

Sweet pearl millet and sweet sorghum have high nitrogen uptake efficiency under cool and wet climate

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Abstract Nitrogen use efficiency is a key factor for the economic and environmental sustainability of farms. It comprises the ability for crops to recover applied fertilizer N in their aboveground biomass, called *fertilizer N uptake efficiency* (NupE). Sweet pearl millet [*Pennisetum glaucum* (L.) R.BR.] and sweet sorghum [*Sorghum bicolor* (L.) Moench] are C4 annual crops known for their capacity to produce high yield under N-limiting conditions, suggesting high NupE. A field study was conducted for 2 years on sandy loam soils in eastern Canada. The NupE was determined using a ¹⁵N-tracing approach. Comparisons were made for both species in regards to their response to (1) increasing mineral N rate (0–160 kg N ha⁻¹), (2) mineral versus organic N (liquid swine and dairy cattle manures), and (3) single versus split N application. For mineral N treatments, NupE ranged from 54 to 82 %, which is greater than

values generally reported for conventional crops such as corn. Moreover, NupE increased with N rates. These findings suggest that both species expressed N luxury consumption. The NupE was lower with organic than with mineral N, and a larger proportion of N remained in the soil, suggesting that N immobilization occurred. The presumed losses (unrecovered ¹⁵N) were <24 kg N ha⁻¹ in all mineral and most organic N treatments. Splitting N application had little effect on NupE. Our results confirm that sweet pearl millet and sweet sorghum are highly efficient at recovering fertilizer N and, with adequate residue management, may represent a low environmental risk.

Keywords Nitrogen-15 · Nitrogen uptake efficiency · Sweet pearl millet · Sweet sorghum · Animal slurry

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Introduction

Sweet pearl millet [*Pennisetum glaucum* (L.) R.BR.] and sweet sorghum [*Sorghum bicolor* (L.) Moench] are annual C4 grasses that have been studied in eastern Canada for the last decade because of their potential to replace or complement grain corn (*Zea Mays* L.) for ethanol production. These species can be exploited similarly to sugarcane, by extracting the sugar-rich juice from their stalks and fermenting it into ethanol (Yu et al. 2012).

In previous experiments under the wet and cool conditions of eastern Canada, sweet pearl millet and sweet sorghum produced aboveground biomass up to 19 and 20 Mg DM ha⁻¹, respectively (Dos Passos Bernardes et al. 2015; Thivierge et al. 2015). The mineral N rate (ammonium nitrate) that maximized ethanol yield for sweet pearl millet and sweet sorghum in eastern Canada varied from 78 to 91 kg N ha⁻¹ (Leblanc et al. 2012; Thivierge et al. 2015). In comparison, grain corn, the current feedstock used for ethanol production in eastern Canada, yielded an average of 6.9 Mg DM ha⁻¹ from 2008 to 2012 (FADQ 2015). Nitrogen rates (ammonium nitrate) required to maximize corn grain and ethanol yield in the same area varied from 128 to 285 kg N ha⁻¹ (Liang et al. 1996; Gagnon and Ziadi 2010). Therefore, the high DM yields obtained from sweet pearl millet and sweet sorghum with relatively low N rates suggest that these species may have high *fertilizer N uptake efficiency* (NupE; proportion of applied fertilizer N recovered in the aboveground biomass at harvest).

Using ¹⁵N-labelled fertilizers, several authors in eastern Canada and the United States demonstrated that for corn, NupE (1) varied from 28 to 60 % (Reddy and Reddy 1993; Liang and MacKenzie 1994; Tran et al. 1997; Stevens et al. 2005; Nyiraneza et al. 2010), (2) decreased with increasing mineral N rate (Liang and MacKenzie 1994; Tran et al. 1997), and (3) was lower with animal manure than with mineral N (Carter et al. 2010). We are not aware of any study on sweet pearl millet and sweet sorghum using this technique.

The aims of the present study were (1) to determine NupE in sweet pearl millet and sweet sorghum, and (2) to compare the response of these species to (i) increasing rates of mineral N, (ii) mineral versus organic N sources (liquid swine and dairy manures), and (iii) to single versus split application of mineral N. Work was carried out at two sites under wet and cool conditions in eastern Canada.

Materials and methods

Sites and species description

Crops were grown at Sainte-Anne-de-Bellevue (Québec, Canada, 45°26'N, 73°56'W), located in the Mixedwood Plains (MWP) ecozone, in 2011 and

2012, and at Saint-Augustin-de-Desmaures (Québec, Canada, 46°44'N, 71°31'W), located in the Boreal Shield (BS) ecozone, in 2010 and 2011. A complete description of ecozones of Canada is found in the National ecological framework for Canada (ESWG 1995). The two ecozones were chosen because they provide the possibility to grow sweet pearl millet and sweet sorghum as bioenergy crops using their sweet juice, or as forage crops, under two contrasting temperature regimes. Mean daily temperatures from seeding to harvest were 21.5 °C in 2011 and 21.9 °C in 2012 at the MWP site, and 19.3 °C in 2010 and 18.6 °C in 2011 at the BS site. During the same period, cumulated rainfall were 359 mm in 2011 and 188 mm in 2012 at MWP, and 282 mm in 2010 and 390 mm in 2011 at BS. The corn heat units (CHU) cumulated from seeding to harvest, a climatic index calculated as per Brown and Bootsma (1993), were 2207 CHU in 2011 and 2250 CHU in 2012 at MWP, and 2095 CHU in 2010 and 1940 CHU in 2011 at BS. Soil types were a St. Bernard sandy loam (coarse-loamy, mixed, nonacid, calcareous, frigid Eutrochrept) at the MWP site, and a St. Antoine sandy loam (fine-loamy, mixed, acid, frigid Haplorthod) at the BS site. Additional characteristics of the experimental sites are presented in Thivierge et al. (2015). At both sites, sweet pearl millet hybrid CSSPM7 and sweet sorghum hybrid CSSH45 (AERC Inc., Delhi, Ontario, Canada) were compared.

Experimental set-up and crop management

A split-plot factorial design with four replicates was used at each site, with species as the main factor (main plots) and N fertilization treatments as the sub-factor (subplots). The complete description of the experimental design can be found in Thivierge et al. (2015).

Briefly, each subplot was 6.3 m² in size at the MWP site, and 9.7 m² at the BS site. Row spacing was 0.18 m in all cases. Seedbed preparation consisted of mouldboard ploughing in the fall and harrowing twice in the spring to prepare seedbed. The seeding rate was 10 kg ha⁻¹ of pure live seeds. Seeding was performed at a depth of 2.5 cm using a Wintersteiger plot seeder (Wintersteiger, Salt Lake City, UT). The growing seasons were from June 10 (seeding) to Sept. 9 (harvesting) at the BS site in 2010 (91 days), from June 6 to Aug. 31 at the BS site in 2011 (86 days),

from June 16 to Sept. 7 at the MWP site in 2011 (84 days), and from June 7 to Aug. 30 at the MWP site in 2012 (85 days).

Bentazone (3-isopropyl-1*H*-2.1.3-benzothiadiazin-4(3*H*)-one 2.2-dioxide) was applied at a rate of 1.08 kg active ingredient ha⁻¹ between the three- and six-leaf stage for both species to suppress dicotyledonous weeds. At the BS site, glyphosate (N-(phosphonomethyl-glycine) was applied in spring 2010 (0.89 kg active ingredient ha⁻¹) to control weeds. Hand weeding of the sweet sorghum plots was done at the 10-leaf stage at both sites, while it was not necessary in sweet pearl millet plots.

Fertilization treatments

The fertilization treatments included calcium ammonium nitrate (27-0-0) at rates of 40, 80, 120 and 160 kg N ha⁻¹, with 40 kg N ha⁻¹ broadcast at seeding and the remaining of N sidedressed at the four-leaf stage for treatments with 80, 120 and 160 kg N ha⁻¹. A 0 N control treatment was included. The mineral N treatments will be hereafter referred to as 0, 40, 80-S, 120-S, and 160-S, where S stands for split application. Two organic N sources, liquid swine manure (LSM) and liquid dairy manure (LDM), were broadcasted at seeding, at a target rate of 80 kg ha⁻¹ total N, based on preliminary analysis (treatments hereafter referred to as LSM80 and LDM80). The actual amounts of N applied with LSM were 87.6 kg N ha⁻¹ in 2011 and 75.0 kg N ha⁻¹ in 2012 at the MWP site, and 86.4 kg N ha⁻¹ in 2010 and 86.7 kg N ha⁻¹ in 2011 at BS. The actual amounts of N applied with LDM were 79.7 kg N ha⁻¹ in 2011 and 80.8 kg N ha⁻¹ in 2012 at MWP, and 89.8 kg N ha⁻¹ in 2010 and 85.5 kg N ha⁻¹ in 2011 at BS. The main characteristics of the slurries can be found in Thivierge et al. (2015). The last N fertilization treatment was calcium ammonium nitrate broadcast at 80 kg N ha⁻¹ at seeding (single-application treatment, referred to as 80) for comparisons with the organic N sources (80 vs. LSM80 and LDM80) and with the split application (80 vs. 80-S). Phosphorus (triple superphosphate; 40 and 20 kg P₂O₅ ha⁻¹ at MWP and BS sites, respectively) and potassium chloride (60 and 40 kg K₂O ha⁻¹ at MWP and BS sites, respectively) were applied annually to the plots receiving mineral N, based on soil analyses and local recommendations (CRAAQ 2010). Phosphorus and K were not added to the plots

fertilized with organic N sources since the manures provided these nutrients.

In the center of each subplot, a 1-m² microplot was delineated and was fertilized with the same N source and rate than the rest of the subplot, but using a ¹⁵N-enriched fertilizer. The subplots fertilized with 160 kg ha⁻¹ of mineral N did not receive ¹⁵N-enriched fertilizer because of scientific evidence that such a high N rate is not suitable for sweet pearl millet and sweet sorghum (Geng et al. 1989; Barbanti et al. 2006; Leblanc et al. 2012). The microplots receiving mineral N treatments (40, 80, 80-S, and 120-S) were fertilized with double-labelled ammonium nitrate (NH₄NO₃ at 5 at.% ¹⁵N). A concentrated ¹⁵N solution was prepared in the laboratory. For each microplot, the appropriate amount of that solution was completed to 2 L with distilled water, which was applied evenly to the microplot using a watering can. To ensure a rapid infiltration of the solution into the soil, an additional 2 L of distilled water was immediately applied to the microplot. The actual atomic percentage excesses (APE) of the concentrated ¹⁵N solutions were 4.85 and 5.19 % at seeding and 4-leaf stage, respectively, at the BS site in 2010, 5.18 and 5.23 % at BS in 2011, 5.10 and 5.26 % at MWP in 2011, and 5.20 and 5.17 % at MWP in 2012.

Microplots receiving organic N treatments were fertilized with slurries to which ammonium sulfate ((¹⁵NH₄)₂SO₄ at 99 at.% ¹⁵N) was added to raise the APE of the NH₄-N fraction of the slurries to approximately 5 % without unbalancing its NH₄-N/total N ratio. This allowed tracing the fate of slurry-derived NH₄-N. The detailed analyses of the ¹⁵N-labelled slurries, including APE, are given in Table 1. The actual excess ¹⁵N added to the system with each fertilization treatment is detailed in Table 2.

All organic amendments applied at seeding were manually broadcast on the soil surface and incorporated into the top 5 cm of soil with hand tools within two hours of application to minimize ammonia volatilization (Rochette et al. 2001).

Samplings and analyses

The LSM and LDM were analyzed for N forms and concentrations on a composite subsample that was collected during manure application. Total N concentration of manures was determined by acid digestion whereas the concentrations of NH₄-N and NO₃-N were

Table 1 Selected characteristics of the ^{15}N -labelled liquid manures used in the experiment

Site	Manure type	Year	$\text{NH}_4\text{-N}$ (mg L^{-1})	$\text{NO}_3\text{-N}$ (mg L^{-1})	Total N (g L^{-1})	APE ^a (% of $\text{NH}_4\text{-N}$)	Application rates		
							Volume (L m^{-2})	Total N (kg ha^{-1})	$\text{NH}_4\text{-N}$ (kg ha^{-1})
Mixedwood plains	LSM ^b	2011	6013	0.71	8.92	3.03	1.0	89.2	60.1
		2012	3731	2.82	7.05	4.07	1.0	70.5	37.3
	LDM ^b	2011	1611	0.00	3.56	2.88	2.5	88.9	40.3
		2012	1066	1.38	3.27	3.97	2.5	81.6	26.7
Boreal shield	LSM ^b	2010	6546	2.31	9.11	3.71	1.0	91.1	65.5
		2011	5931	0.82	8.99	3.14	1.0	89.9	59.3
	LDM ^b	2010	1599	1.29	2.57	2.71	3.5	89.8	56.0
		2011	928	0.70	3.01	2.47	2.5	75.2	23.2

^a APE atomic percentage excess (at.% ^{15}N)

^b LSM liquid swine manure, LDM liquid dairy manure

Table 2 Excess ^{15}N added to the system, at two sites and for two growing seasons

Site	Mixedwood Plains (mg N m^{-2})		Boreal Shield (mg N m^{-2})	
	2011	2012	2010	2011
Treatment				
40	200	207	196	205
80-S ^a	408	414	403	413
120-S	625	619	612	626
80	417	418	384	419
LSM80 ^b	182	152	243	186
LDM80 ^b	116	106	152	57

^a S, N rate was split-applied with 40 kg N ha^{-1} at seeding and the rest sidedressed at the 4-leaf stage

^b LSM80 liquid swine manure applied at a target rate of 80 $\text{kg total N ha}^{-1}$, LDM80 liquid dairy manure applied at a target rate 80 $\text{kg total N ha}^{-1}$

determined by KCl extraction, as described in Chantigny et al. (2007). The $\text{NH}_4\text{-N}$ concentration in the acid digestates and $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ concentrations in KCl extracts were measured with an automated continuous-flow injection analyzer (Model QuickChem 8000 FIA+, Lachat Instruments, Loveland, CO).

Both crop species were harvested at the maximum growth stage that it was possible to reach at each site, but before the risk of lodging was too high, in order to use their sweet juice for ethanol production. At MWP, the site with the warmest temperature, the harvesting stage corresponded to 3/4 of the inflorescence emerged on at least 75 % of the stalks. At BS, the site with the coldest temperature, the harvesting stage corresponded to 3/4 of the inflorescence emerged on at

least 50 % of the stalks. Determination of DM yield is detailed in Thivierge et al. (2015). The central section (36 cm \times 36 cm) of each microplot was harvested to avoid edge effects, and stalks were cut 5 cm above the soil surface. The harvested biomass was cut into 10-cm pieces and mixed. A 250-g subsample was taken and dried at 55 °C until constant weight to determine DM concentration, and ground to 1 mm (Whiley mill, Standard model 3, Arthur H. Thomas Co., Philadelphia, PA) to determine total N concentration by dry combustion (LECO CNS-1000, Leco Corp., St. Joseph, MI).

Three 30-cm soil cores were collected with a stainless steel auger (2 cm diam.) in the center of each microplot. Each core was divided in 10-cm increments and mixed to make one composite sample per depth

for each microplot. Soil samples were air dried and ground to 0.15 mm (ball mixer-mill MM400, Retsch, Germany) to determine total N concentration.

Stable isotope ratio ($^{15}\text{N}/^{14}\text{N}$) was determined in aboveground crop biomass, soil, and manure samples with a continuous flow isotope ratio mass spectrometer (Model Optima, VG IsoTech, Middlewich, Cheshire, UK) after sample combustion to N_2 at 1020 °C with a combustion analyzer (Model NA1500, Carlo Erba Strumentazione, Rodano, Milan, Italy). Soils from the 0 N control plots were analyzed to determine natural ^{15}N abundance at each site.

Calculations and statistical analyses

The crop N uptake (kg ha^{-1}) was calculated as the product of crop DM yield and its N concentration. The ^{15}N fertilizer N uptake efficiency (NupE) in the aboveground biomass (Eq. 1) and ^{15}N recovery in the soil (0–30 cm) (Eq. 2) were calculated as per Muñoz et al. (2004) and Nyiraneza et al. (2010), respectively:

$$\begin{aligned} \text{NupE in aboveground biomass (\%)} \\ = 100 \times \text{N uptake (kg ha}^{-1}\text{)} \\ \times (a - c) / \text{N applied as fertilizer (kg ha}^{-1}\text{)} \\ \times (b - c); \end{aligned} \quad (1)$$

$$\begin{aligned} ^{15}\text{N recovery in soil (\%)} = 100 \times \text{Soil N content} \\ \times (s - c) / \text{N applied as fertilizer (kg ha}^{-1}\text{)} \\ \times (b - c). \end{aligned} \quad (2)$$

where a is the ^{15}N abundance in aboveground biomass harvested in the microplot, b is the ^{15}N abundance in the fertilizer, c is the ^{15}N natural abundance measured in the soil of the control plot, and s is the ^{15}N abundance in soil harvested in the microplot. The amounts of ^{15}N recovered in plant aboveground biomass and soil were summed to calculate N budgets. Similar calculations were made for manure treatments (LSM80 and LDM80), and results were expressed on the basis of $\text{NH}_4\text{-N}$ applied with the manures, as the organic fraction of manure N was not labelled with ^{15}N . The ^{15}N unaccounted for in the soil–plant system was considered to represent environment loss, assuming that ^{15}N remaining in soil below 30 cm was negligible.

The proportion of crop N uptake that derived from the fertilizer was calculated as per Fillery and Recous (2001; Eq. 3):

$$\begin{aligned} \text{Fertilizer-derived N (kg ha}^{-1}\text{)} \\ = \text{N uptake (kg ha}^{-1}\text{)} \times (a - c) / (b - c). \end{aligned} \quad (3)$$

The contribution of soil to crop N uptake was taken as the difference between total N uptake and the fertilizer-derived N uptake (Fillery and Recous 2001).

The analysis of variance was carried out separately for each site, using the MIXED procedure in SAS (SAS Institute 2003) for the dependent variables: N concentration, N uptake, fertilizer-derived N uptake, soil-derived N uptake, and NupE. Treatment replication was considered random effect, whereas years, species, and N treatments were considered fixed effects. Data normality was verified using the UNIVARIATE procedure, using the Shapiro–Wilk test (Shapiro and Wilk 1965) to determine whether the residuals were normally distributed. The homogeneity of variance was verified visually with graphics of residuals. Standard error of means (SEM) is reported in Tables.

Quantitative contrasts were used to test the effect of increasing mineral N rate on plant N concentration, N uptake, fertilizer-derived N uptake, soil-derived N uptake, and NupE. Contrast analysis was also used to test the effect of N source (80 vs. LSM80 and LDM80), to compare the two organic N sources (LSM80 vs. LDM80), and to compare the effect of split versus single application of mineral N (80-S vs. 80). Statistical significance was postulated at $P \leq 0.05$.

Results and discussion

Differences between species and response to increasing rates of mineral N

The average N concentration in aboveground biomass was similar for both species at the MWP site, whereas sweet pearl millet had a greater N concentration than sweet sorghum at the BS site (Table 3). The range of N concentrations reported here (5.3–12.6 g N kg^{-1} at MWP and 8.5–19.4 g N kg^{-1} at BS) echoed those reported for sweet sorghum by Han et al. (2011) in Northern China (7.5–10.2 g N kg^{-1} DM at 40 days

Table 3 Nitrogen concentration (g N kg⁻¹ DM) for sweet pearl millet and sweet sorghum grown on sandy loam soils at the Mixedwood Plains and Boreal Shield sites in response to eight fertilization treatments

	Mixedwood Plains		Boreal Shield	
Species	2011-2012		2010-2011	
Pearl millet	7.6		13.1	
Sorghum	7.6		11.4	
SEM ^a	0.18		0.48	
N treatment	2011	2012	2010	2011
0	5.3	5.9	9.5	8.8
40	5.9	6.4	8.5	10.2
80-S ^b	8.0	7.7	12.4	12.8
120-S	10.0	9.8	15.9	16.2
160-S	12.6	11.3	15.4	19.4
80	7.3	7.4	13.3	12.5
LSM80 ^c	5.7	6.9	10.8	10.8
LDM80 ^c	5.4	5.6	9.8	9.6
SEM	0.50	0.51	1.19	0.84
Fixed effects and interactions (<i>P</i> values)	2011-2012		2010-2011	
Year	0.611		0.270	
Species	0.981		0.014	
Year × species	0.091		0.106	
N treatment	< 0.001 ^d		< 0.001	
Year × N treatment	0.028		0.023	
Species × N treatment	0.501		0.076	
Year × species × N treatment	0.316		0.428	
Contrasts (<i>P</i> values)				
Mineral N ^e Linear	< 0.001		< 0.001	
Mineral N Quadratic	0.001		0.189	
Mineral N Cubic	0.206		0.002	
80 vs. Organic N ^f	< 0.001		< 0.001	
LDM80 vs. LSM80	0.026		0.127	
80 vs. 80-S	0.132		0.718	

^a SEM standard error of the means

^b S, N rate is split-applied with 40 kg N ha⁻¹ at seeding and the rest sidedressed at the 4-leaf stage

^c LSM80 liquid swine manure applied at a target rate of 80 kg total N ha⁻¹, LDM80 liquid dairy manure applied at a target rate 80 kg total N ha⁻¹

^d Bold font highlights a significant effect, at the 0.05 level

^e Mineral N: treatments 0, 40, 80-S, 120-S and 160-S

^f Organic N: LSM80 and LDM80

after anthesis) and by Ceotto et al. (2014) in Italy (4.7–15.9 g N kg⁻¹ DM). Crop N concentration responded similarly to N fertilization rate in both species, as indicated by the absence of interaction between species and N treatments, or species and years (Table 3). However, there was an interaction between N treatments and years at both sites. As a general trend, there was a linear increase in plant N concentration with increasing mineral N rate. Increase in plant N concentration with N rate was previously observed in sweet pearl millet (Leblanc et al. 2012) and in fiber sorghum (Barbanti et al. 2006), but it is observed for the first time in sweet sorghum. At the MWP site, this increase in N concentration was more acute in 2011 (5.3–12.6 g N kg⁻¹ DM) than in 2012 (5.9–11.3 g N kg⁻¹ DM). Also, the lower N concentration of the LSM80 treatment in 2011 than in 2012 (5.7 and 6.9 g N kg⁻¹ DM, respectively) might have contributed to the interaction between N treatments and years. At the BS site, N concentration levelled off at 120 kg N ha⁻¹ in 2010 and reached a maximum with 160 kg N ha⁻¹ in 2011.

In regards to crop N uptake, there was an interaction between species and years at the MWP site (Table 4): the difference in crop N uptake between years was larger for sweet pearl millet (110 vs. 159 kg N ha⁻¹) than for sweet sorghum (113 vs. 133 kg N ha⁻¹). At the BS site, there were significant differences between species (153 kg N ha⁻¹ for sweet pearl millet vs. 130 kg N ha⁻¹ for sweet sorghum) and between years (167 kg N ha⁻¹ in 2010 vs. 117 kg N ha⁻¹ in 2011), but without interaction. There were no interaction between N treatments and years, or N treatments and species, at any site. However, at both sites, N uptake increased linearly with mineral N rates. An increase in N uptake with N rate was previously observed in sweet sorghum (Cosentino et al. 2012; Ceotto et al. 2014) and in corn (Tran et al. 1997), but was not observed before for sweet pearl millet. Crop N uptake ranged from 74 to 217 kg N ha⁻¹ at MWP and from 76 to 202 kg N ha⁻¹ at BS (Table 4). These values are in the range (100–270 kg N ha⁻¹) reported for different varieties of sweet sorghum (Barbanti et al. 2006; Propheter and Staggenborg 2010; Han et al. 2011). Nitrogen uptake of 216 and 290 kg N ha⁻¹ were also reported for grain sorghum (Lemaire et al. 1996; Ra et al. 2012). Propheter and Staggenborg (2010) compared many annual and perennial grasses used for energy production and found greater nutrient

removal rates with annual grasses, particularly sweet sorghum, than with perennial grasses. This difference could be attributed to the capacity of perennial grasses to store minerals into the crown and rhizomes for overwintering.

For all experimental treatments, the crop N uptake was superior to the quantity of N applied, resulting in a negative N balance. The capacity for sweet pearl millet and sweet sorghum to efficiently capture soil N could be attributable to morphological characteristics of their root system. For instance, Wang et al. (2005) demonstrated in corn that root morphology influences the crop's capacity to compete for soil N. Moreover, mycorrhizal associations have been observed in grain pearl millet and grain sorghum roots (Bagayoko et al. 2000), and grain sorghum was found to have a greater ability than corn to directly absorb organic N (Okamoto and Okada 2004). In the last decade, some authors discovered that grain sorghum and grain pearl millet roots have the capacity to exudate secondary metabolites that act as nitrification inhibitors in soil. This phenomenon, known as *biological nitrification inhibition*, keeps fertilizer N in the form of NH₄-N in soils for a longer period, which improves crop N uptake and N₂O, while reducing N losses through leaching and denitrification (Zakir et al. 2008; Subbarao et al. 2013).

In the present study, the fact that N uptake continued to increase linearly up to the highest N rate (160 kg ha⁻¹) at both sites (Table 4) was unexpected, as Thivierge et al. (2015) reported that, for the same sites, the maximum DM yield was reached with 107–121 kg N ha⁻¹. This finding suggests that these species expressed a phenomenon called N luxury consumption. This phenomenon occurs when N application to meet physiological needs (i.e. to achieve maximum growth) is exceeded: N accumulation in the plant therefore increases without affecting yield (Isfan et al. 1995). Nitrogen luxury consumption was described as a competitive strategy, as it deprives neighboring plants of a limiting nutrient (De Mazancourt and Schwartz 2012). It was previously observed in sweet sorghum by Cosentino et al. (2012).

The proportion of crop N uptake derived from the fertilizers (fertilizer-derived N, Table 5) amounted to 36 % on average at the MWP site, and 33 % at the BS site. When considering only the mineral N treatments, an average of 39 and 36 % of crop N uptake was derived from the fertilizer at MWP and BS,

Table 4 Nitrogen uptake (kg N ha^{-1}) for sweet pearl millet and sweet sorghum grown on sandy loam soils at the Mixedwood Plains and Boreal Shield sites in response to eight fertilization treatments

Species	Mixedwood Plains		Boreal Shield	
	2011	2012	2010	2011
Pearl millet	110	159	174	132
Sorghum	113	133	159	101
SEM ^a	3.3	8.6	19.5	5.4
N treatment	2011-2012		2010-2011	
0	74		76	
40	102		103	
80-S ^b	144		148	
120-S	190		199	
160-S	217		202	
80	131		177	
LSM80 ^c	95		126	
LDM80 ^c	77		102	
SEM	9.0		16.7	
Fixed effects and interactions (<i>P</i> values)	2011-2012		2010-2011	
Year	0.002^d		0.001	
Species	0.044		0.033	
Year × species	0.015		0.378	
N treatment	<0.001		<0.001	
Year × N treatment	0.273		0.370	
Species × N treatment	0.147		0.108	
Year × species × N treatment	0.233		0.416	
Contrasts (<i>P</i> values)				
Mineral N ^e Linear	<0.001		<0.001	
Mineral N Quadratic	0.889		0.321	
Mineral N Cubic	0.084		0.081	
80 vs. Organic N ^f	<0.001		<0.001	
LDM80 vs. LSM80	0.045		0.140	
80 vs. 80-S	0.146		0.080	

^a SEM standard error of the means

^b S, N rate is split-applied with 40 kg N ha^{-1} at seeding and the rest sidedressed at the 4-leaf stage

^c LSM80 liquid swine manure applied at a target rate of 80 $\text{kg total N ha}^{-1}$, LDM80 liquid dairy manure applied at a target rate 80 $\text{kg total N ha}^{-1}$

^d Bold font highlights a significant effect, at the 0.05 level

^e Mineral N: treatments 0, 40, 80-S, 120-S and 160-S

^f Organic N: LSM80 and LDM80

Table 5 Fertilizer- and soil-derived N, and fertilizer N uptake efficiency (NupE) in sweet pearl millet and sweet sorghum grown on sandy loam soils at the Mixedwood Plains and Boreal Shield sites in response to six fertilization treatments

	Mixedwood Plains			Boreal Shield		
	Fertilizer- or manure-derived ^a N (kg N ha ⁻¹)	Soil-derived N (kg N ha ⁻¹)	NupE ^b (% of applied N)	Fertilizer- or manure-derived ^a N (kg N ha ⁻¹)	Soil-derived N (kg N ha ⁻¹)	NupE ^b (% of applied N)
Species						
Pearl millet	46.0 (36) ^c	80.2	61.4	49.9 (34)	98.4	61.1
Sorghum	42.2 (35)	79.1	55.4	43.9 (32)	92.7	58.2
SEM ^d	2.31	4.79	2.86	4.30	7.84	4.53
Year ^e						
1	44.1 (42)	61.9	56.9	46.4 (27)	122.4	54.1
2	44.1 (31)	97.4	60.0	47.5 (41)	68.8	65.1
SEM	2.31	4.79	2.86	4.30	8.23	4.53
N treatment						
40	22.8 (22)	79.3	57.0	23.2 (22)	80.5	54.4
80-S ^f	60.8 (41)	86.8	75.9	57.1 (38)	91.3	69.0
120-S	98.4 (52)	91.9	82.0	91.8 (46)	106.8	72.7
80	53.8 (41)	77.1	67.3	64.6 (37)	112.2	75.4
LSM80 ^g	18.6 (20)	76.2	37.8	26.4 (21)	99.4	39.5
LDM80 ^g	10.2 (13)	66.6	30.7	18.5 (18)	83.2	46.7
SEM	3.72	6.30	5.10	8.37	12.90	8.36
Fixed effects and interactions						
Year	0.973	<0.001	0.321	0.802	0.001	0.052
Species	0.148	0.822	0.081	0.215	0.494	0.546
Year × species	0.363	0.064	0.196	0.564	0.093	0.277
N treatment	<0.001 ^h	0.003	<0.001	<0.001	0.087	<0.001
Year × N treatment	0.147	0.773	0.627	0.247	0.258	0.520
Species × N treatment	0.711	0.231	0.838	0.874	0.836	0.969
Year × species × N treatment	0.953	0.167	0.709	0.703	0.950	0.608
Contrasts						
Mineral N ⁱ linear	<0.001	0.045	<0.001	<0.001	0.046	0.006
Mineral N quadratic	0.961	0.827	0.153	0.942	0.826	0.280
80 versus organic N ^j	<0.001	0.275	<0.001	<0.001	0.057	<0.001
LDM80 versus LSM80	0.025	0.122	0.156	0.067	0.199	0.330
80 versus 80-S	0.059	0.118	0.089	0.204	0.097	0.347

^a In the case of LDM (liquid dairy manure) and LSM (liquid swine manure), the *manure*-derived N is presented

^b NupE, nitrogen fertilizer uptake efficiency, was calculated as the amount of ¹⁵N recovered in plant aboveground biomass based on total ¹⁵N applied with mineral fertilizers and NH₄-¹⁵N applied with animal manures

^c Numbers in parentheses are % of total N uptake

^d SEM standard error of the means

^e Years 1 and 2 represent 2011 and 2012, respectively, at the MWP site, and 2010 and 2011, respectively, at the BS site

^f S, N rate is split-applied with 40 kg N ha⁻¹ at seeding and the rest sidedressed at the 4-leaf stage

^g LSM80, liquid swine manure applied at a target rate of 80 kg total N ha⁻¹, LDM80 liquid dairy manure applied at a target rate 80 kg total N ha⁻¹

^h Bold font highlights a significant effect, at the 0.05 level

ⁱ Mineral N: treatments 40, 80-S, and 120-S

^j Organic N: LSM80 and LDM80

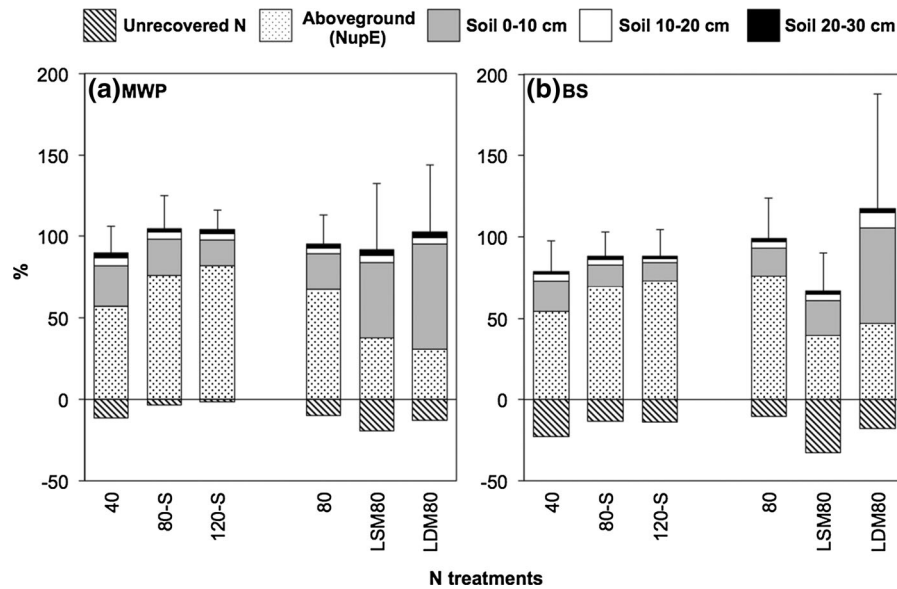


Fig. 1 Partitioning of the applied ¹⁵N in the soil–plant system for sweet pearl millet and sweet sorghum at the Mixedwood Plains (a) and Boreal Shield (b) sites, in response to six fertilization treatments. Vertical bars represent standard deviation of the proportion of the ¹⁵N recovered in the soil–plant system. S, N rate is split-applied with 40 kg N ha⁻¹ at seeding

and the rest side-dressed at the 4-leaf stage. NupE, fertilizer N uptake efficiency; LSM80, liquid swine manure applied at a target rate of 80 kg total N ha⁻¹; LDM80, liquid dairy manure applied at a target rate 80 kg total N ha⁻¹. For LSM80 and LDM80, results are expressed based on applied NH₄-N, as manure organic N was not labelled with ¹⁵N

respectively. Under similar climatic conditions, Tran et al. (1997) measured that on average 46 % of N uptake in grain and silage corn was derived from mineral fertilizers at harvest, while Nyiraneza et al. (2010) reported values between 24 and 66 % for silage corn fertilized with 160 kg mineral N ha⁻¹. These previous studies and the present one indicate that the main source of N to the crops generally comes from the soil reserve, in agreement with several previous studies with various crops, reviewed by Stevens et al. (2005). There were not any differences in fertilizer-derived N between species or between years. At both sites, there was a significant effect of N treatments, with a linear increase in fertilizer-derived N along with mineral N rate. Indeed, the amount of fertilizer N recovered by the crops increased substantially (+76 kg N ha⁻¹ at MWP and +69 kg N ha⁻¹ at BS) with mineral N rate from 40 to 120 kg ha⁻¹ (Table 5), and the contribution of fertilizer N to crop N uptake increased from 22 to 52 % at the MWP site and from 22 to 46 % at BS, confirming the positive response of crops to fertilizer N rate.

Interestingly, the amount of soil-derived N also increased linearly with applied mineral N rate, but to a

much smaller extent (+13 kg N ha⁻¹ at MWP and +26 kg N ha⁻¹ at BS). The increase in amounts of soil-derived N with increasing N rates was also observed in corn by Reddy and Reddy (1993), who suspected either an increased mineralization of soil organic matter, or a pool substitution between ¹⁴N from the soil and ¹⁵N from the fertilizer. The amount of soil-derived N was affected by years at both sites: 62 and 97 kg N ha⁻¹ in 2011 and 2012, respectively, at MWP; 122 and 69 kg N ha⁻¹ in 2010 and 2011, respectively, at BS. This was expected since soil N supply depends on inter-annual variations in climatic factors affecting soil N mineralization.

The NupE (proportion of applied fertilizer N recovered in crop biomass), was similar for sweet pearl millet and sweet sorghum at both sites (Table 5). There was no significant difference between years at any site, although the difference was almost significant at the BS site ($P = 0.052$) with 54 % in 2010 and 65 % in 2011. At both sites, the fertilizer N rate had a linear effect on NupE in both species, which ranged from 54 to 82 % (Table 5; Fig. 1). These NupE are relatively high since many studies on corn reported NupE ranging from 28 to 60 % (Reddy and Reddy

1993; Liang and MacKenzie 1994; Tran et al. 1997; Stevens et al. 2005; Nyiraneza et al. 2010). Moreover, in corn, it has been demonstrated that NupE stays relatively constant (Stevens et al. 2005) or decreases as mineral N rate increases (Liang and MacKenzie 1994; Tran et al. 1997). In our case the linear increase in NupE with fertilizer N rate could relate to N luxury consumption. Indeed, N luxury consumption has been positively related to soil N availability (Tripler et al. 2002). In agreement, our results suggest that the phenomenon was amplified with higher N fertilization rates. The continued increase in crop N uptake to the highest N rate (160 kg ha⁻¹) and the strong linear relationship ($P < 0.001$) between fertilizer-derived N uptake and N rates at both sites (Tables 4, 5) corroborate this finding. A concern stemming from possible N luxury consumption is the management of N-rich residues. After extraction of juice from the stalks, the residues can be either returned to the field or fed to cattle. In either case, care must be taken to avoid environmental loss of N from the field or animal health problems.

As a result of the high NupE, the amounts of residual fertilizer found in soil (0–30 cm) at harvest were generally low with the mineral fertilizer (Fig. 1). For all treatments, 72–90 % of the residual N was recovered in the top 10 cm of soil. This is in agreement with previous studies demonstrating that applied N does not significantly migrate at depth during the growing season wherever it is applied as LSM (Morvan et al. 1997; Chantigny et al. 2014), LDM (Muñoz et al. 2004), or mineral fertilizers (Reddy and Reddy 1993; Nyiraneza et al. 2010).

In the present study, the small proportion of fertilizer N recovered in the 20–30 cm soil layer (2.1–3.5 % at MWP, and 1.5–2.3 % at BS; Fig. 1) suggested that little applied N was present at greater depths, and that N not recovered in the top 30 cm of soil and in crop aboveground biomass was actually lost. In regards to the amount of unrecovered N, there was an interaction between N treatment and species at the MWP site ($P = 0.018$). However, the differences among N treatments and between species were very small, as unrecovered N ranged from 0 to 12 kg N ha⁻¹. At the BS site, there was an interaction between N treatments and years ($P = 0.029$). The main difference between years was for the treatment 120-S, with higher amount of unrecovered N in 2010 than in 2011 (24 vs. 10 kg N ha⁻¹, respectively).

Overall, the unrecovered N at BS was low and ranged from 6 to 24 kg N ha⁻¹ for the mineral N treatments. This further confirms the high efficiency of sweet pearl millet and sweet sorghum to capture applied N. These results are in accordance with previous studies on sweet sorghum, where the sum of nitrate leaching and ammonia volatilization with mineral fertilization rates of 60 and 120 kg N ha⁻¹ were 7.1 and 7.7 kg N ha⁻¹, respectively, in Cosentino et al. (2012), and 18.2 and 19.1 kg N ha⁻¹, respectively, in Barbanti et al. (2006). In corn (Reddy and Reddy 1993; Tran et al. 1997), unrecovered fertilizer N increased from 14 to 103 kg ha⁻¹ with N rates from 60 to 200 kg N ha⁻¹. Therefore, the direct environmental impact from N fertilization of sweet pearl millet and sweet sorghum may be lower than that of corn, especially at higher N rates. Lemaire et al. (1996) reached similar conclusions when comparing the risk of nitrate leaching under grain sorghum and corn.

Response to mineral and organic N sources

At both sites, plant N concentration and N uptake were lower with organic N sources than with the mineral N source for both species (Tables 3, 4). This was expected since the fertilizer value of manures reported by Thivierge et al. (2015) varied from 15 to 52 %, indicating that manure N was less available to the crops than N from the mineral fertilizer. As a result, NupE was smaller with manures (34 % of manure NH₄-N at MWP and 43 % at BS) than with the equivalent mineral N treatment (67 % of applied N at MWP and 75 % at BS; Table 5). The lower uptake of manure-derived N was expected because only 31 and 72 % of manure total N was readily available as ammonia-N (Table 1). This is in agreement with Carter et al. (2010) who observed that the apparent NupE in orchardgrass (*Dactylis glomerata* L.) and reed canarygrass (*Phalaris arundinacea* L.) was greater with mineral fertilizer N than with LDM.

Different factors could explain why crop N uptake was lower with manures than with mineral N. Ammonia loss may have occurred through volatilization following manure application. However, the short 2-h delay elapsed between application and incorporation likely limited those losses (Rochette et al. 2001). It is also possible that N immobilization occurred as highly decomposable carbon was applied with the manures (Kirchmann and Lundvall 1993). This

hypothesis is supported by the fact that most of the manures (LDM80 and LSM80 at MWP, and LDM80 at BS) resulted in a larger proportion of N recovered in the top soil layers than mineral N treatments (Fig. 1). Morvan et al. (1997) reported that 15–35 % of the $\text{NH}_4\text{-N}$ from LSM could be immobilized. Moreover, they argued that immobilization of manure N is favored when crops are not actively growing at the time of manure application, as in our experiment, because ammonia-N remains available to microorganisms for a longer period of time. The immobilized N could eventually be lost and have possible negative impact on the environment, if not taken up by subsequent crops (Chantigny et al. 2014; Sieling et al. 2014).

At the BS site, the proportion of applied N presumably lost to the environment (unrecovered N) was larger for manures than for the mineral fertilizer (18 % of $\text{NH}_4\text{-N}$ for LDM and 33 % for LSM versus 10 % of applied N for treatment 80, $P = 0.006$), and was larger for LSM than for LDM ($P = 0.012$, Fig. 1). There were no significant differences in unrecovered N between manures and mineral treatment at the MWP site (13 % of $\text{NH}_4\text{-N}$ for LDM, 19 % for LSM, and 10 % for mineral treatment 80, $P = 0.172$). The amount of unrecovered N was lower than 20 kg N ha^{-1} for all organic treatments for all sites and years, except for LSM80 in 2010 at the BS site where it reached 32 kg N ha^{-1} .

At the MWP site, LSM80 resulted in slightly greater plant N concentration, N uptake, and fertilizer-derived N than LDM80 (Tables 3, 4, 5). However, NupE were similar between manures at both sites. At the BS site, the proportion of N recovered in the 0–10 cm soil layer was greater for LDM than for LSM ($P < 0.001$). Immobilization of soil N following manure application could also explain these differences between manures, as LDM may induce greater N immobilization, and therefore be less available to crop, than LSM (Kirchmann and Lundvall 1993).

Response to single and split application of mineral N

Splitting N application between seeding and the 4-leaf stage, rather than applying all N at seeding, did not affect plant N concentration and N uptake at any site (Tables 3, 4). In comparison with corn, Abassi et al. (2013) found an increase in N concentration and N

uptake with split N application, whereas Dordas et al. (2008) did not observe any difference. The split N application had no effect on NupE and on the proportion of crop N uptake derived from the soil at any site, and had only a small effect ($P = 0.059$) on fertilizer-derived N at MWP, where the split application increased N uptake by 7 kg when compared to the single application (Table 5). Finally, the split N application slightly lowered the proportion of unrecovered N at MWP (10 % for single application versus 3 % for split, $P = 0.037$, Fig. 1), and the proportion of N recovered in the soil at BS (24 % for single application vs. 19 % for split, $P < 0.001$, Fig. 1). Along with the finding by Thivierge et al. (2015) that splitting N did not significantly increase crop yields in sweet pearl millet and sweet sorghum, the small differences that were observed in the present study do not justify recommending split N application for these crop species in eastern Canada, at least with N rate of 80 kg ha^{-1} .

Conclusion

This investigation confirmed the high NupE of sweet pearl millet and sweet sorghum under wet and cool conditions in eastern Canada. The NupE ranged from 54 to 82 % of applied mineral N, values much higher than those reported for other crops such as corn. Moreover, N uptake was large for both species at both sites and at all N rates, indicating that these crops have the ability to efficiently recover N from fertilizers and from the soil reserve. Luxury N consumption likely occurred in both species and, consequently, N-rich residues remaining after extraction of juice from the stalks will need to be managed with care to prevent further N losses. More research is required to identify the factors explaining the peculiar ability of sweet pearl millet and sweet sorghum to recover N from soil and fertilizer, such as morphological characteristics of their root system, possible mycorrhizal associations, and biological nitrification inhibition. Crop N uptake and NupE were lower with animal manures than mineral fertilizer, presumably because of soil N immobilization in the days following manure application. Because of the very high NupE, the presumed environmental losses (unrecovered N at harvest) were low for all treatments including manures. Overall, these findings together with the low N requirements

(e.g. Thivierge et al. 2015) indicate that sweet sorghum and sweet pearl millet could be successfully grown in eastern Canada with little risk to the environment provided that their N-rich residues are properly managed.

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