Potential utilization of native prairie grasses from western Canada as ethanol feedstock

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Jefferson, P. G., McCaughey, W. P., May, K., Woosaree, J. and McFarlane, L. 2004. **Potential utilization of native prairie grasses from western Canada as ethanol feedstock**. Can. J. Plant Sci. **84**: 1067–1075. The utilization of native grass species for potential biomass feedstocks of the emerging ethanol industry requires more information about their cellulose and hemicellulose concentration. Ten native species were grown at seven sites across the prairie region of western Canada for two to four growing seasons. Northern wheatgrass, *Elymus lanceolatus,* produced high concentrations of cellulose (363 g kg^{-1}) but low concentrations of hemicellulose (266 g kg^{-1}). Green needlegrass, *Nasella viridula*, produced high concentrations of both constituents (351 and 307 g kg^{-1}). Four warm-season grasses, big bluestem, *Andropogon gerardii*, little bluestem, *Schizachyrium scoparium*, prairie sandreed, *Calamovilfa longifolia*, and switchgrass, *Panicum virgatum*, had 346, 342, 340 and 338 g kg^{-1}, respectively, concentrations of cellulose and also exhibited a positive response to temperature that resulted in increased hemicellulose concentration. Accumulated thermal time (degree day base 10°C) was correlated to hemicellulose concentrations in the warm-season grasses but not for cool-season grasses. Holocellulose (cellulose + hemicellulose) concentration differences varied among site-years but warm-season grasses were more stable in hollocellulose concentration than cool-season grasses.

Key words: Biomass, native grasses, cellulose, hemicellulose, biofuel

Jefferson, P. G., McCaughey, W. P., May, K., Woosaree, J. et McFarlane, L. 2004. **Utilité éventuelle des graminées indigènes des plaines de l'ouest du Canada pour la production d'éthanol**. Can. J. Plant Sci. **84**: 1067–1075. Avant que l'industrie naissante de l'éthanol puisse se servir des graminées indigènes comme matière première, on doit en apprendre davantage sur la teneur en cellulose et en hémicellulose de ces dernières. Les auteurs ont cultivé dix espèces indigènes à sept endroits dans les prairies de l'Ouest canadien pendant 2 à 4 saisons végétatives. Le chiendent nordique, *Elymus lanceolatus*, produit une grande quantité de cellulose (363 g par kg), mais peu d'hémicellulose (266 g par kg). Le stipe vert, *Nasella viridula*, synthétise les deux en abondance (351 et 307 g par kg). Quatre graminées de la belle saison, soit le barbon de Gérard (*Andropogon gerardii*), le schizachyrium à balais (*Schizachyrium scoparium*), le calamovilfa à longues feuilles (*Calamovilfa longifolia*) et le panic raide (*Panicum virgatum*), contenaient respectivement 346, 342, 340 et 338 g de cellulose par kilo et réagissent positivement à la température en accroissant leur production d'hémicellulose. Le nombre d'unités thermiques accumulées (degrés-jours au-dessus de 10 °C) présente une corrélation positive avec la concentration d'hémicellulose chez les graminées de la saison chaude mais pas chez celles de la saison froide. La concentration d'holocellulose (cellulose + hémicellulose) varie d'une année et d'un site à l'autre, mais elle est plus stable chez les graminées de la saison chaude que chez celles de la saison froide.

Mots clés: Biomasse, graminées indigènes, cellulose, hémicellulose, biocarburant

Native grass species of the Canadian prairie provinces, the northern reaches of the Northern Great Plains of North America, are of renewed interest for re-vegetation of marginal or degraded farmland (Wark et al. 1995). Switchgrass, *Panicum virgatum* L., is native to western Canada, and has been identified by the US Department of Energy for development as a herbaceous biomass fuel crop (Vogel and Jung 2001). Its use also has been proposed in Canada for both biomass fuel and fiber production, particularly as an alternative crop on marginal soils that would require low inputs of nutrients and management (Samson and Omeilan 1998). Samson (1991) proposed that 35 million acres (14.6 million ha) of marginal cropland in the prairie region of western Canada could be seeded to switchgrass for biomass production. Biomass fuel production utilizes microbial enzyme technology to convert the cellulose and hemicellulose contained in plant cell walls to constituent sugars and then ferment those sugars to produce ethanol (Vogel and Jung 2001). The cellulose concentration of several native grasses is higher than several introduced grass species (Smoliak and Bezeau 1967). However, the cellulose and hemicellulose concentrations of switchgrass and other native grasses were unknown for the prairie region.

Native rangelands of the prairie provinces are dominated by cool-season grasses (Budd et al. 1987) that exhibit the three-carbon (C_3) photosynthetic biochemistry. However, many warm-season grasses that exhibit the four-carbon (C_4) photosynthetic biochemistry are found in the region, particularly in southern Manitoba and southeastern Saskatchewan. At some locations, the occurrence of warm-season grasses is favored by soil type or other edaphic factors. Warm-season grasses have a higher proportion of vascular tissue and cellulosic content than cool-season grasses (Van Soest 1982). Switchgrass is a C_4 or warm-season grass. Jefferson et al. (2002) reported that switchgrass could be successfully seeded for reclamation or forage production at southern latitudes of the Canadian prairie region. Its cellulose and hemicellulose concentrations have not been compared to other, more common, cool-season native grasses such as northern wheatgrass, *Elymus lanceolatus*, western wheatgrass, *Pascopyrum smithii*, or green needlegrass, *Nassella viridula*. These cool-season grasses have exhibited a wider range of adaptation in western Canada than native warm-season grasses (Jefferson et al. 2002).

Our study objective was to evaluate cultivars of 10 grass species that were selected for adaptation to North Dakota or Montana environments at seven sites in the prairie region of western Canada by determining cellulose and hemicellulose concentrations and holocellulose yield. A secondary objective was to compare the cellulose and hemicellulose concentrations of cool-season species to those of warm-season species.

MATERIALS AND METHODS

For a complete description of site characteristics see the report of Jefferson et al. (2002). In brief, seed of 12 native grass cultivars were obtained from the USDA-NRCS Plant Materials Centre in Bismarck, North Dakota. The cultivars and grasses were: Dacotah switchgrass, *Panicum virgatum* L; Tomahawk indiangrass, *Sorghastrum nutans* (L.) Nash; Badlands little bluestem, *Schizachyrium scoparium* (Michx.) Nash; Bison big bluestem, *Andropogon gerardii* Vitman; Goshen and ND-95 prairie sandreed, *Calamovilfa longifolia* (Hook.) Scribn.; and Killdeer sideoats grama, *Bouteloua curtipendula* (Michx.) Torr. We used Alderson and Sharp (1994) for nomenclature of native grasses. Seed of several cool-season native species was also obtained, namely: Rodan and Rosana western wheatgrass, *Pascopyrum smithii* (Rydb.) A. Love; Lodorm green needlegrass, *Nasella viridula* (Trin.) Barkworth (syn. *Stipa viridula* Trin.); and Critana northern wheatgrass, *Elymus lanceolatus* (Scribn. & J.G. Sm.) [syn. *Agropyron dasystachyum* (Hook.) Scribn.& J.G. Sm.] (USA common name is thickspike wheatgrass). One introduced grass, ND-691 mammoth wildrye [*Leymus racemosus* (Lam.) Tzvelev (Syn. *Elymus giganteus* Vahl.)], from the former USSR was included for study because it was deemed to have wildlife habitat potential. Mammoth wildrye has been seeded for soil conservation on sand dunes and other dry sites in Washington state (Alderson and Sharp 1994). Each of these species occurs in native rangeland of the Canadian prairie provinces (Budd et al. 1987) but switchgrass and indiangrass were described as occurring rarely.

These grasses were seeded in replicated, randomized complete block design trials at five sites in western Canada in 1992 or 1993. The sites were: Brandon, Manitoba; Swift Current, Saskatchewan; Melfort, Saskatchewan; Lethbridge, Alberta; and Vegreville, Alberta. An additional site was seeded at Brandon to compare clay soil vs. sandy soil. An irrigated site was seeded at Swift Current to allow a comparison of species under minimal water stress. Sites were sampled in late September ot early October in 1994 and 1995 at all sites with additional samples in 1996 and 1997 at the two sites in Swift current. Sub-samples were ground to a 1-mm particle size and fiber properties determined. Cellulose concentration was determined by the method of Crampton and Maynard (1938). Hemicellulose concentration was determined by the difference between neutral detergent fiber and acid detergent fiber concentrations (Goering and Van Soest 1970). Ash concentration was determined by standard laboratory procedure by combustion at 600°C for 24 h (Association of Official Analytical Chemists 1990). The factors that affect biochemistry of lignocellulose degradation are not well known. However, holocellulose (cellulose + hemicellulose) would be the primary source of fermentable sugars for ethanol production. Holocellulose concentration and holocellulose yield (holocellulose concentration (biomass) (Hopkins et al. 1995) were also determined.

Daily weather data, consisting of maximum (T_{max}) and minimum (T_{min}) temperatures and rainfall amounts, were collected at each site. Daily mean temperature (T_{mn}) was calculated from the average of T_{max} and T_{min}. Monthly mean T_{mn}, T_{max}, and T_{min} and the monthly total precipitation were calculated for each site-year combination. The irrigation amounts at the Swift Current irrigation site were included in the precipitation data for that site. We calculated thermal time accumulation for June, July and August by summing degree days with a base temperature of 10°C (Sanderson and Wolf 1995).

The statistical analysis started with an ANOVA including replication, grass entry, site, and year effects in a General Linear Model of SAS (SAS Institute, Inc. 1985). The combined analysis indicated that the site × year × grass interaction was significant ($P \leq 0.05$) so each site-year combination was analyzed separately with replication and grass species as the only factors in the model. When the grass effect was significant ($P \leq 0.05$) in the ANOVA, a Least Significant Difference (LSD) was used for mean separation of the grasses. For prairie sandreed and western wheatgrass, the two cultivars were averaged for presentation of species means. Cool- and warm-season grasses were compared with a least square mean contrast. Correlation and regression statistics were calculated with JMP software (SAS Institute, Inc. 1995).

In order to examine stability or consistency of fiber concentrations across locations, species concentration was regressed against the site mean according to the technique of Findlay and Wilkinson (1963). The slope of this regression relationship describes the stability of a species in response to changes in the environment. A slope near the value of 1.0 indicates a species that responds to environmental variation in a fashion typical of all entries for the trait of interest. A slope above 1.0 indicates a species that responds positively as the environment becomes more conducive for the trait. A slope below 1.0 indicates a species that responds negatively as the environment becomes more conducive for the trait. A high overall mean for the trait of interest is also desirable.

RESULTS AND DISCUSSION

Cellulose concentration of native grasses differed at every site and year sampled (Table 1). Western wheatgrass consistently exhibited the lowest cellulose concentration at every site-year except Vegreville in 1995. Cellulose concentration of this grass was 12% below the site mean at each location. The grass species exhibiting the highest cellulose concentration varied among site years. At the Brandon sites, little bluestem, sideoats grama and northern wheatgrass had the highest cellulose concentrations. At Lethbridge, big bluestem and little bluestem had the highest cellulose concentrations. At Melfort and Vegreville, northern wheatgrass had the highest cellulose concentration. At Swift Current-dryland, northern wheatgrass and green needlegrass had the highest cellulose concentrations. At Swift Current-irrigation, prairie sandreed, green needlegrass, northern wheatgrass, and big bluestem exhibited the highest cellulose concentrations. No one species was consistently superior in cellulose concentration. However, some species exhibited a more consistent performance than others. Northern wheatgrass had among the highest cellulose concentrations in 15 of a possible 18 site-years and thus, on average, was the highest. Green needlegrass was among the highest cellulose concentrations in 9 of 18 site years. Little bluestem appeared to be very inconsistent in cellulose concentration as it ranged from highest at Brandon-clay in 1994 and Lethbridge in 1995 to lowest at Vegreville in 1994.

Cool- and warm-season grasses differed in cellulose concentration at 14 site-years (Table 1). Warm-season grasses had higher cellulose concentration at 8 site-years while cool-season grasses had higher cellulose concentration at 6 site-years.

Prairie sandreed had the highest hemicellulose concentrations at 6 site-years, sideoats grama at 4 site-years, green needlegrass at 5 site-years, and little bluestem at 2 site-years (Table 2). Mammoth wildrye exhibited the lowest hemicellulose concentrations at 13 site-years, northern wheatgrass at 2 site-years, western wheatgrass at 2 site-years and prairie sandreed and indiangrass at 1 site-year each. The ranking of northern wheatgrass and little bluestem hemicellulose concentration ranged from highest to lowest and indicate the instability of these species.

Warm-season grasses had higher hemicellulose concentrations than cool-season grasses at 13 site-years (Table 2) and tended to exhibit higher hemicellulose concentration at Brandon-clay in 1994 (P = 0.07). The differences ranged from 23% at Brandon-sand in 1995 to 10% at Lethbridge in 1995. The cool- and warm-season grasses were not different in hemicellulose concentration at Melfort and Vegreville, two sites that were considered marginal for warm-season grass adaptation (Jefferson et al. 2002).

The cellulose and hemicellulose concentrations for switchgrass are similar to values reported by Jung and Vogel (1992) for Nebraska. These authors reported cellulose concentrations of 297 and 367 g kg^{-1} for leaf and stem fractions at heading stage. Our values from whole plant samples ranged from 297 to 382 g kg^{-1}. Similarly, these authors reported hemicellulose concentrations of 336 and 312 g kg^{-1} for leaf and stem fractions while our values

Table 1. Cellulose concentration by site and year of 10 native grasses grown for 2 to 4 yr at five sites in western Canada

	Brandon-clay		Brandon-sand		Lethbridge		Melfort		Swift Current-dryland				Swift Current-irrigation				Vegreville	
	1994	1995	1994	1995	1994	1995	1994	1995	1994	1995	1996	1997	1994	1995	1996	1997	1994	1995
	(g kg^{-1})																	
Cool-season																		
Northern wheatgrass	331	396	366	372	359	357	384	336	413	371	386	386	397	334	350	348	402	372
Green needlegrass	321	403	317	364	356	362	336	332	368	345	387	358	362	349	348	346	373	348
Mammoth wildrye	327	407	349	344	322	350	312	331	360	348	349	351	352	317	350	353	370	342
Western wheatgrass	291	312	283	290	302	305	298	275	308	280	289	304	317	276	291	287	335	288
Warm-season																		
Big bluestem	343	408	326	369	372	401	–	269	333	339	371	356	315	317	344	373	379	290
Sideoats grama	329	413	327	365	358	377	–	–	308	370	–	339	308	254	317	319	352	295
Prairie sandreed	299	369	315	347	339	407	312	284	343	367	348	353	293	297	321	345	370	310
Little bluestem	360	392	334	368	350	414	–	–	326	336	368	356	326	298	340	370	356	335
Indiangrass	344	371	362	–	346	394	–	–	332	320	–	–	317	301	–	382	352	271
Switchgrass	316	403	309	356	353	382	–	–	316	327	346	334	304	297	328	372	359	309
Grass Prob > F	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01
$LSD_{0.05}$	36	17	15	17	24	20	26	32	16	17	18	17	19	32	13	13	30	18
cv %	7.7	3.1	3.2	3.4	5.0	3.8	5.7	7.4	3.6	3.6	3.6	3.4	3.3	7.3	5.7	2.6	5.8	3.9
Cool-season mean	318	380	329	342	335	343	332	318	362	336	353	350	357	319	335	333	370	337
Warm-season mean	332	393	329	361	353	396	–	276	326	343	358	348	310	294	330	360	361	302
Contrast Prob > F	0.02	<0.01	NS	<0.01	<0.01	<0.01	–	<0.01	<0.01	<0.01	<0.01	0.03	<0.01	0.01	NS	<0.01	NS	<0.01

Table 2. Table 2. Hemicellulose concentration by site and year of 10 native grasses grown for 2 to 4 yr at five sites in western Canada

	Brandon-clay		Brandon-sand		Lethbridge		Melfort		Swift Current-dryland				Swift Current-irrigation				Vegreville	
	1994	1995	1994	1995	1994	1995	1994	1995	1994	1995	1996	1997	1994	1995	1996	1997	1994	1995
	(g kg^{-1})																	
Cool-season																		
Northern wheatgrass	276	269	263	238	269	242	279	262	278	279	275	252	280	268	262	252	279	278
Green needlegrass	306	333	306	336	310	297	312	297	297	309	324	310	303	289	291	290	305	317
Mammoth wildrye	245	292	244	256	227	235	235	256	256	257	249	258	241	226	236	242	252	260
Western wheatgrass	266	271	265	260	262	285	263	284	279	256	258	267	270	250	252	247	290	272
Warm-season																		
Big bluestem	293	322	274	319	281	302	–	280	292	290	293	299	294	273	291	301	284	271
Sideoats grama	288	346	304	362	292	312	–	–	335	317	–	322	338	287	312	353	288	284
Little bluestem	308	353	321	334	310	308	–	–	317	310	292	309	318	307	307	321	286	291
Prairie sandreed	245	333	294	340	334	293	296	271	341	336	322	325	340	315	292	297	301	281
Indiangrass	297	308	294	–	268	307	–	–	315	302	–	–	313	281	–	312	261	261
Switchgrass	270	329	287	327	283	306	–	–	314	297	388	314	309	284	302	310	277	294
Grass Prob > *F*	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	NS
$LSD_{0.05}$	28	25	20	12	51	22	12	19	16	16	14	25	25	30	15	8	18	NS
cv %	6.9	5.7	4.9	2.8	12.3	5.4	3.0	4.6	3.6	3.8	3.2	5.8	5.0	7.4	3.8	1.9	4.4	11.7
Cool-season mean	273	291	269	272	267	265	272	277	274	275	276	272	273	258	260	258	281	282
Warm-season mean	283	332	296	336	295	305	–	275	319	308	324	314	319	291	301	316	283	280
Contrast Prob > *F*	0.07	<0.01	<0.01	<0.01	<0.01	<0.01	–	NS	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	NS	NS

ranged from 270 to 329 g kg^{-1}. Lemus et al. (2002) reported 361 g kg^{-1} cellulose and 316 g kg^{-1} hemicellulose concentration means for 20 switchgrass cultivars grown in Iowa. These values are very similar given differences in sites, cultivars, and analytical techniques between other reports (Jung and Vogel 1992; Lemus et al. 2002) and our study. This suggests that the cellulose concentration of switchgrass is stable across its range of adaptation.

Smoliak and Bezeau (1967) reported 421 g kg^{-1} cellulose concentration for western wheatgrass compared to 296 g kg^{-1} in our results. Their values may be higher because they sampled from a native rangeland site in southern Alberta rather than seeded pure stands as in our study. They may have sampled more standing dead litter than we did in this study. As litter weathers, the concentration of ligno-cellulosic components increase due to the loss of soluble cell contents. Weather conditions may have also contributed to the differences between our results and those of Smoliak and Bezeau (1967).

Across 16 site-years, mean hemicellulose concentration of warm-season grasses responded positively to increasing degree days (Fig. 1B), while concentration in cool-season grasses did not (Fig. 1A). Sanderson and Wolf (1995) reported a similar positive relationship between neutral or acid detergent fiber concentration and degree days for switchgrass grown in Texas. They reported a larger response for NDF (b = 0.272 for Alamo switchgrass grown at Stephenville, Texas) than for ADF (b = 0.218) (Sanderson and Wolf 1995), which suggests that hemicellulose would increase with degree days although they did not report hemicellulose concentrations. Because hemicellulose concentration is calculated from the difference between NDF and ADF, we concluded that our results for several warm-season grasses are similar to Sanderson and Wolf's (1995) results. The range of degree day values in our study was only 30% of the values reported by Sanderson and Wolf (1995), presumably due to the higher latitude and shorter, cooler growing season in western Canada compared to Texas. Cool-season grasses are capable of growth at temperatures between 0 and 10°C so we also correlated hemicellulose concentrations for northern wheatgrass, green needlegrass, mammoth wildrye and western wheatgrass to degree days base 0°C. There was no significant correlation for degree days base 0°C and hemicellulose concentration of cool-season grasses (data not shown).

Increased temperature has been reported to increase the lignification of grass cell walls but it also contributes to advancing phenological stage, i.e., a decline in leaf proportion and an increase in stem proportion of the biomass (Van Soest 1982). While these species were all harvested at the end of the growing season, we have no estimate of leaf:stem ratios. Warmer summer temperatures may have produced lower leaf:stem ratio and higher hemicellulose concentrations in the warm-season grasses and contributed to this temperature relationship. In eastern Canada, switchgrass yield and phenological development were closely correlated to time (day after May 01) in 2 yr (Madakadze et al. 1999). These relationships were likely also dependent on thermal time.

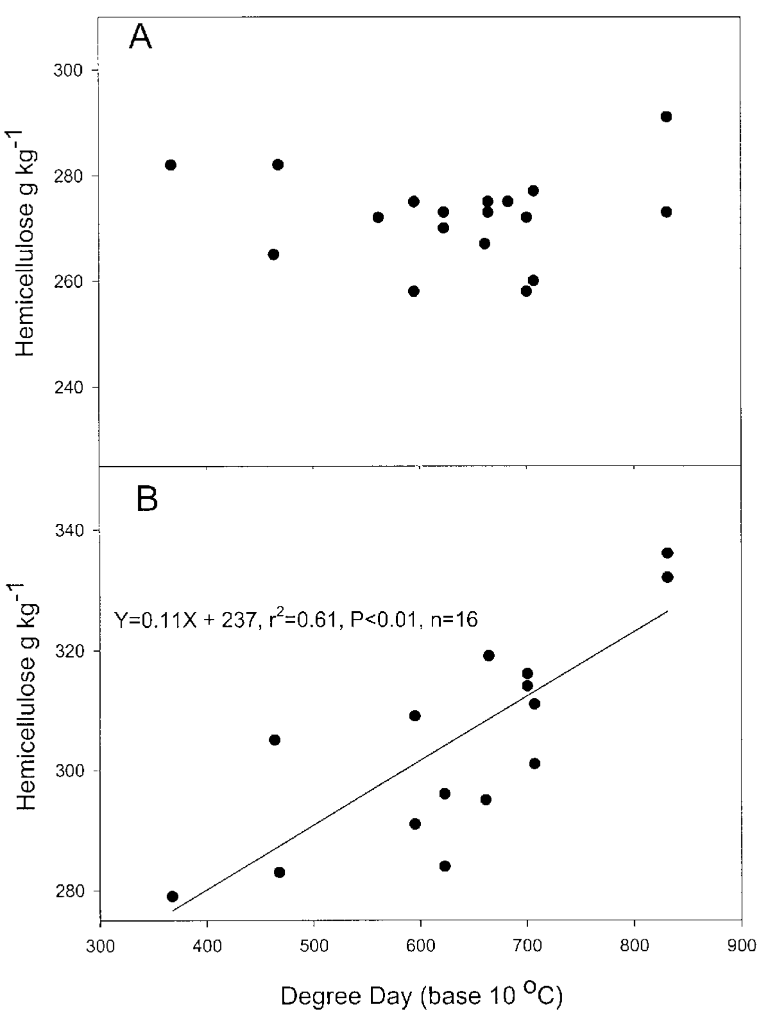

Fig. 1. Mean hemicellulose concentration as influenced by accumulated degree day (base 10°C) at each site-year for: (A) cool-season grass mean at each site-year (n = 18) and (B) warm-season grass mean at each site-year (n = 16).

Holocellulose concentration varied among the grasses in every year at every location (Table 3). Green needlegrass had the highest holocellulose concentration among the grass species at 5 site-years, northern wheatgrass had the highest concentration at 3 site-years, sideoats grama at 3 site-years, little bluestem at 3 site-years, prairie sandreed at 2 site-years, and indiangrass at one site-year. The advantage of green needlegrass and northern wheatgrass can be partly attributed to their performance at Vegreville and Melfort, sites where warm-season grasses are not adapted (Jefferson et al. 2002). Western wheatgrass exhibited the lowest holocellulose concentrations at 13 site-years while mammoth wildrye was lowest at 3 site-years.

Hollocellulose concentration was higher from warm- than cool-season grasses at 12 site-years (Table 3). It was higher for cool-season grasses at Melfort in 1995 but, as mentioned above, this site was probably marginal for warm-season grass production.

Western wheatgrass exhibited the lowest cellulose concentration response (slope) as the environment became more favorable for cellulose production (Fig. 2A) while big bluestem and sideoats grama exhibited the greatest response. Northern wheatgrass, green needlegrass and mammoth wildrye also had slopes below 1.0. The other warm-season species clustered around a slope of 1.0, which indicates a stable response for this variable. Northern wheatgrass had the highest mean cellulose concentration.

Northern wheatgrass and western wheatgrass exhibited the lowest response to environment for hemicellulose concentration with slopes about zero (Fig. 2B) while prairie sandreed, sideoats grama and switchgrass exhibited the highest responses with slopes above 1.0. Mammoth wildrye exhibited a slope near 1.0 but had the lowest mean hemicellulose concentration.

Northern wheatgrass and western wheatgrass also exhibited the lowest slopes for holocellulose concentration (Fig. 2C), while sideoats grama, big bluestem, switchgrass and prairie sandreed had the highest. Green needlegrass had a stable slope (b = 0.82) and the highest holocellulose concentration (661 g kg^{-1}). The highest slope for holocellulose concentration was observed for sideoats grama (b = 1.65) while big bluestem, switchgrass, prairie sandreed and little bluestem all had slopes above 1.0. Taken together, these results suggest that the warm-season grasses are more responsive to environmental conditions, such as warmer summer temperatures, which increase hemicellulose concentration, than cool-season grasses.

Table 3. Holocellulose (cellulose + hemicellulose) concentration by site and year of 10 native grasses grown for 2 to 4 yr at five sites in western Canada

	Brandon-clay		Brandon-sand		Lethbridge		Melfort		Swift Current-dryland				Swift Current-irrigation				Vegreville	
	1994	1995	1994	1995	1994	1995	1994	1995	1994	1995	1996	1997	1994	1995	1996	1997	1994	1995
	(g kg^{-1})																	
Cool-season																		
Northern wheatgrass	607	665	629	610	628	599	671	598	690	651	660	638	678	602	612	600	681	650
Green needlegrass	627	736	623	700	666	659	648	629	664	654	711	667	666	638	639	636	678	665
Mammoth wildrye	572	700	594	601	549	584	547	587	610	605	598	609	593	543	586	595	622	602
Western wheatgrass	558	584	547	551	564	590	561	558	582	536	548	573	586	526	543	534	625	560
Warm-season																		
Big bluestem	637	731	600	688	653	703	–	550	625	629	664	655	609	590	635	674	663	561
Sideoats grama	617	759	631	727	656	689	–	–	643	687	643	654	646	541	629	668	640	579
Little bluestem	669	745	655	703	660	719	–	–	643	680	–	662	611	604	628	665	656	601
Prairie sandreed	541	695	610	688	647	700	–	555	684	672	691	685	667	613	630	666	657	616
Indiangrass	640	678	649	–	614	706	–	–	648	621	–	–	630	572	–	693	614	533
Switchgrass	586	731	596	683	635	688	–	–	630	624	684	649	614	582	630	682	636	603
Grass Prob > *F*	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.04	<0.01
$LSD_{0.05}$	49	37	48	39	36	34	30	41	24	21	29	33	29	54	36	18	41	54
cv %	5.5	3.7	3.5	2.7	4.0	3.5	3.3	4.8	2.6	2.3	3.1	3.5	3.2	6.4	4.1	2.0	4.4	6.3
Cool-season mean	591	671	598	615	602	608	607	593	636	611	629	622	631	577	595	591	651	619
Warm-season mean	615	723	623	698	644	701	–	552	645	652	670	661	629	584	630	675	644	582
Contrast Prob > *F*	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	–	0.01	<0.01	<0.01	<0.01	<0.01	0.07	0.07	<0.01	<0.01	NS	NS

Holocellulose yield differences among the grasses (Table 4) was strongly influenced by biomass differences (Jefferson et al. 2002). Mammoth wildrye produced the highest holocellulose yield at 9 site-years, green needlegrass at 5 site-years, northern wheatgrass and western wheatgrass at one site-year each. They frequently produced higher holocellulose yield than most of the warm-season grasses, particularly at Brandon-clay, Lethbridge, and Swift Current-irrigation in 1994 or the first year of sampling. It is interesting to note at these sites that the difference between cool- and warm-season grasses decreased each year. At Swift Current-irrigation for example, the cool-season grasses were clearly superior in holocellulose yield in 1994 but inferior to big bluestem, prairie sandreed, and switchgrass by 1997. This rank order change over time might be due to more rapid establishment of the cool-season grasses in the first year after seeding (Jefferson et al. 2002) or perhaps to the ability of the warm-season grasses to produce more biomass under low nutrient conditions (Samson 1991) since no fertilizer was applied at any site.

Cool-season grasses produced higher hollocellulose yields at 9 site-years while warm- season grasses produced more at only one site-year (Table 4). These differences reflected forage production differences (Jefferson et al. 2002) rather than hollocellulose concentration differences as described above.

Ash concentration varied among grasses at every site-year (Table 5), but there were few consistent trends. For example, mammoth wildrye had the highest ash concentration at Lethbridge but the lowest at Melfort and Swift Current-dryland. Prairie sandreed had the highest ash concentration at two site-years and the lowest at 6 other site-years. Little bluestem had the lowest concentration at 5 site-years. Low ash concentration is desirable if the biomass will be used for co-firing with other fuels for energy production. High biomass ash concentration can result in slagging and fouling of combustion chambers (Lemus et al. 2002). Ash concentration of switchgrass ranged from 52 to 72 g kg^{-1} among 3 yr at one site in Iowa, USA (Lemus et al. 2002), while it ranged from 62 to 123 g kg^{-1} among our 16 site-years. Our ash concentrations were higher but typical of semiarid environments such as Swift Current.

Cool-season grasses had higher ash concentration than warm-season grasses at 9 site-years while the reverse was true at 2 site-years (Table 5). Lower ash concentration of warm-season grasses combined with greater hollocellulose concentration would be an advantage over cool-season grasses.

Switchgrass, big bluestem, little bluestem, and prairie sandreed had the highest concentrations of cellulose and hemicellulose. Samson and Omielan (1998) had proposed that other native warm-season grasses could be seeded for biofuel feedstock production along with switchgrass. They recommended prairie sandreed, big bluestem, and little bluestem as potential candidates, and our results confirm that these species do exhibit similar cellulose and hemicellulose concentrations to switchgrass at southern locations of the Canadian prairies. Northern wheatgrass had high concentrations of cellulose but not hemicellulose. Green needlegrass, a cool-season grass species, also had high concentrations and should be considered a viable candidate for

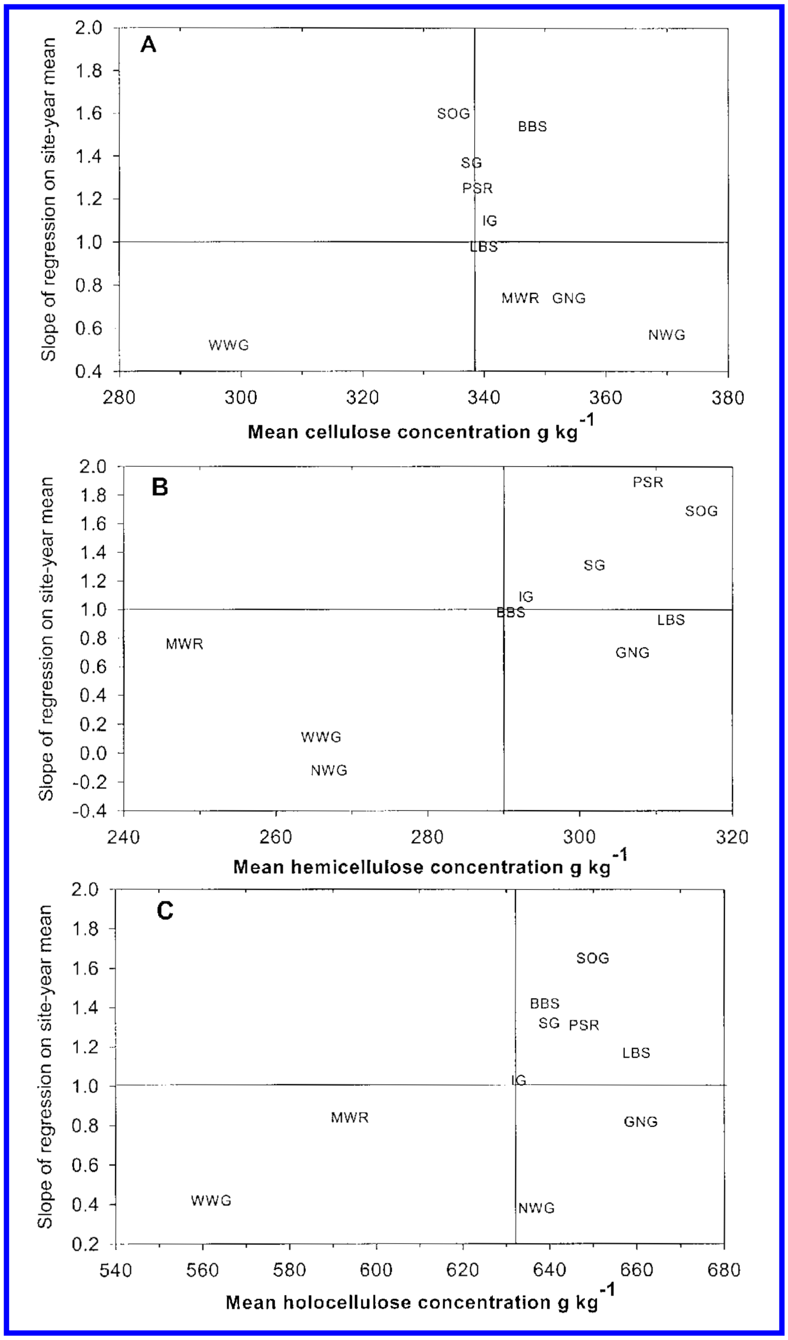

Fig. 2. Slope of regression of cellulose (A), hemicellulose (B) and holocellulose (C) concentration of each species on site mean relative to overall species mean. Species abbreviations used as symbols are: BBS, big bluestem; GNG, green needlegrass; IG, indiangrass; LBS, little bluestem; MWR, mammoth wildrye; NWG, northern wheatgrass; PSR, prairie sandreed; SG, switchgrass; SOG, sideoats grama; and WWG, western wheatgrass. Reference lines are drawn at slope 1.0 and overall mean concentration.

biomass production. It may be useful to include green needlegrass at northern latitude locations, such as Melfort, where warm-season grasses are not as well adapted. Western wheatgrass was not well-suited to biomass production for biofuel feedstocks.

As no biomass-based ethanol production industry yet exists on the Canadian prairies, we must speculate about the potential economics of native grasses for the production of holocellulose. If biomass feedstocks are priced on a mass basis, then cool-season grasses will clearly have an advantage over warm-season grasses. However, if prices are set on the basis of hollocellulose concentration, then warm-season grass species will be favored over cool-season grasses, particularly at southern locations where the warm-season grasses are adapted for biomass production. Hollocellulose concentrations in warm-season grass species were remarkably stable over site-years (Table 3; Fig. 2c) compared to wide variations in hollocellulose yield (Table 4). This result would be important information to industry prospectus and business plan development for biomass-based ethanol production.

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Table 4. Holocellulose (cellulose + hemicellulose) yield of 10 native grasses grown for 2 to 4 yr at five sites in western Canada

	Brandon-clay		Brandon-sand		Lethbridge		Melfort		Swift Current-dryland				Swift Current-irrigation				Vegreville	
	1994	1995	1994	1995	1994	1995	1994	1995	1994	1995	1996	1997	1994	1995	1996	1997	1994	1995
Cool-season	(g kg^{-1})																	
Northern wheatgrass	3.33	3.52	0.72	0.68	8.46	2.88	5.00	1.18	1.92	1.42	0.89	0.99	4.15	3.05	2.06	0.78	2.25	0.31
Green needlegrass	5.20	6.03	0.95	1.83	6.88	4.62	3.26	2.28	1.14	1.90	1.44	0.98	5.19	4.75	1.88	1.83	2.77	0.53
Mammoth wildrye	6.90	8.73	1.35	1.38	12.42	9.07	3.55	1.66	1.61	1.77	0.68	0.94	5.87	4.14	4.43	2.87	3.39	0.70
Western wheatgrass	5.63	4.18	0.93	1.05	8.47	4.22	4.12	1.18	1.95	1.60	0.91	1.14	4.65	3.49	1.94	1.18	2.18	0.24
Warm-season																		
Big bluestem	4.60	5.53	1.16	1.45	3.26	4.52	–	0.74	0.41	0.97	0.79	1.07	0.57	2.75	3.76	4.12	0.69	0.17
Sideoats grama	2.39	3.20	0.62	0.86	3.35	3.70	–	–	0.14	0.27	–	0.09	0.20	0.43	1.15	1.88	1.26	0.30
Little bluestem	5.05	6.18	1.24	1.46	4.40	3.34	–	–	0.15	0.88	0.35	0.38	0.07	0.32	0.33	0.43	1.46	0.36
Prairie sandreed	2.25	6.92	0.75	1.37	6.98	6.95	0.38	0.49	0.46	1.08	1.00	1.71	0.40	1.54	2.76	4.63	1.33	0.39
Indiangrass	3.26	4.48	0.96	–	4.60	4.48	–	–	0.12	0.36	–	–	0.18	0.48	–	0.69	0.54	0.15
Switchgrass	3.83	6.42	1.21	1.76	5.18	5.57	–	–	0.66	1.17	0.54	0.66	1.09	3.03	3.34	4.49	0.28	0.32
Grass Prob > *F*	<0.01	<0.01	<0.01	NS	<0.01	<0.01	–	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	0.05	<0.01	<0.01	<0.01
$LSD_{0.05}$	1.30	1.30	0.23	NS	1.68	1.45	–	0.44	0.44	0.68	0.47	0.58	1.05	1.40	NS	1.62	0.58	0.17
cv. %	20.2	16.8	10.6	24.9	16.4	19.7	–	26.8	33.2	40.2	37.9	40.6	30.5	38.4	56.1	42.7	22.7	34.5
Cool-season mean	5.26	5.61	0.99	1.23	9.06	5.20	3.98	1.57	1.65	1.67	0.98	1.01	4.96	3.86	2.58	1.66	2.65	0.44
Warm-season mean	3.56	5.45	0.99	1.38	4.63	4.76	–	0.61	0.32	0.79	0.67	0.78	0.42	1.42	2.27	2.71	0.95	0.28
Contrast Prob > *F*	<0.01	NS	NS	NS	<0.01	NS	–	<0.01	<0.01	<0.01	0.06	NS	<0.01	<0.01	NS	<0.01	<0.01	0.02

Table 5. Ash concentration of 10 native grasses grown for 2 to 4 yr at five sites in Western Canada

	Brandon-clay		Brandon-sand		Lethbridge		Melfort		Swift Current-dryland				Swift Current-irrigation				Vegreville	
	1994	1995	1994	1995	1994	1995	1994	1995	1994	1995	1996	1997	1994	1995	1996	1997	1994	1995
Cool-season	(g kg^{-1})																	
Northern wheatgrass	129	116	103	86	122	85	85	82	67	87	81	84	82	97	92	82	69	74
Green needlegrass	131	112	113	73	107	100	83	70	82	82	55	73	90	89	96	86	69	76
Mammoth wildrye	130	101	80	58	154	102	67	56	62	53	51	56	80	65	60	68	48	68
Western wheatgrass	145	138	126	102	125	94	95	90	77	91	76	84	108	102	107	90	70	86
Warm-season																		
Big bluestem	110	91	113	83	84	70	–	88	79	91	72	74	82	71	63	60	53	81
Sideoats grama	139	106	139	98	104	90	–	–	114	99	–	66	105	145	110	79	80	90
Little bluestem	81	67	72	67	92	55	–	–	78	65	62	65	97	85	54	59	62	76
Prairie sandreed	168	80	104	57	92	60	80	89	62	57	44	45	61	75	56	50	60	66
Indiangrass	119	113	99	–	124	67	–	–	84	96	–	–	88	105	–	58	85	104
Switchgrass	123	83	114	72	109	79	–	–	81	92	62	76	84	90	66	66	81	88
Grass Prob > *F*	0.01	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
$LSD_{0.05}$	34	12	26	7	36	19	10	10	12	12	9	13	17	29	14	11	12	15
cv %	18.7	8.0	11.4	4.2	21.9	16.1	8.1	8.1	10.3	10.7	10.2	12.7	11.8	21.6	12.3	10.8	12.8	13.1
Cool-season mean	134	117	105	80	127	95	82	74	72	78	66	74	90	88	89	82	64	76
Warm-season mean	123	90	107	75	101	70	–	88	83	83	60	65	86	95	70	62	70	84
Contrast Prob > *F*	NS	<0.01	NS	<0.01	<0.01	<0.01	–	<0.01	0.01	NS	<0.01	<0.01	0.01	NS	<0.01	<0.01	NS	NS

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