BioEnergy Research
https://doi.org/10.1007/s12155-018-9897-0

Shrub Willow Biomass Production Ranking Across Three Harvests in New York and Minnesota

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Abstract

Shrub willow has potential for being a viable dedicated bioenergy crop in temperate northern latitudes of the USA. Selection of high-producing willow cultivars is critical for economic viability and long-term sustainability of willow production systems. Long-term trials are needed in different geographic areas to better understand genetic by environment interactions on biomass yield for greater profitability and to enhance future breeding efforts. Field trials were conducted in two contrasting environments, northern New York and southern Minnesota, to explore changes in shrub willow yield ranking over three harvest cycles across a range of cultivars and diversity groups. Overall, the MN site produced higher, more stable biomass yields than the NY site due primarily to more productive soils, warmer climate, and less weed pressure. However, between-site differences in willow biomass yield were nominal after the second harvest cycle. Yield variability among the top five willow cultivars at each harvest was significantly less than variability among all cultivars regardless of site. Shrub willow cultivars identified in the top-ranking groups were different between sites. Results show that willow can be a viable long-term crop for sustained biomass feedstock production across a wide range of soils and climates but proper cultivar selection is critical for biological and economic success.

Keywords Shrub willow · Short rotation woody crop · Biomass yield ranking · Willow cultivars

Introduction

Agricultural landscapes are under pressure to produce a wide variety of products and values including food, energy, bio-based products, and critical ecosystem services. Consequently, a multifunctional approach to agriculture is being suggested that temporally and spatially integrates annual and perennial food crops and biomass feedstock on the landscape [1]. Second generation bioenergy and biomaterial feedstocks can provide part of the solution to this issue, since they provide temporal diversification (i.e., perenniality) and spatial integration (e.g., may be grown on land that is of poorer quality and more marginal areas than those required for food production) [1]. The perennial nature of these crops creates opportunities for multiple environmental benefits that address water quality concerns and improves biodiversity on the landscape [2], especially when they are intentionally integrated across the landscape [3]. Biomass crops also have potential to be a feedstock for liquid fuels, novel bio-product streams, and to meet energy demands in electricity and residential heating sectors [4].

In temperate northern latitudes, shrub willows (*Salix* spp.) are gaining acceptance as a viable woody biomass crop. Willow is in the Salicaceae family and is characterized by high CO_2 exchange rates, high light-use efficiencies, and high photosynthetic capacities compared to other woody species [5]. Shrub willow is cultivated in a short-rotation coppice (SRC) system comprising high-density plantings in a single or twin row configuration. Harvest is typically on a 3-year interval with as many as seven harvest cycles, making the system viable for over 20 years. Of the 330 species of willow in the world, the shrub willows *Salix viminalis*, *S. eriocephala*, *S. miyabeana*, *S.* x *dasyclados*, and *S. purpurea* are among the most promising shrub willow species for biomass production [5]. In the USA, research on willow began in New York in the mid-1980s. By the 1990s, a large and diverse breeding population of willow germplasm had been assembled along with technical expertise to perform controlled pollinations [6, 7]. Willow germplasm was collected from

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Published online: 20 February 2018

natural stands across the Northern United States or acquired from other breeding programs, including varieties from Canada, China, Japan, Sweden, Ukraine, New Zealand, Russia, and the Western United States. Breeding of new cultivars was initiated in 1998 and several of these high-yielding varieties have been deployed in regional yield trials and for commercial deployment in partnership with private nurseries.

As a perennial species, willow can have multiple environmental benefits. For example, willow has been shown to positively affect species diversity, especially on landscapes with lower habitat diversity such as in agricultural landscapes [8, 9]. SRC shrub willow plantations established on abandoned farmland and harvested in a 3- to 5-year cycle provide adequate habitat for early successional bird species [10] and support a wide diversity of soil microarthropods [11]. Biological characteristics and management of willow create a structurally diverse habitat for an array of species and protect soil and water resources [8, 12, 13]. Life cycle analysis of willow biomass crops has shown that it is capable of sequestering C while producing biomass for renewable energy, and the net energy ratio for chipped willow biomass delivered to an end user is in the range of 18:1 to 43:1 [14, 15]. In a study of six energy cropping systems, Boehmel et al. [16] found that willow was among the best cropping systems for generating high biomass and energy yields in an efficient and environmentally benign way.

The North Central and North Eastern regions of the USA are considered to be excellent environments for dedicated production of shrub willow given existing infrastructure for both forestry and agriculture [7, 17]. However, matching the right willow cultivar with the right environment is a key factor for ensuring high biomass yield and quality traits, especially across large geographic areas [18]. For example, a genotype by environment interaction was noted for willow biomass yield in an evaluation of 18 cultivars, including a number of recently developed cultivars, over one harvest cycle in New York [19] and Saskatchewan [20]. Earlier studies also indicated that there was a strong genetic by environment interaction across a range of sites in the Northeast and Midwest United States [21–23].

Despite willow's high productivity and provision of multiple environmental benefits, its high cost of production has limited deployment. Cost can be lowered by significant improvements in yield and production efficiency and by valuing environmental benefits [7, 24]. Shrub willow has great potential as a dedicated bioenergy crop, but commercialization and adoption by growers and end users will depend upon the identification and selection of high-yielding cultivars with biomass chemistry and quality amenable to conversion to biofuels and bioenergy [19]. For example, a 17% increase in yield of willow can increase the internal rate of return of the system by over 50% from 5.5 to 8.3% [25].

Long-term trials are needed to better understand genetic by environment interactions on biomass yield in a way that provides information to inform breeding efforts and production system design [26]. The objective of this research is to assess rank variation in shrub willow biomass production over three harvest cycles in two contrasting environments.

Material and Methods

Field research was conducted at two sites, one in Minnesota (North Central USA) and one in New York (North East United States). At the Minnesota site, field trials were established in 2006 at the Agricultural Ecology Research Farm, which is part of the University of Minnesota Southern Research and Outreach Center in Waseca, Minnesota (44°03′48″ N latitude; 93°32′42″ W longitude). Soils and climate information for this site are presented in Table 1. The experimental site has no artificial drainage and has a National Commodity Crop Productivity Index (NCCPI) of 0.8. The NCCPI model uses criteria that relate directly to the ability of soils, landscapes, and climates to foster crop productivity with index values ranging from 0 (least productive) to 1 (most productive) [27]. The field site was in a long-term corn–soybean rotation prior to establishment. The site was field cultivated on May 12, 2006, and again on June 2, 2006, to prepare the seedbed for planting.

At the New York site, field trials were conducted on private land in the town of Constableville, NY (43° 33′ 30.81″ N 75° 31′ 18.47″ W), that has a 0.2 NCCPI. Soils and climate information for this site are presented in Table 1. The experimental site was in pasture for the previous 10–15 years and had not been actively grazed or harvested for forage in the 2 to 3 years prior to the willow trial. Consequently, vegetation cover was quite heavy and a substantial seed bank had built up on this site over time. Existing vegetation was cut, bailed, and removed from the site in the fall of 2005. A mixture of herbicides including glyphosate at 1.7 kg ai ha^{-1}, 2,4 D at 0.6 kg ai ha^{-1}, and triclopyr at 0.3 kg ai ha^{-1} was applied to the regrowing vegetation in the fall of 2005. The site was moldboard plowed and disked in the spring of 2006 prior to planting.

The trial in NY included 30 willow cultivars. However, only 24 of these 30 cultivars were planted in MN because of limitations on available plant material. All willow cultivars were developed by the State University New York College of Environmental Science and Forestry with the exception of S25, SX61, SX64, and SV1 that were originally from University of Toronto and Ontario Ministry of Natural Resources. A complete list of willow cultivars used at each site is found in Table 2. The same experimental design was deployed at both the Minnesota and New York field sites. As much as possible, common crop management practices were applied at both sites, although there were some differences in weed control practices. The experimental design was a

Table 1 Shrub willow cultivars and diversity grouping planted at the Waseca, MN, and Constableville, NY, research sites

Epithet	Cultivar ID	Diversity group[1]
S25	S25	ERIO
NA	9837-077	ERIO
NA*	9832-49	ERIO
NA	00X-032-094	ERIO
NA	00X-026-082	ERIO
SX61	SX61	MIYA
SX64	SX64	MIYA
Cicero	9870-001	MIYA
Marcy	9870-023	MIYA
Sherburne	9871-031	MIYA
Canastota	9970-036	MIYA
NA	94001	PUR
Fish Creek	9882-34	PUR
Wolcott	9882-41	PUR
Oneonta	9879	PM
Oneida	9980-005	PM
Millbrook	99217-015	PM
Saratoga	99217-023	PM
Verona*	99201-002	VM
Otisco*	99201-007	VM
Fabius	99202-004	VM
Tully Champion	99202-011	VM
Taberg*	99202-043	VM
Owasco	99207-018	VM
Truxton	99207-020	VM
Erie*	99208-038	VM
Preble*	01X-2658-015	VM
Onondaga	99113-012	KP
Allegany	99239-015	KP
SV1	SV1	VCC-HYB

[1] ERIO = *S. eriocephala*; KP = *S. koriyanagi*; MIYA = *S. miyabeana*; PM = *S. purpurea* x *S. miyabeana*; PUR = *S. purpurea*; VCC-HYB = *S.* x *dasyclados* (*S. viminalis* x *S. cinerea* x *S. caprea*); VM = *S. viminalis* x *S. miyabeana*

NA epithet not currently assigned

*Planted at NY site only

randomized complete block with four replications. Plot size was 6.9 m × 7.9 m allowing for three double rows of willow. Willow cuttings were spaced 60 cm apart within the row and 75 cm between rows with 150 cm between each set of twin rows resulting in 13 plants along each row. Un-rooted cuttings were planted May 31 through June 1, 2006, in NY and June 6, 2006, in MN. Each cutting was 25 cm long and planted at least 20–23 cm into the soil.

Weed control at the Minnesota site consisted of a post-emergence application of clopyralid (3,6-dichloro-2-pyridinecarboxylic acid) at 0.28 kg ai ha^{-1} plus sethoxydim (2-[1-(ethoxyimino)butyl]-5-[2-(ethylthio)propyl]-3-hydroxy-2-cyclohexen-1-one) at 0.31 kg ai ha^{-1} on July 10, 2006, and June 20, 2007. In New York, the trials were sprayed with a mixture of oxyflourfen (2-chloro-1-(3-ethoxy-4-nitrophenoxy)-4-(trifluoromethyl)benzene) at 1.1 kg ai ha^{-1} and 6-chloro-*N*,*N′*-diethyl-1,3,5-triazine-2,4-diamine at 2.3 kg ai ha^{-1} following planting and applications of Fluazifop-P-butyl (Butyl (*R*)-2-[4-[[5-(trifluoromethyl)-2-pyridinyl]oxy] phenoxy]propanoate) at 0.42 kg ai ha^{-1} in 2006 and 2007. Plots were also mechanically cultivated on July 22, 2006, and June 7, 2007, at the Minnesota site and hand weeding was performed as needed to keep plots weed-free throughout the duration of the study. Mowing between the rows and some hand weeding were conducted at the New York site on several occasions in 2007. Despite these actions, weed pressure at the NY site was very high.

Willow plants at both sites were coppiced after leaf desiccation following the first growing seasons. Harvests occurred every 3 years after leaf desiccation when the root system was 4, 7, and 10 years old while aboveground biomass was 3 years old at each harvest. Above ground plant material was hand harvested at 5–10 cm height above ground in a 2.28 m by 5.49 m area in the middle double row of each plot comprising 18 plants. Above ground plant material was harvested manually and weighed either as whole stems or the stems were chipped and weighed. A subsample was collected, weighed, and dried at 60 °C to a constant weight to determine moisture content. Dry biomass per hectare was calculated by scaling up the area harvested to per hectare basis. At the MN site, 112 kg ha^{-1} nitrogen was applied as urea in the spring of 2007 and 2010 as the plants were re-growing after being harvested. No fertilizer was applied in 2013. At the NY site, nitrogen was applied in the spring of 2010 and 2013. No nitrogen fertilizer was applied in 2007 due to high weed pressure.

Plot level data were excluded from the dataset if plant mortality during the first rotation was greater than 50%. All data were checked for outliers and tested for homogeneity of error variances. Shrub willow cultivars were ranked from highest to lowest yielding at each site and harvest as well as overall yield after three harvest cycles. Spearman's rank correlation coefficients were calculated at each site for the variables of rank after the first, second, and third harvests along with overall yield after three harvest cycles using PROC CORR procedure in SAS 9.4 [26, 28].

Results

Average growing season precipitation across the entire study period was 55 cm for the Waseca, MN, and 61 cm at Constableville, NY (Table 2). Conversely, average growing season growing degree units across the study period was

Table 2 Precipitation and growing degree units and deviation from normal during May–September and soil characteristics at Waseca, MN, and Constableville, NY, field sites

	Study site	
	Waseca, MN Precipitation (deviation from average)	Constableville, NY
	cm	
2006 (establishment)	53.9 (+ 2.0)	60.8 (− 0.4)
2007	63.5 (+ 11.6)	40.6 (− 20.6)
2008	43.2 (− 8.7)	67.4 (+ 6.2)
2009 (harvest 1)	27.9 (− 24.0)	56.0 (− 5.2)
2010	87.9 (+ 36.0)	72.4 (+ 11.2)
2011	45.7 (− 7.5)	67.7 (+ 6.5)
2012 (harvest 2)	36.8 (− 16.4)	48.0 (− 13.2)
2013	56.9 (+ 2.4)	65.5 (+ 4.3)
2014	57.1 (+ 2.6)	65.1 (+ 3.9)
2015 (harvest 3)	80.4 (+ 25.9)	68.2 (+ 7.0)
Growing degree units[a] (deviation from average)		
2006 (establishment)	2549 (+ 132)	1472 (− 55.5)
2007	2523 (+ 105)	1442 (− 85.5)
2008	2425 (+ 6)	1282 (− 245.5)
2009 (harvest 1)	2273 (− 146)	1237 (− 290.5)
2010	2581 (+ 162)	1679 (+ 151.5)
2011	2481 (− 91)	1770 (+ 242.5)
2012 (harvest 2)	2687 (+ 197)	1703 (+ 175.5)
2013	2519 (+ 49)	1527 (− 0.5)
2014	2235 (− 235)	1447 (− 80.5)
2015 (harvest 3)	2609 (+ 139)	1704 (+ 176.5)
Soil Classification	Nicollet clay loam (fine-loamy, mixed, superactive, mesic Aquic Hapludolls)	Empeyville loam (coarse-loamy, isotic, frigid Aquic Fragiorthods)
Soil organic matter (%)	5.1	8.2
Soil pH	5.8	5.7

[a] 10 to 30 °C base temperatures, May 1 until first frost

2488 in MN and 1526 in NY. Precipitation was 7.5–24.0 mm below the average during the 2008–2009 and 2011–2012 growing seasons in MN. At the NY site, precipitation was 5.2–20.6 mm below average in 2007, 2009, and 2012 growing seasons. Precipitation was 11.6–36.0 cm above the 10-year average in MN in 2007, 2010, and 2015 and 11.2 cm above average in NY in 2010. Growing degree units were 91–235 units below average during the 2009, 2011, and 2014 growing season at the MN site, while growing degree units were 55.5–290.5 units below normal 2006–2009 and 2014 growing seasons in NY.

In MN, cultivar “SV1” did not successfully establish and a severe infestation of a poplar and willow borer (*Cryptorhynchus lapathi*) resulted in significant post-establishment plant mortality (> 75%) of “94,001” shrub willow cultivar. Because of abnormally poor stands and/or productivity at this site, the aforementioned cultivars were removed from the dataset prior to analysis. Shrub willow survival for all other cultivars was > 90 and > 70% across all cultivars at the MN and NY sites, respectively.

New York Site

Shrub willow biomass yield across all the cultivars at the first harvest cycle averaged 18.1 Mg ha^{-1} (6.0 Mg ha^{-1} year^{-1}) while yield of the top five cultivars averaged 27.8 Mg ha^{-1} (9.3 Mg ha^{-1} year^{-1}), an increase of 53.6% (Table 3). Yield variability among the top five willow cultivars was less than that among all cultivars (27.2 vs 49.7%). The top five willow cultivars were Preble, Otisco, SV1, Fabius, and Oneonta at the first harvest cycle (Table 4).

Table 3 Mean yield and coefficient of variation across willow cultivars for all cultivars and top-ranking 1, 3, and 5 cultivars at Waseca, MN, and Constableville, NY, sites for the first, second, and third harvests and for the cumulative yield after three harvests

Site	Number of cultivars	First harvest yield Mg ha^{-1} (coefficient of variation)	Second harvest yield	Third harvest yield	Total yield
New York	All	18.1 (49.7)	25.5 (42.8)	25.0 (42.9)	68.5 (39.8)
	Top 5	27.8 (27.2)	34.2 (13.9)	35.6 (14.7)	90.8 (16.1)
	Top 3	28.9 (21.1)	34.9 (20.6)	36.5 (16.9)	91.5 (14.7)
	Top 1	30.7	36.3	37.5	93.0
Minnesota	All	25.9 (16.2)	28.8 (21.8)	24.1 (27.8)	77.5 (21.4)
	Top 5	29.5 (9.3)	35.6 (9.2)	32.0 (13.0)	94.9 (9.3)
	Top 3	29.9 (7.1)	36.1 (7.4)	33.3 (9.9)	98.8 (6.8)
	Top 1	30.4	37.1	34.6	101.1

Shrub willow biomass yield at the second harvest cycle averaged 25.5 Mg ha^{-1} (8.5 Mg ha^{-1} year^{-1}) among all cultivars. Biomass yield of the top five shrub willow cultivars at the second harvest cycle averaged 34.2 Mg ha^{-1} (11.4 Mg ha^{-1} year^{-1}), an increase of 34% over the average yield of all cultivars (Table 3). As with the first harvest cycle, yield variability was less among the top five willow cultivars compared to variability among all cultivars (13.9 vs 42.8%). The top five willow cultivars were Millbrook, Oneida, SX64, Wolcott, and SX61 at the second harvest cycle (Table 4). Among the top 5, 3, and 1 cultivars, yield increased on average 20% from first to second harvest cycle but across all cultivars yield increased 40%. None of the top five willow cultivars at the end of the first rotation were in the top five at the end of the second rotation (Table 4).

Average biomass yield of all shrub willow cultivars was 25.0 Mg ha^{-1} (8.3 Mg ha^{-1} year^{-1}) at the end of the third harvest cycle, which was similar to the second harvest (Table 3). As with previous harvests, selecting the top 5, 3, or 1 cultivars increased yield 42–50% over the average yield of all cultivars and significantly reduced yield variability (Table 3). The yield of the top set of cultivars was similar to the second rotation yields. The top five willow cultivars were Fabius, SX64, SX61, Wolcott, and Fish Creek at the third harvest cycle (Table 4). Spearman's rank correlation coefficient was 0.87 ($p < 0.001$) between harvest cycles 2 and 3 and 0.59 ($p < 0.0006$) for harvest cycles 1–3. Among the top five highest yielding willow cultivars at the end of the second cycle, three cultivars were within the top five at the end of the third rotation: SX61, SX64, and Wolcott.

Average total cumulative shrub willow biomass yield over three harvest cycles was 68.5 Mg ha^{-1} (7.6 Mg ha^{-1} year^{-1}) at the end of the third harvest cycle (Table 3). Total biomass yield of the top five shrub willow cultivars averaged 90.8 Mg ha^{-1} (10.1 Mg ha^{-1} year^{-1}), an increase of 32% over the average yield of all cultivars (Table 3). The top five cultivars in cumulative yield were Millbrook, SX61, Preble, Otisco, and Fabius (Table 4). Spearman's rank correlation coefficients between total cumulative willow yield over three rotations and yield at harvest cycles 1, 2, and 3 were 0.83 ($p < 0.0001$), 0.90 ($p < 0.0001$), and 0.88 ($p < 0.0001$), respectively, among all willow cultivars.

Minnesota Site

Average yield of all shrub willow cultivars at the Waseca, MN, site was 25.9 Mg ha^{-1} (8.6 Mg ha^{-1} year^{-1}) at the first harvest cycle (Table 3). Yield of the top five willow cultivars averaged 29.5 Mg ha^{-1} (9.8 Mg ha^{-1} year^{-1}), an increase of 13.9% over the average yield of all cultivars. There was only a small increase in yield among the top five, three or one cultivars, similar to the NY site. Yield variability was less among the top five willow cultivars compared to variability among all cultivars (9.3 vs 16.2%). The top five willow cultivars at the MN site were SX64, Fabius, Oneida, SX61, and Marcy at the end of the first harvest cycle (Table 5).

Average yield of all willow cultivars at the second harvest was 28.8 (9.6 Mg ha^{-1} year^{-1}) (Table 3), a yield increase of 11% from the first harvest. Average shrub willow biomass yield at the second harvest increased 23–29% when selecting the top 5, 3, or 1 cultivars compared to the average yield of all cultivars. Among the top five willow cultivars, there was a 22–34% increase from the first to second harvest periods. As in the first harvest, yield variability among the top five cultivars was lower compared to variability among all cultivars. The top five willow cultivars at the MN site were Oneida, Oneonta, Marcy, SX61, and SX64 at the end of the second harvest cycle (Table 5). Among all cultivars at the MN site, Spearman's rank correlation coefficient between harvest 1 and 2 was 0.82 ($p < 0.0001$). Among the top five highest yielding willow cultivars at the end of the first cycle, all but one (Fabius) were within the top five at the end of the second rotation. Fabius was the only cultivar in the top five in the first rotation that did not show a yield increase of at least 14% and as a result, Fabius' ranking dropped to tenth in the second rotation.

Table 4 Yield and rank of willow cultivars in Constableville, NY, for three 3-year harvests and total yield. The top five willow cultivars are in italics

Cultivar	Yield after 1st harvest (Mg ha^{-1})	Rank	Yield after 2nd harvest (Mg ha^{-1})	Rank	Yield after 3rd harvest (Mg ha^{-1})	Rank	Total yield after three harvests (Mg ha^{-1})	Rank
Preble	30.7	1	32.4	7	27.7	15	90.7	3
Otisco	28.5	2	29.4	15	32.1	8	90.0	4
SV1	27.4	3	30.4	14	27.5	17	85.3	9
Fabius	26.4	4	31.9	9	37.5	1	89.8	5
Oneonta	24.9	5	32.4	6	30.0	12	87.3	8
Millbrook	24.2	6	36.3	1	32.6	7	93.0	1
Tully Champion	22.8	7	25.5	19	25.9	19	74.2	17
SX61	22.8	8	32.6	5	35.4	3	90.8	2
Saratoga	22.3	9	30.9	13	31.2	9	84.4	10
Taberg	22.2	10	31.3	12	30.1	11	83.7	11
Allegany	21.1	11	23.1	22	27.0	18	71.2	18
Oneida	21.0	12	34.7	2	32.9	6	88.6	6
Verona	20.7	13	24.0	20	25.3	20	70.0	19
Marcy	20.4	14	32.4	8	28.3	14	81.0	14
Owasco	19.6	15	22.7	23	19.7	23	62.0	23
Fish Creek	19.1	16	29.3	16	33.0	5	81.5	13
Erie	18.2	17	31.6	11	28.4	13	78.1	15
SX64	18.1	18	33.9	3	36.6	2	88.5	7
Truxton	17.4	19	18.9	25	15.7	25	51.9	25
Cicero	15.9	20	27.9	17	23.7	21	67.5	20
94001	14.8	21	23.9	21	27.6	16	66.2	21
Sherburne	14.6	22	31.7	10	30.4	10	76.7	16
Wolcott	14.4	23	33.2	4	35.3	4	82.9	12
Onondaga	14.0	24	22.7	24	18.9	24	55.6	24
Canastota	13.4	25	27.6	18	22.4	22	63.3	22
S25	8.6	26	8.2	26	6.0	29	22.8	27
9837-77	7.8	27	6.0	28	6.3	27	20.1	28
00X-026-082	6.4	28	4.3	29	7.2	26	17.9	29
00X-032-094	6.2	29	6.5	27	6.1	28	25.7	26
9832-49	1.5	30	2.2	30	0.2	30	13.9	30

At the third harvest cycle, average yield of all shrub willow cultivars at the Waseca, MN, site was 24.1 Mg ha^{-1} (8.0 Mg ha^{-1} year^{-1}) (Table 3). This represented a slight reduction in yield compared to the second harvest and may been the result of no nitrogen application in 2013 at the start of the third rotation. Selecting the top 5, 3, or 1 cultivars increased yield 31–44% compared to the average yield of all cultivars. Variability in yield among the top yielding cultivars was less than that of all cultivars. The top five willow cultivars at the MN site were Oneonta, Oneida, Wolcott, SX64, and Fish Creek at the end of the third harvest cycle (Table 5). Wolcott was one of the only cultivars that showed an increased in yield from the second to third rotation (6.6%), which contributed to being ranked third. Two cultivars ranked third (Marcy) and fourth (SX61) in the second rotation have large decreases in yield (28.8–35.3%) which made them being ranked ninth (Marcy) and 15th (SX61) in the third rotation. Spearman's rank correlation coefficients were 0.76 ($p < 0.0001$) and 0.69 ($p < 0.0002$) among all willow cultivars between harvests 2–3 and 1–3, respectively. Among the top five highest yielding willow cultivars at the end of the second cycle, three cultivars were within the top five at the end of the third rotation: Oneonta, Oneida, and SX64.

Table 5 Yield and rank of willow cultivars in Waseca, MN, for three 3-year harvests and total yield. The top five willow cultivars are in italics

Cultivar	Yield after 1st harvest (Mg ha^{-1})	Rank	Yield after 2nd harvest (Mg ha^{-1})	Rank	Yield after 3rd harvest (Mg ha^{-1})	Rank	Total yield after three harvests (Mg ha^{-1})	Rank
SX64	30.4	1	34.9	5	30.6	4	95.8	3
Fabius	30.0	2	30.3	10	27.7	6	88.1	6
Oneida	29.4	3	37.1	1	34.5	2	101.1	1
SX61	28.8	4	34.9	4	22.6	15	86.4	8
Marcy	28.7	5	35.1	3	25.0	9	88.9	5
Oneonta	28.7	6	36.1	2	34.6	1	99.4	2
Millbrook	27.3	7	30.7	9	24.3	11	82.3	9
Fish Creek	27.2	8	32.6	6	29.7	5	89.5	4
Canastota	26.9	9	31.4	8	23.4	12	81.7	10
Wolcott	26.9	10	28.8	14	30.7	3	86.4	7
Truxton	26.9	11	29.6	13	22.8	14	79.2	13
Cicero	26.8	12	29.9	12	22.2	16	78.9	14
Saratoga	26.6	13	27.1	16	26.5	7	80.3	11
Owasco	26.5	14	24.8	20	19.4	21	70.8	20
9832-49	26.4	15	25.7	19	18.8	22	70.9	19
Sherburne	25.8	16	30.0	11	22.8	13	78.7	15
Otisco	25.5	17	20.4	21	20.1	19	66.0	21
9837-77	23.9	18	27.1	17	20.2	18	71.2	18
Tully Champion	22.7	19	31.4	7	25.6	8	79.8	12
Allegany	22.5	20	28.5	15	20.5	17	71.6	17
Onondaga	22.0	21	26.4	18	24.7	10	73.1	16
00X-032-094	20.8	22	18.2	24	12.9	23	51.9	23
00X-026-082	20.3	23	19.3	23	10.4	24	49.9	24
S25	19.8	24	20.3	22	19.6	20	59.7	22

The average cumulative biomass yield of all willow cultivars was 77.5 Mg ha^{-1} (8.6 Mg ha^{-1} year^{-1}) at the end of three harvest cycles (Table 3). Total cumulative yield after three harvest cycles followed trends observed in the first, second, and third harvest cycles whereby yield increased and variability decreased when selecting the top five, three, or one cultivars compared to all cultivars (Table 3). The top five cultivars for cumulative yield were Oneida, Oneonta, SX64, Fish Creek, and Marcy (Table 4). Spearman's rank correlation coefficients were 0.45 ($p < 0.0001$), 0.41 ($p < 0.0001$), and 0.49 ($p < 0.0001$) between cumulative total yield and harvests 1, 2, and 3, respectively, indicating less stability and worse rank predictions compared to within harvest rank predictions. The correlation coefficients in MN were much lower than the same values for the NY site. Three, four, and four of the top five cultivars at harvests 1, 2, and 3, respectively, were also in the top five cultivars for highest cumulative yield.

Discussion

In the first rotation, shrub willow biomass yield was 43% greater among all cultivars and 6% greater among the top five cultivars at the MN compared to NY sites. Relatively small differences in biomass yield between sites among the top five cultivars are likely due to the inclusion of improved cultivars that have been subjected to some degree of screening and selection pressure before being deployed for testing [29]. Biomass yield tended to increase from first to second harvest at both sites. However, this effect was more dramatic at the NY site where yield across all cultivars increased 41% compared to 11% at the MN site. Three cultivars in NY (Sherburne, Wolcott, and Canastota) more than doubled their yields between the first and second rotation. Volk et al. [30] reported a 23% increase in yield of commercial willow cultivars from first to second harvest cycles in New York. However, the Volk et al. [30] study included older unimproved willow cultivars compared to the present study. In a larger study, to evaluate if yield data from the first harvest yield data can be used to predict yield at the second harvest across five sites, Sleight et al. [29] found that where willow establishment and management was good (e.g., MN site), there is a smaller chance of increased yields in the second year as opposed to sites that were more marginal or had poor establishment because of weeds or other factors (e.g., NY site) where chances of increased yield are high. Furthermore, Sleight et al. [29] noted a first harvest willow biomass yield of approximately 11.4 $Mgha^{-1}$ $year^{-1}$ as the breakeven point where yield change between rotations becomes zero. In the present study, first harvest biomass yield was 3.0 in NY and 8.6 Mg ha^{-1} $year^{-1}$ in MN across all the cultivars with no single cultivar at either site reaching the 11.4 Mg ha^{-1} $year^{-1}$ level. At the NY site, all but three of the cultivars increased yield from the first to second rotation while at the MN site, all but five cultivars increased. All three of the cultivars at the NY site and three of the five cultivars at the MN site with reduced yield from first to second harvest were *S. eriocephala* cultivars. *S. eriocephala* is a native species in the eastern half of North America with many years of field trial data. In NY, these cultivars have consistently underperformed relative to other cultivars [19, 22, 23]. However, yields of cultivars of *S. eriocephala* have performed quite well in other trials with yields approaching 20 Mg ha^{-1} $year^{-1}$, most notably in Quebec and New Brunswick in eastern Canada [31]. In a recent study of native willow species in a nursery setting [32] on a coal mine site in eastern Canada, *S. eriocephala* cultivars were consistently among the best performers. Interest in using native material in certain situations where biomass yield is not the primary focus continues to generate interest in *S. eriocephala* and other native species.

Changes in shrub willow biomass yield between the second and third harvests were slightly different at the two sites. At the MN site, biomass yield across all the cultivars decreased almost 18% and there were 13 cultivars where yield decreased by more than 20%. The yield of the top five cultivars dropped by about 5%. No N fertilizer was applied at the start of the third rotation in MN, and the GDD in second year of the third rotation (2014) were almost 10% below the 10-year average and the lowest of any year during this trial. Both of these factors may have contributed to the trend of lower yields in the third rotation in MN. At the NY site, yield decreased by 4% across all cultivars but the top five cultivars increased yield by almost 11%. N fertilizer was applied at this site in the third rotation and precipitation and GDD were more typical of the 10-year average. Both growing season precipitation and GDD have been positively correlated with willow yield [22]; however, the precise impact of these weather factors is hard to determine because yield was only every third year.

There are only a few studies where willow yield has been tracked over three rotations consistently (see Table 1 in [26]) and the reported patterns between the second and third rotation are variable, as is the case in this study. Among the reported trials [26], two of them showed yield increases, five had yield decreases, and six had changes in yield of less than 10%. There are a number of factors that could impact changes in yield in the third and later rotations including factors noted above such as nutrient management decisions and weather patterns. Other factors may include the re-sprouting ability of different cultivars, the amount of damage caused to stools, and root systems during harvesting operations (i.e., compaction or pulling of stools). Developing a better understanding on willow yield dynamics over multiple rotations and associated causal factors is important because the projected lifespan of this crop is greater than 20 years.

There was greater variability in shrub willow biomass yield among all cultivars at the NY compared to MN sites, especially in the first two rotations. Variation remained greater at the NY site in the third rotation, but the difference in variability was smaller. The lower variability at the MN was likely due to environmental differences and potentially cropping history. The NY site is characterized by having shallow and lower quality soils associated with the low NCCPI value (0.2) compared to deep and productive soils with a high NCCPI value (0.8) at the MN site. The NY site had one of the lowest environmental scores among ten willow trial locations while Waseca had the second highest score [22]. Growing-degree days at the NY site were also about one third lower than the MN site. The NY site is a marginal site for agriculture and is at the fringe of the agriculture-forest interface in that part of the state and has not been in active agriculture for a few years and had not been plowed for 10–15 years. As a result, there were established perennial weeds and a more extensive seed bank at the NY site thereby contributing to greater weed pressure compared to the MN site, but it was not consistent across the study area. There was clearly a more variable response among cultivars to stressful growing conditions in NY, indicating that there were a smaller number of cultivars suitable for the marginal conditions experienced in NY.

Strong weed pressure early in the trial likely influenced first harvest biomass yield at the NY location, especially considering differential response among willow cultivars to weed competition during establishment. For example, Fish Creek and Wolcott (*S. purpurea* cultivars) were ranked 16th and 23rd, respectively, at the first harvest. By the third harvest, Fish Creek and Wolcott cultivars were ranked fifth and fourth, respectively. Delayed growth of *S. purpurea* cultivars during establishment has been noted in other trials when there is strong weed pressure during establishment [33, 34]. *S. purpurea* cultivars have an upright stem form and hold their leaves close to stems resulting in less shading and poor competition with weeds for light compared to other cultivars. Once these cultivars become established, however, production can increase dramatically. For example, yield of Wolcott and Fish Creek increased 130.5 and 53.4% from first to second harvest, respectively.

The goal of any biomass production system is to deploy the cultivars that will be the most stable and productive over multiple rotations. To that end, Spearman's rank correlation was used to explore stability in yield rank among all shrub willow cultivars across harvest cycles. Furthermore, the top five, three, and one most productive willow cultivars were identified at each harvest to provide more insight into rank changes over time with respect to biomass yield after each harvest for high yielding cultivars.

Relatively poor correlation in willow rank between the first and second and first and third harvest periods at the NY site is likely due to competition from weeds during the first harvest cycle which results in differential response among willow cultivars to weed pressure. However, there was good correlation in biomass yield between the second and third harvest cycles. At other field sites in NY, the correlation coefficients between the first and third rotations at two sites were 0.91 and 0.83 [26]. The quality of the sites was better and weed pressure was less at these sites compared to the present study, which probably contributed to greater stability over time. Deviations in weather conditions at more marginal sites have the potential to have greater impact on yields of crops than sites that have better quality. At MN, rank correlation among all cultivars was relatively stable across harvest cycles likely due to better site and management conditions. However, rank correlation among all cultivars was poor when comparing total cumulative yield to the first, second, or third harvest cycle, but much of this difference is related to changes in ranking among lower producing willow cultivars.

At the NY site, the top five shrub willow cultivars changed in each rotation with the exception of one cultivar in the third rotation suggesting that consistency in yield rank across all cultivars, as measured by Spearman's rank correlation, was influenced by lower yielding cultivars. Conversely, yield rank was generally consistent among the top five cultivars at the MN site suggesting greater influence of higher yielding cultivars on Spearman's rank correlation among all cultivars. Sleight and Volk [26] noted stable yield rankings within a site over multiple harvest periods suggesting that selection of cultivars after one rotation at a given site can be an effective strategy to identify top performing willow cultivars. At the NY site, lack of stability in the top five cultivars suggests that long-term data is necessary to identify stable and productive willow cultivars at this site. This may be related to the more marginal site conditions at this NY location and the weed pressure during the establishment period. At sites where there are weather conditions, weed pressure, or other issues that negatively impact willow establishment, extra care needs to be taken in selecting cultivars based on first rotation data.

As expected, willow cultivars identified in the top-ranking groups were different between sites, reinforcing the importance of making cultivar section based on local climate and soil conditions. The top five cultivars in MN based on total yield were ranked from 6th to 14th in NY. These two sites were intentionally selected in different regions to explore how a common suite of cultivars would perform under different conditions. While expected, these results emphasize the importance of selecting cultivars based on trials from a similar region. A previous study [26] noted that selecting the top cultivars based on data from a different site could result in a reduction of yield of 7–14%, which over the life of a perennial crop like willow could have a negative impact on the potential for financial returns from the system.

Between the two sites, there were a few willow cultivars that were ranked relatively high regardless of site. For example, Fabius was ranked fifth in cumulative yield in MN and sixth at NY. This triploid hybrid (*S. viminalis* x *S. miyabeana*) had the best average yield across ten sites [22] and was the top ranked cultivar on better quality sites. On lower quality sites, a different *S. viminalis* x *S. miyabeana* triploid hybrid (Tully Champion) was the best performer. The NY site in this study was an exception to that broader analysis because Fabius consistently did well, but there were several other lower quality sites included in the broader analysis [22]. Fabio et al. [22] only included first rotation data in their analysis, including data from the two sites in this study, which may influence some of the results especially if there are changes in ranking of cultivars across rotations as occurred in this study. There is a need to build datasets over multiple rotations and sites for long-term perennial crops like willow to better understand their performance so cultivar recommendations can be made.

Deploying a diverse mix of diverse cultivars could provide additional resilience and stability in yields over time [35]. In both locations, at least three diversity groups were represented in the top five willow cultivars, regardless of harvest year, suggesting that a high-yielding yet diverse mixture could be successfully deployed, but care needs to be taken when selecting cultivars to minimize the impact on overall yield over time. As new cultivars are developed from a focused breeding and selection program, yield stability will likely increase among the top suite of cultivars thereby providing a broader array of cultivars to choose from in the future. While there is variation in ranking at these site, the difference in yield among the top cultivars is small (average of top five in MN was 94.9 Mg ha^{-1} and in NY was 90.0 Mg ha^{-1}) suggesting that there is a suite of cultivars that can be consistently productive at this marginal site and that the majority of these cultivars are ones that have been subjected to screening pressure as part of the breeding and selection process.

Conclusions

We explored productivity and changes in yield rank among shrub willow cultivars between two contrasting environments over three harvest cycles. The primary factors influencing cultivar productivity and stability were environment and management during establishment years. Environment drove differences in variability and rank stability among willow cultivars across harvest cycles. Weed competition during the first harvest cycle significantly reduced willow biomass yield at the NY compared to the MN site. However, willow productivity was similar between sites in both the second and third harvest cycles showing good willow yield can be achieved on marginal sites. Rank stability among the top five willow cultivars was greater at the MN compared to NY site. Results also suggest that the top ranked cultivars comprised a mix of diversity groups thereby supporting a mixed planting strategy aimed at increasing genetic diversity and reducing production risk within a field. Overall, results show that willow can be a viable long-term crop for sustained biomass feedstock production across a wide range of soils and climate. However, cultivar selection is critical and highly dependent on environment and management.

Acknowledgments Support for the initial establishment of the trial in NY was provided by USDA AFRI and the New York State Energy Research and Development Authority (NYSERDA). T.A. Volk is a co-inventor on the patents for the following willow cultivars that are included in this trial: Tully Champion (US PP 17,946), Fish Creek (US PP 17,710), Millbrook (US PP 17,646), Oneida (US PP 17,682), Otisco (US PP 17,997), Canastota (US PP 17,724), Owasco (US PP 17,845), and Preble (US PP 24,537).

Funding Essential funding to maintain and monitor these plots over the past several years was provided by the North Central Regional Sun Grant Center at South Dakota State University through a grant provided by the US Department of Energy Bioenergy Technologies Office under award number DE-FC36-05GO85041 and the University of Minnesota Southern Research and Outreach Center.

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