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RESEARCH ARTICLE

Yield-height correlation and QTL localization for plant height in two lowland switchgrass populations

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Abstract Switchgrass (*Panicum virgatum* L.), as a model herbaceous crop species for bioenergy production, is targeted to improve biomass yield and feedstock quality. Plant height is a major component contributing to biomass yield. Accordingly, the objectives of this research were to analyze phenotypic variation for biomass and plant height and the association between them and to localize associated plant height QTLs. Two lowland switchgrass mapping populations, one selfed and another hybrid population established in the field at Perkins and Stillwater, Oklahoma, were deployed in the experiment for two years post establishment. Large genetic variation existed for plant biomass and height within the two populations. Plant height was positively correlated with biomass yield in the selfed population (r = 0.39, $P < 0.0001$) and the hybrid population ($r = 0.41$, $P < 0.0001$). In the selfed population, a joint analysis across all environments revealed 10 QTLs and separate analysis for each environment, collectively revealed 39 QTLs related to plant height. In the hybrid population, the joint analysis across overall environments revealed 35 QTLs and the separate analysis for each environment revealed 38 QTLs. The findings of this research contribute new information about the genetic control for plant height and will be useful for future plant breeding and genetic improvement programs in lowland switchgrass.

Keywords yield-height, QTL localization, lowland switchgrass

1 Introduction

Switchgrass (*Panicum virgatum* L.) is a model cellulosic herbaceous feedstock species selected for bioenergy feedstock production by the United States Department of Energy in 1991[1]. It is a perennial, C_4, highly polymorphic and wind pollinated polyploid species exhibiting disomic inheritance in tetraploid forms[2–8]. With the base chromosome number x = 9, switchgrass ploidy levels ranging from diploid ($2n$ = 2x = 18) to duodecaploid ($2n$ = 12x = 108) have been reported[2]. Lowland and upland ecotypes are the two dominant phenotypic and ecotypic groups in switchgrass[9]. The lowland ecotypes are exclusively tetraploid ($2n$ = 4x = 36) while the upland ecotypes are tetraploid ($2n$ = 4x = 36) or octaploid ($2n$ = 8x = 72) with reportedly rare hexaploids ($2n$ = 6x = 54)[2,9,10]. Aneuploidy was reportedly more common in octaploids than in tetraploids[11]. In the south-central Great Plains of the USA, lowland switchgrass plants are taller and produce more biomass than upland plants[12,13].

Plant height is an important yield component that influences biomass yield significantly. Past studies reported that biomass yield was strongly and positively correlated with plant height[14,15]. Lemus et al.[14] analyzed correlations between traits including selected agronomic traits and cell wall components in 20 upland switchgrass populations over four years (1998–2001) in southern Iowa. They observed significant positive correlation of biomass yield and plant height (r = 0.85, $P < 0.0001$). Sripathi et al.[15] reported positive correlation based on a study conducted in greenhouse conditions (r = 0.76, $P < 0.001$) and in a field experiment (r = 0.82, $P < 0.001$). Bhandari et al.[16] reported narrow-sense heritability estimates for lowland switchgrass biomass yield at 0.17, 0.14, and 0.24, based on half-sib families, full-sib/half-sib families, and midparent-progeny regression, respectively. They also

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reported narrow-sense heritability estimates for lowland switchgrass plant height at 0.14 and 0.53, based on full-sib/half-sib families and parent-progeny regression, respectively[16]. However, little information is available on the genetic control for plant height in switchgrass. Identification of genomic regions (i.e., quantitative trait loci (QTL)) for the important traits based on DNA marker genetic maps could provide useful information for understanding its genetic structure and in improving plant height resulting in high yielding cultivars in switchgrass.

Linkage mapping studies in switchgrass have been previously reported[5,17–21]. Missaoui et al.[17] constructed a restriction fragment length polymorphism based linkage map in a population derived from two outbred parents 'Alamo' AP13 (a tetraploid lowland genotype) and 'Summer' VS16 (a tetraploid upland genotype). Okada et al.[5] constructed complete linkage maps of two lowland switchgrass genotypes based on SSR and STS markers. Their mapping population consisted of 238 full-sib F_1 progeny of a cross between selected genotypes of switchgrass 'Kanlow' as the female parent and 'Alamo' as the male parent. Liu et al.[18] constructed a complete genetic map of 18 linkage groups in a lowland switchgrass population derived from selfing a heterozygous parent using SSR markers. Dong et al.[19] constructed a linkage map for a hybrid population derived from a cross between NL94 (♀) and SL93 (♂). Lowry et al.[20] reported an integrated map by combining male (Alamo-A4) and female (Kanlow-K5) maps together for their QTL work on more than 50 traits in switchgrass.

Serba et al.[22], recently reported detection of QTLs for biomass yield and plant height in switchgrass based on parental linkage maps constructed using the AP13 × VS16 population, which was initially reported by Missaoui et al.[17]. They identified four QTLs for biomass yield across 10 environments and five QTLs for plant height across eight environments. They also reported more than 30 QTLs for each of the two traits in single environments and more than 50 epistatic QTLs in each trait. Lowry et al.[20] observed a major QTL for biomass, centered on marker nfsg262, on LG 9b and multiple QTLs for tiller traits (including tiller width, tiller length, tiller mass, and leaf area). Switchgrass has high genetic diversity[23], therefore, more work is needed to better understand genetic structure for biomass, plant height, and many other traits contributing to biomass yield and quality. We have developed two mapping populations, one being derived from selfing a northern lowland genotype 'NL94 LYE 16×13', and another from crossing 'NL94 LYE 16×13' and 'SL93 7×15'[18,21]. Using the two populations, Dong et al.[19] reported QTLs for reproductive maturity. Obviously, our mapping populations were different from the populations used by Serba et al.[22] and Lowry et al.[20]. The objectives of the present study were to analyze phenotypic variation for biomass and plant height, compute yield-height correlation, and localize QTLs associated with the plant height based on linkage maps developed in the two Oklahoma State University (OSU) populations.

2 Materials and methods

2.1 Plant materials

Two mapping populations, including a first-generation selfed population of 'NL94 LYE 16×13' (NL94) and a hybrid population derived from NL94 (♀) × 'SL93 7×15' (SL93) (♂), were used in this study. The NL94 parent shared in the two populations was originally selected from the OSU northern lowland (NL) breeding population growing in a low yield environment (LYE), while SL93 was selected from the OSU southern lowland (SL) breeding population[18,24]. Those two parents were each grown in 30 cm diameter pots in a greenhouse at the OSU Agronomy Research Station in the summer of 2007[18]. Two pots, one pot each from the two parents, were moved to a large growth chamber in the OSU Controlled Environmental Research Laboratory in October 2007, just before anthesis, to produce a hybrid full-sib mapping population[18]. A total of 456 progeny were obtained from the NL94 parent[18]. Out of the 456 progeny from NL94, Liu et al.[18] identified 279 as selfed progeny constituting the selfed population and 177 as hybrids between the two parents forming the hybrid population.

2.2 Experimental design, establishment and management

To collect phenotypic data, two field trials were established in 2011, one at the OSU Cimarron Valley Research Station, Perkins (PKS) and the other at the OSU Agronomy Research Station, Stillwater (STW), OK[19]. The experimental design used at both locations was a randomized complete block with three replications. Each replication constituted 443 plots, encompassing 265 plots of the selfed population, 176 of the hybrid population, and two parents. Each plot consisted of three ramets of one genotype. The spacing between two neighboring rows and two adjacent plants in a row was 107 cm. To minimize border effects, border rows were maintained in each location.

The plants were transplanted in STW and PKS in May 16–17 and June 1–7, 2011, respectively[19]. The protocol for managing the plantings in the establishment year was described[19]. To facilitate data collection in 2012 and 2013, white posts were put on the west end of each field at intervals every 10 rows in the month of March. In May of the two years, urea at 67.2 $kg \cdot hm^{-2}$ N was applied to the fields and the weedy plants were removed either by spot-spraying with Roundup or hand-weeding.

2.3 Field data collection and analysis

Prior to the harvest of plant biomass and after plants became dormant, the plant height (cm) was measured from the base of a plant to the top of its panicle. The plants were cut at 10 cm height from the ground surface using a Single Row Silage Chopper (John Deere, Moline, Illinois) in the winters of 2012 and 2013 for biomass yields. Three-tiller samples collected from each plot were weighed for fresh weight, dried at 55°C in a forced air oven for 3 to 7 days, and again weighed for dry weight to calculate dry matter percent which was used to calculate biomass yield. The statistical data analysis for phenotypic traits, plant biomass yield and plant height, was carried out using SAS software, Version 9.4 of the SAS System[25].

2.4 Linkage maps

QTL mapping for the selfed population obtained from NL94 was carried out using the linkage map previously developed by Liu et al.[18]. This linkage map, consisting of 499 marker loci, was constructed with progeny population of 139 selfed plants from a single heterozygous parent unlike the two separate male and female maps reported by Okada et al.[5] or by Serba et al.[22]. Additional 79 SSR markers were integrated into this linkage map[21]. QTL mapping for the hybrid population obtained from NL94 (♀) and SL93 (♂) was carried out using the linkage map developed by Dong et al.[19]. This linkage map, consisting of 178 SSR markers, was constructed with 176 hybrid progeny population[19].

2.5 QTL analysis procedures

QTL analysis was carried out separately for each of the two populations. In each population, the analysis was carried out separately for each environment (a combination of year and location) as well as across all environments (abbreviated as Allenv). PKS12, STW12, PKS13, and STW13 refer to the four separate environments. The software program MapQTL 6[26] was used in the data analysis using a map file, a locus genotype file, and a quantitative trait file all in text formats. For the selfed population, the locus genotype file was converted into data format of F_2 population type[19]. CP population type was used for the hybrid population. Quantitative trait files were prepared for each of the four environments and across all environments by taking mean values averaged over three replications. Both interval mapping (IM) and multiple QTL model (MQM) mapping employed in the analysis used regression algorithm. The other calculation options in QTL analysis included fit dominance for F_2 (IM) as yes, mapping step size 1, maximum number of neighboring markers 5, maximum number of iterations 200, functional tolerance value 10^{-8}, $P = 0.02$ for automatic cofactor selection (ACS) and number of permutations 1000. The initial search for QTL was performed using the IM approach. The genome-wide LOD threshold was obtained by a 1000-permutation test at the relative cumulative value of 0.95 (or a close value). The permutation test, being based on actual data, was considered to avoid the problem of non-normal data[27]. The QTLs detected by IM were used as cofactors for the MQM mapping. The ACS was performed using initial cofactor markers detected at significant LOD peaks as a fixed set, extending with all markers on a single linkage group or a subset of the linkage group, and continued using the same procedure for each linkage group.

3 Results and discussion

Across all environments, biomass yield and plant height both showed significant variation between genotypes in both the selfed and hybrid populations (Table 1). Year, location, and replication had significant effects on biomass yield and plant height with an exception that location was non-significant for biomass yield in the selfed population.

Table 1 ANOVA for the biomass yield (g per plant) and plant height (cm) across all environments for each of the selfed and the hybrid populations using GLM procedure

Sources of variation	Selfed population				Hybrid population			
	df	Biomass yield	df	Plant height	df	Biomass yield	df	Plant height
Year	1	****	1	****	1	****	1	****
Location	1	NS†	1	****	1	****	1	****
Genotype	264	****	264	****	175	****	175	****
Replication	2	***	2	****	2	****	2	****
Year×location	1	****	1	****	1	****	1	NS
Year×genotype	264	NS	263	****	175	NS	175	***
Location×genotype	258	****	257	****	175	****	175	**
Year×location×genotype	257	NS	251	NS	175	NS	175	NS

Note: **** Significant at the 0.0001 probability level; *** Significant at the 0.001 probability level; ** Significant at the 0.01 probability level; † Non-significant at the 0.05 probability level.

The year×location interaction was significant for the biomass yield in both populations while it was significant for plant height in the selfed population only. The year×genotype interaction was significant only for the plant height in both populations but not for the biomass yield. The location×genotype interaction was significant for both traits in both populations but the year×location×genotype interaction was not significant for both traits (Table 1). Previous study by Serba et al.[22] also reported significant effect of environment on biomass yield and plant height. Biomass yield of hybrid population was more than three times that of the selfed population and plant height was also higher for the hybrid population (Table 2). Coefficient of variation (CV) calculated as a ratio of standard deviation to population mean and expressed as percentage, was used to compare variation of biomass yield and plant height. Yield variation was higher compared to plant height in both populations (Table 2).

Both biomass yield and plant height were significantly different between the plant genotypes in each environment (Table 3). Biomass yield and plant height were higher in 2013 compared to 2012 in PKS whereas the results were different in STW (Table 4). The critical growth period in switchgrass includes the months of May, June, July, and August and any sharp departure from the normal rainfall and solar radiation activities in these months can impact plant growth[28]. In our study, the rainfall in May, June, and July for year 2012 was excessively low compared to the 30-year means (Table 5). The year 2012 was the second growing year and 2013 was the third year in this study. Previous reports indicated switchgrass production in the second growing year is about 2/3 of its full potential and productions in the subsequent years are at full capacity[29]. Biomass yield values in 2012 were higher in STW than PKS in both populations; however, the difference did not exceed LSD values (Table 4). Biomass yields were higher in PKS in 2013 in both populations. Plant height was taller in PKS than STW in each environment (Table 4). Stillwater had a greater soil moisture deficit in the upper 40.6 cm soil than Perkins when compared to an average for 15 years (1999–2013) for both 2012 and 2013[30]. The soil moisture deficit was observed for all active growing months May, June, and July in 2012, while deficit was observed for June to July in 2013[30].

Based on results across all environments, biomass yield was positively correlated with plant height for both the selfed population ($r = 0.39$, $P < 0.0001$) and the hybrid population ($r = 0.41$, $P < 0.0001$). The estimated regression lines of the biomass yield on the plant height for the selfed and the hybrid populations showed an increase of 1 cm plant height leads to an increase of biomass yield by 4 g per plant in the selfed population and by 8 g per plant in the hybrid population. Similarly, the plant height explained 15% and 17% variation in the biomass yields in the selfed ($R^2 = 0.15$) and the hybrid populations ($R^2 = 0.17$), respectively. The correlation analysis by Serba et al.[22] indicated

Table 2 Summary of the biomass yield (g per plant) and plant height (cm) across all environments for each of the selfed and the hybrid populations

Parameter	Selfed population		Hybrid population	
	Biomass yield	Plant height	Biomass yield	Plant height
NL94 (P1)	980	206	980	206
SL93 (P2)			1259	203
Population mean	401	184	1469	212
LSD (0.05)	211.42	12.73	367.64	13.17
R^2	0.53	0.80	0.66	0.84
CV/%	65.85	8.63	31.26	7.75

Table 3 ANOVA for the biomass yield and plant height in Perkins, OK (PKS) and Stillwater, OK (STW) from 2012 to 2013

Sources of variation	Selfed population								Hybrid population							
	Biomass yield				Plant height				Biomass yield				Plant height			
	df	2012	df	2013	df	2012	df	2013	df	2012	df	2013	df	2012	df	2013
PKS																
Genotype	258	****	258	****	256	****	254	****	175	****	175	****	175	****	175	****
Replication	2	NS†	2	**	2	****	2	****	2	****	2	****	2	****	2	****
STW																
Genotype	263	****	264	****	263	****	262	****	175	****	175	****	175	****	175	****
Replication	2	***	2	*	2	****	2	**	2	**	2	*	2	NS	2	NS

Note: **** Significant at the 0.0001 probability level; *** Significant at the 0.001 probability level; ** Significant at the 0.01 probability level; * Significant at the 0.05 probability level; † Non-significant at the 0.05 probability level.

Table 4 Summary of the biomass yield and plant height at Perkins, OK (PKS) and Stillwater, OK (STW) from 2012 to 2013

Location	Parameter	Selfed population				Hybrid population			
		Biomass yield		Plant height		Biomass yield		Plant height	
		2012	2013	2012	2013	2012	2013	2012	2013
PKS	NL94 (P1)	922.33	1277.00	203.67	225.00	922.33	1277.00	203.67	225.00
	SL 93 (P2)					1332.00	1783.33	206.33	242.33
	Polulation mean	275.08	538.32	169.57	213.11	1366.98	2028.51	205.70	248080
	LSD	419.54	638.64	32.59	24.72	767.84	1018.41	34.67	21.40
	R^2	0.46	0.46	0.62	0.69	0.60	0.52	0.56	0.63
	CV	95.07	73.94	11.98	7.23	34.98	31.26	10.50	5.36
	RMSE	261.52	398.05	20.31	15.41	478.15	634.18	21.59	13.32
	SE	10.48	16.14	0.99	0.84	26.86	32.35	1.16	0.78
	Maximam	3511	3757	313	292	3223	4611	329	312
	Minimum	1	12	86	125	4	110	9	179
	N	750	732	717	693	528	528	526	521
STW	NL94 (P1)	886.67	833.00	181.00	215.67	886.67	833.00	181.00	215.67
	SL 93 (P2)					1021.33	897.67	159.67	204.33
	Polulation mean	401.32	392.86	162.93	193.14	1398.95	1080.35	175.82	218.89
	LSD	299.17	236.36	20.26	18.51	547.11	429.71	23.96	20.49
	R^2	0.59	0.56	0.63	0.72	0.50	0.51	0.59	0.63
	CV	46.47	37.51	7.75	5.98	24.35	24.77	8.49	5.83
	RMSE	186.51	147.35	12.63	11.54	340.69	267.59	14.92	12.76
	SE	8.43	6.48	0.61	0.64	17.11	13.63	0.83	0.74
	Maximam	2226	1351	220	250	2782	3458	220	265
	Minimum	3	14	107	130	25	126	115	140
	N	781	784	765	767	528	528	526	526

Table 5 Monthly total precipitation (cm) at Perkins and Stillwater, OK from 2012 to 2013 compared with 30-year average (1981–2010)

Month	Perkins, OK			Stillwater, OK		
	2012	2013	30-year mean	2012	2013	30-year mean
January	2.4	4.5	3.4	2.4	2.5	3.4
February	6.1	8.4	4.3	7.4	7.9	4.2
March	11.5	1.4	8.0	10.0	2.8	8.0
April	12.9	13.0	8.8	15.6	13.5	8.9
May	2.8	17.8	13.8	2.8	15.8	13.5
June	7.4	10.5	12.6	5.5	10.0	12.2
July	0.7	15.4	7.4	0.2	14.1	7.7
August	8.6	12.1	7.0	6.7	6.5	7.6
September	3.4	4.9	10.1	2.8	4.3	10.1
October	2.2	6.4	8.4	1.5	4.8	8.2
November	1.7	3.0	6.4	1.1	4.1	6.2
December	1.5	2.2	4.7	1.1	1.6	4.6
Total	61.1	99.5	94.8	57.3	88.0	94.6

Source: Mesonet – monthly rainfall table.

20% biomass variation was accounted for by plant height, and this is comparable with our result of 17% in the hybrid population. In the correlation analysis for each environment separately, significant positive correlation was observed in each environment (Table 6). The histograms for biomass yield and plant height in the selfed population showed most of the progeny values were distributed toward the left side of parental values indicating the substantial effect of inbreeding depression. In contrast, the histograms for biomass yield and plant height in the hybrid population indicated most of progeny values were toward right side of both parents which showed a large amount of hybrid vigor.

In the selfed population with the joint analysis across all environments, a total of 10 QTLs associated with seven LGs were detected (Table 7). The phenotypic variance explained (PVE) by each of these QTLs ranged from 4.9% to 7.1%. In 2012 at Perkins, no QTLs were detected. In 2012 at Stillwater, 12 QTLs associated with six LGs were identified with PVEs 4.4% to 11% each QTL. In 2013 at Perkins, eight QTLs associated with eight LGs were detected with PVEs 3.8% to 10.2%. In 2013 at Stillwater, 19 QTLs associated with eight LGs were detected with PVEs 1.9% to 14.5%. In LG 1b, The QTLs between sww-162 and sww-196 were observed at STW12, at STW13, and across all environments. In LG 2a, the QTLs between sww-938 and Millet-MPGD25 were observed consistently at STW12, PKS13, and STW13. The QTLs between PVE-425/426 and PVCA-219/220 in LG 4a and between sww-556 and PVAAG-3139/3140 in LG 5b were observed consistently at STW12 and STW13. In LG 9b, the QTLs between PVCAG-2615/2616 and sww-585 were observed at STW13 and across all environments (Table 7).

In the hybrid population with the joint analysis across all environments, a total of 35 QTLs associated with 14 LGs were detected (Table 8). The PVEs by these QTLs ranged from 0.6% to 8.8%. In 2012 at Perkins, six QTLs were identified associated with six LGs. The PVEs of each QTL ranged from 5.4% to 10.6%. In 2012 at Stillwater, five QTLs were identified associated with four LGs. The PVEs ranged from 5.2% to 13.3%. In 2013 at Perkins, 22 QTLs were identified associated with 12 LGs. The PVEs ranged from 1.5% to 8.5%. In 2013 at Stillwater, five QTLs were identified associated with five LGs. The PVEs ranged from 8.1% to 13.7%. In LG 1a, The QTLs between PVCAG-2537/2538 and sww-1667 were common among the environments PKS12, PKS13 and all environments. In LG 1b, QTLs between PVGA-1401/1402 and PVCA-179/180 were common among PKS12, PKS13, and Allenv. In the same LG 1b, QTLs between PVCAG-2987/2988 and PVCAG-2361/2362 were common among PKS12, STW12 and Allenv. Also in LG 1b, QTLs between sww-177 and sww-2320 were common among STW12, PKS13, and STW13. In LG 2a, the QTLs between sww-532 and nfsg-052 were consistent between STW12 and Allenv.

QTLs at 0 to 5 cM position on LG 1b were common between selfed and hybrid populations. QTLs at 42.7 to 49.3 cM position and at 54.6 to 55.6 cM positions on LG 2a were common between selfed and hybrid populations. QTLs at 65.8 to 65.9 cM positions on LG 3a were common between selfed and hybrid populations. QTLs on LG 4a of selfed population were common with QTLs of 4a-b of

Table 6 The Pearson correlation coefficient (r) between biomass yield and plant height

Year	Location	Selfed population		Hybrid population	
		r	N	r	N
2012	PKS	0.41****	717	0.38****	526
2012	STW	0.29****	765	0.43****	526
2013	PKS	0.33****	693	0.20****	521
2013	STW	0.33****	767	0.30****	526

Note: N is the number of observations used in the calculation. **** Significant at the 0.0001 probability level.

Table 7 QTLs identified by multiple QTL mapping (MQM) for plant height in a selfed population of NL94 lowland switchgrass

Environment†	SN	Linkage group	Position of LOD peak/cM	LOD peak	Left locus	Right locus	Phenotypic variance explained (PVE)/%
STW12	1	1b	34.5	7.9	PVGA-1735/1736	sww-2320	8.8
	2	1b	48.6	8.1	sww-162	sww-196	9.1
	3	2a	47.2	9.6	sww-1805	PVCA-765/766	11.0
	4	2a	90.1	5.2	PVAAG-3245/3246	nfsg-129_282	5.3
	5	3b	0.0	4.8	PVE-977/978	PVCAG-2393/2394	5.1
	6	3b	15.9	5.3	PVCAG-2393/2394	PVGA-2105/2106	5.7
	7	3b	50.5	4.4	PVGA-1201/1202	PVGA-1853/1854	4.7
	8	4a	3.7	4.8	PVE-425/426	PVCA-219/220	5.0

(*Continued*)

Environment†	SN	Linkage group	Position of LOD peak/cM	LOD peak	Left locus	Right locus	Phenotypic variance explained (PVE)/%
	9	5a	50.9	4.9	PVGA-1357/1358	PVGA-1971/1972	4.4
	10	5a	56.2	5.0	PVGA-1971/1972	PVAAG-2861/2862	4.5
	11	5b	62.3	7.7	sww-556	PVAAG-3139/3140	8.5
	12	5b	91.6	6.0	sww-2250	PVE-571/572	6.5
PKS13	1	1b	24.8	5.2	PVGA-1271/1272_345	sww-2115	3.8
	2	2a	46.2	10.7	sww-1805	PVCA-765/766	10.2
	3	2b	50.4	5.7	PVE-775/776	PVE-413/414	5.0
	4	3a	65.2	7.3	PVE-169/170	sww-2503	6.8
	5	6b	103.0	6.9	sww-1749	sww-3053_180d	6.1
	6	8a	67.8	6.2	5005_B08	PVCA-979/980	5.4
	7	8b	49.0	4.8	PVGA-1149/1150	PVCA-541/542	4.1
	8	9b	87.2	4.5	PVE-613/614	sww-466	3.8
STW13	1	1b	2.0	5.7	PVCAG-2361/2362	PVAAG-2143/2144	2.7
	2	1b	18.9	4.7	PVGA-1401/1402	sww-2271	1.9
	3	1b	48.6	22.1	sww-162	sww-196	14.5
	4	2a	49.3	9.7	sww-938	Millet-MPGD25	5.0
	5	3b	103.3	4.6	PVGA-1665/1666	PVAAG-3029/3030	1.9
	6	4a	0.6	7.3	PVGA-1135/1136	PVE-425/426	3.7
	7	4a	3.7	13.5	PVE-417/418	PVGA-1637/1638	7.9
	8	5a	85.1	12.1	PVE-361/362	sww-2387	6.5
	9	5b	63.0	10.5	sww-556	PVAAG-3139/3140	5.6
	10	5b	80.2	7.1	Millet-MPGD19	sww-2250	3.5
	11	9a	66.5	4.5	PVGA-1605/1606_160	sww-463	2.2
	12	9a	76.9	5.1	sww-2364	sww-2285	2.5
	13	9a	96.8	7.3	sww-651	PVAAG-3091/3092	3.7
	14	9a	105.1	5.7	PVCAG-2517/2518	PVCAG-2281/2282	2.8
	15	9b	63.8	9.3	nfsg-299	PVGA-1225/ 1226_168	4.7
	16	9b	95.3	6.0	PVGA-1153/1154	5008_B05	2.9
	17	9b	99.7	7.5	5008_B05	nfsg-202	3.8
	18	9b	119.9	5.0	PVCAG-2487/2488	PVE-219/220	2.4
	19	9b	147.0	4.4	PVCAG-2615/2616	sww-585	2.1
Allenv	1	1b	48.6	6.0	sww-162	sww-196	6.9
	2	2a	55.6	6.2	PVE-625/626	sww-393	7.1
	3	2b	11.0	5.3	sww-1534	PVE-831/832	5.9
	4	3a	102.1	4.6	PVGA-1387/1388	PVCAG-2239/2240	5.1
	5	7a	61.1	5.4	PVGA-1869/1870	sww-2167	6.0
	6	7a	73.7	5.8	sww-2876	PVGA-2139/2140	6.5
	7	9a	12.0	4.7	PVE-281/282	PVE-1135/1136	5.2
	8	9a	28.5	5.5	sww-125_208	PVE-953/954	6.1
	9	9b	123.9	5.7	PVE-487/488	PVCA-19/20	6.5
	10	9b	147.0	4.4	PVCAG-2615/2616	sww-585	4.9

Note: † PKS, location Perkins, OK; STW, location Stillwater, OK; The numbers 12 and 13 in column 1 represent the years 2012 and 2013, respectively; Allenv, overall environments.

Table 8 QTLs identified by multiple QTL mapping (MQM) for plant height in a hybrid population of NL94 × SL93 lowland switchgrass parents

Environment†	SN	Linkage group	Position of LOD peak/cM	LOD peak	Left locus	Right locus	Phenotypic variance explained (PVE)/%
PKS12	1	1a	1.0	5.2	PVCAG-2537/2538	sww-1667	6.4
	2	1b	37.3	8.1	PVAAG-2987/2988	PVCA-179/180	10.6
	3	2b	24.1	6.4	PVGA-2079/2080	PVCAG-2647/2648	8.1
	4	3b	17.2	5.0	PVE-977/978	PVGA-1201/1202	7.0
	5	4a-b	63.8	4.4	PVCA-219/220	PVCA-793/794	5.4
	6	5b	40.2	5.7	PVAAG-3163/3164	PVCAG-2535/2536	7.1
STW12	1	1b	28.9	6.8	PVAAG-2987/2988	PVGA-1401/1402	8.2
	2	1b	68.8	8.4	PVCA-179/180	sww-2320	10.3
	3	2a	19.4	9.7	sww-532	nfsg-052	12.3
	4	7a	0.0	9.6	PVCAG-2503/2504	PVAAG-3051/3052	13.3
	5	9b	27.4	4.5	sww-2377	nfsg-200	5.2
PKS13	1	1a	3	7.9	PVCAG-2537/2538	sww-1667	3.0
	2	1b	1	4.4	sww-2034	sww-2405_160	1.6
	3	1b	23.862	6.3	sww-2405_160	PVCAG-2361/2362	2.3
	4	1b	43.264	6.2	PVCAG-2361/2362	PVCA-179/180	2.2
	5	1b	72.827	9.6	PVCA-179/180	PVAAG-3353/3354	3.6
	6	2b	24.082	9.7	PVE-781/782	PVCAG-2647/2648	3.8
	7	3a	16.691	19.6	PVAAG-3315/3316	PVCA-55/56	8.7
	8	3a	62.485	6.8	PVCA-55/56	PVCAG-2297/2298	2.0
	9	3b	5	5.2	PVE-977/978	PVCAG-2393/2394	1.8
	10	3b	20.334	14.4	PVCAG-2393/2394	PVGA-1201/1202	6.5
	11	3b	87.739	7.8	PVGA-1599/1600	sww-323	2.9
	12	4a-b	16.698	9.7	PVAAG-2979/2980	sww-1795	3.8
	13	4a-b	65.673	4.8	PVGA-1637/1638	PVCA-793/794	1.7
	14	5a	30.874	12.5	PVE-1369/1370	PVGA-1813/1814	5.1
	15	5a	44.283	12.9	PVGA-1813/1814	PVCAG-2197/2198	4.9
	16	5b	53.701	6.8	PVAAG-3163/3164	PVCAG-2535/2536	2.4
	17	6b	11.411	8.2	sww-1889	sww-1749	3.1
	18	7a	13.017	17.8	PVAAG-2881/2882	sww-1742	7.9
	19	7a	43.418	7.9	sww-2167	sww-348	3.0
	20	8b	20.091	5.2	sww-1862	nfsg-219	1.9
	21	8b	30.752	5.4	nfsg-219	PVGA-2005/2006	1.9
	22	9b	0	4.3	PVGA-1663/1664	nfsg-021	1.5
STW13	1	1b	52.964	6.29	PVGA-1401/1402	sww-2320	11.9
	2	2b	23.806	4.36	PVGA-2079/2080	sww-573	8.1
	3	5b	37.212	7.18	PVAAG-3163/3164	PVCAG-2535/2536	13.7
	4	6b	11.411	5.6	PVAAG-3017/3018	sww-1813	10.5
	5	7a	0	6.78	PVCAG-2503/2504	PVAAG-3253/3254	13.1
Allenv	1	1a	2.0	10.0	PVCAG-2537/2538	sww-1667	1.7
	2	1b	0.0	4.2	sww-2034	sww-2405_160	0.6
	3	1b	22.9	11.8	sww-2405_160	PVCAG-2361/2362	2.1
	4	1b	37.3	20.8	PVCAG-2361/2362	PVCA-179/180	4.5
	5	2a	17.4	32.5	sww-532	nfsg-052	8.8
	6	2a	50.6	6.6	PVCA-765/766	PVAAG-3245/3246	0.9

(*Continued*)

Environment†	SN	Linkage group	Position of LOD peak/cM	LOD peak	Left locus	Right locus	Phenotypic variance explained (PVE)/%
	7	2b	23.8	29.1	PVE-781/782	PVCAG-2647/2648	7.3
	8	3a	9.7	4.1	PVAAG-3315/3316	PVCA-55/56	0.6
	9	3b	6.0	16.8	PVE-977/978	PVCAG-2393/2394	3.4
	10	3b	17.3	30.0	PVCAG-2393/2394	PVGA-1201/1202	7.7
	11	3b	31.4	4.5	PVGA-1201/1202	PVGA-1727/1728	0.6
	12	3b	87.7	16.0	PVGA-1599/1600	sww-323	3.1
	13	4a-b	10.0	5.7	PVAAG-2979/2980	sww-1795	0.8
	14	4a-b	63.8	7.1	PVCA-219/220	PVCA-793/794	0.9
	15	5a	30.9	9.5	PVCAG-2167/2168	PVGA-1813/1814	1.7
	16	5a	50.8	12.5	PVGA-1649/1650	PVCAG-2197/2198	2.3
	17	5a	70.0	5.8	sww-389	sww-2387	1.0
	18	5b	2.2	17.4	PVGA-1773/1774	PVGA-1243/1244	3.5
	19	5b	39.2	9.5	PVAAG-3163/3164	PVCAG-2535/2536	1.6
	20	6b	13.8	14.4	sww-1889	sww-1749	2.7
	21	7a	11.0	14.6	PVAAG-3051/3052	PVGA-1869/1870	2.9
	22	7a	12.0	14.5	PVGA-1869/1870	sww-1742	2.8
	23	7a	37.9	5.6	PVGA-2139/2140	sww-348	0.8
	24	8b	0.0	5.7	PVCA-979/980	sww-1862	0.9
	25	8b	20.5	16.6	sww-1862	nfsg-219	3.3
	26	8b	33.4	6.9	nfsg-219	PVGA-1275/1276	1.1
	27	9a	0.0	5.0	sww-463	PVAAG-3091/3092	0.8
	28	9a	22.7	9.5	PVGA-1513/1514	PVCA-863/864	1.7
	29	9a	26.2	7.1	PVCA-863/864	nfsg-137	1.2
	30	9a	45.3	4.9	nfsg-137	PVCA-17/18	0.8
	31	9b	0.0	9.4	PVGA-1663/1664	PVCAG-2487/2488	2.0
	32	9b	23.6	12.0	PVCAG-2487/2488	sww-2377	2.2
	33	9b	26.6	10.1	sww-2377	PVCA-7/8	2.3
	34	9b	30.7	20.6	PVCA-7/8	nfsg-202	5.5
	35	9b	34.6	6.8	nfsg-202	5008_B05	1.1

Note: † PKS, location Perkins, OK; STW, location Stillwater, OK; The numbers 12 and 13 in column 1 represent the years 2012 and 2013, respectively; Allenv, overall environments.

hybrid population. QTLs at 50.8 to 51.6 cM on LG 5a were common between the selfed population and the hybrid population. QTLs at 27.2 to 29.2 cM on LG 9a were common between selfed and hybrid population.

Serba et al.[22] observed a QTL for plant height at LG VIIa-f (f refers to female linkage group) at 1.0 cM position which was consistent with our findings at environments STW12 and STW13 for the hybrid population. The QTL in the same group at 35.3 cM position was consistent with our findings at environments PKS13 and Allenv for hybrid population. The QTL detected by their study at LG IVa-f at 21.3 cM position is consistent with our findings at environment PKS13 for the hybrid population. Similarly, their QTL at IXb-f at 16.2 cM position is consistent with our QTL at environment Allenv for the hybrid population. Their QTL observation at IVb-m (m refers to male linkage group) at 16.4 cM is consistent with our QTL at environment PKS13 and Allenv for hybrid population. Their QTL at IVa-m 64 cM position is consistent with our findings at environments PKS12 and Allenv for the hybrid population. Their QTLs at LG Va-m at 40.6 cM, 43 cM, and 52.0 cM are consistent with our findings at environment PKS13 for the hybrid population. The QTL observed by Serba et al.[22] at LG IXb-m at 122.1 cM position is consistent with our QTL for Allenv for the selfed population. The other QTLs observed by Serba et al.[22] were not consistent with our results. Lowry et al.[20] observed a QTL for plant height at LG 2a at 63.17 cM, at

LG 3b at two positions 58 cM and 61 cM, at LG 4a at 37 cM, and at LG 9b at 76 cM. Our findings were not consistent with the results of Lowry et al.[20].

4 Conclusions

Two lowland switchgrass mapping populations were studied in this experiment. A large genetic variation existed for plant biomass and height within the two populations. This study confirmed that plant height was significantly correlated with biomass yield in lowland switchgrass. The data analysis in the selfed population across all environments revealed 10 QTLs on seven LGs. The separate analysis of the selfed population in each environment revealed 39 QTLs among which three QTLs appeared consistently in two environments and one appeared consistently in three environments. The data analysis with the hybrid population across all environments revealed 35 QTLs across 14 LGs. The markers tightly linked to the QTLs have the potential to be used in marker assisted selection for breeding improvement programs in switchgrass.

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Compliance with ethics guidelines Shiva O. Makaju, Yanqi Wu, Michael P. Anderson, Vijaya G. Kakani, Michael W. Smith, Linglong Liu, Hongxu Dong, and Dan Chang declare they have no conflicts of interest or financial conflicts to disclose.

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