DOI: 10.1002/agj2.21202

ARTICLE

Biofuels

Harvest frequency and harvest timing following a freeze event effects on yield and composition of switchgrass

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Assigned to Associate Editor Meetpal Kukal.

Funding information

Bioenergy Research Initiative of the North Carolina Department of Agriculture and Consumer Services Miguel S. Castillo D | Travis W. Gannon | Pe

Perejitei E. Bekewe

Abstract

Constant supply of biomass from the field is limited by the seasonality of production of warm-season grasses in the transition U.S. region. Delaying harvest after occurrence of freeze may be an alternative to extend the biomass supply period of switchgrass (Panicum virgatum L.) in North Carolina. The objectives of this study were to evaluate the effects of harvest frequency (HF) and harvest timing at the end of the growing season (HT) on switchgrass biomass yield, nutrient (N, P, and K) removal, and dry matter (DM) and ash concentrations. Treatments were the factorial combination of two HF (clipped once [1X] or twice [2X] per season) and three HT (before freeze in October, after first freeze in November, and late winter in February). Delaying harvest after occurrence of freeze did not affect total annual biomass vield for the 2X treatment (average of 15.5 Mg ha⁻¹), whereas for 1X yield declined from 14.4 to 10.1 Mg ha⁻¹ when harvest was delayed from October to February. Ash concentration declined from 29 g kg⁻¹ in October to 14 g kg⁻¹ in February. The DM concentration level reached in February was lowest (893 g kg⁻¹) and it would be considered safe for storage of biomass. Nutrient removal was consistently greater for 2X than 1X (ranging from 43 to 137, 3.6 to 25.1, and 54 to 213 kg ha⁻¹ for N, P, K, respectively). Delaying harvest of switchgrass after a freeze event is feasible when clipping twice a year to extend the window of biomass supply.

1 | INTRODUCTION

Finite fossil fuel reserves, government mandates for use of renewable energy, and controversy associated with the use of land dedicated to animal-agriculture vs. bioenergy production, have turned attention to the use of grasses as potential bioenergy feedstocks. Switchgrass is a C_4 warmseason perennial grass native to the North American prairies with potential to be used as a forage and bioenergy crop (Vogel, 2004). Positive attributes that make switchgrass a

Abbreviations: DM, dry matter; HF, harvest frequency; HT, harvest timing.

candidate bioenergy feedstock include high biomass production (McLaughlin & Adams Kszos, 2005; Parrish & Fike, 2005), greater biomass yield per unit of N uptake than C_3 grasses (Brown, 1978; Friesen & Cattani, 2017), adaptation to marginal lands that are less suitable for row crop production (Castillo et al., 2020; Jung et al., 1988), and drought tolerance (Barney et al., 2009; Liu et al., 2015). In addition, multiple harvest events per year successfully enable dual-use of switchgrass for forage and bioenergy (Bekewe et al., 2020; Burns et al., 1984; Mosali et al., 2013; Richner et al., 2014; Sanderson et al., 1999).

A major logistical issue concerning the use of biomass is its storage, especially when it is characterized by seasonal

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availability (Rentizelas et al., 2009). Delaying the harvest timing at the end of the growing season is an agronomic practice that may help achieve year-round supply of switchgrass biomass from the field directly to the bio-processing facilities in North Carolina. To the best of our knowledge, there are limited reports of biomass yield responses for switchgrass when harvest is delayed to late winter and after several freeze events in the upper Southeast U.S. transition region, and none using cultivar BoMaster of switchgrass. Working with energycane (Saccharum spp. hybrid) and elephantgrass (Pennisetum purpureum Schum.) in Florida, Na et al. (2014) reported that extending the harvest period up to 60 d after first freeze did not affect biomass yield for energycane, whereas biomass yield was $\approx 30\%$ lower for elephantgrass. Delaying the harvest timing at the end of the growing season from autumn to winter and to spring months resulted in $\approx 16-40\%$ less harvestable biomass, with greater reduction in years with above-average winter snowfall, in Iowa, Pennsylvania, Arkansas, and Texas (Adler et al., 2006; Ashworth et al., 2017; Sanderson et al., 1999; Wilson et al., 2013). In Pennsylvania, approximately 90% of the yield reduction was attributed to lodging and subsequent difficulties of field equipment to pick up the biomass (Adler et al., 2006). Delaying the harvest timing at the end of the growing season also affects the quality of the biomass (Na et al., 2014; Sanderson & Wolf, 1995); however, the quality of the feedstock for bioenergy use is defined by the postharvest conversion process and the utilization pathway, and it may drastically change with crop and biomass partition (Monti et al., 2008). For instance, the concentrations of water, minerals, and ash should be as low as possible for combustion processes.

The Southeast U.S. transition zone is a region characterized by the climate intersection of the cool-humid North and the warm-humid South. Information on the effects of in-season harvest management and harvest timing at the end of the growing season is critical to assess whether delayed harvest after freeze events can serve as an alternative for year-round supply of switchgrass biomass directly from the field to the bio-processing plants in this region. The objectives of this experiment were to quantify the effects of in-season harvest frequency and harvest timing at the end of the growing season on aboveground biomass yield, nutrient (N, P, and K) removal, and tissue dry matter (DM) and ash concentrations for switchgrass.

2 | MATERIALS AND METHODS

2.1 | Experimental site, plot management, and weather

The experiment was conducted for two growing seasons (2016–2017 and 2017–2018) at the Central Crops Research

Core Ideas

- Clipping twice per year allows delay of biomass harvest from October to February with no total annual yield loss.
- Dry matter and ash concentrations were not different between one- and two-time harvests in November and February.
- The dry matter concentration level reached in February would be safe for storage of the biomass.
- Nitrogen, P, and K removals were greater for twovs. one-time clippings.

Station, Clayton, NC (35°40′ N, 78°29′ W). A mature stand (>8 yr) of switchgrass cultivar BoMaster, originally planted from seeds, was used for this experiment. 'BoMaster' was released because of greater biomass yield and cellulose concentration compared with cultivars Alamo and Cave-in-Rock (Burns et al., 2008). Management of the experimental area prior to initiation of this project consisted of maintenance fertilization and a single-clipping event at the end of the growing season, followed by residue-burning in February of each year. The accumulated biomass from the 2015 growing season was clipped and removed from the plots in late September 2015 in preparation for this experiment.

The soil series at the experimental site is a Wedowee sandy loam (fine, kaolinitic, thermic Typic Kanhapludult). Soil samples for initial characterization were collected to a depth of 20 cm on February 2016 and analyzed at the Agronomic Division of the North Carolina Department of Agriculture and Consumer Services (NCDA&CS). Soil pH was 6.2, and Mehlich-3 extractable P and K concentrations were 622 and 205 mg kg⁻¹, respectively. According to the NCDA&CS soil test index system (Hardy et al., 2014), the P and K concentrations were classified as very high and high, respectively; therefore, soil amendments were not recommended. Fertilizer N was broadcasted in a single application in mid-April at a rate of 134 kg N ha⁻¹ using a granular formulation of pre-mixed urea-ammonium sulfate blend (340 g N kg⁻¹; 26% polymercoated urea plus 8% ammonium sulfate) in 2016, and at rate of 101 kg N ha⁻¹ using a granular formulation of ammonium nitrate (340 g N kg⁻¹) in 2017. Although N fertilization rates varied between years, the applied N rates in our study were in the higher range, or above, of the rates for which switchgrass responses to fertilizer N application have been reported in the region (Brejda, 2000; Obour et al., 2017; Wang et al., 2018). The dates of the last freeze events in the spring before the initiation of the growing season were 10 Apr. 2016 and 23 Apr. 2017. Total rainfall values were 1,591, 1,327, and 1,253 mm in 2016, 2017, and the 30-yr average, respectively. Monthly

rainfall and monthly average daily maximum and minimum temperatures data are presented in Figure 1. Dates of first freeze at the end of the growing season and harvest events are reported in Table 1.

2.2 | Treatments and experimental design

Treatments were the factorial combination (2×3) of harvest frequency (HF) and harvest timing at the end of the growing season (HT). The two HF levels were full-season growth (1X)and two harvests per season (2X). The HT treatment consisted of harvesting at three sampling dates based on occurrence of the first freeze. The three HT levels were before freeze occurrence in October (36 and 30 d before first freeze in 2016 and 2017, respectively), after first freeze in November (6 and 13 d after first freeze in 2016 and 2017, respectively), and late winter in February (88 and 94 d after first freeze in 2017 and 2018, respectively. The specific dates for HT treatments and first freeze events are presented in Table 1; hereafter, the specific HT treatments will be referred to as October, November, and February. For the 1X treatment, a single harvest occurred based on HT treatments. For the 2X treatment, there were two harvests per season. The first harvest occurred half-way through the active growing season for switchgrass in North Carolina on 22 and 15 June in 2016 and 2017, respectively, and the regrowth (second harvest) was harvested at the end of the growing season based on HT treatments.

The experimental design was a split-plot design with the main-plot factor arranged in a complete randomized block design replicated three times. The main-plot factor was HF and subplot factor was HT. Treatments in 2017 were imposed on the same corresponding experimental units as in 2016. The experimental unit size was 4.9-m wide by 4.9-m long with 2.4-m wide alleys between plots.

2.3 | Response variables

Biomass samples were collected by clipping a 7.5-m^2 area (2.5-m wide by 3-m long) to 15-cm stubble height using hedge trimmers. By using this approach, the area from which samples were collected was centered at the plots and at least 1.5-m away from the borders of the plots to minimize border effects. The clipped material was weighed fresh in the field



FIGURE 1 Monthly rainfall for 2016, 2017, and 30-yr average, and average temperatures (T. max. and T. min. for 2016 and 2017) at the Central Crops Research Station, Clayton, NC (35°40′ N, 78°29′ W). Total rainfall values were 1591, 1327, and 1253 mm in 2016, 2017, and the 30-yr average, respectively

and a subsample (\approx 1 kg fresh weight) was used to determine DM concentration. The DM concentration was estimated by drying the subsamples at 60 °C until constant weight using an air-forced drier. Dried subsamples were ground with a Thomas Wiley mill model (Thomas Scientific) to \leq 1-mm screen in preparation for tissue analyses.

Biomass yield was estimated by multiplying the DM concentration by the corresponding fresh weight of each treatment. For the 2X treatment, biomass yield was the summation of two harvest events, that is, harvested at mid-season in June plus the regrowth harvested according to HT treatments. After

TABLE 1 Sampling and freeze event dates during the experimental period at the Central Crop Research Station, Clayton, NC (35°40′ N, 78°29′ W)

	Harvest before		Harvest after	Second harvest
Season	freeze	First freeze	first freeze	after freeze
2016–2017	6 Oct. 2016	11 Nov. 2016	17 Nov. 2016	7 Feb. 2017
2017–2018	5 Oct. 2017	4 Nov. 2017	17 Nov. 2017	6 Feb. 2018

the last harvest event in February, all plots were cleared by clipping the standing biomass to the target stubble height and the clipped material was removed from the plots.

The removals of N, P, and K by the harvested tissue were calculated by multiplying tissue nutrient concentrations by the biomass yield values. For the 2X HF treatment, total seasonal nutrient removal was calculated by adding the removal values of the mid-season June harvest with the corresponding values of the end-of-season HT treatments. Tissue N concentration was determined by gas chromatography with a model NA1500s2 elemental analyzer from CE Elantech Instruments (CE Elantech) (AOAC, 1990; Campbell & Plank, 1992). Total tissue P and K concentrations were determined with inductively coupled plasma–optical emission spectrometry (ICP–OES) (Spectro Arcos EOP, Spectro Analytical) (Donohue & Aho, 1992; adapted USEPA, 2001). Ash concentration was determined using a muffle furnace and the loss on ignition methodology (Thiex et al., 2012).

2.4 | Statistical analysis

Analysis of variance was set up considering the effects of HF, HT, year, and all two- and three-way interactions as fixed effects; year was considered as repeated measured with compound symmetry as the covariance structure based on smaller AIC value; block was considered a random effect. Biomass yield and nutrient removal values for the 2X treatment correspond to the summation of the mid-season June sampling event and the end-of-season HT. For DM and ash concentrations analysis, the data for the 2X treatment correspond to the values of the plant regrowth after the mid-season June clipping harvested at the end of the growing season based on HT treatments. Analyses were performed using the GLIMMIX procedure of SAS (SAS Institute). When an interaction effect was significant, simple effects were analyzed using the SLICE procedure of SAS. All means reported are least square means and separation of means was done using the LINES option of LSMEANS. Plots of model residuals were used to check for normality. Treatment differences were declared significant if P < .05.

3 | **RESULTS AND DISCUSSION**

3.1 | Biomass yield

There was a significant HF × HT interaction effect. Biomass yield was not different between 1X and 2X (\approx 14.5 Mg ha⁻¹) in October; however, delaying the harvest to November and February resulted in lower biomass yield only for 1X (Figure 2). Biomass yield of switchgrass 1X was approximately 22 and 29% lower in November and February,



FIGURE 2 Biomass yield of 'BoMaster' switchgrass as a function of harvest frequency (one clipping [1X] and two clippings [2X] per season) and harvest timing at the end of the growing (before freeze in October, after freeze in November and February) in Clayton, NC ($35^{\circ}40'$ N, $78^{\circ}29'$ W). Data are means of 2 yr and three replicates per year. *P* values correspond to harvest frequency effects at each harvest timing. Lowercase letters were used to compare harvest timing mean values by harvest frequency treatments; means followed by the same lowercase letter are not different

respectively, compared with when it was harvested in October (14.4 Mg ha⁻¹). In contrast, biomass yield remained constant from October to February for the 2X clipping (average of 15.5 Mg ha⁻¹).

Temperature patterns, dates of freeze occurrence, and field sampling dates where consistent between the 2 yr in this experiment (Figure 1; Table 1). Biomass yield of switchgrass can vary widely due to several factors such as environment, crop management, and genotype (Casler & Boe, 2003; Fike et al., 2006a; Lemus et al., 2002; Seepaul et al., 2014). Across several cultivars and locations, biomass yield of switchgrass was either not different or up to $\approx 30\%$ greater for a twoclipping per year system compared with clipping once per year (Burns et al., 2010; Fike et al., 2006b; Guretzky et al., 2011). The 2X treatment in our experiment did not result in biomass yield differences compared with 1X when the endof-season harvest occured before freeze, like October in our experiment. The benefit of the 2X treatment is that it allows to extend the end-of-season harvest of switchgrass from October to February with no penalty loss for biomass yield.

Yield losses due to delayed harvest at the end of the growing season have been reported to range from 16 up to $\approx 40\%$ among several locations (Arkansas, Iowa, Pennsylvania, and Texas) and cultivars of switchgrass in the United States (Adler et al., 2006; Ashworth et al., 2017; Sanderson et al., **TABLE 2**Removal of N, P, and K of 'BoMaster' switchgrass as afunction of harvest frequency (one clipping [1X] and two clippings[2X] per season) and harvest timing at the end of the growing (beforefreeze in October, after freeze in November and February)

	Harvest frequency		
Harvest timing	1X	2X	P value ^a
	——kg ha	-1	
	N removal		
Oct.	_	-	-
Nov.	-	-	-
Feb.	_	-	-
Mean	42.7	136.7	<.01
SE	10.7		
	P removal		
Oct.	15.4a	25.1	<.01
Nov.	7.1b	24.8	<.01
Feb.	3.6b	21.4	<.01
Mean	-	-	
SE	1.6		
	K removal		
Oct.	_	-	-
Nov.	_	-	-
Feb.	_	-	-
Mean	54	213	<.01
SE	23.4		

Note. Means followed by the same lowercase letters within a column are not significantly different.

^a*P* value for the difference between harvest frequency treatments within harvest timing.

1999; Wilson et al., 2013). Plant tissue losses, mainly leaf losses, but also observed difficulty by the harvesting equipment to pick up stems, are factors attributed to the lower biomass yield when the end-of-season harvest is delayed to winter or spring months; however, the extent of the losses is site-specific (Adler et al., 2006; Na et al., 2014). In Pennsylvannia, switchgrass biomass yield decreased up to 43% when harvest was delayed from fall to spring (Adler et al., 2006). In northern Florida, delaying harvest to 60 d after first freeze resulted in unchanged yield values for energycane; however, the yield of elephantgrass was approximately 30% lower (Na et al., 2014). Canopy lodging, although not severe, and abscised leaves, were observed in our study especially in the November and February harvests. Difficulty to pick up the plant material did not play a role in our experiment because we hand-picked up the clipped biomass; however, on a commercial scale operation the magnitude of uncollected biomass will depend on the type of harvesting equipment.

5

3.2 | Nutrient removal

There was a HF effect for N and K removal. Removal of N and K were at least twofold greater for 2X (136.7 and 212.6 kg ha⁻¹ for N and K, respectively) than to 1X (42.7 and 54 kg ha⁻¹ for N and K, respectively) (Table 2). Lower N and K removed in the harvested tissue is desirable to increase efficiency of bioconversion processes such as pyrolysis (Trendewicz et al., 2015; Wilson et al., 2013). Values for N and K removal like our study have been previously reported in the literature for switchgrass (Ashworth et al., 2017; Lemus et al., 2008; Lindsay et al., 2018; Reynolds et al., 2000). Examining the 2X harvest frequency treatment in our study, approximately 80% for N and 89% for K of the total seasonal removal was accounted by the mid-season clipping in June (data not shown).

Phosphorus removal ranged from ~ 4 to 25 kg ha⁻¹ (Table 2). There was a HF \times HT interaction effect on P removal. The interaction effect occurred because delaying the end-of-season harvest resulted in lower P removal for the 1X treatment (from 15.4 to 3.6 kg ha^{-1} in October and February, respectively) but not for 2X (average of 23.8 kg ha^{-1}) (Table 2). In general, P removal followed a similar trend to N and K removal, that is, lower nutrient removal was observed for 1X compared with 2X harvest frequency. Similar P removal values to those found in our study have been reported in the literature when switchgrass was harvested during the frost-free period (Ashworth et al., 2017; Seepaul et al., 2014). Silveira et al. (2013) reported greater P removal values than in our study, at about 56 kg ha^{-1} , for 'Alamo' switchgrass; however, greater P removal values reported by Silveira et al. (2013) were most likely due to more frequent clipping (clipped every 6 wk from May until November) and because switchgrass was grown on a manure-impacted soil with approximately 232 mg kg^{-1} Mehlich 1-P concentration in the Ap (0–15 cm) horizon in southern Florida.

3.3 | Biomass dry matter concentration and ash

For the mid-season June harvest of 2X treatment, the DM concentrations ranged from 236 to 284 g kg⁻¹ and the concentration of ash ranged from 39 to 67 g kg⁻¹. For the regrowth tissue harvested at the end of the growing season based on HT treatments, there was a significant HF × HT interaction effect for DM concentration (Figure 3). The concentration of DM was greater for 1X (466 g kg⁻¹) than 2X (415 g kg⁻¹) in October but not different in November (average of 516 g kg⁻¹) and February (average of 893 g kg⁻¹). Overall, DM concentration increased approximately twofold



FIGURE 3 Dry matter concentration of 'BoMaster' switchgrass as a function of harvest frequency [one clipping [1X] and two clippings [2X] per season)] and harvest timing at the end of the growing (before freeze in October, after freeze in November, and February) in Clayton, NC ($35^{\circ}40'$ N, $78^{\circ}29'$ W). Data are means of 2 yr and three replicates per year. *P* values correspond to harvest frequency effects at each harvest timing. Lowercase letters were used to compare harvest timing mean values by harvest frequency treatments; means followed by the same lowercase letter are not different

when harvest timing at the end of the growing season was delayed from October (averaged across HF treatments of 453 g kg⁻¹) to after freeze in February (averaged across HF treatments of 893 g kg⁻¹), and it was intermediate in November (515 g kg⁻¹). The DM concentration levels reached in February are considered safe for storage of the biomass. Higher DM concentrations of biomass at harvest time is desirable because it can reduce the cost of artificial drying and risk of spoilage when the biomass is stored or transported (Lewandosky & Kicherer, 1997).

The five ash components of Si, K, Ca, S, and Cl are generally thought to have an effect on biomass conversion (Bakker & Elbersen, 2005). There were significant main effects of HF and HT on ash concentration. Ash concentration was greatest in October (32 g kg⁻¹), intermediate in November (27 kg⁻¹), and lowest in February (17 g kg⁻¹). Ash concentration was greatest for 2X (29 g kg⁻¹) and lowest for 1X (23 g kg⁻¹). Ash concentration of switchgrass has been reported to be at least threefold greater in the leaves than the stems (Monti et al., 2008). Lower ash concentration due to delayed harvest at the end of the season may be attributed to leaf losses (Adler et al., 2006); however, further study with ash is needed to specify the extent of leaf loss and to quantify which elements change over time. Decreasing ash concentration in the harvested tissue during winter has been previously reported for several grasses including switchgrass in a temperate environment (Adler et al., 2006) and elephantgrass and energycane in a subtropical environment (Na et al., 2014).

4 | SUMMARY AND CONCLUSIONS

Delaying the end-of-season harvest of switchgrass from October (before freeze) to November or February for the 2X clipping management had no negative effect on total annual biomass yield (average yield of 15.5 Mg ha^{-1}). In contrast, biomass yield decreased up to 29% when the harvest at the end of the season was delayed from October to February for the 1X treatment. Nutrient removal (N, P, and K) was consistently greater for 2X treatment compared with 1X. The DM and ash concentrations for the mid-season June harvest of 2X treatments ranged from 236 to 284 g kg⁻¹ and from 39 to 67 g kg^{-1} for DM and ash, respectively. For the regrowth tissue harvested at the end of the growing season, DM and ash concentrations were not different for after freeze harvests (i.e., November and February) between 1X and 2X. The DM concentration level reached in February, for both 1X and 2X treatments, would be safe for storage of the biomass after field harvest. These are important findings that demonstrate how a 2X clipping management strategy enables biomass supply from the field directly to the biorefineries in North Carolina during the winter months while reducing the need of drying and potentially the need for storage. In conclusion, clipping switchgrass biomass twice per year, like the 2X treatment in our experiment, represents an opportunity for biomass producers to extend the window of biomass supply from October to February directly from the field to the biorefineries.

AUTHOR CONTRIBUTIONS

Raul Rivera: Conceptualization; Data curation; Formal analysis; Investigation; Methodology; Project administration; Resources; Software; Supervision. Miguel S. Castillo: Conceptualization; Data curation; Formal analysis; Funding acquisition; Investigation; Methodology; Project administration; Resources; Software; Supervision; Validation; Visualization; Writing – original draft; Writing – review & editing. Travis W. Gannon: Conceptualization; Funding acquisition. Perejitei E. Bekewe: Investigation; Methodology; Project administration.

ACKNOWLEDGMENTS

The authors express their appreciation to Consuelo Arellano for providing insight with statistical analyses, to Stephanie Sosinski, Diego Contreras, and Izamar Gonzales for their excellent support with field and laboratory activities, and to the personnel of the Central Crops Research Station

7

CONFLICT OF INTEREST

The authors report no conflicts of interest.

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How to cite this article: Rivera-Chacon, R., Castillo, M. S., Gannon, T. W., & Bekewe, P. E. (2022). Harvest frequency and harvest timing following a freeze event effects on yield and composition of switchgrass. *Agronomy Journal*, 1–8. https://doi.org/10.1002/agj2.21202