	17.6a
October	11.7a	7.3e	10.2bc	8.6d	9.5cd	10.9ab
December	5.1b	7.2a	5.3b	3.8c	4.6b	7.2a
–g Ca kg^{-1}–						
May	6.5a	3.6c	4.4b	6.4a	3.1d	4.7b
June	5.8a	3.0d	4.2b	5.6a	3.7c	4.2b
July	5.1a	2.9d	4.0b	5.3a	3.0d	3.6c
October	4.8a	2.9bc	3.1b	5.1a	2.5c	4.8a
December	4.6a	2.3b	2.5c	4.8a	2.6b	4.9a
–g Mg kg^{-1}–						
May	6.1a	4.4c	4.1cd	5.8b	2.8e	3.9d
June	5.8a	3.6c	3.6c	5.1b	2.2e	3.7d
July	4.8a	3.5c	3.3c	4.4b	1.9d	3.1c
October	4.1a	2.8b	2.5c	4.2a	1.8d	2.5c
December	3.4a	2.4d	2.4d	3.0b	1.4e	2.7c

Values for a given element on a given month followed by same letter are not significantly different at $P=0.05$

kleingrass, giant miscanthus, switchgrass, and weeping lovegrass, respectively. By October, N concentration had dropped by 50% in all grass species with switchgrass showing the lowest concentration at 6.9 $\mathrm{g\,kg^{-1}}$. Between October and December, N content dropped by more than 50% in switchgrass and kleingrass but only by 22% to 38% in weeping lovegrass, giant miscanthus, and giant reed. Across years, tissue P concentration ranged between 2.03 and 2.83 $\mathrm{g\,kg^{-1}}$ in May. In all grass species, P concentration dropped 25% to 48% from May to October and by ≥50% from October to December, diminishing to approximately 1.0 $\mathrm{g\,kg^{-1}}$ by harvest time. Across years, K concentration ranged from 15.9 to 22.7 $\mathrm{g\,kg^{-1}}$ in May, with weeping lovegrass having the lowest concentration. Biomass K concentration dropped by up to 50% by October in nearly all species to <12 $\mathrm{g\,kg^{-1}}$. The drop in K contents between October and December averaged 49% in all species except switchgrass which showed slight changes. In all species, K concentration by harvest time in winter ranged from 2.3 to 4.3 $\mathrm{g\,kg^{-1}}$. Similar to other nutrients, Ca and Mg concentrations declined from May through December. However, changes in concentration of these two elements were relatively small, <20% between October and December. Giant reed and Johnsongrass maintained the highest concentration of Ca and Mg throughout the season while weeping lovegrass maintained the lowest concentration.

A notable pattern observed with giant reed was a significant amount of leaf loss between biomass sampling in October and December (Table 4). The number of leaves per stem of giant reed declined from 36 during the fall to 6 by winter. The percentage of biomass in leaves was 20% and 32% in October and 15% and <3% in December for switchgrass and giant reed, respectively (Table 5). Average leaf weight dropped drastically for giant reed from 1.39 g in October to 0.75 g in winter (Table 4). In switchgrass, a smaller reduction in leaf weight between October and December was observed. Except for stem moisture content in October, moisture content within leaves and stems of giant reed was higher than that within leaves and stems of switchgrass. In general, nutrient concentrations were higher in leaves and stems of giant reed ($P=0.05$) than in leaves and stems of switchgrass during both the October and December sampling times. Leaves also had comparatively higher mineral element concentrations than stems for the two grasses (Table 4). Based on the leaf mass to stem mass ratio, harvesting giant reed in the fall compared to winter would result in a higher biomass yield but the biomass would have much greater nutrient concentrations.

Table 4 Average leaf and stem weights, moisture content, and nutrient concentration of switchgrass and giant reed in October and December

Fraction	October		December	
	Giant reed	Switchgrass	Giant reed	Switchgrass
Leaves stem^{-1}	36a	7b	6a	6a
Leaf mass stem mass^{-1}	0.32a	0.20b	0.02b	0.15a
Leaf moisture (%)	57.6a	43.8b	12.5a	4.9b
Stem moisture (%)	51.1a	50.6a	43.3a	20.6b
–g plant^{-1}–				
Leaf	50.0a	1.86c	4.5a	1.1b
Stem	154a	9.2b	174a	7.4b
–g N kg^{-1}–				
Leaf	23.0a	11.6b	15.6a	7.6b
Stem	10.3a	5.5b	3.8a	1.4b
–g P kg^{-1}–				
Leaf	2.46a	1.27b	1.69a	1.10b
Stem	1.47a	1.22b	1.25a	1.11b
–g K kg^{-1}–				
Leaf	11.7a	5.7b	6.3a	2.3b
Stem	10.0a	7.1b	6.0a	4.5b
–g Ca kg^{-1}–				
Leaf	7.9a	4.5b	6.4a	5.9a
Stem	3.1a	2.7a	1.0a	1.2a
–g Mg kg^{-1}–				
Leaf	6.8a	4.3b	4.6a	3.8b
Stem	2.7a	2.6a	2.2b	2.5a

Values for with different letters on a row for a given element are significantly different at $P=0.05$

Table 5 Removal of N, P, K, Ca, and Mg in biomass harvested after frost (December) at Burneyville, Oklahoma, USA

Year	Giant reed	Switchgrass	Kleingrass	Johnsongrass	Weeping lovegrass	Miscanthus
–kg N kg^{-1}–						
2009	228b	75def	104d	85de	327a	20g
2010	162c	40e	74efg	54fg	118cd	31g
–kg P kg^{-1}–						
2009	25ab	16c	12cd	4fg	29a	3g
2010	23b	11cd	5ef	2fg	10de	4fg
–kg K kg^{-1}–						
2009	121bc	136abc	110c	38de	162a	24e
2010	149ab	141abc	44de	23e	61d	43de
–kg Ca kg^{-1}–						
2009	136a	66b	61b	53b	120a	20c
2010	68b	24c	20cd	21c	17c	18c
–kg Mg kg^{-1}–						
2009	111a	63b	51b	32c	58b	14d
2010	53b	32c	20cd	15d	13d	9d

Values for a given element followed by same letter are not significantly different at $P=0.05$

Nutrient Removal

Year by species interactions for nutrient removal were significant ($P<0.01$, Table 5) and similar to biomass yield interactions. Therefore, means were reported by year and species. In 2009, weeping lovegrass removed the highest amounts of N at 327 kg N ha^{-1} followed by giant reed at 228 kg N ha^{-1} (Table 5). Giant miscanthus removed the lowest amount, 20 kg N ha^{-1} (Table 5). Other species removed N at rates ranging from 75 to 104 kg N ha^{-1} in 2009; whereas N removal in 2010 ranged from 31 to 162 kg N ha^{-1}. Phosphorus removal rates varied by year and species. In 2009, P removal rates were greatest for giant reed and weeping lovegrass (25 and 29 kg P ha^{-1}, respectively) followed by switchgrass and kleingrass (16 and 12 kg P ha^{-1}, respectively), while giant miscanthus and Johnsongrass had the least (3 and 4 kg P ha^{-1}, respectively). In 2010, P removal rates were greatest for giant reed (23 kg P ha^{-1}), followed by switchgrass and weeping lovegrass (11 and 10 kg P ha^{-1}, respectively) followed by kleingrass, giant miscanthus, and Johnsongrass (5, 4, and 2 kg P ha^{-1}, respectively). Potassium removal rates also varied by year and species. Amount of K removed in 2009 and 2010 were similar for giant reed, switchgrass, and Johnsongrass and averaged removed 136, 134, and 30 kg ha^{-1}, respectively. Weeping lovegrass and kleingrass had 80% to 268% greater removal in 2009 (162 and 110 kg K ha^{-1}) compared to 2010 (44 and 61 kg K ha^{-1}), respectively. Giant miscanthus removed 80% less in 2009 compared to 2010 (24 and 43 kg K ha^{-1} in 2009 and 2010, respectively).

Calcium removal followed similar trend in that 2009 removal rates were 11% to 606% greater than 2010. In 2009, Ca removal rates were greatest for giant reed and weeping lovegrass (136 and 120 kg Ca ha^{-1}, respectively) followed by switchgrass, kleingrass, and Johnsongrass (66, 61, and 53 kg Ca ha^{-1}), while giant miscanthus had the least 20 kg Ca ha^{-1}. In 2010, Ca removal was greatest for giant reed (68 kg Ca ha^{-1}), while the other species did not differ ranging from 17 to 24 kg Ca ha^{-1}. Magnesium removal also followed similar trend in that 2009 removal rates were 55% to 346% greater than 2010. In 2009, Mg removal rates were greatest for giant reed (111 kg Mg ha^{-1}), followed by switchgrass, kleingrass, and weeping lovegrass (51 to 63 kg Mg ha^{-1}). Johnsongrass and giant miscanthus removed the least amount (32 and 14 kg ha^{-1}, respectively). In 2010, Mg removal was greatest for giant reed (53 kg Mg ha^{-1}) followed by switchgrass and kleingrass (20 to 32 kg Mg ha^{-1}), while Johnsongrass, weeping lovegrass, and giant miscanthus removed the least (9 to 15 kg Mg ha^{-1}).

Discussion

Giant reed showed tremendous potential for biomass yield. Higher biomass yield than the other grasses in the first year after establishment may have been due in part to its relatively larger vegetatively propagated transplants. These large transplants led to faster growth and an almost 100% transplant success in the establishment year of 2008. In the year after establishment, rhizomes had spread in the plot increasing tiller density. Had giant reed retained its leaves after a killing frost, biomass yield at harvest in winter may have been 30% higher. Leaf loss upon senescence appeared to be biased towards larger leaves as the average weight of a giant reed leaf in winter was 50% lower than that in the fall. High wind speeds, commonly experienced in the Great Plains, may lead to preferential loss of older and larger lower leaves compared to relatively young and smaller upper leaves. The tall height of giant reed, up to 3–4 m at physiological maturity, may have compounded leaf loss problems because of displacement and vibration from its vertical position during windy conditions [28]. Despite being a C-3 species growing on sandy soil in this southern environment, biomass yield of giant reed was higher than that of the other C-4 species grasses. Physiological and morphological traits that may support high biomass yields of giant reed including high net photosynthetic CO_2 uptake rates of 37 μmol m^{-2} s^{-1}, lack of light saturation, and little photoinhibition have been reported [29]. Giant reed grows in dense clumps, produces stems from dense, knotty rhizomes, and tolerates a wide range of soil conditions and types, surviving under wet and dry conditions [2].

Switchgrass biomass yields were similar between years and averaged 17.8 Mg ha^{-1}. These biomass yields were comparable to that reported before for the southeastern and south-central USA [16, 18, 30, 31], but higher than those reported for Midwestern states [7, 22]. Concentration of N in biomass sampled in July or December also were comparable to those reported earlier [16, 18, 30]. Concentration of P was comparable to those reported for Alamo receiving 38 kg Nha^{-1} in Tennessee [16] and 112 kg Nha^{-1} in south-central Oklahoma [18]. Changes in nutrient concentration from fall to winter were comparable to those found by others [32]. Nutrient removal in harvested biomass was comparable to those found by others for a single-cut, after-frost harvest system [16, 18, 30, 31].

Unlike other findings indicating higher biomass yield for giant miscanthus than switchgrass [33], poor establishment combined with adverse soil and environmental conditions contributed to low biomass yields for giant miscanthus in this study. It has been reported that giant miscanthus response to precipitation is better than that of switchgrass [33]. This study site had sandy soils and received low precipitation and registered high temperatures during the establishment year. The low precipitation and high summer temperatures in 2008 (Table 1) led to low establishment success in giant miscanthus which affected plant density, growth and yield in subsequent production years. High biomass yields in weeping lovegrass may be related to its ability to rapidly establish and develop a thick canopy. The average biomass yield of 21 Mg ha^{-1} was higher than 7.9 Mg ha^{-1} reported elsewhere for a single-cut, after-frost harvest system in northern Oklahoma [13, 14]. During the year of transplanting, rapid establishment appeared to give it an edge in resource use against weeds. A challenge with use of weeping lovegrass, however, was its susceptibility to harvest damage. It was observed that harvesting weeping lovegrass with the flail harvester partially uprooted these plants. Despite previous research showing that weeping lovegrass does well on sandy soils [34], the damage observed with mechanical harvesting may have been compounded by growing of these grasses on the sandy loam soil, where ability of roots to anchor the plant was lessened. Declines in biomass yields of weeping lovegrass from 2009 to 2010 likely resulted from compounding effects of mechanical damage, winter damage, and reduced rainfall. Harvesting weeping lovegrass after frost may have contributed to reduced yield in the second season as shown by other findings where harvesting after mid-fall was reported to predisposes the grass to winter damaged and reduced yield the following season [35].

Kleingrass produced relatively high biomass yields in the single-cut, after-frost harvest system despite its adaptation to multiple defoliations. Kleingrass has been shown to produce more than 6.0 Mg DM ha^{-1} $year^{-1}$ in multiple harvest forage systems in Texas [15]. Early flowering and maturity of kleingrass could enhance nutrient remobilization to underground organs, but may limit growth and biomass yields relative to other perennial grasses. A concern with kleingrass like that with weeping lovegrass was that harvesting of this densely tillered grass during the first year contributed to reduced productivity of the stands during the second year. Weeping lovegrass, switchgrass, and kleingrass grew more as bunchgrasses, with a distinct base from which tillers arose, unlike in weeping lovegrass and kleingrass, lack of harvest damage in switchgrass during the first year may have been due to a combination of better rooting depth and an open rather than a closed cluster of tillers that made harvesting easier and less likely to disturb the rooting system. Despite being considered a noxious weed, Johnsongrass may have potential for use as a bioenergy crop. Johnsongrass competes strongly with crops. In one study, Johnsongrass height, relative growth rate, and unit leaf area was 3-fold, 1.5-fold and 4-fold higher, respectively than that of cotton [36]. With a 10-fold greater root biomass and larger leaf area after 8 weeks of growth, Johnsongrass showed potential for greater resource use efficiency than cotton [36]. These qualities indicate a potential for Johnsongrass to use scarce resources efficiently for biomass production. In this study, Johnsongrass spread laterally with rhizomes and produced tillers in the inter-row spaces throughout the growing season, but senescenced earlier and experienced more lodging than the other grasses. Biomass produced in this study was lower than 9.7 Mg ha^{-1} $year^{-1}$ biomass yield obtained under a three-cut per year forage production in Mississippi [37]. The concentrations of P and K in the grasses were comparable to those found elsewhere [38]. Early onset of senescence may be positive for energy production, as the harvested material will have lower nutrient levels at harvest as shown for P and K in this study. However, early senescence also may have contributed to the increased lodging of Johnsongrass by the time of winter harvest.

The high nutrient concentrations early in the season for all species were attributed to predominance of young vegetative tissues. In late summer months, changing plant morphology with increased proportion of older tissue, reduced plant-nutrient demand and possible dilution due to increased biomass from rapid vegetative growth may explain reduced tissue element concentrations. Decreased mineral concentrations from fall to winter may be attributed to remobilization of nutrients from aboveground to belowground tissue during senescence. Similar scenarios has been widely reported by other researchers who found significant reduction in tissue element concentrations as harvest is delayed from fall to winter months [18, 19, 22, 36].

Low biomass nutrient concentration in 2010 especially for kleingrass and Johnsongrass which showed relatively early senescence was likely a result of a 1-month delayed harvest in 2010 compared to 2009. The delay may have led to nutrient leaching as reported previously for K [39]. The relatively high nutrient concentration in giant reed compared to other species during most of the season was possibly due to its botanical characteristics. Unlike all the other species, giant reed is a C-3 grass, a group that lacks bundle sheath cells as found in C-4 grasses, thus, allowing for larger mesophyll cells in C-3 plants [40, 41] and may potentially increase cytoplasmic content including mineral elements. High level of nutrients in giant reed is not surprising as previous studies [21, 42, 43] have shown that C-3 grasses have higher mineral elements than C-4 grasses.

Conclusions

Giant reed, switchgrass, weeping lovegrass, and kleingrass may have potential for biomass production under the climatic conditions prevalent in the southern Great Plains. On this sandy soil, harvesting weeping lovegrass and kleingrass in winter for 2 years after establishment, caused reduced yields in the third year. Establishment of giant miscanthus on the sandy soil in this study was problematic and more evaluation is required before its true potential as a bioenergy crop in semi-arid environments like southern Oklahoma is known. Giant reed, a C-3 species, appeared to be a strong alternative to switchgrass, the leading perennial C-4 grass candidate for biomass energy production. However, its higher nutrient removal rates under annual harvesting of this grass would increase fertilization requirements over time. Therefore, based on these results, it appears that switchgrass may be the best suited perennial grass for the southern Great Plains due to its relatively high biomass yield and relatively low nutrient removal compared to the other species evaluated. Future research on giant reed should aim at high yields and reduced nutrient removal through increased leaf retention and increased remobilization of nutrients at senescence.

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